

Teagasc

GRANGE AND ATHENRY RESEARCH CENTRE

Research Report 2004

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BEEF AND SHEEP PRODUCTION RESEARCH

Grange Research Centre Dunsany, Co. Meath

and

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Research Programme for 2004

The 2004 research report contains information on the programme conducted at both Grange and Athenry Research Centres. This is the first year that research reports from both centres are jointly presented. This report outlines the main findings for the 2004 research programme.

The primary aim of the beef and sheep research programmes is to undertake research which underpins competitiveness and innovation in the agricultural sector. Highlights from the 2004 programme are summarised in the following section.

Highlights from the 2004 Beef and Sheep Research Programme

Live animal scoring and carcass grades

Studies involving the bull progeny of the suckler herd at Grange showed significant positive relationships between both muscular scores taken on the live animals prior to slaughter and carcass conformation scores with killing-out rate, carcass meat proportion and carcass value. Carcass value was calculated as the sum of the commercial values of each separate fat trimmed cut. The carcasses graded mainly U and R for conformation and 2, 3 and 4 L for fatness. Despite the close similarity in carcass grades, when averaged over four separate experiments, a 1 unit improvement in conformation (scale 1 to 5) increased carcass value by 9c per kg, while a 1 unit increase in fatness (scale 1 to 5) decreased value by 8c per kg.

Value of genetic index differences in beef bulls

A new programme of genetic improvement for Irish beef cattle is being developed by the Irish Cattle Breeding Federation (ICBF). Progeny of Limousin bulls of low and high genetic index for growth were compared for growth, feed intake and carcass traits. The progeny of the high index bulls grew faster than the progeny of low index bulls. About two thirds of the “extra” carcass weight from the high index progeny came from higher liveweight gain and one third came from a higher kill-out proportion. There was no difference between the progeny groups in feed intake or in carcass conformation but the high index progeny had more compact carcasses as indicated by carcass measurements scaled for carcass weight. High index progeny had a lower proportion of hindquarter but there was no difference in carcass composition between progeny groups. The effects of genetic index were similar for male and female progeny. The effects of genetic index for breeds other than the Limousin and across breeds were less consistent.

Weanling response to energy and protein supplementation

Grass silage is the most common basal feed for Irish cattle. It is often variable in quality and is supplemented as necessary to achieve the desired rate of gain. Barley and beet pulp are commonly used supplements. On an ‘as fed’ basis these do not differ greatly in net energy content but do differ in total crude protein and in the nutritionally important protein fractions. Thus, inclusion of a protein source such as soyabean meal would be indicated for a pulp-based supplement. At the same level of offered supplementary dry matter, silage intake was lower for pulp than barley but addition of a protein source with pulp brought silage intake up to the level of that for barley. Addition of protein to barley had no effect on silage intake. There was a growth response to protein with both barley and pulp but it was much greater with pulp. Because of compensatory growth at pasture by those with lower weight gains to winter, there was an inverse relationship between gain in winter and gain subsequently at pasture. Thus, the liveweight differences between the groups were less at the end of the following grazing season than at the end of winter immediately following the treatments. However, the overall extent of liveweight recovery was low (ca. 30%) and the degree of recovery did not vary with winter feeding

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treatment. Depending on feed costs and the value of liveweight gain the results permit calculation of the optimum level of concentrate supplementation with silage in winter and relative value of barley and pulp as concentrate ingredients.

Conserving high-moisture wheat grain

Cereal grains are important sources of nutrients for ruminants in many livestock production systems, and may need to be stored for up to twelve months before being fed. During this time both quantitative and qualitative losses must be minimised, with fungal growth in particular needing to be prevented. Grain with a dry matter (DM) content ≥ 860 g/kg can be stored for extended durations. However, the duration of safe storage progressively decreases and/or the requirement for additional protective treatments increases as the DM concentration at harvesting decreases.

An experiment quantified the conservation characteristics of high moisture wheat grains, stored anaerobically, following contrasting processing and additive treatments. High moisture wheat stored anaerobically preserved successfully, with rolled grain fermenting more extensively than whole grain but at the cost of a decrease in starch content and organic matter digestibility. Acid additives, urea or bacterial inoculants containing *Lactobacillus buchneri* each altered the characteristics of the conserved grain. A final conclusion on the optimal choice of additive will be made when in-silo losses and aerobic stability data are added to the above.

Bull beef production from grazed pastures

Bull production systems which finish cattle off pasture are potentially more competitive than those with a long indoor finishing period. This study determined the performance response of bulls to concentrate supplementation at pasture, and compared performance at pasture with that of bulls finished indoors. Bulls offered no concentrate (No Meal) had a mean carcass weight (CW) of 292 kg (0.52 of mean live weight) after 214 days at pasture. Cattle offered 25% of their pasture allowance as concentrate (25% Meal) produced 35 kg more CW compared with 66 kg for cattle offered 50% Meal. Bulls finished indoors on *ad lib* concentrate for the second half the grazing season produced 68 and 61 kg more CW than their contemporaries which remained at pasture on No Meal and 25% Meal diets, respectively. In contrast, bulls finished indoors on *ad lib* concentrate for 214 days had a mean CW of 404 kg (0.56 of mean live weight). Finishing bulls indoors for the second half of the grazing season appeared to be the most cost effect management option.

Confirmation of dietary background of beef

A critical component of any strategy to overcome beef-scapes and ensure future competitiveness and sustainability will be an ability to guarantee quality, including authenticity, of beef. Use of DNA technology provides information about the identity of an animal from a beef sample taken after slaughter. This technology cannot provide information about the feed consumed by the animal over its lifetime nor can it confirm information relating to the birth place or subsequent movement of the animal. Results of two feasibility studies suggest that the analysis of natural stable isotope compositions of carbon, nitrogen and sulphur is one potential tool for the verification of the geographical origin and feeding history of beef cattle. Beef reared in the USA (23 samples) and Brazil (10 samples) was, isotopically, different from northern European beef (35 samples), mainly because of contrasting proportion of plants with C_3 and C_4 photosynthetic pathways in the cattle diets. Combined C, N and S stable isotope ratio analysis also separated organically (15 samples) and conventionally (17 samples) produced Irish beef, even though underlying mechanisms are not fully understood at present.

Results to date confirm the potential of this technology to discriminate beef based on its dietary history and geographical location. The ongoing and planned research in this project will identify sources of error in the identification/authentication of beef and will also test its power under less extreme dietary situations such as those typical of Irish beef production.

Production and human health benefits of omega-3 polyunsaturated fatty acid and conjugated linoleic acid-enriched beef

Previous research has demonstrated that beef produced from forage-based systems in particular, can have a fatty acid composition more compatible with dietary recommendations for improved human health. In addition, ruminant fat is the highest natural source of conjugated linoleic acid (CLA), a novel fatty acid with health-enhancing properties. To fully capture the added value of Irish grass-fed beef as a healthy, functional food, the concentration of beneficial fatty acids must be optimised such that beef becomes a significant source of these fatty acids in the human diet. In addition, the relevance of beef CLA to human health in terms of its contribution to total dietary consumption, bio-availability and efficiency must be determined. In a comparison of cattle breeds, muscle from early maturing animals had a higher concentration of CLA than from later maturing animals reflecting the lower fatness of the latter. Increasing bodyweight appears to differentially affect the fatty acid composition of muscle from early and late maturing cattle. The animal phase of a subsequent study concerned with optimising the concentrate of CLA in tissue of grazing animals has been completed. Analysis of fatty acids and delta-9 desaturase gene expression in tissue is in progress as is measurement of colour stability and lipid oxidation of striploin. A high-CLA minced beef product has been prepared for feeding to ob-ob mice, a rodent model of human obesity, diabetes and insulin resistance. The increased concentrations of healthy fatty acids that will be possible as a result of this research will redress the negative consumer perception in relation to beef and promote the consumption of grass-fed beef in particular.

Progesterone Regulated Uterine Gene Expression in Cattle

In a previous large scale field study at Athenry a positive association between low progesterone on days 5, 6 and 7 after AI and subsequent early embryo loss was shown. The current project, as part of a biotechnological approach to determine the molecular basis for this loss, has examined the association between the concentration of progesterone and gene expression in the cow uterus. This study shows that sub optimal concentrations of progesterone as early as day 7 after oestrus can adversely affect the uterine expression of genes encoding the proteins retinol- and folate-binding protein that transport Vitamin A and folic acid to the developing embryo. This is the first time that changes in the expression of these genes this soon after oestrus has been associated with the concentration of systemic progesterone in cattle. Inadequate expression of these genes is likely to be associated with inadequate retinol- and folate- binding protein secretion resulting in inadequate nutrition of the embryo and therefore early embryo death.

Uterine protein expression

In cows, and particularly high yielding dairy cows, inadequate systemic progesterone in the first few days after AI results in an increased rate of early embryo loss. We have shown that the genes encoding the uterine transport proteins, retinol- and folate-binding protein are very sensitive to changes in progesterone concentrations at day 7 after oestrus. Changes in gene expression, however, are often not translated into changes in protein expression. The objective of this project is to determine if retinol- and folate-binding proteins are secreted into the uterus in the first few days after AI and if this secretion rate is dependent on the systemic concentration of progesterone. To date, uterine flushings have been collected from a total of 30 dairy cows and, from these, two groups with significantly different mean concentrations of progesterone have been selected. Sensitive assays for bovine retinol- and folate-binding protein have been developed and are being

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used to determine concentrations of these proteins in blood and uterine flushings and their relationship with systemic progesterone.

Ovarian gene expression

Corpus luteum (CL) function is central to farm animal reproduction. Inadequate progesterone production by the developing CL and/or premature CL regression are associated with embryo loss while persistent CLs that fail to regress result in irregular oestrous cycles. An improved understanding of the molecular mechanisms regulating CL regression has long term implications for improving bovine reproductive efficiency. In this project using a 434 character ovarian specific cDNA array a total of 15 differentially expressed genes have been identified in non-regressed compared to spontaneously regressed bovine CLs. The nature of the identified genes suggests that CL regression is a complex multi-mechanistic process that involves alterations in extracellular matrix remodeling, oxygen radical metabolism, apoptosis and steroid biosynthesis. This new information is providing further insights into the genes and mechanisms regulating the lifespan of the CL and the underlying causes of luteal insufficiency.

Negative Energy Balance

Negative energy balance (NEB) is a severe metabolic disorder affecting high yielding dairy cows during the early postpartum interval which can impair cow fertility and animal health but the precise mechanisms remain unknown. The objective of this study is to investigate the effects of NEB on global patterns of gene expression in metabolic and reproductive tissues. Dairy cows were managed to provide two groups with an average EB status difference of 4UFLs between the groups. Preliminary analysis of metabolic profiles (glucose, BHB, NEFA, urea, cholesterol triglycerides) suggest that cows in the severe NEB group were metabolically compromised. Total RNA has been isolated and quality assessed from metabolic (liver) and reproductive tissues (uterine and oviduct). This RNA will be used in QRT-PCR and cDNA array analysis to determine the effects of NEB on gene expression and how this may impact on cow fertility and cow health.

Maternal Environment of the Early Cow Embryo

Reproductive wastage is now the primary reason for involuntary culling in Irish dairy herds and early embryo loss has been identified as the greatest problem. During their first 16 days cattle embryos are dependent nutritionally on the fluids in the reproductive tract for their development and survival but despite that there is little or no available information on the composition of these fluids or what factors affect this composition. This project developed an *in vivo* approach that allowed measurement of the composition of cattle oviduct and uterine fluids. The pH and concentrations of energy substrates (glucose, lactate and pyruvate), ions (chloride, phosphate, sulphate, sodium, calcium, magnesium and potassium) and 20 amino acids and how these change with stage of the oestrous cycle was determined. This baseline information, much of which is now known for the first time has, and is currently being published in peer reviewed papers and in an end of project report.

Nutrition and cattle fertility

Reproductive efficiency is a major factor affecting production efficiency in both the national dairy and beef cowherds. Data from this laboratory and elsewhere has clearly show that early embryo death before about day 16 of gestation is the major cause of low conception rates. There is some evidence of significant and repeatable differences between animals in their ability to establish and sustain pregnancy as well as genetic variability for heifer pregnancy rate and dairy cow sustainability. An experiment was undertaken to establish the endocrine, molecular and genetic bases for these apparent differences in embryo survival rate. To date a total 69 heifers were inseminated on 4 occasions over a 9-month period. From this the repeatability of conception rate was calculated as 0.183 ± 0.003 . From these heifers two groups of animals, phenotypically

divergent for conception rate have been established are currently been endocrinologically and physiologically characterised.

Suckler Beef Production

In a comprehensive study, five suckler cow genotypes were compared and their progeny taken to slaughter following which their carcasses were dissected into meat, fat and bone. The five cow breed types were LF (Limousin x Friesian), LLF (Limousin x (Limousin x Friesian)), L (Limousin), C (Charolais) and SLF (Simmental x (Limousin x Friesian)). The results (LF = 100) for the first three calf crops for the spring born bull progeny of LF, LLF, L, C and SLF slaughtered at 15 months of age were pre-weaning growth rate 100, 82, 81, 86, 95 ; carcass produced per day of age 100, 94, 94, 99 and 101 ; carcass conformation scores 100, 104, 115, 111 and 111 ; carcass meat proportions 100, 101, 104, 100 and 100 ; carcass fat proportion 100, 97, 80, 93, 99 and carcass bone proportion 100, 97, 93, 104 and 97. These results show the superior pre-weaning gains of the LF and SLF progeny due mainly to superior milk production of their dams and which tended to be reflected in carcass produced per day of age. The pure bred (C and L) and SLF progeny had better conformation. Meat yield was greatest for L progeny as a result of the expected lower bone content in addition to less fat, which was not expected.

Interaction of fertiliser N level and grazing management in a dairy calf-to-beef system

Well managed, moderately intensive, dairy calf to beef system use in excess of 200kg fertiliser N/ha. Under the Nitrate Directive and the Rural Environment Protection Scheme (REPS), the total permitted N input is 260kg/ha (fertiliser plus organic). Thus total permitted fertiliser N is in the range 100-140kg/ha depending on stocking rate. When this is allocated between grazing and silage production the amount available for grazing is in the range 50-60kg/ha. Previous work has shown that when yearling steers directly follow calves in a rotational grazing system, the performance of steers is reduced but the performance of calves is improved compared with separate or mixed grazing. It would be desirable to retain the good calf performance from leader/follower grazing but avoid the adverse effect on yearling performance. A study compared animal performance on swards given high (as for intensive systems) and low (as for extensive REPS systems) levels of fertiliser N and three grazing managements. The result showed that swards which received only fertiliser N in Spring gave the same animal performance as swards that were continually fertilised during the grazing season but stock carrying capacity was lower. Operating a split rotation and/or providing extra grass resulted in a small improvement in animal performance. Small differences in performance during the grazing season were compensated for during finishing the following winter, resulting in the same slaughter weight for all treatment groups. Grazing calves half a rotation ahead of yearlings was no advantage compared to grazing them immediately ahead but providing extra grass for the yearlings gave some benefits and merits further investigation.

Fungi on baled silage on midland farms

A survey was conducted to investigate the extent and identity of visible fungi growing on the surface of baled silage throughout the winter-feeding season on farms in the midlands. Between mid-November 2003 and mid-March 2004, baled silage was surveyed on 50 farms along a route between Cavan town and Thurles (*ca.* 150 km). A detailed questionnaire was completed on each farm visited - records were taken of the number of bales present, harvesting and storage characteristics of the bales, type and extent of damage to the plastic wrap and the means used to prevent and remedy damage to the plastic wrap on the bales. Two bales in readiness for feeding were examined in detail on each farm (total of 100 bales examined).

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Fungal growth was observed on 90/100 (0.9) bales. The proportion of bales affected with fungal growth (n = 20 bales/month) ranged from 0.75 (January) to 1.0 (March). Throughout the winter feeding season, the mean proportion of the bale surface area affected ranged from 0.04 in November to 0.09 in January, with mean proportion coverage of 0.07 over the five months. On average, there were 6 visible fungal colonies on each affected bale and this ranged from one to 21 colonies per bale. A total of 582 fungal colonies were sampled, resulting in 862 isolates. The most frequently isolated fungus was *Penicillium roqueforti* (0.41) and this was present on 78/100 (0.78) bales examined. Other frequently isolated fungi included yeasts, *Schizophyllum commune* and mucoraceous moulds. Visible damage to the plastic film (including repaired damage) was recorded on 49/100 (0.49) bales. Of the 90 bales that were affected with fungal growth, those that had damaged film had proportionally higher mean fungal coverage (0.10) than where the film appeared undamaged (0.04) and this difference was statistically significant ($P < 0.05$).

Plastic film use on farms

European Union policy strongly encourages a sustainable and multifunctional agriculture. Therefore, in addition to providing European consumers with quality food produced within approved systems, agriculture must also contribute positively to the conservation of natural resources and the upkeep of the rural landscape. Plastics are widely used in agriculture and their post-use fate on farms must not harm the environment - they must be managed to support the enduring sustainability of farming systems. A survey was conducted to estimate the quantities of flexible plastic film used on Irish farms and their destinations post primary use. The survey was a supplement to the Teagasc National Farm Survey. It found that nationally, estimated total (mean) plastic use was 4242 t (4.7 kg/ha) for new pit-silage sheets, with corresponding values of 8617 t (21 kg/ha) for baled silage stretch-film, 1059 t (2.5 kg/ha) for baled silage twine/netting, 221 t (56 kg/ha) for maize mulch and 3505 t (1.1 kg/ha) for fertiliser bags. A range of post primary uses of these plastics occurred which differed depending on the primary use of the plastic and the type of plastic.

Housing animals

Restricted spatial allowances in slatted floor facilities may present a challenge to the ability of an animal to cope with housing stress. The effect of varied space allowances on feed intake, performance and immunity of finishing bulls was studied. Housing finishing bulls at a reduced space allowance (1.2 vs. 2.4 and 4.2m²) did not alter average daily feed intake (ADFI), but reduced performance (ADG) without causing substantial effects on immune function.

Animal transport

A series of studies that had been conducted to evaluate the effects of 1) transport by land and sea journeys (roll-on roll-off) and 2) stocking density on the welfare of cattle transported within Ireland, from Ireland to Spain and from Ireland to Italy under conditions outlined in Directive 91/628/EEC. A further study (November 2003) examined the effects of transport on the welfare of weanling heifers transported from Ireland to Spain according to a new proposed regulation COM (2003) 425. It is concluded that within the conditions of the transport studies, transport had no adverse effect on animal welfare. The results of these transport studies made a significant scientific contribution to the debate at National and EU level during the discussions that took place in Ireland and Brussels during the first 6-months of 2004.

Parasite resistance in Suffolks and Texels

Suffolk and Texels show a large difference in parasite resistance. The overall objective of this study was to examine the reasons why this disparity between breeds exists both at a genetic and immunological level. On the basis of previous analysis we had shown that in the Suffolk breed

variation in exon 2 of the DRB1 gene in the major histocompatibility complex was significantly associated with faecal egg count (FEC). Detailed genetic analysis of the alleles at this locus showed that one allele was associated with reduced FEC while two other alleles were associated with increased FEC relative to the most frequent allele in a purebred Suffolk population. Crossbred progeny from Suffolk rams, carrying either the low FEC allele or one of the high FEC alleles and the reference allele, were produced in 2004 and evaluated for FEC during the late-summer and early-autumn periods. The results confirmed the ranking of the alleles with respect to FEC in the purebred Suffolks but the numerical difference in FEC was not statistically significant, although it represented a 30% reduction in FEC. Further evaluation of the role of exon 2 of DRB1 gene is warranted to establish definitively the role of these alleles. Real-time PCR studies were undertaken to evaluate the expression of 6 genes involved in the cytokine complex responsible for immunological responses. Results to date show differential expression between Suffolk and Texel sheep – the direction of change depends on the gene.

Prion Proteins

The objective of this project was to establish whether there was any association between genotype at the prion protein locus (PrP gene) responsible for resistance/susceptibility to scrapie and important production traits. During the year the evaluation of the effects of PrP genotype on lamb growth rate and on the traits used in the Lean Meat Index, generated from the national sheep breed improvement programme operated by The Department of Agriculture and Food, was completed. This involved progeny testing pedigree Suffolk, Texel and Charollais rams with contrasting PrP genotypes. The results show that PrP genotype has no effect on lamb growth traits or on backfat depth measured by ultrasonography. In the Suffolk and Charollais breeds significant associations between PrP genotype and muscle depth were detected. The sign of the association differed between the breeds and the absolute size of the effects was quite small. It was concluded that there are no important negative associations between PrP genotype and important lamb performance traits and consequently the national breeding programme for resistance to scrapie will have no detrimental effect on lamb performance.

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May 2004

Grange Research Centre

BEEF PRODUCTION

SUCKLER BEEF SYSTEMS

Quality suckler beef from low and high input grassland management systems

The standard spring-calving suckling system operated at Grange was compared with a Rural Environment Protection Scheme (REPS) compatible system. The objective was to compare different systems of suckler beef production in order to provide blueprints for production systems. In each of the two systems, the cow herd consisted of five breed types; Limousin x Friesian, Limousin x (Limousin x Friesian), Limousin, Charolais and Simmental x (Limousin x Friesian). The herds were mainly third and fourth calvers. An easy-calving Limousin bull (AI) was used on heifers which were bred to calve at two years old, while Charolais sires (AI initially, then a stock bull) were used on the cows. In each system, the male progeny were left intact and taken to slaughter at 15 months of age. The heifers were destined for slaughter at about 20 months of age in November/December. The breeding season commenced in early May. The plans for the two systems were as follows: 1) standard system: 0.65 ha/cow unit, 225 kg fertiliser nitrogen (N)/ha, 2 silage cuts, and 2) REPS compatible system: 0.82 ha/cow unit, 88 kg N/ha with only one silage cut taken.

In both systems a cow unit consisted of a cow plus the progeny to slaughter plus replacements. The grassland area was randomly divided into four equal sections, two of which were assigned to each system resulting in four herds. The actual numbers of cows (plus progeny and replacements) were 46 and 38 for the standard and REPS systems, respectively. The herds spent from April to late October at pasture and in winter, were offered grass silage, conserved from within the systems. Similar paddock grazing programmes were operated in each system. In the standard system the plan (Table 1) involved cutting 0.37 ha/cow unit (57% of the total area) in May (high quality silage for the progeny) with a further 0.23 ha/cow unit (35% of the total area) cut in late July (lower quality silage for the cows). In the REPS systems the plan was to cut 0.48 ha/cow unit (58% of the area) as first cuts and take no second cut. In the REPS system it was planned that up to 50% of the silage area would be harvested early (in May) to provide high quality silage for the progeny. The remainder, to be harvested two weeks later, was to provide a higher yield of lower quality material, adequate for the cows. The actual quantities of nitrogen applied were 218 and 94 kg/ha on the standard, and REPS systems, respectively.

Table 1: Planned grass conservation programme/cow unit for the standard and REPS systems

	Planned		
	<u>Date</u>	<u>ha/cow unit</u>	<u>% total area</u>
<u>Standard system</u>			
First cut early	May 20/27	0.37	57
Second cut	July 20/27	0.23	35
<u>REPS system</u>			
First cut early	May 20/27	0.24	29
First cut late	June 3/10	0.24	29

Cows averaged about 570 kg liveweight at the start of the grazing season (Table 2). Body condition scores averaged 2.23 (scale 0 to 5) at the start of grazing, having experienced a small loss in body condition in the period from calving to the start of the grazing season. Liveweight gains of the cows over the entire grazing season were 114 and 89 kg for those in the standard and REPS systems, respectively. Corresponding cow body condition score gains over the entire season were 0.35 and 0.30. Calf daily liveweight gains from birth to weaning (housing) were 1144 and 1093 g for those in the standard and REPS systems, respectively.

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The bulls born in 2003 were slaughtered on 8th July 2004 having received a grass silage/concentrate diet from weaning to slaughter. Total concentrate intake per animal over the 239 day period from weaning to slaughter was 1328 kg (5.5 kg/head daily). The heifers received silage plus 1 kg of concentrates per head daily in winter, spent a second grazing season at pasture and were slaughtered on 6th December 2004 having received a total 388 kg of concentrates per head during the last 96 days, of which the final 46 days were on silage plus concentrates. Performance of the bulls and heifers to slaughter and carcass composition is presented in Table 3. The only differences recorded was that bulls in the REPS system had greater ($P<0.05$) slaughter weights than those in the standard system while heifers in the REPS system had a lower ($P<0.05$) carcass fat proportion than those in the standard system.

Table 2: Performance of cows and calves during the grazing season in the two suckler systems

	System	
	<u>Standard</u>	<u>REPS</u>
<u>Cows</u>		
Weight post-calving, (kg)	577	596
Weight at turnout, (kg)	553	585
Weight change: turnout to June (kg)	62	45
Weight change: June to housing (kg)	52	44
Weight change: turnout to housing (kg)	114	89
Body condition score post-calving	2.38	2.33
Body condition score at turnout	2.17	2.28
Condition score change: turnout to June	0.17	0.05
Condition score change: June weaning	0.18	0.25
Condition score change: turnout to weaning	0.35	0.30
<u>Calves</u>		
Weight at birth (kg)	48.0	47.7
Weight at housing (kg)	267	255
Age at housing (days)	190	190
Daily gains birth to housing (g)	1144	1093

Table 3: Performance of the progeny in the two systems

	Bulls			Heifers		
	<u>Standard</u>	<u>REPS</u>		<u>Standard</u>	<u>REPS</u>	
Birth wt (kg)	50	53	NS	46	44	NS
Weaning wt (kg)	262	280	NS	247	254	NS
Slaughter wt (kg)	570	597	*	535	524	NS
Carcass wt (kg)	328	344	NS	292	288	NS
Killing out rate (g/kg)	577	576	NS	547	550	NS
Age at slaughter (days)	452	452	-	609	609	-
Liveweight gain/d (g)	1150	1200	NS	804	790	NS
Carcass gain/d (g)	726	758	NS	480	474	NS
Hind quarter (g/kg)						
Meat	775	777	NS	761	769	NS
Fat	38	37	NS	61	52	*
Bone	187	186	NS	178	180	NS

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Performance of the progeny of five suckler cow breed types

In Ireland, suckler dams are increasingly being sourced from within the suckler herd. Approximately 85% of mature suckler cows are bred to late-maturing continental sire breeds, 46% of which are Charolais. Thus, sourcing replacements from within the suckler herd would result in a high proportion of continental, mainly Charolais genes, due to the extensive use of Charolais sires. Continual breeding using just one breed would lead to loss of the heterosis advantage. Two important determinants of profitability in suckler beef production are high progeny weight gains and the production of animals (or carcasses) suitable for the highest priced markets. The objectives of this study were to determine the effect of suckler cow breed type on (a) the efficiency of beef output and (b) carcass quality.

The Grange spring-calving suckler herd, consisting mainly of second and third parity animals, was used in this study. Five cow breed types were examined: LF (Limousin x Friesian), LLF (Limousin x Limousin x Friesian), L (Limousin), C (Charolais) and SLF (Simmental x Limousin x Friesian). An easy-calving Limousin A.I. sire was used for first calvers (small proportion) which were bred to calve at two years of age while Charolais sires were used for second and third calvers. The progeny used were born in spring 2003 and spent from April until November at pasture with their dams. They were then housed in a slatted floor shed. The bulls were slaughtered on 8th July 2004, at approximately 15 months of age. The heifers were returned to pasture in April for a second grazing season and were slaughtered at about 20 months of age on 6th December 2004. Liveweights were recorded at regular intervals. Carcass data collected included standard EU carcass classification (conformation and fat) and kidney and channel fat weights. Meat, fat and bone weights were recorded following dissection of the hindquarter (8 rib pistola) from one side of each carcass. The statistical procedure used the SAS Proc GLM model with dam breed, sire and birth day as a covariate. Student's Newman Kuels multiple range test was used to compare the means.

The bull progeny of LF cows had greater daily gains from birth to weaning (DGBW) than the remaining four groups which were similar (Table 4). Daily gains from birth to slaughter (DGBS) of the bull progeny of LF and SLF were similar but greater than the L and C progeny. The L and C progeny had similar DGBS while values for LLF progeny were intermediate and not significantly different than any other group. Values for the carcass produced per day of age for the LF, LLF, L, C and SLF cows' bull progeny were 783 (100), 756 (97), 709 (91), 675 (86) and 773 (99) g, respectively. There was no significant effect of cow breed type on carcass conformation. There were some differences in fat scores but not in pistola fat proportions.

There was no significant difference between the heifer progeny of the different dam breeds in DGBW or DGBS (Table 5). Values for carcass produced per day of age (NS) for the heifer progeny of LF, LLF, L, C and SLF cows were 497 (100), 460 (93), 458 (92), 481 (97) and 489 (98) g, respectively. There were no differences between the progeny of the different cow types in carcass fat scores but the proportion of fat in the hindquarter was lower for the heifer progeny of the C cows than all other groups except L which were intermediate. Kidney plus channel fat weight showed a similar trend.

Table 4: Mean values for liveweight, carcass weight, killing-out rate, weight gains and carcass traits of bull progeny of five beef cow breed types

Dam breed	LF	LLF	L	C	SLF	s.e.		Significance
						n=11	n=2	
No. of animals	8	8	9	2	11			
Slaughter weight (kg)	624 ^a	584 ^{ab}	550 ^b	545 ^b	600 ^a	12.9	28.3	**
Cold carcass (kg)	354.5	342.4	320.5	305.0	349.6	8.45	18.53	*
Killing-out rate (g/kg)	570	586	582	561	583	4.9	10.7	NS
Daily gain to weaning (g)	1238 ^a	956 ^b	912 ^b	966 ^b	1070 ^b	47.6	104.3	***
Daily gain to slaughter (g)	1263 ^a	1186 ^{ab}	1102 ^b	1089 ^b	1210 ^a	28.1	61.6	**
Carcass weight/day of age (g)	783	756	709	675	773	18.1	39.8	*
Carcass conformation score ¹	3.35	3.80	3.87	3.59	3.84	0.151	0.331	NS
Carcass fat score ²	2.70 ^a	2.18 ^a	1.85 ^b	2.16 ^{ab}	2.11 ^b	0.123	0.269	**
Kidney + channel fat (kg)	7.9 ^a	5.3 ^b	4.7 ^b	4.0 ^b	5.7 ^b	0.55	1.21	**
Body condition score	2.40 ^a	1.71 ^a	1.70 ^a	1.67 ^{ab}	1.65 ^b	0.151	0.330	*
Carcass hind quarter (g/kg)	464	478	471	491	472	6.1	13.6	NS
Pistola meat (g/kg)	751	773	771	748	768	5.2	11.3	*
Pistola fat (g/kg)	68	59	54	61	56	4.2	9.2	NS
Pistola bone (g/kg)	181 ^a	168 ^b	174 ^{ab}	191 ^a	175 ^a	3.5	7.6	*
Meat to bone ratio	4.2 ^b	4.6 ^a	4.5 ^a	4.0 ^b	4.4 ^{ab}	0.11	0.23	*

¹Scale 1 to 5 (best conformation) ²Scale 1 to 5 (fattest); ^{abc}Within rows means with different superscripts differ from one another (P<0.05); *P<0.05; **P<0.01; ***P<0.001; NS = Not significant.

Table 5: Mean values for liveweight, carcass weight, killing-out rate, weight gains and carcass traits of heifer progeny of five beef cow breed types

Dam breed	LF	LLF	L	C	SLF	s.e.		Significance
						N=12	N=5	
No. of animals	8	7	5	12	7			
Slaughter weight (kg)	555	507	503	533	553	13.7	18.5	*
Cold carcass (kg)	303	280	279	293	298	7.8	10.4	NS
Killing-out rate (g/kg)	546	552	553	549	539	5.4	7.2	NS
Daily gain to weaning (g)	1006	886	862	956	1020	48.9	60.2	NS
Daily gain to slaughter (g)	832	768	754	796	1020	21.7	29.1	NS
Carcass weight/day of age (g)	497	460	458	481	489	12.9	17.3	NS
Carcass conformation score ¹	3.07	2.98	3.25	3.34	3.05	0.15	0.20	NS
Carcass fat score ²	3.19	3.23	3.01	3.03	3.58	0.15	0.21	NS
Kidney + channel fat (kg)	7.4 ^b	6.4 ^{ab}	5.4 ^a	5.1 ^a	7.7 ^b	0.66	0.88	*
Body condition score	2.76 ^b	2.51 ^{ab}	1.99 ^a	1.94 ^a	2.75 ^b	0.237	0.318	*
Carcass hind quarter (g/kg)	493 ^b	496 ^{ab}	503 ^a	505 ^a	489 ^b	4.1	5.4	*
Pistola meat (g/kg)	743	739	768	761	735	8.4	11.3	*
Pistola fat (g/kg)	78 ^b	80 ^b	57 ^{ab}	51 ^a	84 ^b	7.5	10.0	**
Pistola bone (g/kg)	180	181	176	188	181	3.9	5.2	NS
Meat to bone ratio	4.2	4.1	4.4	4.1	4.1	0.11	0.15	NS

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The effect of sire muscularity on bull progeny scanned eye muscle measurements, muscular scores and carcass conformation

Conformation is an important determinant of the price obtained for carcasses. There is substantial variation both within and between breeds for conformation. This has implications in the selection of animals with genetically superior carcass traits for breeding. The objectives of this study were to determine the effect of sire conformation on ultrasonic muscle measurements, visual muscular scores and carcass conformation scores of bull progeny at weaning and at slaughter.

Four Charolais sires were examined over 2 years, one of average and one of good conformation in each year. In year one the expected progeny difference (EPD) values for the sire of good (MDO) conformation were 49.2, 1.16 and 0.0 and for the average sire were (CF47) 49.6, 1.0 and -0.09 for growth, conformation and fatness respectively. In year two corresponding figures for the good (CF46) sire were 53.3, 1.13 and -0.11 and for the average (CF41) sire, 44.6, 1.04 and -0.44. The number of progeny from MDO, CF47, CF46 and CF41 were 13, 16, 18 and 17 respectively. The bull progeny from five cow breed types were used in this study: LF (Limousin x Friesian), LLF (Limousin x (Limousin x Friesian)), L (Limousin), C (Charolais) and SLF (Simmental x (Limousin x Friesian)). The progeny were offered a grass silage plus concentrate diet from weaning (8 months) until slaughter at approximately 450 days of age (July). The animals were ultrasonically scanned for eye muscle area, maximum muscle depth and average muscle depth at the 12th-13th rib and the 3rd lumbar using an Aloka 500v ultrasound unit. Visual muscular scores were assigned using the Signet muscular scoring system (3 scorers) which examines characteristics at three locations (roundness of hindquarter, width of rump and width /thickness of loin). Other data collected included liveweights, carcass weights and standard EU carcass classification (conformation and fat). Meat, fat and bone weights were recorded following dissection of the eight rib Italian pistola from one side. The factors included in the SAS General Linear Model Procedure were sire conformation, dam breed and year with age as a covariate.

Ultrasonically scanned muscle areas and depths were greater for the progeny of sires of good conformation at both weaning and slaughter (Table 6). Signet muscular scores at weaning and slaughter were also significantly larger for sires of good conformation. Carcass weight per day of age was similar for sires of average and good conformation. Carcass conformation scores were better for sires of good conformation as was meat proportion in the pistola. Sires of average and good conformation had similar pistola fat proportions. The proportion of bone in the pistola was lower for sires of good conformation.

In conclusion, ultrasonically scanned muscle measurements and visual muscular scores taken at weaning and slaughter were greater for the progeny of sires of good conformation indicating the usefulness of live animal scanning and muscular scoring in the selection of sires of good conformation at an early age. Carcass conformation score of the progeny reflected that of their sires. Pistola meat content was greater for the sires of good conformation.

Table 6: Mean values for ultrasonic muscle measurements, Signet muscular scores and carcass data for progeny of Charolais sires of average and good conformation (Least squares mean \pm s.e.)

		Sire conformation		Significance
		Average	Good	
<i>Weaning</i>	Number of animals	33	31	
	Area 12-13 th rib (cm ²)	60.1 \pm 1.23	65.7 \pm 1.23	**
	Max. depth 12-13 th rib (cm)	7.0 \pm 0.08	7.3 \pm 0.09	**
	Average depth 12-13 th rib (cm)	5.2 \pm 0.07	5.5 \pm 0.07	**
	Area 3 rd lumbar (cm ²)	40.3 \pm 1.01	44.6 \pm 1.02	**
	Max. depth 3 rd lumbar (cm)	5.5 \pm 0.08	5.8 \pm 0.08	*
<i>Slaughter</i>	Signet muscular score (cm)	19.5 \pm 0.58	21.2 \pm 0.59	**
	Area 12-13 th rib (cm ²)	95.3 \pm 1.63	102.1 \pm 1.63	**
	Max. depth 12-13 th rib (cm)	8.6 \pm 0.13	8.9 \pm 0.13	*
	Average depth 12-13 th rib (cm)	6.8 \pm 0.09	7.2 \pm 0.09	**
	Area 3 rd lumbar (cm ²)	67.4 \pm 1.41	71.7 \pm 1.42	*
	Max. depth 3 rd lumbar (cm)	22.9 \pm 0.82	26.0 \pm 0.83	**
<i>Carcass</i>	Signet muscular score	23.0 \pm 0.72	25.9 \pm 0.73	**
	Carcass weight / day of age (g)	714 \pm 9.1	732 \pm 9.2	NS
	Carcass conformation score ¹	3.16 \pm 0.090	3.48 \pm 0.091	**
	Pistola meat (g/kg)	753 \pm 2.9	765 \pm 2.9	**
	Pistola fat (g/kg)	55 \pm 2.0	52 \pm 2.0	NS
	Pistola bone (g/kg)	192 \pm 2.2	184 \pm 2.2	**

¹Scale 1 to 5 (best conformation) *P<0.05; **P<0.01; ***P<0.001.

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Residual feed intake, feed conversion ratio, growth and body composition traits in pedigree beef bulls

As maintenance energy and feed costs are a considerable proportion of the total costs of beef production, accordingly beef breeding selection strategies need to focus on improving feed efficiency without negatively altering performance or carcass traits. Traditionally feed efficiency was expressed as the ratio of weight gain to feed intake (FCR) but selection for this measure can lead to an increase in mature size and thus maintenance requirements. An alternative measure of feed efficiency proposed is residual feed intake (RFI) or net feed efficiency, which is the difference between actual energy intake (EI) and calculated EI required for bodyweight maintenance and liveweight gain (negative or lower values desirable). This measure is largely independent of growth and maturity patterns. As heritability estimates range from 0.16 to 0.43 the concept of RFI can be used to identify efficient bulls. The objective of this study was to characterise RFI in Irish pedigree performance tested bulls and evaluate the relationship between RFI and growth and carcass traits.

Data were obtained on a total of 255 Charolais (CH) and 432 Limousin (LM) pedigree bulls that completed a performance test at the Irish bull testing station in Tully between January 1998 and June 2004. There were 28 batches of bulls tested in total. Expected EI (UFV/day) was calculated by regressing average daily EI (intake period min. 98 to max. 168 days) on average daily liveweight gain (ADG) and mid-test liveweight^{0.75} with a model which included batch using the GLM procedure of SAS. The RFI for each bull was calculated as actual EI minus the expected EI predicted from the regression model generated for each batch. FCR was calculated as average daily EI divided by ADG. Relationships between RFI and FCR and between performance and ultrasound measures of eye muscle area and fat cover were

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determined using partial correlation coefficients (corrected for batch) using the CORR procedure of SAS. Within breed, bulls were then ranked by RFI and separated into low, medium and high groups that were < 0.5 SD, ± 0.5 SD and > 0.5 SD respectively, from the mean RFI. These data were analysed using GLM with a model that included RFI group, and batch as a covariate. Final test-weight per day of age was used as an indicator of mature weight.

Unlike FCR, there was no significant relationship between RFI and weight per day of age or ADG but RFI and EI was significantly correlated (Table 7).

Table 7: Partial correlations of RFI and FCR and, performance and carcass traits for Limousin and Charolais bulls

Parameter	Breed	RFI	FCR
Wt. per day of age (kg)	LM	0.03 (NS)	-0.17 (***)
	CH	-0.05 (NS)	0.12 (*)
ADG (kg/day)	LM	-0.00 (NS)	-0.62 (***)
	CH	0.00 (NS)	-0.68 (***)
EI (UFV/day)	LM	0.49 (***)	0.16 (**)
	CH	0.49 (***)	0.24 (***)
FCR (UFV / kg gain)	LM	0.43 (***)	-
	CH	0.41 (***)	-
Fat area (cm ²)	LM	0.10 (*)	-0.00 (ns)
	CH	0.19 (**)	0.09 (ns)
Muscle area (cm ²)	LM	0.05 (NS)	0.05 (ns)
	CH	-0.07 (NS)	0.08 (ns)

While growth rates and weight per day of age were similar, high RFI bulls consumed more feed per day and had a higher FCR ($P<0.001$) than low RFI bulls (Table 8). There was no effect of RFI on ultrasound estimates for LM but fat area was lower ($P<0.01$) in the low RFI than the high RFI CH sires.

Table 8: Comparison of low, medium and high RFI

Parameter	Breed	RFI			s.e.	Significance
		Low	Med	High		
RFI (kg)	LM	-0.48	0.00	0.55	0.020	***
	CH	-0.62	0.00	0.57	0.026	***
Wt per day of age (kg)	LM	1.46	1.47	1.48	0.012	NS
	CH	1.66	1.62	1.62	0.018	NS
ADG (kg)	LM	1.58	1.62	1.61	0.022	NS
	CH	1.80	1.80	1.77	0.030	NS
EI (UFV /day)	LM	8.2	8.7	9.3	0.08	***
	CH	9.1	9.6	10.1	0.11	***
FCR (UFV /kg gain)	LM	5.28	5.48	5.95	0.080	***
	CH	5.10	5.40	5.79	0.076	***

Results suggest that RFI is an alternative measure of feed efficiency, which is independent of growth traits and mature weight.

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RMIS No. 4936

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DAIRY CALF TO BEEF SYSTEM

Effects of separate or mixed feeding of silage and concentrates on carcass traits of beef cattle

An experiment on diet mixing was described previously (Research Report, 2003).

The study aimed to:

- (1) compare separate and mixed feeding of silage and concentrates when the target concentrate proportions in the dietary dry matter (DM) were 0.4 and 0.8
- (2) ascertain if there were interactions between feeding method (separate or total mixed ration (TMR) and dietary concentrate level)
- (3) determine the response to increasing levels of concentrates in the diet, and
- (4) ascertain the effects of duration of the finishing period on performance.

Finishing steers (117 Friesians and Charolais x Friesians) were used in 6 treatments (18/treatment) and a pre-experimented slaughter group (n = 9). The treatments were:

- (1) Silage only (SO)
- (2) Silage and concentrates at target 0.6:0.4 DM ratios fed separately (LS)
- (3) Silage and concentrates at target 0.6:0.4 DM ratios fed mixed (LM)
- (4) Silage and concentrates at target 0.2:0.8 DM ratios fed separately (HS)
- (5) Silage and concentrates at target 0.2:0.8 DM ratios fed mixed (HM)
- (6) Concentrates *ad libitum* + 1kg/day silage DM (AL)

The animals were housed in two slatted floor sheds. One shed fitted with Calan-Broadbent doors accommodated 84 animals. These were fed individually throughout. The second shed accommodated 24 animals in groups of 4 by treatment. Group intakes of these were recorded. Half of the animals in each feeding treatment were slaughtered after 105 day (E) and the rest were slaughtered after 175 day (L). Every effort was made to ensure that concentrate intake was similar for the corresponding separate and mixed treatments. The concentrate allowance was fed once daily to the separate groups. The silage analysis (g/kg) was: DM 201, *in vitro* DM digestibility 674, ash 93 and pH 3.9. The calculated NE was 0.70 UFV. The concentrate formulation was (g/kg): rolled barley 870, hi-pro soyabean meal 67.5, molasses 47.5, minerals/vitamins 15. The calculated NE value was 1.14 UFV. At slaughter, carcass weight, carcass grades, weight of perirenal plus retroperitoneal fat and standard carcass measurements were recorded. The 6-10th ribs joints were separated into muscle, fat and bone. The data were analysed using a model that had terms for treatment, slaughter time, treatment x slaughter time, mixing, mixing x concentrate level and the linear and quadratic effects of concentrate level. Data on growth and slaughter traits were presented previously (Research Report, 2003). This report contains the data on carcass traits.

The effects of concentrate level on absolute carcass measurements are shown in Table 9. Both leg length and circumference of round increased with increasing concentrate level, with both the linear and quadratic components significant but carcass depth and leg width were unaffected by concentrate level. There was a quadratic (but not linear) effect of concentrate level on carcass length and there was a linear effect on leg thickness. When scaled for carcass weight all measurements decreased with increasing concentrate level with both the linear and quadratic components significant for all. There was no significant concentrate level x feeding method interaction and there was no significant effect of feeding method on any of the measurements either absolutely or scaled for carcass weight. There was no significant feeding treatment x duration of finishing interaction. Other than leg width which was unaffected, all the absolute measurements were significantly greater for the longer duration of finishing. Scaled for carcass weight all measurements were significantly lower for the longer duration of finishing.

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As with carcass weight, weight of both fore and hind quarters and ribs joint increased with increasing concentrate level and both the linear and quadratic components were significant (Table 10). As a proportion of carcass side weight, pistola weight decreased linearly with increasing concentrate level and fat depth increased with increasing concentrate level with both the linear and quadratic components being significant. Ribs joint composition was also significantly affected by concentrate level. The proportions of both fat depots and total fat increased, and the proportions of total muscle and bone decreased, with increasing concentrate level and the linear and quadratic components were significant for all ($P < 0.07$ for total muscle quadratic). The relationship of *m. longissimus* proportion with concentrate level was quadratic only, while that for "other muscle" proportion was linear only (decreased with increasing concentrate level).

There was a significant concentrate level x feeding method interaction for ribs joint weight in that it was greater for TMR at the low but not at the high concentrate level. Otherwise, there were no concentrate level by feeding method interactions and there was no significant effect of feeding method on carcass traits or ribs joint composition. There were no significant concentrate level x duration of finishing interactions. Fore and hind quarter weights and fat depth all increased with increasing duration of finishing but ribs joint weight was not significantly affected. The proportions of pistola in the side decreased with increasing length of finishing period. The proportions of subcutaneous fat and bone were not significantly affected by duration of finishing. Otherwise, intermuscular fat and total fat proportions increased significantly while *m. longissimus*, "other muscle" and total muscle proportions decreased significantly with increasing length of finishing.

Because the linear and quadratic components of the concentrate effects on intake and growth were significant, quadratic models were fitted to the data (Table 11). Silage intake decreased at an increasing rate with increasing concentrate level. The models accounted for proportionately, 0.84 and 0.74 of the variation for silage and total intakes, respectively. Daily live weight and carcass gains decreased at a decreasing rate with increasing concentrate level and the models accounted for proportionately 0.74 and 0.75 of the variation for live weight and carcass gains, respectively. Carcass fat score increased at a decreasing rate with increasing concentrate level but the model only accounted for proportionately 0.20 of the variation. For carcass conformation score, where only the linear component was significant in the analysis of variance, the model only accounted for proportionately 0.08 of the variation. Linear regressions of various carcass traits on carcass conformation and fat scores are shown in Table 12. As breed type would be expected to affect these relationships, the relationships are shown separately for the two breed types. Daily liveweight gain was not significantly related to carcass conformation score but daily carcass gain was, although the model accounted for only 0.10 to 0.25 of the variation. Kill-out proportion was significantly related to carcass conformation with the model accounting for 0.44 to 0.53 of the variation. Both the pistola proportion and the proportion of muscle in the ribs joint were also significantly related to carcass conformation but the models accounted for only a small proportion of the variation (0.08-0.09 for pistola proportion and 0.11 to 0.14 for ribs joint muscle proportion).

Fat depth and ribs subcutaneous fat proportion were moderately related to carcass fat score with the models accounting for proportionately 0.45 to 0.58 of the total variation. Although significant, the relationship between ribs intermuscular fat proportion and carcass fat score was poor with only proportionately 0.07 to 0.08 of the variation accounted for by the model. The relationship between fat score and ribs total fat proportion was intermediate between that for subcutaneous fat and intermuscular fat proportions with the models accounting for proportionately 0.22 to 0.30 of the variation. Perirenal plus retroperitoneal fat as a proportion of carcass weight was not significantly related to carcass fat score.

Table 9: Effects of concentrate level, feeding method and duration of finishing on carcass measurements and carcass measurements scaled for carcass weight of finishing steers

	Feeding Treatment (F)										Duration (D)					Significance					
	SO	LS	LM	HS	HM	AL	s.e. ₁	S	L	s.e. ₂	F	D	F x D	M ¹	M x C ⁴	L ⁵	Q ⁶				
<u>Carcass measurements (cm)</u>																					
Carcass length	134.1	136.9	137.8	137.7	134.9	135.8	1.02	134.2	138.2	0.60	*	***	NS	NS	NS	NS	NS	*			
Carcass depth	51.2	51.3	52.2	51.0	51.7	51.0	0.64	50.8	52.0	0.38	NS	*	NS	NS	NS	NS	NS	NS			
Leg length	72.4	74.3	73.2	74.8	73.8	73.2	0.55	73.0	74.2	0.32	*	**	NS	NS	NS	NS	*	*			
Leg width	44.5	45.0	45.8	45.5	45.7	45.7	0.45	45.0	45.7	0.27	NS	NS	NS	NS	NS	NS	NS	NS			
Leg thickness	26.6	27.8	28.3	27.8	28.0	28.4	0.28	27.5	28.2	0.16	***	**	NS	NS	NS	NS	**	P<0.06			
Circumference of round	112.5	117.2	116.6	119.2	117.0	118.7	0.91	114.0	119.7	0.54	***	***	NS	NS	NS	NS	***	*			
<u>Carcass measurements (cm/kg)</u>																					
Carcass length	0.525	0.442	0.439	0.416	0.412	0.398	0.0059	0.457	0.421	0.0035	***	***	NS	NS	NS	NS	***	***			
Carcass depth	0.201	0.166	0.166	0.155	0.158	0.150	0.0031	0.173	0.158	0.0018	***	***	NS	NS	NS	NS	***	***			
Leg length	0.284	0.241	0.235	0.228	0.228	0.216	0.0029	0.249	0.228	0.0016	***	***	NS	NS	NS	NS	***	***			
Leg width	0.174	0.145	0.146	0.138	0.140	0.134	0.0023	0.153	0.139	0.0014	***	***	NS	NS	NS	NS	***	***			
Leg thickness	0.104	0.090	0.090	0.084	0.085	0.083	0.0014	0.093	0.086	0.0008	***	***	NS	NS	NS	NS	***	***			
Circumference of Round	0.440	0.377	0.370	0.360	0.357	0.348	0.0049	0.387	0.363	0.0029	***	***	NS	NS	NS	NS	***	***			

For n = 18; ²For n = 54; ³Method of feeding (separate or TMR); ⁴Method of feeding by concentrate level interaction; ⁵Linear effect of concentrate level; ⁶Quadratic effect of concentrate level.

Table 10: Effects of concentrate level, method of feeding and duration of finishing on carcass traits and ribs joint composition of finishing steers

Weight of (kg)	Feeding Treatment (F)										Duration (D)				Significance Of							
	SO	LS	LM	HS	HM	AL	s.e. ¹	S	L	s.e. ²	F	D	FxD	M	MxC	L	Q					
Fore quarter	67.1	82.9	83.9	89.3	88.1	93.4	1.23	78.1	90.2	0.73	***	***	NS	NS	NS	***	***					
Hind quarter (pistola)	62.0	72.9	73.8	76.6	75.5	78.4	1.17	70.1	76.3	0.69	***	***	NS	NS	NS	***	***					
Ribs joint	6.40	7.76	8.24	8.72	8.50	8.78	0.170	8.10	8.03	0.100	***	NS	NS	NS	*	***	***					
Pistola (g/kg side)	481	468	471	464	461	459	2.9	474	461	1.7	***	***	NS	NS	NS	***	NS					
Fat depth (mm)	4.9	9.1	9.7	10.4	10.6	11.3	0.62	8.3	10.4	0.37	***	***	NS	NS	NS	***	***					
Ribs joint composition (g/kg)																						
Subcutaneous fat	33	56	55	61	64	63	3.7	54	57	2.2	***	NS	NS	NS	NS	***	***					
Intermuscular fat	129	153	160	176	179	182	7.7	146	181	4.5	***	***	NS	NS	NS	**	*					
<i>M.longissimus</i>	208	192	198	196	197	202	4.8	203	194	2.9	NS	*	NS	NS	NS	NS	*					
Other muscle	401	393	386	376	372	369	6.9	395	371	4.1	**	***	NS	NS	NS	*	NS					
Total fat	163	209	215	237	242	245	9.3	200	238	5.5	***	***	NS	NS	NS	***	**					
Total muscle	609	584	584	573	569	571	8.2	598	565	4.8	**	***	NS	NS	NS	*	P<0.07					
Total bone	227	207	201	190	189	184	4.1	202	197	2.4	***	NS	NS	NS	NS	***	**					

¹For n = 18; ²For n = 54. See Table 1 footnotes also.

Table 11: Regressions ($Y = a + b_1X + b_2X^2$) of silage dry matter (DM) intake, total dry matter intake, daily gains and carcass grades on concentrate level (kg/day)

<u>X = Concentrate level¹</u>	<u>Intercept</u>		<u>Regression coefficients</u>				<u>R²</u>
	<u>a</u>	<u>s.e.</u>	<u>b₁</u>	<u>s.e.(b₁)</u>	<u>b₂</u>	<u>s.e.(b₂)</u>	
Silage intake (kg/DM/day)	7.23	0.206	-0.438	0.0866	-0.0136	0.0081	0.84
Total intake (kg/DM/day)	7.23	0.206	0.562	0.0866	-0.0136	0.0081	0.74
Daily gain to 97 days (g)	221	79.2	280	37.8	-18	3.7	0.71
Overall daily gain (g)	222	58.7	221	28.0	-14	2.8	0.74
Carcass gain (g/day)	148	34.0	123	16.2	-7	1.6	0.75
Fat score	2.99	0.153	0.226	0.0729	-0.017	0.0072	0.20
Conformation	1.98	0.181	0.05	0.086	0.001	0.0085	0.08

¹Using values of 4.0, 8.0 and 9.5kg DM/day for low concentrates, high concentrates and *ad libitum* concentrates, respectively.

Table 12: Regressions ($Y = a + bX$) of daily gains and carcass traits on carcass conformation and carcass fat scores

<u>X = Conformation score</u>	<u>Breed¹</u>	<u>a</u>	<u>s.e. (a)</u>	<u>b</u>	<u>s.e.(b)</u>	<u>Sig.</u>	<u>R²</u>
Daily live weight gain (g)	F	764	240.2	90	98.9	NS	-0.003
	C	516	192.4	147	81.5	NS	0.04
Kill-out (g/kg)	F	449	10.1	27	4.2	***	0.44
	C	444	11.2	37	4.7	***	0.53
Carcass gain (g/day)	F	201	132.5	143	54.6	*	0.10
	C	97	101.3	184	42.9	***	0.25
Pistola (g/kg side)	F	451	8.9	9	3.7	*	0.09
	C	438	9.3	9	3.9	*	0.08
Ribs muscle (g/kg)	F	533	20.4	26	8.4	**	0.14
	C	500	22.9	27	9.7	**	0.11
Ribs bone (g/kg)	F	262	10.2	-25	4.2	***	0.39
	C	246	11.8	-21	5.0	***	0.23
<u>X = Fat score</u>							
Fat depth	F	-6.2	1.72	4.3	0.51	***	0.58
	C	-6.6	2.47	4.8	0.69	***	0.47
Ribs subcutaneous fat (g/kg)	F	-2.4	9.5	21	2.8	***	0.56
	C	-36	14.1	26	3.9	***	0.45
Ribs intermuscular fat (g/kg)	F	81	29.5	20	8.7	*	0.08
	C	100	37.6	24	10.5	*	0.07
Ribs total fat (g/kg)	F	58	30.3	43	8.9	***	0.30
	C	63	44.4	50	12.4	***	0.22
Perirenal + retroperitoneal fat (g/kg carcass)	F	24	11.0	3	3.2	NS	-0.01
	C	23	12.4	5	3.5	NS	0.18

¹F = Friesian, C = Charolais x Friesian

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Comparison of barley and sugar beet pulp with and without added protein as supplements for weanling steers

The optimum growth rate for young steers in winter depends on the response to, and cost of, feed in winter and the extent to which the animals express compensatory gain subsequently at pasture. In Irish beef systems the basal winter diet of cattle is grass silage which may be variable in quality. This is usually supplemented with concentrates to achieve the desired rate of gain.

On a dry matter (DM) basis, the net energy of molassed sugar beet pulp (MSBP) is proportionately about 0.94 that of barley but as MSBP is higher in DM, on an “as fed basis” their net energy values are broadly similar. These two feeds are widely used as supplements with silage for young growing cattle and depending on price they are substituted for each other on a fresh weight basis. While they may be of similar energy value on a fresh weight basis, crude(cp) protein content and the main protein fractions are lower for pulp.

The objectives of this study with young growing steers were (1) to determine the response to increasing levels of supplementary MSBP as a supplement to silage, (2) to compare MSBP and barley, and (3) to ascertain if there was a response to the inclusion of soya bean meal as a protein source with MSBP and barley.

One hundred and fifty four weanling steers of mixed breed type (70 Belgian Blue x Friesians, 28 Angus x Friesians, 28 Limousin x Friesians and 28 Friesians) were blocked on weight and breed type and assigned from within blocks to 7 equal treatment groups of 22 animals each. The treatments were:

1. Silage only (SOO)
2. Silage plus a low level of pulp (LPO)
3. Silage plus a low level of pulp plus soya bean meal (LPS)
4. Silage plus a high level of pulp (HPO)
5. Silage plus a high level of pulp plus soya bean meal (HPS)
6. Silage plus a high level of barley (HBO)
7. Silage plus a high barley plus soya bean meal (HBS)

Low and high pulp levels were 1.5 and 3.0 kg, respectively per head daily. The high barley level was also 3.0 kg per head daily. In the treatments with soya bean meal, 0.2 kg /day of soya bean meal replaced 0.2 kg /day of low pulp and 0.4 kg of soya bean meal replaced 0.4 kg/day of high pulp or barley. All animals were given 70 g/day of an appropriate mineral/vitamin premix. It was dusted on the silage for the silage only group and was added to the concentrates for the supplemented groups. The treatments commenced on November 20 and lasted for 125 days until March 25. From 49 and 105 days, 11 representative blocks of animals were accommodated in tie-up stalls and individual silage intakes were recorded for 56 days. At the end of the 125-day winter treatment period the animals were weighed on two consecutive days, scored for body condition (scale 1 to 5) and put to pasture together for a 148-day grazing period. At pasture they grazed behind calves in a leader/follower rotational grazing system. The experiment terminated on August 20.

The mean analysis of the feeds used are shown in Table 13. Pulp was 40 g/kg higher in DM concentration than barley. Barley and pulp had similar CP concentrations on an “as fed basis” but differed by about 14 g/kg on a DM basis. Low pulp supplied 1.31 UFL (net energy for growth) plus 139 g CP, and high barley supplied 2.73 UFL plus 297 g CP per head daily. Adding soya bean meal to pulp or barley had a negligible effect on UFL supply but increased CP supply by 75 g/day with low pulp, 151 g/day with high pulp and 148 g/day with high barley. Compared with high pulp, high barley supplied 0.12 more UFL per day. The supply

of CP was broadly similar for pulp and barley (278 and 297 g/day without soya bean meal and 429 and 445 g/day with soya bean meal).

Silage intakes, both absolute values and per kg live weight, for the individually fed animals are shown in Table 14. There was a significant linear effect of pulp level for all measures of intake. Compared with silage only, in the absence of soya bean meal the first increment of pulp reduced silage DM intake by 0.36 kg/day while the second increment reduced it by a further 1.20 kg/day. Per kg liveweight, the first pulp (without soya bean meal) increment reduced silage DM intake by 3.5 g and the second increment reduced it by a further 5.0 g. The effect of soya bean meal was not significant but there was a significant pulp level by soya bean meal interaction such that inclusion of soya bean meal increased intake with high but not with low pulp.

All measures of intake were significantly higher for barley than for pulp, and while the concentrate type by soya bean meal interaction was not significant, inclusion of soya bean meal greatly reduced the intake difference between high pulp and barley. When soya bean meal was absent silage intake was 1.02 kg DM/day higher for barley than for the same level of pulp whereas when soya bean meal was present the difference was only 0.20 kg DM/day. Per kg mean live weight, silage intakes was 2.5 g/day higher for barley than pulp in the absence of soya bean meal compared with only 0.3 g/day higher when soya bean meal was present.

Live weight gains are shown in Table 15. For the period from 49 days to the end of winter and for the winter period as a whole there were significant linear and quadratic effects of pulp level. During the first 49 days live weight gain on silage only was 339 g/day. This fell to 231 g/day afterwards to the end of the winter. The higher initial value probably reflects an increase in gut contents after the animals were housed. Mean responses to the first and second pulp increments for the winter period as a whole were 330 and 128 g/day, respectively. Although differences were not significant over the first 49 days, there was a significant response to soya bean meal with both levels of pulp and barley for the entire winter period. There was a pulp level by soya bean meal interaction for the period from 49 days to the end of the winter. This was due to a higher response to soya bean meal at the high than at the low pulp level (389 v. 193 g/day). There was also a concentrate type by soya bean meal interaction for the entire winter period due to a higher response with pulp than barley (275 v. 70 g/day). Overall responses in winter to soya bean meal with low and high pulp and with barley were 167, 275 and 70 g/day, respectively. The superiority of barley over the same level of pulp was 241 g/day in the absence of soya bean meal and 36 g/day when soya bean meal was included.

During the first month at pasture there was a significant linear effect of pulp level and a significant effect of soya bean meal. Thereafter, while the pulp level effect tended to be close to significance ($P < 0.08$), there were no other significant effects. In the first month after turn out live weight gain was 101 g/day lower for those on the low pulp (mean of with and without soya bean meal) than for those on silage only. There was further reduction of 95 g/day for those on high pulp (mean of with and without soya bean meal). For the grazing season overall, live weight gain on low pulp (mean of with and without soya bean meal) was 95 g/day lower than those on silage only and the gain of those on high pulp was 27 g/day lower again. In the first month after turn out, live weight gain was significantly lower for those that had received soya bean meal in winter, with values of 138, 273 and 30 g/day for low pulp, high pulp and barley, respectively. This trend continued to the end of the grazing season although after the first month the differences were not statistically significant. For the grazing season as a whole, inclusion of soya bean meal in winter reduced gains at pasture for low and high pulp by 34 and 28 g/day, respectively. Mean liveweight gain at pasture was 30 g/day greater for pulp than barley.

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For the entire 273 day experimental period there was a significant linear effect of pulp level comprising 100 g/day for the first increment and a further 44 g/day for the second increment. There was also a significant effect of soya bean meal comprising of 57, 111 and 1 g/day with low pulp, high pulp and barley, respectively. Barley was 102 g/day superior to pulp in the absence of soya bean meal but when soya bean meal was included there was no difference.

Starting live weight was the same for all treatments (Table 16). There was a significant linear effect of pulp level on all live weights to the end of grazing but the quadratic component never attained significance. There were no significant pulp level or concentrate type by soya bean meal interactions for any of the live weights. Both soya bean meal and concentrate type significantly affected end of winter but not end of grazing season live weights. By the end of winter the animals on low pulp were (mean of with and without soya bean meal) 41 kg heavier than those on silage only. The additional weight advantage for the high pulp (mean of with or without soya bean meal) was 16 kg. The winter live weight responses to soya bean meal were 21 kg with low pulp, 33 kg with high pulp and 9 kg with barley. The winter live weight advantage to barley over the same level of pulp was 29 kg in the absence of soya bean meal and 5 kg when soya bean meal was included. Body condition score reflected live weight and was significantly affected by pulp level, soya bean meal and concentrate type. It increased linearly with increasing pulp level, was significantly higher when soya bean meal was included and was significantly higher for barley than for the same level of pulp. By the end of the experiment, only the linear effect of pulp level was significant but there were numerical differences in favour of soya bean meal (16 and 30 kg for low and high pulp, respectively), and of barley over pulp (27 kg) in the absence of soya bean meal.

Compared with silage only, low and high pulp (means of with and without soya bean meal) increased end of winter weight by 41 and 57 kg, respectively. Of these, 27 (66%) and 39 (68%) kg were still present at the end of the experiment. Inclusion of soya bean meal increased mean end of winter live weight by 21 kg and of this 16 kg (75%) was still present at the end of the experiment. Compared with pulp, barley increased mean end of winter weight by 11 kg and of this 8 kg (74%) was still present by the end of the experiment. These figures indicate that only about one quarter to one third of the differences in weight at the end of the winter were compensated for and there was little difference between the different sources of winter weight gain (pulp level, soya bean meal, concentrate type) in the extent of compensation.

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RMIS No. 5075

Table 13: Mean analysis¹ of feeds offered to weanling steers

	<u>Silage</u>	<u>Barley</u>	<u>Pulp</u>	<u>Soya bean meal</u>
Dry matter (g/kg)	210	828	868	871
Crude protein (g/kg)	143	99	93	469
Oil A	-	8.4	2.9	15.6
Ash (g/kg)	75.3	21.0	75.0	62.3
Lactic acid (g/kg)	120	-	-	-
NH ₃ N (g/kg total N)	65	-	-	-
Acid detergent fibre (g/kg)	309	51	141	53
In vitro DM digestibility	731	894	916	908
PH	3.6	-	-	-
UFL ²	0.82	0.91	0.87	1.04

¹Values for silage are on a dry matter basis, values for barley, pulp and soya bean meal are on a fresh basis.

²Unite Fourragere Lait - net energy value for growing cattle.

Table 14: Silage dry matter intakes of weanlings offered different concentrate supplements

Per day (kg)	Treatment										Significance					
	SOO	LPO	LPS	HPO	HPS	HBO	HBS	s.e. ¹	L ²	Q ³	S ⁴	P x S ⁵	C ⁶	C x S ⁷		
Days 0 to 28	3.46	3.20	3.13	1.93	2.57	2.82	2.77	0.209	***	P<0.08	NS	*	***	NS		
Days 29 to 56	4.08	3.63	3.64	2.49	3.36	3.63	3.57	0.223	***	NS	NS	*	***	*		
Days 0 to 56	3.77	3.41	3.38	2.21	2.97	3.23	3.17	0.212	***	NS	NS	*	**	**		
Per kg live weight (g)																
Days 0 to 28	17.3	14.9	15.2	9.4	12.2	12.4	12.7	0.55	***	*	P<0.08	*	***	NS		
Days 29 to 56	19.1	14.7	15.1	10.4	12.7	12.9	13.1	0.58	***	NS	NS	*	**	**		
Days 0 to 56	18.3	14.8	15.0	9.8	12.2	12.3	12.5	0.51	***	NS	NS	*	***	***		

¹For n = 11. ²Linear effect of pulp level; ³Quadratic effect of pulp level; ⁴Soya bean meal effect; ⁵Pulp level x soya bean meal interaction; ⁶Concentrate type effect. There was no significant concentrate type x soya bean meal interaction.

Table 15: Live weight gains of weanlings and yearlings offered different concentrate supplements in winter

Live weight gains (g/day) for	Treatment										Significance					
	SOO	LPO	LPS	HPO	HPS	HBO	HBS	s.e. ¹	L ²	Q ³	S ⁴	P x S ⁵	C ⁶	C x S ⁷		
Days 0 to 49	339	511	637	737	838	786	834	66.6	***	NS	NS	NS	NS	NS		
Days 49 to 125	231	527	720	502	891	868	947	43.6	***	**	***	*	***	*		
Days 0 to 125 ⁸	274	521	688	595	870	836	906	32.9	***	**	***	NS	***	**		
Days 125 to 154	864	832	694	804	531	605	575	69.8	*	NS	*	NS	NS	NS		
Days 154 to 203	1009	989	952	928	839	886	769	58.5	P<0.08	NS	NS	NS	NS	NS		
Days 203 to 273	1080	939	907	982	944	966	955	81.4	NS	NS	NS	NS	NS	NS		
Days 125 to 273 ⁹	1043	965	931	935	907	919	862	55.2	P<0.08	NS	NS	NS	NS	NS		
Days 0 to 273 ¹⁰	691	762	819	779	890	881	882	34.6	**	NS	*	NS	NS	NS		

¹For N = 22; ²Linear effect of pulp level; ³Quadratic effect of pulp level; ⁴Soya bean meal effect; ⁵Pulp level x soya bean meal interaction; ⁶Concentrate type effect; ⁷Concentrate type x soya bean meal interaction; ⁸Entire winter period; ⁹Entire grazing period; ¹⁰Entire period.

Table 16: Live weights and body condition scores of weanlings and yearlings offered different concentrate supplements in winter

Live weight (kg) on	Treatment										s.e. ¹	L ²	Significance		
	SOO	LPO	LPS	HPO	HPS	HBO	HBS	S ³	C ⁴						
Day 0 (Nov 20)	187	187	187	187	187	187	187	187	187	187	6.5	NS	NS	NS	
Day 49 (Jan 08)	204	212	219	223	228	225	228	228	228	228	7.8	*	NS	NS	
Day 125 (Mar 25)	222	252	273	262	295	291	300	300	300	300	8.9	****	**	*	
Body condition score (Day 125) ⁵	2.05	2.32	2.57	2.52	2.75	2.86	2.91	2.91	2.91	2.91	0.070	****	**	****	
Day 154 (Apr 23)	247	276	294	285	311	309	317	317	317	317	9.3	****	*	NS	
Day 203 (Jun 11)	300	329	348	334	361	360	360	360	360	360	10.6	****	P<0.08	NS	
Day 273 (Aug 20)	376	395	411	400	430	427	428	428	428	428	14.2	**	NS	NS	

¹For n = 22; ²Linear effect of pulp level; ³Soya bean meal effect; ⁴Concentrate type effect; ⁵Scale 1 (poorest) to 5 (best). There was no significant quadratic component for pulp level, no significant pulp level x soya bean meal interaction and no significant concentrate type x soya bean meal interaction.

Comparison of Belgian Blue x Friesian and Charolais x Friesian steers for beef

In recent years the Belgian Blue has been replacing the Charolais for cross breeding in the dairy herd. There is a paucity of data on the relative merits for beef production of these two breeds when crossed on Holstein - Friesian dairy cows. A total of 84 (48 Belgian Blue x Friesians and 36 Charolais x Friesians) male animals were purchased as calves and reared together to slaughter. The Belgian Blue (BB) were the progeny of 3 known AI bulls (36) and unidentified bulls (12). The Charolais (CH) were the progeny of one known AI bull (12) and unidentified bulls (24). It was presumed that the unidentified bulls were stock bulls. This broadly reflects the relative usage of AI and stock bulls of the two breeds in Irish dairy herds. Mean birth dates, mean arrival dates at Grange, and mean arrival weights for BB and CH were February 27 and February 16, March 23 and March 11, and 62 and 67 kg, respectively. The calves were conventionally reared and turned out to pasture on May 31. They grazed ahead of yearling steers in a leader/follower rotational grazing system. No concentrates were fed after turnout but 1 kg/day of concentrates was fed from castration on September 20 to housing on November 29. During the first winter the animals were offered medium quality silage *ad libitum* plus 1 kg concentrates per head daily until turnout on March 29. During the second grazing season they followed calves in leader/follower rotational grazing system. The animals were housed on October 9 but finishing did not start until November 7. Finishing over the second winter was on silage *ad libitum* plus 5 kg concentrates per head daily. They were slaughtered on March 20 but final weighing was on March 13.

In the first winter the animals were accommodated in 12 pens in a slatted floor shed (8 BB per pen and 6 CH per pen) giving 6 observations per breed type for the measurement of feed intake. Prior to turnout at the start of the second grazing season individual silage intakes were measured for two weeks on 26 BB and 22 CH. These same animals were housed again in May, July and September for the measurement of grass intake. Fresh grass was harvested daily, treated with formic acid as for silage conservation and a weighed allowance was offered daily for 14 days to each animal in a tie-up shed. Refusals were weighed back and discarded daily. After slaughter, cold carcass weights, carcass grades and routine carcass measurements were recorded. The right side was divided into a pistola hind quarter and fore quarter. The 6 – 10th rib joint was removed and dissected into its component tissues. Live weights and live weight gains are shown in Table 17. The Charolais crosses were 12 days older and 5 kg heavier at arrival. This difference increased to 11 kg at turnout and to 24 kg at first housing due to a significantly higher growth rate by CH from first turnout to first housing. Thereafter, while growth differences were not significant, values tended to be higher for the BB crosses. Nevertheless, the CH were still 26 kg heavier ($P < 0.05$) at the start of finishing and were 19 kg heavier at slaughter. There were no significant differences between the breed types in slaughter weight per day of age and in carcass weight per day of age which were 11 g (1.3%) and 6 g (1.3%) higher for the CH.

Silage intake in the first winter and grass intake in the second grazing season did not differ significantly between the breed types but tended to be lower for the CH crosses (Table 18). When account is taken of the fact that the CH were heavier by adjusting intake for live weight, the difference was significant with the CH having a 6.3% lower grass intake per kg live weight. Slaughter traits and carcass measurements are shown in Table 19. There was no significant difference between the breed types in kill out proportion, but because the CH crosses were 19 kg heavier at slaughter, their carcass weight was 10 kg heavier (both differences not significant). Conformation score was significantly better for CH but fat score was significantly lower for the BB. Fat depth reflected fat score in that it was also lower for the BB (difference not significant) but kidney plus channel fat scaled for carcass weight was significantly lower for CH. *M.longissimus* area scaled for carcass weight was similar for the two breed types. None of the carcass measurements scaled for carcass weight differed significantly between the breed types but all the values were numerically lower for CH indicating a trend towards greater carcass compactness in line with their superior carcass conformation score. There were no significant differences between the breed types in the

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proportion of side weight in the pistola or in the composition of the ribs joint although there was a trend towards lower fat proportions in the BB in line with their lower fat score (Table 20). It is concluded that lifetime daily live and carcass growth rates were similar for the two breed types but the CH achieved these with a slightly lower feed intake. Carcass conformation was better and indicators of carcass compactness tended to be better for CH, but carcass fat score was lower and other indicators of carcass fatness tended to be lower for BB crosses. Internal fat however, as indicated by kidney plus channel fat, was higher for the BB crosses. There were no difference between the breed types in weight distribution between the fore and hind quarters or in ribs joint composition.

Table 17: Lifetime live weights and live weight gains of Belgian Blue x Friesian (BB) and Charolais x Friesian (CH) steers

<u>Live weights (kg) at:</u>		<u>BB</u>	<u>s.e.</u>	<u>CH</u>	<u>s.e.</u>	<u>Significance</u>
Arrival ¹		62	1.3	67	1.6	*
First turnout (May 31)		111	2.9	125	3.5	**
First housing (Nov. 29)		253	5.3	277	6.3	**
Second turnout (March 29)		346	8.0	364	9.4	NS
Start of finishing (Nov. 7)		516	6.5	542	7.7	*
Slaughter (March 13) ²		637	8.7	656	10.3	NS
<u>Live weight gains (g/day) for:</u>	<u>Days</u>					
First turnout to first housing	182	778	18.0	838	21.2	*
First housing to second turnout	120	771	47.7	718	56.5	NS
Second turnout to second housing	224	761	34.1	797	40.3	NS
Finishing period ³	126	936	44.1	871	52.2	NS
First to second turnout	302	775	22.7	790	26.8	NS
Second turnout to slaughter ³	350	824	19.5	824	23.0	NS
First turnout to end of finishing	652	802	12.0	808	14.2	NS
Arrival to slaughter ⁴	-	792	11.8	797	14.2	NS
<u>No days from</u>						
Arrival to slaughter ⁴	-	727	2.3	739	2.7	***
Birth to slaughter ⁴	-	751	2.2	763	2.6	**
<u>Per day of age (g)</u>						
Slaughter weight	-	849	11.5	860	13.6	NS
Carcass weight	-	458	6.9	464	8.2	NS

¹March 23 and March 11 for BB and CH, respectively. ²The animals were slaughtered on March 20 but final weighing was on March 13, ³To March 13, ⁴March 20.

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Table 18: Dry matter intakes of Belgian Blue x Friesian (BB) and Charolais x Friesian (CH) steers

	BB	s.e.	CH	s.e.	Significance
Silage intake in first winter (kg) ¹	3.68	0.289	3.57	0.289	NS
For 2 weeks pre turnout ²	5.03	0.101	4.80	0.109	NS
Grass intake in May ³	7.57	0.116	7.38	0.126	NS
Grass intake in July ³	6.27	0.112	6.26	0.122	NS
Grass intake in September ³	9.04	0.168	8.69	0.182	NS
Mean grass intake (kg) ³	7.63	0.096	7.44	0.104	NS
Mean grass intake (g/kg LW) ³	18.5	0.034	17.4	0.372	NS

¹Measured on 6 pens per breed type for 17 weeks, ²Silage intake for 22 and 26 animals for BB and CH, respectively, ³Measured indoors on 22 and 26 animals per breed, LW = Liveweight.

Table 19: Slaughter traits and carcass measurements for Belgian Blue (BB) and Charolais x Friesian (CH) steers

	BB	s.e.	CH	s.e.	Significance
Carcass weight (kg)	344	5.2	354	6.2	NS
Kill-out (g/kg)	539	2.0	538	2.3	NS
Conformation	2.38	0.08	2.66	0.09	*
Fat score	3.08	0.12	3.48	0.14	*
Kidney + channel fat (g/kg) ¹	38.9	1.11	32.4	1.33	**
Fat depth (mm)	10.6	0.70	11.5	0.83	NS
<i>M. longissimus</i> area (cm ² /kg) ¹	0.258	0.0044	0.252	0.0053	NS
<u>Carcass measurements (cm/kg)¹</u>					
Carcass length	0.404	0.0055	0.396	0.0066	NS
Carcass width	0.148	0.0021	0.143	0.0025	NS
Leg length	0.216	0.0029	0.212	0.0035	NS
Leg width	0.134	0.0017	0.131	0.0020	NS
Leg thickness	0.083	0.0011	0.083	0.0012	NS
Round circumference	0.357	0.0042	0.349	0.0050	NS

¹Of carcass

Table 20: Pistola proportion and ribs joint composition of Belgian Blue x Friesian (BB) and Charolais x Friesian (CH) steers

	BB	s.e.	CH	s.e.	Significance
Pistola (g/kg side)	466	2.4	465	2.8	NS
<u>Ribs composition (g/kg)</u>					
Subcutaneous fat	51	2.7	54	3.2	NS
Intermuscular fat	142	5.1	148	6.1	NS
<i>Longissimus thoracis</i>	216	3.7	215	4.4	NS
Other muscle	404	4.6	399	5.5	NS
Total fat	193	7.1	203	8.4	NS
Total muscle	620	6.1	613	7.2	NS
Total bone	187	3.0	184	3.5	NS

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Comparison of progeny of two Belgian Blue bulls reared as bulls and steers

Calves by artificial insemination from two progeny tested Belgian Blue bulls (KIC and EWN) and out of Holstein - Friesian cows were sourced on dairy farms through insemination records. Calves were reared normally on milk replacer, hay and concentrates and turned out to pasture in early May. In late September, 16 calves per bull of similar mean birth date were selected for the comparison. Within bull they were blocked on weight and half were castrated. They remained at pasture until housing in early November. On housing in a slatted shed, the bulls were offered grass silage and 4 kg concentrates per head daily until two months before slaughter when concentrates were increased to 6 kg/day. The mean duration of the finishing period was 328 days. During the first winter, the steers were offered silage and 1 kg concentrates per head daily until turnout in late March for a second grazing season. They were housed in early October for finishing on silage and 2 kg/day concentrates until two months before slaughter when it was increased to 6 kg/day. The mean duration of finishing was 162 days. After slaughter, perinephric plus retroperitoneal fat was weighed, carcasses were graded and a number of linear carcass measurements were made. The ribs joint (6-10th) was dissected into subcutaneous fat, intermuscular fat, bone, *m. longissimus et thoracis* and other muscle. When this project commenced, AI bulls genetic values were expressed in index form relative to the breed mean = 100 with a standard deviation of 10. Subsequently, the method of expression changed to units of trait on an across breed basis. When the bulls were selected for comparison there were indices differences of 9 units of growth, 5 units of conformation and 25 units of kill-out in favour of KIC (Table 21). EWN had a leanness advantage of 32 units of index. When the genetic values were converted to units of trait the differences between the bulls changed. The growth difference converted to a progeny difference of 3.8 kg carcass weight, the conformation difference now favoured EWN and the large kill-out difference almost disappeared.

Table 21: Genetic values for two Belgian Blue Bulls (Index and Units of Trait)

Index	Growth	Conf.	Lean	Kill-out	Reference
EWN	104	106	128	90	Progressive Genetics
KIC	113	111	96	115	Beef Sire Panel, 1999
<u>Units of Trait</u>					
EWN	30.8	1.07	-0.68	3.48	Grogan, 2001
KIC	34.6	0.97	-0.25	3.58	Genetic Values for Beef AI Bulls

Mean birth dates of the calves were February 13 and February 15 for KIC and EWN, respectively. Mean intervals from birth to slaughter of the bulls and steers were 603 and 771 days, respectively. Mean intervals from birth to slaughter for KIC and EWN progeny were 688 and 686 days, respectively. There were no significant difference in liveweight between the progeny of the two bulls at any time. The progeny of KIC grew faster from arrival to first housing, but subsequently the EWN progeny caught up and mean liveweight gain from arrival to slaughter was identical for the two progeny groups. As the interval from arrival to slaughter was 11 days shorter for EWN progeny, their slaughter weight per day was slightly higher (954 v 942 g/day). Carcass weight per day followed the same trend (536 v 519 g/day). By chance, the steers were heavier than the bulls at first turnout as calves. There was no difference at first housing (shortly after castration of the steers) but thereafter the bulls were always significantly heavier than the steers. Slaughter weight per day was 1056 and 840 g for bulls and steers, respectively. Corresponding carcass weights per day were 598 and 457g (Table 22).

Except for conformation which was better ($P < 0.08$) for EWN, there were no significant differences in slaughter traits between the progeny of the two bulls (Table 23) and neither were there any differences in the carcass measurements. Because slaughter weight was fixed

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there was no difference between bulls and steers and neither was the carcass weight difference significant. Kill-out proportion was significantly better, fat score and perinephric plus retroperitoneal fat proportion were significantly lower, and *m. longissimus* area scaled for carcass weight was significantly greater for bulls. There was no difference in carcass measurements other than carcass depth which was greater for steers. In line with slaughter and carcass weight there was no difference between KIC and EWN progeny in side weight or pistola proportion (Table 24). However, there was a difference in ribs composition with KIC having significantly more fat and less muscle than EWN. Bulls had a lower proportion of pistola than steers. They also had less fat and more muscle in the ribs joint. There were no biologically important interactions between sire and sex.

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Table 22: Liveweights and liveweight gains and carcass gains for progeny of two Belgian Blue bulls

	Bull (B)		Sex (S)		s.e.d.	Significance		
	KIC	EWN	Bull	Steer		B	S	B x S
<u>Liveweights (kg) at:</u>								
First turnout	103	98	90	111	3.9	NS	**	NS
First housing	226	205	218	214	8.2	NS	NS	NS
Second turnout	340	334	358	316	10.3	NS	**	NS
Final weighing	499	489	553	435	12.1	NS	***	NS
Silage intake (kg/day)	3.59	3.75	-	-	0.069	NS	-	-
Arrival to slaughter (days)	656	645	572	729	2.3	*	***	NS
Birth to slaughter (days)	688	685	603	771	2.34	NS	***	NS
<u>Liveweight gains (g/day)</u>								
Arrival – first housing	713	615	685	685	19.2	*	NS	NS
Arrival – slaughter	848	848	949	747	12.3	NS	***	NS
Slaughter weight per day (g)	942	954	1056	840	13.7	NS	***	NS
Carcass weight per day (g)	519	536	598	457	7.1	NS	***	NS

Table 23: Slaughter data and carcass measurements for progeny of two Belgian Blue bulls

	Bull (B)		Sex (S)		Significance			
	KIC	EWN	Bull	Steer	s.e.d.	B	S	B x S
	Slaughter weight (kg)	609	609	606	612	12.5	NS	NS
Carcass weight (kg)	335	342	344	333	6.4	NS	NS	NS
Kill-out (g/kg)	550	561	567	544	1.6	**	***	NS
Conformation	3.03	3.33	3.48	2.88	0.111	P<0.08	***	NS
Fat score	3.25	2.99	2.83	3.41	0.114	NS	***	NS
Perinephric and retroperitoneal fat (g/kg) ¹	23.0	24.5	15.5	32.0	0.88	NS	***	*
<i>M. longissimus</i> area (cm ² /kg) ¹	0.246	0.246	0.254	0.238	0.0050	NS	NS	NS

Carcass measurements (cm)

Carcass length	136.0	135.2	135.0	136.2	1.01	NS	NS	NS
Carcass depth	48.4	47.6	46.7	49.3	0.39	NS	NS	***
Leg depth	71.2	71.4	70.5	72.1	0.72	NS	NS	NS
Leg width	45.7	44.9	44.8	45.8	0.52	NS	NS	NS

¹Of carcass weight**Table 24: Side weight, pistola proportion and ribs composition of progeny of two Belgian Blue bulls**

	Bull (B)		Sex (S)		Significance			
	KIC	EWN	Bull	Steer	s.e.d.	B	S	B x S
Side weight (kg)	169.0	172.6	172.7	168.9	2.25	NS	NS	NS
Pistola (g/kg side)	442	450	426	465	3.8	NS	***	NS
Ribs composition (g/kg)								
Total fat	144	127	100	166	7.8	*	***	NS
Total muscle	676	697	724	656	8.3	***	***	NS
Total bone	180	175	177	179	3.9	NS	NS	NS

Comparison of progeny of Belgian Blue and Charolais bulls of known and unknown genetic index

Bulls in AI approved for widespread use generally have genetic indices for growth, carcass and reproductive traits. Thus, the relative merits of their progeny in beef systems can be estimated. Calves sired by stock bulls or from unknown sires purchased at livestock marts are of unknown genetic merit. A total of 84 spring-born male calves sired by Belgian Blue and Charolais bulls out of Holstein-Friesian cows were reared to slaughter within the framework of a two year old steer production system. There were 49 Belgian Blue sired calves (18 progeny of EWN, 19 progeny of KIC and 12 from unknown sires (BBM) purchased in small numbers at livestock marts). There were 35 Charolais sired calves (13 progeny of HKI and 22 from unknown sires (CHM) purchased in small numbers at livestock marts. The animals were managed together throughout, and any experimental treatments were balanced across the progeny groups. Calf rearing was by standard procedures and all calves were turned out to pasture together on May 31 where they grazed ahead of yearlings in a leader/follower system of rotational grazing. They remained at pasture until November 29 when they were housed for the first winter and offered silage *ad libitum* plus a mean level of 1 kg/day concentrates until turnout for the second grazing season on March 29. During the second grazing season they grazed behind calves in a leader/follower rotational grazing system. They were housed for finishing on November 7 and over the finishing winter they were offered silage *ad libitum* plus a mean concentrate level of 5 kg/day until final weighing on March 13 and slaughter on March 20. After slaughter, cold carcass weight, carcass grades and routine carcass measurements were recorded. The right carcass side was divided into a pistola hindquarter and forequarter. The ribs joint (6 to 10) was removed and separated into its component tissues.

At insemination the relative (breed mean = 100) breeding values (BVs) of the sires were as follows: KIC-growth 113, conformation 97 and leanness 86, EWN-growth 104, conformation 110 and leanness 128, HKI-growth 136, conformation 120, leanness 113. Since the sires of the mart purchased calves were unknown, it was presumed that they would approximate to the breed mean and that the comparison would demonstrate the differences between breed mean progeny and the progeny of bulls with genetic indices superior to the breed mean. Liveweights of the 5 progeny groups are shown in Table 25. There were differences between the groups in birth date, arrival date and arrival weight which had knock-on effects on subsequent liveweights to slaughter. The mart purchased calves were born earlier, were heavier at arrival and remained heavier to slaughter. Other than at arrival, there was no significant difference in liveweight at anytime between the KIC, EWN and HKI progeny groups. Because of the differences in birth dates and liveweights, it is necessary to compare daily gains to ascertain if there were differences in growth rate between the groups. Liveweight gains together with slaughter and carcass weights per day of age are shown in Table 26. Other than in the first grazing season when the mart purchased calves gained faster (probably a consequence of their earlier birth date and greater arrival weight), there were no significant differences in liveweight gains between the progeny groups. Slaughter weight per day of age differed by less than 4% between the groups and carcass weight per day of age differed by less than 3%.

Slaughter traits and carcass measurements scaled for carcass weight are shown in Table 27. There was no significant difference between the progeny groups in carcass weight, kill-out proportion or carcass measurements scaled for carcass weight. The mart purchased Charolais had significantly better conformation than the three Belgian Blue progeny groups. The progeny of EWN had a significantly lower fat score than all other groups. Kidney plus channel fat proportion was significantly lower for the HKI progeny than for the three Belgian Blue progeny groups. EWN progeny had a significantly larger *m. longissimus* area per kg carcass than the mart purchased Belgian Blue group. Pistola proportion and ribs joint composition are shown in Table 28. Pistola proportion and ribs joint weight did not differ between progeny groups. Ribs composition was generally similar for KIC progeny and both

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mart purchased groups. EWN progeny had significantly lower fat and higher muscle proportions than KIC progeny and both mart purchased groups. HKI progeny ribs composition was intermediate between that of EWN progeny and the other groups.

The outcome of the comparison between KIC and EWN progeny was consistent with the results shown earlier. Previously, slaughter weight was similar and carcass weight was less than 2% greater for EWN, while here, slaughter weight per day was less than 3%, and carcass weight per day was less than 2%, greater for KIC. None of these differences were close to being significant. In both comparisons, kill-out proportion was higher for EWN but again the difference was not significant. The lower fat score of EWN progeny was also consistent between comparisons as was the absence of differences in carcass measurements. Although the differences were not significant, in both comparisons EWN progeny had lower proportions of fat, higher proportions of muscle and similar proportions of bone. Overall, it can be concluded that notwithstanding the differences in growth genetic index, KIC and EWN progeny had similar live and carcass growth rates, but at the same carcass weight EWN progeny were less fat and had more muscle in line with their higher leanness index.

The mart purchased Belgian Blue calves were similar to the KIC progeny. They tended to grow faster initially, probably reflecting their earlier birth date and heavier weight at arrival, but the KIC progeny overtook them with the result that both groups had almost identical slaughter and carcass weights for age. Slaughter traits and ribs composition were also similar. Because they were computed on a within breed basis, the genetic indices for HKI are not directly comparable with those for KIC and EWN. However, as the breed means for the main production traits are similar for Belgian Blue and Charolais, it could be inferred that HKI progeny would have superior growth and conformation to KIC or EWN progeny. This was not the case. HKI progeny were almost identical to KIC and EWN progeny. The performance of the mart purchased Charolais was equal in every respect to that of the HKI progeny. Like the mart purchased Belgian Blues, performance was better in the first grazing season probably because of their earlier birth date and greater arrival weight. The mart purchased group tended to have better conformation and a higher fat score which was paralleled by more fat and less muscle in the ribs joint.

While all reasonable precautions were taken to ensure that the calves were from the sires indicated, sire identify was not confirmed by genotyping. Calves were inspected shortly after birth to confirm breed type and that birth date was in keeping with service date. It is concluded that the growth index differences between KIC, EWN and HKI were not reflected in differences in the growth rate of their progeny. Carcass leanness index differences were evident in the progeny particularly in the ribs joint composition. Mart purchased calves had similar growth rates and slaughter traits to the progeny of high growth index AI bulls.

Table 25: Liveweights of progeny of Belgian Blue and Charolais bulls

	<u>KIC</u>	<u>EWN</u>	<u>BBM</u>	<u>HKI</u>	<u>CHM</u>	<u>s.e.¹</u>	<u>Significance</u>
No. animals	19	18	12	13	22		
Birth date	Mar. 3	Mar. 7	Feb. 11	Feb. 26	Feb. 9	3.7	***
Arrival date	Mar. 23	Apr. 3	Mar. 10	Mar. 21	Mar. 5	3.8	***
<u>Liveweights (kg) at:</u>							
<u>Arrival</u>	56 ^a	64 ^b	71 ^c	62 ^b	71 ^c	2.2	***
1 st Turnout (May 31)	105 ^a	106 ^a	130 ^b	107 ^a	136 ^b	4.9	***
1 st Housing (Nov 29)	245 ^a	234 ^a	294 ^b	245 ^a	297 ^b	8.2	***
2 nd Turnout (Mar 29)	335 ^a	326 ^a	361 ^b	327 ^a	365 ^b	14.3	**
2 nd Housing (Nov 7)	520 ^a	506 ^a	526 ^{ab}	518 ^a	556 ^b	12.8	**
Final weighing (Mar 13)	637	620	650	629	665	17.1	NS
Slaughter	643 ^{ab}	621 ^a	653 ^{ab}	634 ^{ab}	669 ^b	17.4	*
<u>Days from</u>							
Birth to slaughter	749	743	769	751	769	3.7	***
Arrival to slaughter	728	716	740	729	746	3.8	***

¹For N = 12, Values within a row with a common superscript are not significantly different

BBM = Mart purchased Belgian Blue. CHM = Mart purchased Charolais

Table 26: Liveweight gains of progeny of Belgian Blue and Charolais bulls

<u>Liveweight gain (g/day) for:</u>	<u>KIC</u>	<u>EWN</u>	<u>BBM</u>	<u>HKI</u>	<u>CHM</u>	<u>s.e.¹</u>	<u>Significance</u>
First turnout to first housing	774 ^a	703 ^b	899 ^c	759 ^{ab}	885 ^c	30.6	***
First housing to second turnout	751	765	558	685	567	98.0	NS
Second turnout to second housing	826	804	737	853	853	70.6	NS
Second housing to final weighing	928	906	991	879	866	90.5	NS
First turnout to final weighing	817	788	798	801	812	24.6	NS
<u>Per day of age (g)</u>							
Slaughter weight	859	835	851	844	870	23.4	NS
Carcass weight	461	454	457	457	467	14.2	NS

Table 27: Slaughter traits and carcass measurements of progeny of Belgian Blue and Charolais bulls

	<u>KIC</u>	<u>EWN</u>	<u>BBM</u>	<u>HKI</u>	<u>CHM</u>	<u>s.e.¹</u>	<u>Significance</u>
Carcass weight (kg)	345	337	351	344	359	10.6	NS
Kill-out (g/kg)	537	542	356	541	537	3.9	NS
Conformation	2.4 ^a	2.4 ^a	2.4 ^a	2.5 ^{ab}	2.8 ^b	0.16	*
Fat score	3.4 ^{bc}	2.6 ^a	3.3 ^{bc}	3.1 ^{bc}	3.7 ^c	0.23	**
Kidney + channel fat (g/kg)	39.2 ^a	37.8 ^a	40.0 ^c	27.9 ^b	34.9 ^{ab}	1.74	*
<i>M.longissimus</i> (cm ² /kg carcass)	0.25 ^{ab}	0.27 ^a	0.24 ^b	0.26 ^{ab}	0.25 ^{ab}	0.009	*
<u>Carcass measurements (cm/kg)</u>							
Side length	0.41	0.41	0.40	0.41	0.39	0.012	NS
Carcass depth	0.15	0.15	0.14	0.14	0.14	0.004	NS
Leg length	0.22	0.22	0.22	0.22	0.21	0.006	NS
Leg width	0.13	0.14	0.13	0.13	0.13	0.004	NS

Table 28: Pistola proportion and ribs joint composition of progeny of Belgian Blue and Charolais bulls

	<u>KIC</u>	<u>EWN</u>	<u>BBM</u>	<u>HKI</u>	<u>CHM</u>	<u>s.e.¹</u>	<u>Significance</u>
Pistola (g/kg side)	463	470	465	470	463	4.7	NS
Ribs joint weight (g)	9492	8798	9476	8877	9376	356.8	NS
<u>Ribs composition (g/kg)</u>							
Subcutaneous fat	60 ^c	39 ^a	54 ^b	44 ^{ab}	61 ^c	4.9	***
Intermuscular fat	159 ^b	112 ^a	161 ^b	130 ^{ab}	159 ^b	8.8	***
<i>M.longissimus</i>	208 ^a	231 ^b	208 ^a	217 ^{ab}	213 ^a	7.1	*
Other muscle	387 ^a	425 ^b	398 ^a	414 ^{ab}	398 ^a	8.4	**
Total fat	219 ^b	151 ^a	215 ^b	174 ^a	220 ^b	12.0	***
Total muscle	595 ^a	656 ^b	606 ^a	632 ^{ab}	602 ^a	10.5	***
Total bone	186	193	179	194	178	5.8	NS

Monetary value of the genetic merit of AI beef bulls - 2004

The Irish Cattle Breeding Federation (ICBF) Active Beef Bull List for 2004 contained 56 bulls available in AI that had been progeny tested in Ireland. The progeny test data are expressed as expected progeny deviations (EPD) from a common reference base. This is the performance of Holstein-Friesian steers slaughtered at 26 months of age and 350 kg carcass weight with carcass conformation and fat scores of 2.0 and 3.4, respectively. From the (EPDs) for growth and carcass traits, together with the calving survey data, monetary values were estimated. This permits a ranking of the bulls based on the monetary value of their progeny for beef production.

Estimation of monetary value

The valuation of production traits was based on “Breeding Objectives for Beef Cattle in Ireland (Amer, Simm, Keane, Diskin and Wickham, Livestock Production Science (2001) 67: 223-239)”. Briefly, the economic values used were: €1.85 per kg carcass, €30.00 for slaughter animals and €45.00 for weanlings per carcass conformation class, €-15.00 per carcass fat class, €1.40 per kg weaning weight, €3.50 per percentage unit calving difficulty (including an allowance for calf mortality) and €2.00 per day gestation length beyond 279 days. Weanling quality value was estimated as the sum of the estimated weanling weight deviation (reference base 300 kg) valued at €1.40 per kg plus carcass conformation valued at €45.00 per class. This value is mainly relevant to suckled weanlings of good conformation eligible for export to Italy and Spain. Five Belgian Blue bulls (BIG, JOK, NEZ, AVT and FPS) had no calving survey data. Missing values were estimated for these as the Belgian Blue breed means.

Breed mean values

The 56 bulls in the 2004 list comprised 10 Aberdeen Angus (AA), 9 Belgian Blue (BB), 12 Charolais (CH), 7 Hereford (HF), 10 Limousin (LM) and 8 Simmental (SM). Breed means for growth and carcass grades, together with calving survey data and estimated monetary values are shown in Table 29. Mean growth EPD was highest for CH and lowest for AA, mean carcass conformation EPD was highest for BB and lowest for HF, mean carcass fatness EPD was highest for HF and lowest for BB, mean calving difficulty percentage was highest for BB and lowest for HF and mean gestation period was longest for LM and shortest for AA. For carcass production, the mean monetary values for the breeds in descending order were CH, BB, SM, LM, HF and AA. The corresponding order for weanling production monetary value was CH, BB, LM, SM, HF and AA.

Individual bull values

The individual bull values are shown in Table 30. They are ranked within breed in descending order for carcass production value. The first column is the bull breed abbreviation and the second is the bull code. The next three columns are the growth, conformation and fatness EPDs. The next two columns are the percentage of serious calving difficulty and gestation length. The remaining columns are the monetary values. These are in order of carcass value (growth, conformation and fatness), reproduction value (calving difficulty plus gestation length), total value for carcass production (growth plus carcass grades plus calving values), weanling growth value, weanling conformation value and total value for weanling production. The top 20 bulls for carcass production comprised 11 of the 12 Charolais, 8 of the 9 Belgian Blue and one Limousin. For weanling production, the top 20 bulls were the same as for carcass production. Thus, there was little re ranking of the bulls between carcass and weanling production values. There are big differences in value between the best and poorest bulls within any breed. For carcass production the range for CH, BB, SM, LM, HF and AA was €62, €23, €32, €55, €36 and €30, respectively. Generally, the greater the number of bulls listed the greater the range in value within a breed. Because of the within breed range there was considerable overlap between breeds in individual bull values. For example, although the Limousin breed mean ranked below that of the Charolais for carcass production,

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one Limousin bull (PYR) was ranked higher than 2 Charolais bulls. Similarly, although the Hereford breed mean was lower than for the Limousin, 2 Limousin bulls (KHH and CIO) ranked lower than 5 Herefords.

Distribution of value between traits

For both carcass and weanling production, the contributions of growth, carcass grades and calving traits to total value is shown in Table 31 for the breed means. For carcass production, growth value exceeded the value of carcass grades for all breeds, but for weanling production, the value of conformation exceeded that of growth for all breeds except Charolais (a reflection of the high growth value of Charolais). For CH, BB, SM, LM, HF and AA, growth accounted for 74%, 60%, 71%, 57%, 81% and 55%, respectively, of carcass production value. In contrast, the corresponding values of growth as a proportion of weanling value (excluding calving values) were 56%, 44%, 52%, 38%, 48% and 26%.

Limitations

The purpose of genetic evaluation is to identify bulls which produce more valuable progeny. In order to do this, the breeding values for the bulls must be brought to a common denominator. For the beef producer this is the monetary value of the progeny. In choosing a bull, the monetary value should not be considered until after the individual trait EPDs have been examined. This is because a bull which may have a high monetary value could have individual traits that are unacceptable to producers such as a high incidence of calving difficulty or an excessively long gestation. While these are included in the valuation, their importance could be masked by exceptionally good values for other traits such as growth. Because the baseline for calving results is set at zero calving difficulty and 279 days gestation length, all bulls have negative valuations for calving traits. Also, because a Friesian-Holstein steer is the baseline, the values for carcass traits are inflated. This is because of the large superiority of the beef breeds over Friesian-Holsteins in carcass conformation. When the value of this carcass conformation difference is then increased to represent the value of the conformation (shape) of weanlings for the continental live export trade, this exaggerates the weanling production value of the early maturing breeds which are generally not suited to this trade. For this reason, the weanling production value should only be used for calves intended for, and suited to, the continental live export trade. Otherwise, the carcass production value should be used. In estimating the monetary valuations, linearity was assumed for carcass grades. This is not necessarily so in practice as there are thresholds and ceilings, particularly of conformation, above and below which values can change greatly. It was also assumed that the changes in individual traits had the same value for all breeds. This is not the case in practice. For example, a relatively small change in conformation which puts a carcass into a different market outlet could have a large effect on value.

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Table 29: Breed mean EPDs for growth and carcass traits, calving data and monetary value of bulls for carcass and weanling production

Breed	No. Bulls	EPDs for			Calv. Data		Value (€)	
		Grow ¹	Conf. ²	Fat ³	Diff (%) ⁴	Gest (d) ⁵	Carcass ⁶	Weanling ⁷
CH	12	50.88	1.09	-0.05	4.11	284.50	102.04	84.51
BB	9	34.34	1.17	-0.43	4.59	281.78	83.59	72.35
SM	8	29.36	0.73	0.02	3.69	284.00	53.07	45.24
LM	10	21.76	0.96	-0.12	2.98	285.20	48.06	46.44
HF	7	20.54	0.59	0.60	1.91	281.86	34.31	38.79
AA	10	8.92	0.67	0.43	3.14	280.20	16.88	27.60

¹Growth (kg carcass); ²Conformation class units; ³Fat class units; ⁴Calving difficulty (%);
⁵Gestation length (days); ⁶Monetary value for carcass production; ⁷Monetary value for weanling production.

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Table 30: Growth and carcass grades EPDs, calving survey data and monetary values for carcass and weanling production of beef AI bulls

Breed	Bull Code	EPDs ¹			Calving Data ²		Monetary Values (€)					
		Growth	Conf.	Fat	Diff (%)	Gest (d)	CALV ³	CFVL ⁴	TFVL ⁵	WGVL ⁶	WCVL ⁷	TWVL ⁸
CH	CF49	68.5	1.15	0.09	5.0	285	159.88	29.50	130.38	82.20	51.75	104.45
CH	HKI	61.5	1.18	-0.19	4.0	285	152.03	26.00	126.03	73.80	53.10	100.90
CH	HWN	56.0	1.29	-0.18	7.0	283	145.00	32.50	112.50	67.20	58.05	92.75
CH	CF46	53.3	1.13	-0.11	3.6	285	134.16	24.60	109.56	63.96	50.85	90.21
CH	CF44	56.4	1.00	-0.02	3.8	285	134.64	25.30	109.34	67.68	45.00	87.38
CH	CF48	48.9	1.09	0.05	2.8	283	122.42	17.80	104.62	58.68	49.05	89.93
CH	CF43	47.5	1.12	-0.02	2.7	284	121.78	19.45	102.33	57.00	50.40	87.95
CH	CF47	49.6	1.00	-0.09	3.3	284	123.11	21.55	101.56	59.52	45.00	82.97
CH	MDO	49.1	1.16	0.00	4.7	284	125.64	26.45	99.19	58.92	52.20	84.67
CH	CF41	44.4	1.04	-0.13	4.8	286	115.29	30.80	84.49	53.28	46.80	69.28
CH	BSK	38.9	0.93	0.01	3.9	284	99.72	23.65	76.07	46.68	41.85	64.88
CH	KFC	KFC	0.93	0.00	3.7	286	95.43	26.95	68.48	43.80	41.85	58.70
MEAN		50.9	1.09	-0.05	4.1	285	127.42	25.38	102.04	61.06	48.83	84.51
BB	TIY	42.0	1.17	-0.34	6.5	281	117.90	26.75	91.15	50.40	52.65	76.30
BB	BIG	40.1	1.09	-0.32	4.5	282	111.69	21.75	89.94	48.12	49.05	75.42
BB	NRO	37.0	1.22	-0.46	5.5	281	111.95	23.25	88.70	44.40	54.90	76.05
BB	JOK	34.3	1.30	-0.52	4.5	282	110.26	21.75	88.51	41.16	58.50	77.91
BB	VDC	34.6	1.08	-0.69	4.5	282	106.76	21.75	85.01	41.52	48.60	68.37
BB	GUY	27.8	1.25	-0.45	2.3	282	95.68	14.05	81.63	33.36	56.25	75.56
BB	AVT	30.9	1.26	-0.47	4.5	282	102.02	21.75	80.27	37.08	56.70	72.03
BB	NEZ	31.6	1.19	-0.44	4.5	282	100.76	21.75	79.01	37.92	53.55	69.72
BB	FPS	30.8	0.99	-0.21	4.5	282	89.83	21.75	68.08	36.96	44.55	59.76
MEAN		34.3	1.17	-0.43	4.6	282	105.20	21.62	83.59	41.21	52.75	72.35
SM	HRG	36.3	0.82	-0.12	3.8	283	93.56	21.30	72.26	43.56	36.90	59.16
SM	BKY	36.3	0.79	0.21	5.0	284	87.71	27.50	60.21	58.71	35.55	66.76
SM	SCO	30.3	0.82	-0.05	3.5	284	81.41	22.25	59.16	36.36	36.90	51.01
SM	FBX	29.5	0.83	0.11	4.0	284	77.83	24.00	53.83	35.40	37.35	48.75
SM	SKB	30.9	0.82	-0.05	6.2	285	82.52	33.70	48.82	37.08	36.90	40.28
SM	DFI	23.5	0.61	0.08	1.1	284	60.58	13.85	46.73	28.20	27.45	41.80
SM	HWB	26.4	0.50	0.02	2.5	285	63.54	20.75	42.79	31.68	22.50	33.43
SM	DBO	21.7	0.66	-0.05	3.4	283	60.70	19.90	40.80	26.04	29.70	35.84
MEAN		29.4	0.73	0.02	3.7	284	75.98	22.91	53.07	35.24	32.91	45.24
LM	PYR	32.6	1.16	0.03	0.6	286	94.66	16.10	78.56	39.12	52.20	75.22
LM	DAD	28.2	1.07	-0.24	4.0	285	87.87	26.00	61.87	33.84	48.15	55.99
LM	DWB	30.5	0.94	-0.19	4.3	285	87.48	27.05	60.43	36.60	42.30	51.85
LM	BHH	21.0	1.09	-0.13	1.6	284	73.50	15.60	57.90	25.20	49.05	58.65
LM	FL17	22.3	0.99	-0.08	1.7	285	72.16	17.95	54.21	26.76	44.55	53.36
LM	BHO	20.8	0.92	-0.02	2.5	284	66.38	18.75	47.63	24.96	41.40	47.61
LM	DGA	20.1	1.08	-0.07	3.1	287	70.64	26.85	43.79	24.12	48.60	45.87
LM	BHY	13.4	0.78	-0.34	3.6	285	53.29	24.60	28.69	16.08	35.10	26.58
LM	KHH	11.8	0.85	-0.06	3.5	285	48.23	24.25	23.98	14.16	38.25	28.16
LM	CIO	16.9	0.71	-0.14	4.9	286	54.67	31.15	23.52	20.28	31.95	21.08
MEAN		21.8	0.96	-0.12	3.0	285	70.89	22.83	48.06	26.11	43.16	46.44

Table 30. (contd.) Growth and carcass grades EPDs, calving survey data and monetary values for carcass and weanling production of beef AI bulls

Breed	Bull Code	EPDs ¹			Calving Data ²		Monetary Values (€)					
		Growth	Conf.	Fat	Diff (%)	Gest (d)	CALV ³	CFVL ⁴	TFVL ⁵	WGVL ⁶	WCVL ⁷	TWVL ⁸
HF	CKT	26.6	0.61	0.79	0.4	281	55.66	5.40	50.26	31.92	27.45	53.97
HF	RCE	23.8	0.61	0.65	0.3	283	52.58	9.05	43.53	28.56	27.45	46.96
HF	CFB	16.4	0.61	0.58	0.4	281	39.94	5.40	34.54	19.68	27.45	41.73
HF	GDS	26.2	0.52	0.57	6.1	284	55.52	31.35	24.17	31.44	23.40	23.49
HF	BWT	11.2	0.58	0.52	2.5	281	30.32	12.75	17.57	13.44	26.10	26.79
HF	CJO	11.3	0.60	0.62	1.8	282	29.61	12.30	17.31	13.56	27.00	28.26
MEAN		20.5	0.59	0.60	1.9	282	46.73	12.41	34.31	24.65	26.55	38.79
AA	RHD	14.7	0.89	0.46	3.3	281	47.00	15.55	31.45	17.64	40.05	42.14
AA	MTL	7.9	0.79	0.55	1.0	279	30.07	3.50	26.57	9.48	35.55	41.53
AA	LJP	13.1	0.62	0.50	2.6	279	35.34	9.10	26.24	15.72	27.90	34.52
AA	KJE	19.2	0.69	0.36	6.3	281	50.82	26.05	24.77	23.04	31.05	28.04
AA	DVE	8.5	0.68	0.39	1.7	280	30.28	7.95	22.33	10.20	30.60	32.85
AA	BJP	9.2	0.74	0.48	3.5	279	32.02	12.25	19.77	11.04	33.30	32.09
AA	RUH	3.3	0.64	0.55	1.3	281	17.06	8.55	8.51	3.96	28.80	24.21
AA	PBO	-0.3	0.57	0.29	1.6	280	12.20	7.60	4.60	-0.36	25.65	17.69
AA	LHL	1.0	0.50	0.34	1.3	281	11.75	8.55	3.20	1.20	22.50	15.15
AA	RHG	12.6	0.61	0.36	8.8	281	36.21	34.80	1.41	15.12	27.45	7.77
MEAN		8.9	0.67	0.43	3.1	280	30.27	13.39	16.88	10.70	30.29	27.60

¹Growth, carcass conformation and carcass fatness EPDs; ²Incidence of serious calving difficulty (%) and gestation length (days); ³Carcass value (combined growth, conformation and fatness values); ⁴Calving traits value (calving difficulty and gestation length); ⁵Total net value for carcass production; ⁶Weanling growth value; ⁷Weanling conformation value; ⁸Total net value for weanling production (weanling growth value plus weanling conformation value minus reproduction value).

Table 31: Distribution of breed mean bull value (€) between growth, carcass and calving traits

Breed	Carcass production				Weanling production			
	Growth	Carcass	Calving	Total	Growth	Conformation	Calving	Total
CH	94	33	-25	102	61	49	-25	85
BB	64	42	-22	84	41	53	-22	72
SM	54	22	-23	53	35	33	-23	45
LM	40	31	-23	48	26	43	-23	46
HF	38	9	-12	35	25	26	-12	39
AA	16	14	-13	17	11	30	-13	28

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Performance response of bulls to concentrate supplementation at pasture

Following the decoupling of premia, profitability in beef production will depend on the costs of production and the value of the carcass. Costs of production are greatly influenced by the proportion of total gain obtained from grazed pasture relative to that obtained from indoor feeding. Thus, systems which finish cattle off pasture are likely to be more competitive than those with a long indoor finishing period.

Carcass weight (CW) is the major determinant of carcass value. Bulls grow 8-12% faster than steers and produce about 15% more CW at the same age. Their carcasses also have a higher proportion of muscle and better conformation, making them more valuable per unit weight. It would enhance the competitiveness of beef production if the superior performance of bulls could be exploited without the high indoor feeding costs normally associated with bull production systems.

The initial objective of this study was to determine the performance response of bulls to concentrate supplementation at pasture. The second objective was to compare the performance of bulls at pasture with that of bulls finished indoors.

One hundred and twenty 12-14-month-old continental-cross bulls (mean initial liveweight (LW) 405 (s.e. 29.3) kg) were assigned to six treatments. Three treatment groups were offered perennial ryegrass at a daily herbage allowance of 2.3 kg DM/100 kg LW, plus either 0, 3, or 6 kg of concentrates/head/day from 24 April to 24 November, 2004 (214 days). The fourth and fifth treatment groups were offered the same herbage allowance plus either 0 or 3 kg of concentrates/head/day from 24 April to 31 August (129 days), and then offered concentrates *ad libitum* indoors from 1 September to 24 November (85 days). The sixth treatment group was offered concentrates *ad libitum* indoors from 24 April to 24 November (214 days). Ten animals from each group were slaughtered after 129 days and the remaining 60 animals were slaughtered after 214 days.

Bulls at pasture were offered fresh herbage daily and did not have access to the previous day's allowance. The concentrate was a mixture of barley (87.0%), soyabean (6.75%), sugarcane molasses (4.75%) and minerals (1.5%), and was 83% dry matter (DM). Perennial ryegrass herbage on offer had mean digestibility (DMD) and crude protein concentrations of 778 and 163 g/kg DM, respectively.

Liveweight gain (LWG) was calculated at 21 to 28-day intervals. After slaughter, kill-out proportion was calculated as CW divided by final LW. Carcass weight gain (CWG) from 24 April to 31 August was the difference between CW on 31 August and 0.55 times initial LW. CWG from 1 September to 24 November was the difference between final CW and the CW of bulls on 31 August. Carcasses were classified for conformation (1 = worst and 5 = best conformation) using the EU Beef Carcass Classification Scheme.

From 24 April to 31 August the LWG of the bulls offered *ad libitum* concentrates indoors (1.56 kg/day) was higher ($P < 0.05$) than those at pasture (Table 32). The bulls offered 6 kg of concentrates had a higher ($P < 0.05$) LWG (1.26 kg/day) than those offered 3 kg of concentrates (1.10 kg/day). The lowest LWG was achieved by the bulls offered no concentrates at 0.80 kg/day.

CW and CWG of the bulls at pasture increased ($P < 0.05$) with increasing levels of concentrate supplementation. However, the bulls offered *ad libitum* concentrates indoors had a higher ($P < 0.05$) CW (346 kg) and CWG (114 kg) than the bulls at pasture. The carcass conformation score of the *ad libitum* concentrates group (3.6) was higher ($P < 0.05$) than those offered 0 (2.7)

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and 3 (3.0) kg of concentrates but did not differ from the bulls offered 6 kg of concentrates (3.4).

From 1 September to 24 November there was a continued response in LWG to concentrates supplementation at pasture. However, the greatest response was from the bulls initially offered 0 and 3 kg of concentrates and then moved onto *ad libitum* concentrates indoors.

The performance of bulls at pasture increased with concentrate supplementation but with diminishing returns per unit of concentrates. Offering bulls no concentrates for the first half of the season and then *ad libitum* concentrates indoors appeared to be a cost-effective option for bull finishing systems.

Table 32: Performance response of yearling bulls to concentrate supplementation at pasture compared with those on *ad libitum* concentrates indoors

	LW (kg)	LWG (kg/day)	CW (kg)	CWG (kg)	Kill-out (g/kg)	Carcass conform.
<u>24 April to 31 August</u>						
No conc.	492 _c	0.82 _{cd}	268 _c	40 _c	544 _b	2.8 _{bc}
3 kg conc.	516 _{bc}	1.04 _{bc}	293 _{bc}	71 _b	568 _a	3.0 _{ac}
6 kg conc.	559 _b	1.26 _b	311 _b	87 _b	556 _{ab}	3.4 _{ab}
0 kg \square Indoors	485 _c	0.78 _d	263 _c	39 _c	542 _b	2.5 _c
3 kg \square Indoors	549 _b	1.16 _b	309 _b	82 _b	562 _{ab}	2.9 _{bc}
Indoors	628 _a	1.56 _a	346 _a	114 _a	550 _{ab}	3.6 _a
s.e.d.	16.2	0.078	10.5	6.9	7.2	0.21
Significance	**	**	**	**	**	**
<u>1 September to 24 November</u>						
No conc.	562 _d	0.85 _d	292 _d	24 _d	520 _b	2.7 _b
3 kg conc.	599 _{cd}	0.97 _{cd}	327 _c	34 _d	545 _{ab}	3.1 _{ab}
6 kg conc.	649 _{bc}	1.30 _{bc}	358 _{bc}	47 _{cd}	552 _a	3.1 _{ab}
0 kg \square Indoors	665 _b	2.10 _a	360 _b	97 _a	542 _{ab}	3.3 _a
3 kg \square Indoors	699 _{ab}	1.85 _a	389 _{ab}	83 _{ab}	555 _a	3.5 _a
Indoors	722 _a	1.44 _b	404 _a	64 _{bc}	560 _a	3.3 _a
s.e.d.	18.8	0.121	11.2	8.7	9.0	0.20
Significance	**	**	**	**	**	**

Values with the same subscript are not significantly different ($\square = 0.05$)

** = $P < 0.01$

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Seasonal changes in grass yield and persistency

This experiment compared the yield, chemical composition and sward appearance characteristics of six cultivars of perennial ryegrass. Two of the cultivars (Aberdart and Ba11353) were bred for elevated concentrations of water soluble carbohydrates (WSC). There were five diploid cultivars and one tetraploid (Greengold).

The calendar year of 2003 was the third successive season of this study, and the experiment was conducted as planned. The six treatments were Aberdart, Ba11353, Fennema, Aberelan, Spelga and Greengold, there were six replicate blocks and there were four harvests during the season. Plots were fertilised at the appropriate times and harvested in late May, early July, mid-August and mid-October. Visual observations were made for persistence, disease, growth stage, lodging, ground cover, etc. Yields were recorded and samples stored at -18°C prior to being processed and submitted to chemical analysis. All of the chemical analyses except crude protein and fibre measurements are now completed. WSC values were determined on samples dried at 60°C using NIRS (equations from IGER). In addition to the above, samples were taken from each plot at weekly intervals for two or three weeks before the main harvest dates, and assayed (as above) for water soluble carbohydrate (WSC) concentration.

In 2003, overall differences in DM, ash and buffering capacity (Table 33) were relatively small among diploids. Aberdart and Ba11353 tended to have higher organic matter digestibility (OMD) values in the earlier cuts than Spelga. WSC concentrations among the diploids were (on average) in the order Ba11353 > Aberdart > others. The scale of the differences were relatively small - Ba11353 averaged 2.3%units WSC higher than Fennema while Aberdart averaged 1.4%units WSC higher than Fennema. Greengold (tetraploid), tended to have a lower DM (overall average: 161 vs.171 g/kg) concentration than diploids, tended to have higher OMD values compared to the diploids (overall average: 774 vs.758 g/kg), generally had similar buffering capacity (overall average: 394 vs.397 g/kg) and ash (overall average: 85 vs.87 g/kg) values to the diploids, and had a mean WSC concentration of 192 g/kgDM relative to values of 197 and 188 g/kgDM for Ba11353 and Aberdart, respectively, and of 182 g/kgDM for all diploids.

Purity was very high in all treatments (and in all plots) throughout and at the end of the third growing season (Table 34). Morphological effects of inflorescence were most evident in the order May>August>July>October. Growth stage effects were most evident in late May, with Greengold appearing the most vegetative and Spelga having the most advanced inflorescences. Lodging was not an issue at any of the harvests, for any of the six varieties. 'Disease' was evident only on grass at harvest in late May. It included discoloration of vegetation (chlorosis) due possibly to senescence found in the lower leaves of high yielding crops. The extent of bare soil at the end of the harvesting season was very low and treatment effects were not significant.

Table 33: Chemical composition of six cultivars of grass at each of four consecutive harvests during 2003 - also annual averages per cultivar

Treatment	DM g/kg	Ash g/kgDM	OMD g/kg	Buff.cap. mEq/kgDM	NIR WSC g/kgDM
Primary growth - harvested on 27 May, 2003					
Aberdart	189	70	709	491	231
Ba11353	175	77	733	478	231
Fennema	181	72	706	467	211
Aberelan	176	71	698	515	201
Spelga	185	69	692	518	207
Greengold	166	72	743	519	227
Regrowth 1 - harvested on 8 July, 2003					
Aberdart	151	95	801	386	168
Ba11353	166	90	785	366	176
Fennema	154	92	784	380	153
Aberelan	152	94	784	390	167
Spelga	142	92	771	394	165
Greengold	141	93	784	372	163
Regrowth 2 - harvested on 25 August, 2003					
Aberdart	194	91	753	344	192
Ba11353	200	88	740	348	207
Fennema	204	89	741	342	182
Aberelan	208	88	733	337	187
Spelga	198	91	732	341	180
Greengold	187	86	757	331	195
Regrowth 3 - harvested on 15 October, 2003					
Aberdart	146	95	803	361	160
Ba11353	154	92	799	373	175
Fennema	150	97	803	385	149
Aberelan	148	94	803	352	155
Spelga	149	97	801	371	146
Greengold	148	88	814	353	183
Annual averages					
Aberdart	170	88	766	395	188
Ba11353	174	87	764	391	197
Fennema	172	87	758	393	174
Aberelan	171	86	754	399	177
Spelga	168	87	749	406	176
Greengold	161	85	774	394	192

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Table 34: Sward appearance characteristics for six cultivars of grass at each of four consecutive harvests

	Purity (%)	Growth stage	Lodging (%)	Disease (%)	Bare soil (%)
May					
Aberdart	94	3.6	0	13.5	
Ba11353	96	3.2	0	10.5	
Fennema	91	3.5	0	11.0	
Aberelan	90	3.7	0	12.5	
Spelga	94	3.9	0	11.0	
Greengold	92	3.0	0	9.5	
July					
Aberdart	97	1.6	0	0	
Ba11353	97	1.5	0	0	
Fennema	96	1.6	0	0	
Aberelan	96	1.6	0	0	
Spelga	97	1.7	0	0	
Greengold	97	1.5	0	0	
August					
Aberdart	99	2.8	0	1	
Ba11353	99	2.6	0	1	
Fennema	98	2.9	0	1	
Aberelan	99	2.9	0	1	
Spelga	99	2.8	0	1	
Greengold	99	2.8	0	1	
October					
Aberdart	99	1.0	0	1	5.0
Ba11353	99	1.0	0	1	3.8
Fennema	98	1.0	0	1	4.2
Aberelan	99	1.0	0	1	4.8
Spelga	99	1.0	0	1	4.5
Greengold	99	1.0	0	1	5.3

The absolute differences between varieties in WSC concentration could change considerably between weeks (Table 35).

It is concluded that Ba11353 (+2.3% units) and Aberdart (+1.4% units) had slightly higher average WSC concentrations than Fennema (174 gWSC/kgDM) - each are diploid varieties with similar heading dates. This scale of average increase in WSC concentration associated with the 'elevated sugar' varieties (compared to Fennema) was quite modest. The comparable effects within individual harvests were generally similar to the annual average effects. Greengold (tetraploid) had average concentrations of WSC intermediate between Ba11353 and Aberdart. The absolute differences in WSC concentration between varieties could change considerably between weeks. Sward purity was maintained with all varieties while neither disease nor lodging were problems.

Table 35: Weekly concentration of WSC

Sample date	Cultivar	Growth	WSC (g/kgDM)
12-May-03	Aberdart	Primary	249
12-May-03	Aberdove	Primary	261
12-May-03	Fennema	Primary	228
12-May-03	Aberelan	Primary	236
12-May-03	Spelga	Primary	228
12-May-03	Greengold	Primary	261
19-May-03	Aberdart	Primary	186
19-May-03	Aberdove	Primary	257
19-May-03	Fennema	Primary	248
19-May-03	Aberelan	Primary	198
19-May-03	Spelga	Primary	199
19-May-03	Greengold	Primary	237
21-May-03	Aberdart	Primary	189
21-May-03	Aberdove	Primary	202
21-May-03	Fennema	Primary	203
21-May-03	Aberelan	Primary	188
21-May-03	Spelga	Primary	197
21-May-03	Greengold	Primary	217
23-Jun-03	Aberdart	Regrowth 1	151
23-Jun-03	Aberdove	Regrowth 1	161
23-Jun-03	Fennema	Regrowth 1	143
23-Jun-03	Aberelan	Regrowth 1	152
23-Jun-03	Spelga	Regrowth 1	160
23-Jun-03	Greengold	Regrowth 1	155
30-Jun-03	Aberdart	Regrowth 1	134
30-Jun-03	Aberdove	Regrowth 1	148
30-Jun-03	Fennema	Regrowth 1	138
30-Jun-03	Aberelan	Regrowth 1	141
30-Jun-03	Spelga	Regrowth 1	141
30-Jun-03	Greengold	Regrowth 1	142
11 Aug. 2003	Aberdart	Regrowth 2	183
11 Aug. 2003	Aberdove	Regrowth 2	169
11 Aug. 2003	Fennema	Regrowth 2	144
11 Aug. 2003	Aberelan	Regrowth 2	167
11 Aug. 2003	Spelga	Regrowth 2	160
11 Aug. 2003	Greengold	Regrowth 2	181
18 Aug. 2003	Aberdart	Regrowth 2	204
18 Aug. 2003	Aberdove	Regrowth 2	211
18 Aug. 2003	Fennema	Regrowth 2	184
18 Aug. 2003	Aberelan	Regrowth 2	190
18 Aug. 2003	Spelga	Regrowth 2	192
18 Aug. 2003	Greengold	Regrowth 2	202
29 Sept. 2003	Aberdart	Regrowth 3	137
29 Sept. 2003	Aberdove	Regrowth 3	144
29 Sept. 2003	Fennema	Regrowth 3	146
29 Sept. 2003	Aberelan	Regrowth 3	144
29 Sept. 2003	Spelga	Regrowth 3	158
29 Sept. 2003	Greengold	Regrowth 3	151
6 Oct. 2003	Aberdart	Regrowth 3	101
6 Oct. 2003	Aberdove	Regrowth 3	103
6 Oct. 2003	Fennema	Regrowth 3	109
6 Oct. 2003	Aberelan	Regrowth 3	113
6 Oct. 2003	Spelga	Regrowth 3	114
6 Oct. 2003	Greengold	Regrowth 3	105

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RMIS No. 5002

Water-soluble carbohydrate (WSC) concentrations in Ireland and Norway of *Lolium perenne* differing in WSC genotype and receiving varying rates of N fertiliser

Cultivars bred for elevated water-soluble carbohydrate (WSC) concentration may have improved grass ensilability and nutritive value. Increasing rates of application of N fertiliser generally reduce grass WSC concentration, although it is unknown if the response is similar for normal and elevated WSC genotypes or if these factors interact with growing conditions. This experiment evaluated the effects on grass WSC concentration of varying N fertiliser application rates to perennial ryegrass cultivars of high or normal WSC genotype grown in Ireland and Norway.

The experiment was conducted in both Ireland (Grange) and Norway (Saerheim) in two years (Y). It was a split-plot randomised complete block design containing four replicates; replicates consisted of three or four main plots providing for successive harvests (early June (H1), early Aug. (H2) and late Sept.(H3) in Norway and late May (H1), early July (H2), mid-Aug. (H3) and early Oct. (H4) in Ireland). Within main plots, 2 cultivars x 4 (Norway) or 5 (Ireland) rates of inorganic N fertiliser (N_r) were fully randomised. The two diploid intermediate heading perennial ryegrass cultivars (Aberdart: selected for elevated WSC concentration and Fennema: control) were sown as monoculture plots. The rates of N fertiliser (calcium ammonium nitrate (CAN); 275 g N/kg) were equivalent to 0 (N_0), 40 (N_{40}), 80 (N_{80}), 120 (N_{120}) and 160 (N_{160} - Ireland only) kg N/ha. The remaining sub-plots received an application of CAN equivalent to 80 kg N/ha. All plots were harvested at each harvest period but only herbage from the main plots that recently received N_r were sampled.

Grass WSC concentrations are given in Table 36. Aberdart > Fennema in Ireland (169 vs. 158 g/kg DM) and Norway (263 vs. 235 g/kg DM) and Year 1 > Year 2 at both sites. In Ireland, H1 had the highest values while in Norway values were highest for H1 and H2. Values decreased progressively from N_0 to N_{120} in Ireland and N_{40} to N_{120} in Norway. The differences between cultivars in response to N_r were maintained across sites, harvests and years - there were not significant $C \times N_r$, $H \times C \times N_r$, $C \times N_r \times Y$ (except Y1 N_{80} in Norway) or $H \times C \times N_r \times Y$ interactions.

Table 36: Grass WSC concentration (g/kg DM) in Aberdart and Fennema swards at each harvest (H)

Cultivar (C) N rate (N _r)	Aberdart					Fennema					Statistical summary		
	N ₀	N ₄₀	N ₈₀	N ₁₂₀	N ₁₆₀	N ₀	N ₄₀	N ₈₀	N ₁₂₀	N ₁₆₀	Sig.	s.e.	
Ireland - Y1													
H1	242	214	191	164	164	242	207	179	164	153	H	***	3.2
H2	240	201	171	147	128	233	168	174	136	132	N _r	***	3.0
H3	217	185	149	138	127	185	164	141	122	122	C	***	1.9
H4	202	162	148	137	135	181	154	134	116	123	Y	**	2.0
Ireland - Y2													
H1	275	247	209	188	169	238	235	196	175	156	HxN _r	***	6.2
H2	173	168	132	106	106	185	116	111	110	92	HxC	ns	4.2
H3	175	161	155	149	157	153	156	162	143	143	CxN _r	ns	4.2
H4	180	145	140	132	137	160	141	134	132	136	HxCxN _r	ns	8.7
Norway - Y1													
H1	350	364	308	241	-	319	331	300	221	-	H	***	3.8
H2	311	302	248	226	-	288	267	233	197	-	N _r	***	2.8
H3	291	295	230	206	-	246	266	223	204	-	C	***	2.0
											Y	***	2.0
Norway - Y2													
H1	319	269	216	215	-	313	239	174	182	-	HxN _r	***	5.6
H2	304	311	284	267	-	274	271	221	206	-	HxC	NS	4.5
H3	197	209	193	166	-	170	194	161	153	-	CxN _r	NS	3.9
											HxCxN _r	NS	7.4

It is concluded that grass WSC was higher and the difference between cultivars larger in Norway than Ireland. The negative effects of N fertiliser on WSC were similar for the two grasses across a range of conditions.

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RMIS No. 5002

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Perennial ryegrasses bred for normal or elevated sugar contents: estimated ensilability and nutritive value at different levels of N fertiliser input

Nitrogen fertiliser is applied to grassland managed for silage production to promote economically justifiable yields. This has the side effect of making grass more difficult to preserve successfully as silage. It is reflected in a reduction in dry matter (DM) and water soluble carbohydrate (WSC) concentrations, and an increase in buffering capacity (BC). Occasionally, it can also reduce *in vitro* DM digestibility (DMD). Grasses with elevated concentrations of WSC should be easier to preserve as silage and, if this WSC is protected during ensilage and feedout, to improve animal productivity and reduce urinary losses of N. This experiment determined the effects of N fertiliser application rate on the estimated ensilability and nutritive value of perennial ryegrass (*Lolium perenne*) cultivars of high or normal WSC genotype.

A split-plot randomised complete block design experiment was conducted in the first and third years following sowing in September 2000. Two diploid intermediate perennial ryegrass cultivars

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(Aberdart: selected for high WSC concentration; Fennema: control) were sown as monoculture plots. Each of the four replicates consisted of four main plots providing for successive harvests (21 May (H1), 2 July (H2), 20 August (H3) and 8 October (H4) 2001; 22 May (H1), 7 July (H2), 20 August (H3) and 14 October (H4) 2003). Within main plots, 2 cultivars x 5 rates of inorganic N fertiliser (N_r) were fully randomised. The N_r (calcium ammonium nitrate; 275 g N/kg) were equivalent to 0 (N_0), 40 (N_{40}), 80 (N_{80}), 120 (N_{120}) and 160 (N_{160}) kg N/ha and were applied to sub-plots within H1 in mid-March and within H2, H3 and H4 immediately after harvesting H1, H2 and H3, respectively. Main plots received 80 kg N/ha when not receiving N_r . All plots were harvested at each harvest date but only herbage from within main plots fertilised with N_r were sampled and weighed. Data were analysed as a split-split-plot design. Main plot, sub-plot and sub-sub plot factors were harvest, grass and N_r , and year, respectively.

Total DM yields (only recorded in 2001) of 14.39 and 14.70 t/ha for Aberdart and Fennema, respectively, were not different ($P>0.05$) and sward botanical composition appeared similar in 2001 and 2003. In comparison with Fennema, Aberdart had a lower BC (350 vs. 358 (s.e. 2.0; $P<0.01$) mEq/kgDM), pH (5.86 vs. 5.94 (s.e. 0.014; $P<0.001$) and crude protein (CP) (178 vs. 181 (s.e. 1.2; $P<0.05$) g/kgDM), a higher WSC (169 vs. 158 (s.e. 1.9; $P<0.001$) g/kgDM) and ash (106 vs. 104 (s.e. 0.6; $P<0.05$) g/kgDM) but did not differ ($P>0.05$) in DMD (796 vs. 797 (s.e. 1.1) g/kg) or DM (165 vs. 165 (s.e. 0.9) g/kg). Higher N_r increased ($P<0.001$) CP, ash, pH and BC, and decreased ($P<0.001$) DM, WSC and DMD (Table 37). Year influenced the variables measured, but there were no cultivar x N_r or cultivar x N_r x year interactions ($P>0.05$) for any of the variables measured.

Table 37: Composition of Aberdart and Fennema at increasing rates of N fertiliser application (across years & harvests)

		DM g/kg	CP g/kgDM	DMD g/kg	Ash g/kgDM	WSC g/kgDM	BC mEq/kgDM	pH
N_0	Aberdart	198	132	808	103	213	292	5.65
	Fennema	199	133	810	101	197	302	5.69
N_{40}	Aberdart	169	153	792	105	185	330	5.83
	Fennema	172	159	794	104	167	340	5.91
N_{80}	Aberdart	158	180	796	107	162	359	5.87
	Fennema	159	184	798	104	154	372	6.02
N_{120}	Aberdart	150	206	794	108	145	385	5.99
	Fennema	149	209	790	107	137	389	6.05
N_{160}	Aberdart	148	218	790	108	140	382	5.97
	Fennema	147	221	791	106	132	384	6.02
Cultivar (C)	s.e.	0.9	1.2	1.1	0.6	1.9	2.0	0.014
	Sig.	ns	*	ns	*	***	**	***
Rate of N (N_r)	s.e.	1.5	1.8	1.7	1.0	3.0	3.1	0.021
	Sig.	***	***	***	***	***	***	***
C x N_r	s.e.	2.1	2.6	2.5	1.4	4.2	4.4	0.030
	Sig.	NS	NS	NS	NS	NS	NS	NS

In conclusion, the effects of N fertiliser on estimated ensilability and nutritive value were similar for both grasses and followed expected response patterns. The small absolute differences between Aberdart and Fennema in WSC, DM and BC indicate that differences in predicted ensilability were negligible. Similarly, DM, CP and DMD values show that the nutritive value of both grasses was similar.

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Perennial ryegrasses bred for contrasting sugar contents: manipulating fermentation and aerobic stability using wilting and additives (1)

Higher concentrations of water-soluble carbohydrate (WSC) in silage offer ruminant nutrition and environmental attractions. Both successful field wilting and alternative silage additives provide the opportunity to manipulate silage WSC by modifying fermentation and/or improving aerobic stability. This experiment evaluated the fermentation and aerobic stability of silages made from perennial ryegrass cultivars of high or normal WSC genotype that differed in field wilting or additive use.

Aberdart (Ab; bred for high WSC) and Fennema (Fn; normal WSC) perennial ryegrasses were mown on 19 Sept. 2002. Each was precision-chopped and ensiled in laboratory silos (6kg) after a 0 or 24h wilt. The additives applied to grass for three silos per treatment were (1) no additive, (2) Add-SafeR (85% ammonium tetraformate salt; Trouw Nutrition UK Ltd.) at 6ml/kg, (3) *Lactobacillus buchneri*, *L. plantarum* and *Enterococcus faecium* (Pioneer Hi-Bred) at 3ml/kg, (4) Powerstart (*L. plantarum* and *Lactococcus lactis*; Genus plc) at 3ml/kg, (5) and (6) Kofasil Ultra (80g hexamine, 120g sodium nitrite, 150g sodium benzoate, 50g sodium propionate and 600g water/kg; Addcon Agrar GmbH) at 2.5 or 5 ml/kg, (7) treatments 4 + 5, and (8) treatments 4 + 6. Silos were filled, sealed and stored (15°C) for > 100 days. Silage composition (N=3/treatment) and aerobic stability (N=2/treatment) measurements were made and the results subjected to 3-way analysis of variance.

Mean (s.d.) grass WSC and buffering capacity for unwilted and wilted Ab were 172 (6.1) and 178 (11.0) g/kg dry matter (DM) and 374 (22.8) and 364 (23.1) mEq/kgDM, respectively, with corresponding values for Fn of 158 (11.8) and 186 (5.7) g/kgDM and 379 (7.2) and 386 (7.6) mEq/kgDM. Unwilted and wilted silage DM values were 152 and 199 (s.e. 0.5; P<0.001) g/kg, respectively, and cultivar had no significant (P>0.05) effect. Wilting increased lactic acid/fermentation products (Table 38). Fn had a more lactic acid fermentation than Ab. Formic acid promoted the dominance of lactic acid in the unwilted silages and restricted fermentation in the wilted silages (reduced fermentation products; P<0.001). Except for unwilted Ab, the *L.buchneri* additive reduced (P<0.001) lactic acid and increased (P<0.05) acetic acid and ethanol. Powerstart increased lactic acid/fermentation products. Kofasil Ultra promoted a more lactic acid fermentation with unwilted Ab but had minor effect with wilted herbage. Little additivity occurred when Powerstart and Kofasil Ultra were co-applied. Unwilted silages were very stable when exposed to air. Powerstart increased susceptibility to aerobic deterioration while Add-SafeR, *L.buchneri* and Kofasil Ultra (high) improved stability with wilted silages.

Table 38: Chemical composition and aerobic stability of silages

Additive (A)	1		2		3		4		5		6		7		8		Significance				
Cultivar (C)	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	sem	C	A	CxA	
pH	U ⁴	4.6	4.2	3.7	3.8	4.5	4.5	3.8	4.0	4.4	4.6	4.1	4.2	3.8	4.1	4.0	4.1	0.05	0.08	***	***
	W ⁵	4.2	4.0	4.0	4.1	4.5	4.3	3.8	3.9	4.1	4.1	4.2	4.2	3.9	4.0	4.1	4.2				
Lactic acid (g/kg FP ¹)	U	243	605	755	760	278	75	762	737	382	374	547	537	819	686	854	837	19.0	**	***	***
	W	594	771	667	700	291	512	825	825	646	730	559	636	832	829	843	783				
NH ₃ N (g/kg N)	U	88	80	100	107	84	143	60	74	94	111	88	96	59	85	67	72	2.8	**	***	***
	W	109	76	105	99	106	78	51	58	93	78	90	83	60	70	67	70				
Butyric acid (g/kg DM)	U	0	0	0	0	0.5	0.5	1.0	0	0	1.9	10.0	0	1.4	0	0	1.07	**	***	0.06	
	W	10.7	0.5	1.1	0	11.8	3.6	1.0	0	5.5	0	8.1	0	0.4	0	0	1.1				
Duration to temp. rise ²	U	192	192	192	192	192	192	56	135	192	186	192	192	43	43	57	57	2.6	***	***	***
	W	96	88	192	192	192	55	61	192	109	192	192	66	81	192	192					
ATR to d5 ³	U	3	1	2	1	3	2	30	1	2	2	2	2	33	1	17	1	1.3	***	***	***
	W	12	11	2	1	3	3	46	25	1	6	2	1	26	11	2	1				

¹FP=fermentation products (lactic+VFA+ethanol); ²hours; ³accumulated temp. rise to day 5 (⁰C); ⁴unwilt; ⁵wilt; sem = CxA

In conclusion, cultivar had minor effects on ensilability indices, but Fn silages were better preserved. The partial wilt generally promoted a more efficient fermentation but poorer aerobic stability. The most consistent improvement in dominance by lactic acid was from Add-SafeR and Powerstart, but Powerstart silages were prone to aerobic deterioration. Add-SafeR, *L.buchneri* and Kofasil Ultra (high) improved aerobic stability with wilted silages.

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Perennial ryegrasses bred for contrasting sugar contents: manipulating fermentation and aerobic stability of unwilted silage using additives (2)

Grass cultivars bred for elevated concentrations of water-soluble carbohydrate (WSC) could have improved silage preservation but possibly disimproved aerobic stability. Additives can be used to manipulate fermentation and thereby increase silage WSC. They can also influence aerobic stability. This experiment evaluated the fermentation and aerobic stability of unwilted silages made from perennial ryegrass cultivars of high or normal WSC genotype that differed in additive use.

Aberdart (Ab; bred for high WSC) and Fennema (Fn; normal WSC) perennial ryegrasses were mown on 17 June, 2003. Each was precision-chopped and ensiled in laboratory silos (6kg/silo) without wilting. The additives applied to grass, using three silos per treatment, were (1) no additive, (2) and (3) Add-SafeR (85% ammonium tetraformate salt; Trouw Nutrition UK Ltd.) at 3 and 6ml/kg, (4) Biomax SI (*Lactobacillus plantarum*; Chr. Hansen UK Ltd.) at 5ml/kg, (5) Biomax SI at 5 ml/kg + potassium sorbate (KSor; 30g/l) at 5ml/kg, (6) Biomax SI at 5 ml/kg + sodium benzoate (NaBe; 30g/l) at 5ml/kg, and (7), (8) and (9) Bio-Sil (*Lactobacillus plantarum*; Dr. Pieper Technologie- und Produktentwicklung GmbH) at 5 ml/kg alone or with KSor or NaBe at 5ml/kg, (10) KSor at 5ml/kg, and (11) NaBe at 5ml/kg. Silos were filled, sealed and stored (15⁰C) for > 100 days. Silage composition and aerobic stability measurements were made on every silage and the results subjected to 2-way analysis of variance.

Mean (s.d.) grass dry matter (DM), WSC and buffering capacity for unwilted Ab were 143 (12.6) g/kg, 180 (4.8) g/kgDM and 226 (19.7) mEq/kgDM, respectively, with corresponding values for Fn of 141 (12.8) g/kg, 154 (11.6) g/kgDM and 242 (24.4) mEq/kgDM. Lactic acid bacteria on Ab and Fn at harvesting were 6.1 and 6.2 log₁₀ colony forming units/g, respectively. All silages were well preserved. Ab silages had lower NH₃-N (68 vs. 77 g/kgN) and lactic acid/fermentation products (616 vs. 702 g/kg) values and a higher accumulated temperature rise to day 5 (ATR; 27 vs. 23°C) than Fn silages (Table 39). Incremental additions of Add-SafeR restricted (P<0.05) fermentation, improved (P<0.05) aerobic stability (i.e. duration to temp. rise) and reduced (P<0.05) aerobic deterioration (i.e. ATR). Neither of the bacterial inoculants and neither of the salts (KSor or NaBe) altered (P>0.05) fermentation, aerobic stability or aerobic deterioration indices.

Table 39: Chemical composition and aerobic stability of unwilted silages

Additive (A)	pH		Lactic acid g/kg FP ¹		NH ₃ -N g/kgN		Hours to temp. rise		ATR to day 5 ²	
	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn
No additive	3.73	3.83	636	732	59	66	28	25	34	27
Add-SafeR low	3.80	3.87	633	653	104	105	69	79	24	19
Add-SafeR high	4.20	3.97	379	527	146	140	94	128	14	9
Biomax SI	3.70	3.87	683	686	58	72	51	25	25	27
Biomax SI + KSor	3.97	3.93	491	664	52	69	34	22	29	26
Biomax SI + NaBe	3.80	3.90	594	690	56	64	27	34	31	22
Bio-Sil	3.73	3.80	680	739	53	76	22	38	29	24
Bio-Sil + KSor	3.67	3.73	720	764	62	62	32	32	31	24
Bio-Sil + NaBe	3.67	3.80	702	759	53	61	25	29	30	24
KSor	3.70	3.83	662	740	53	67	24	25	26	28
NaBe	3.77	3.73	597	772	51	65	24	21	27	29
s.e.m. (CxA)	0.072		34.0		8.2		14.7		4.9	
Sig. C (cultivar)	NS		***		*		NS		P=0.076	
A	***		***		***		***		P=0.051	
CxA	NS		NS		NS		NS		NS	

¹FP=fermentation products (lactic+VFA+ethanol); ²accumulated temp. rise to day 5

In conclusion, the higher WSC and lower buffering capacity for Ab compared to Fn indicate that Ab had better ensilability indices. The higher lactic acid/fermentation products for Fn silage reflects its higher concentration of lactic acid and lower concentration of both acetic acid and ethanol. The formic acid-based additive had the largest impact on fermentation and was the only additive to consistently and significantly improve aerobic stability and reduce aerobic deterioration.

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Perennial ryegrasses bred for contrasting sugar contents: manipulating fermentation and aerobic stability of wilted silage using additives (3)

Rapid field-drying of grass prior to successful ensilage restricts fermentation and can assist preservation, but can consequently result in silages that are prone to aerobic deterioration at feedout. Additives that directly (e.g. potassium sorbate or sodium benzoate) or indirectly (e.g. formic acid or *Lactobacillus plantarum*, via manipulation of fermentation) alter yeast activity at feedout could modify silage aerobic stability. This experiment evaluated the fermentation and aerobic stability of wilted silages made from perennial ryegrass cultivars of high or normal water soluble carbohydrate (WSC) genotype that differed in additive use. Aberdart (Ab; bred for high WSC) and Fennema (Fn; normal WSC) perennial ryegrasses were mown on 17 June, 2003, and field dried for 24 h. Each was then precision-chopped and ensiled in laboratory silos (5kg/silo). The additives applied to grass, using three silos per treatment, were (1) no additive, (2) and (3) Add-SafeR (85% ammonium tetraformate salt; Trouw Nutrition UK Ltd.) at 3 and 6ml/kg, (4) Biomax SI (*Lactobacillus plantarum*; Chr. Hansen UK Ltd.) at 5ml/kg, (5) Biomax SI at 5 ml/kg + potassium sorbate (KSor; 30g/l) at 5ml/kg, (6) Biomax SI at 5 ml/kg + sodium benzoate (NaBe; 30g/l) at 5ml/kg, and (7), (8) and (9) Bio-Sil (*Lactobacillus plantarum*; Dr. Pieper Technologie- und Produktentwicklung GmbH) at 5 ml/kg alone or with KSor or NaBe at 5ml/kg, (10) KSor at 5ml/kg, and (11) NaBe at 5ml/kg. Silos were filled, sealed and stored (15°C) for > 100 days. Silage composition and aerobic stability measurements were made on every silage and the results subjected to 2-way analysis of variance.

Mean (s.d.) grass dry matter (DM), WSC and buffering capacity for wilted Ab were 372 (26.4) g/kg, 165 (4.8) g/kgDM and 208 (11.0) mEq/kgDM, respectively, with corresponding values for Fn of 383 (27.4) g/kg, 144 (4.8) g/kgDM and 235 (16.0) mEq/kgDM. Lactic acid bacteria on Ab and Fn at harvesting were 7.1 and 7.2 log₁₀ colony forming units/g, respectively. All silages were well preserved and were aerobically very stable. Cultivar had relatively little effect on the variables measured (Table 40), although Ab resulted in lower (P<0.05) lactic acid/fermentation products (653 vs. 678 g/kg) and duration to temperature rise (136 vs. 158 h). Add-SafeR increased final pH and NH₃-N values. Even though it decreased the content of lactic and acetic acids it increased ethanol content. Treatments containing Bio-Sil increased ethanol and reduced acetic acid and the duration to temperature rise. Neither of the bacterial inoculants and neither of the salts altered pH, total fermentation products or the content of lactic acid.

Table 40: Chemical composition and aerobic stability of wilted silages

Additive (A)	pH		Lactic acid g/kg FP ¹		NH ₃ -N g/kgN		Hours to temp. rise		ATR to day 5 ²	
	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn	Ab	Fn
No additive	4.00	4.03	669	702	52	61	139	165	5	2
Add-SafeR low	4.10	4.13	613	623	75	88	192	158	2	4
Add-SafeR high	4.23	4.27	459	466	96	80	192	192	3	2
Biomax SI	4.00	4.00	696	716	59	66	110	145	5	4
Biomax SI + KSor	4.03	4.00	672	733	65	89	107	192	5	2
Biomax SI + NaBe	4.00	4.00	702	695	59	58	129	176	5	3
Bio-Sil	4.00	4.00	665	690	50	54	82	109	9	8
Bio-Sil + KSor	4.03	4.00	645	729	52	63	103	159	6	1
Bio-Sil + NaBe	4.00	4.00	673	710	51	58	78	133	12	4
KSor	4.00	4.00	719	705	63	60	171	124	2	8
NaBe	4.00	4.03	666	692	59	56	192	187	2	1
s.e.m. (CxA)	0.019		23.0		8.1		19.8		2.1	
Sig. C (cultivar)	NS		*		NS		*		P=0.089	
A	***		***		***		***		*	
CxA	NS		NS		NS		*		NS	

In conclusion, the higher WSC and lower buffering capacity of Ab at harvesting gave it an apparent ensilability advantage over Fn. However, preservation was quite similar for both cultivars. The high rate of the formic acid-containing additive had the largest effect on fermentation and improving aerobic stability. The rates of inclusion of sorbate or benzoate salts did not improve aerobic stability under the test conditions prevailing.

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Perennial ryegrasses bred for contrasting sugar contents: manipulating fermentation and aerobic stability of unwilted and wilted silages using additives (4)

Aberdart (Ab; bred for high WSC) and Fennema (Fn; normal WSC) perennial ryegrasses were mown on 22 July, 2003. Each was then precision-chopped without wilting or after 24h field drying, and ensiled in laboratory silos (6 kg unwilted grass or 5kg wilted grass/silo). The additives applied to grass, using three silos per treatment, were (1) no additive, (2) and (3) Add-SafeR (85% ammonium tetraformate salt; Trouw Nutrition UK Ltd.) at 3 and 6ml/kg, (4) Biomax SI (*Lactobacillus plantarum*; Chr. Hansen UK Ltd.) at 5ml/kg, (5) Biomax SI at 5 ml/kg + potassium sorbate (KSor; 30g/l) at 5ml/kg, (6) Biomax SI at 5 ml/kg + sodium benzoate (NaBe; 30g/l) at 5ml/kg, and (7), (8) and (9) Bio-Sil (*Lactobacillus plantarum*; Dr. Pieper Technologie- und Produktentwicklung GmbH) at 5 ml/kg alone or with KSor or NaBe at 5ml/kg, (10) KSor at 5ml/kg, and (11) NaBe at 5ml/kg. Silos were filled, sealed and stored (15⁰C) for > 100 days. Silage composition and aerobic stability measurements were made on every silage and the results subjected to 2-way analysis of variance.

This experiment was partially funded by the European Union under the Fifth Framework Programme(QLK5-CT-2001-04980).

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Enumeration of yeast on unwilted and wilted grass and on the resultant silages made using additives with contrasting modes of action

Both first- and second-cut perennial ryegrasses (monocultures of Aberdart or Fennema) were precision-chopped without wilting or after 24h field drying, and ensiled in laboratory silos (6 kg unwilted grass or 5kg wilted grass/silo). The additives applied to grass, using three silos per treatment, were (1) no additive, (2) and (3) Add-SafeR (85% ammonium tetraformate salt; Trouw Nutrition UK Ltd.) at 3 and 6ml/kg, (4) Biomax SI (*Lactobacillus plantarum*; Chr. Hansen UK Ltd.) at 5ml/kg, (5) Biomax SI at 5 ml/kg + potassium sorbate (KSor; 30g/l) at 5ml/kg, (6) Biomax SI at 5 ml/kg + sodium benzoate (NaBe; 30g/l) at 5ml/kg, and (7), (8) and (9) Bio-Sil (*Lactobacillus plantarum*; Dr. Pieper Technologie- und Produktentwicklung GmbH) at 5 ml/kg alone or with KSor or NaBe at 5ml/kg, (10) KSor at 5ml/kg, and (11) NaBe at 5ml/kg. Silos were filled, sealed and stored (15⁰C) for > 100 days.

The mean number of colony-forming units (cfu) of yeast on precision-chop harvested Aberdart and Fennema were log₁₀ cfu/g 3.07 and 2.94 (s.e. 0.109; P>0.05) for first-cut grass, respectively,

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and correspondingly 0.83 and 0.61 (s.e. 0.151; $P>0.05$) for second-cut grass. The mean number of yeast on unwilted and wilted grass were \log_{10} cfu/g 3.38 and 2.18 (s.e. 0.109; $P<0.001$) for first-cut grass, respectively, and correspondingly 1.15 and 0.29 (s.e. 0.151; $P<0.001$) for second-cut grass. There was not a significant ($P>0.05$) interaction between grass cultivar and wilting. Yeast counts were higher on silage than on the parent grass (Table 41). In general, the formic acid-based additive was the most effective at reducing yeast counts, and would thus be expected to improve the aerobic stability of silage during feedout.

Table 41: Yeast counts on first- and second-cut silages made using contrasting additive treatments

	Additive										s.e.	Sig	
	None	Add-SafeR		Inoculant A		Inoculant B			Sorbate	Benzoate			
		Low	High	Alone	+	+	Alone	+	+				
Cut 1	3.69	0.38	0	4.32	3.11	3.29	3.63	4.12	3.99	4.03	2.50	0.256	**
Cut 2	1.44	0.31	0.45	0.59	0.91	1.15	1.14	0.98	0.39	0	0.56	0.355	NS

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Perennial ryegrasses bred for contrasting sugar contents: manipulating fermentation and aerobic stability of unwilted and wilted silages using additives (5)

An experiment was commenced in May 2004 to test the following null hypotheses: (1) grass bred for elevated WSC content will no more readily undergo a lactic acid dominant primary fermentation or risk producing higher concentrations of ethanol than the grass of normal WSC content, (2) unwilted high WSC grass will not require specific additive treatment to retain a high concentration of residual WSC, and (3) silage made from grass of elevated WSC content will not be inherently more unstable when exposed to air than comparable silage made with normal WSC grass. Furthermore, aerobic deterioration will not reduce its elevated WSC content, and thus will not necessitate specific additive treatment to enhance aerobic stability.

Aberdart (Ab; bred for high WSC) and Fennema (Fn; normal WSC) perennial ryegrasses were mown in May 2004. Each was then precision-chopped without wilting or after 24h field drying, and ensiled in laboratory silos (6 kg unwilted grass or 5kg wilted grass/silo). The additives applied to grass, using three silos per treatment, were (1) no additive, (2) Add SafeR (formic acid based) at 3 l/t, (3) lactic acid bacterial (LAB) inoculant alone, (4) LAB inoculant plus sodium benzoate at 200g/t, (5) LAB inoculant plus sodium benzoate at 400 g/t and (6) LAB inoculant plus sodium benzoate at 800 g/t. The silos were opened after over 100 days storage. The silages were subjected to aerobic stability assessment and are being subjected to chemical analyses.

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Investigating the effects of elevated sugar contents in grass for beef cattle

The aim of this experiment was to quantify the effects of varying the levels of WSC (water-soluble carbohydrate) in perennial ryegrass on intake, growth, digestibility, estimated microbial protein production and nitrogen excretion of growing cattle. It was a proof-of-concept study using protein-responsive animals, aimed at mimicking the sugar concentrations that could occur if comparable grasses (i.e. same species with similar heading date) of very contrasting sugar concentration existed and were used optimally. The treatments compared were 1) grass alone, 2) grass + 3% sugar, 3) grass + 6% sugar, 4) grass + 9% sugar and 5) grass +12% sugar. Seventy-five continental crossbred steers were used in a randomised complete block design experiment. Grass was direct-cut with a single-chop flail harvester, mixed in a feeder wagon and had 0, 3, 6, 9 or 12% sugar added (on a dry matter basis). Treated grass was then individually offered *ad libitum* through Calan doors to the steers. Intakes, liveweight gain, digestibility, urinary losses and estimated microbial protein production were measured.

The zero-grazed grass offered had a mean composition of: dry matter (DM) 203 (30.9) g/kg, *in-vitro* DMD 687 (50.2) g/kg, *in-vitro* OMD 709 (45.9) g/kg, ash 148 (36.1) g/kgDM, crude protein 160 (27.4) g/kgDM, neutral detergent fibre (NDF) 495 (19.1) g/kgDM, acid detergent fibre (ADF) 259 (17.3) g/kgDM and WSC 148 (24.8) g/kgDM. Cattle fed zero-grazed grass with incremental levels of supplemental WSC had an increase in intake to the first increment (30g/kg grass DM) of WSC, but not thereafter (Table 42). Orthogonal contrasts showed a positive (P=0.007) linear relationship between DMI (kg/d) and level of added WSC. A positive linear trend (P=0.002) existed between increasing WSC addition (X) (g/kg grass DM) and DMI (Y) (g/kg bodyweight) and this relationship is best described by the equation: $Y = 0.014(\text{s.e. } 0.0044)X + 18.65(\text{s.e. } 0.324)$, $r = 0.33$, $P=0.002$., derived from regression analysis. There was no significant effect (P>0.05) of increasing WSC content on liveweight gain, but relative to the control (no added WSC), increasing levels of WSC tended (P=0.054) to increase liveweight gain. A positive linear trend (P=0.027) existed between LWG and increasing levels of WSC.

Table 42: Liveweight gain (LWG) and dry matter intakes (DMI) for the experimental treatments

	Level of added WSC (g/kg of grass DM)					s.e.d.	Sig	L	Q	C
	0	30	60	90	120					
DMI(kg/d)	6.63 ^a	7.08 ^{bc}	6.91 ^{ab}	7.36 ^c	7.33 ^c	0.207	**	***	NS	NS
DMI (g/kg bw)	18.5 ^c	19.3 ^{abc}	19.1 ^c	20.3 ^a	20.2 ^{ab}	0.50	**	***	NS	NS
DMI (g/kg bw ^{0.75})	80.6 ^c	84.5 ^{abc}	83.4 ^c	88.3 ^a	88.2 ^{ab}	2.25	**	***	NS	NS
LWG (kg/d)	0.77	0.94	0.84	0.95	0.95	0.073	NS	*	NS	NS
LWG (g/kg bw)	2.16	2.55	2.34	2.61	2.50	0.181	NS	NS	NS	NS

L=linear, Q=quadratic, C=cubic, ^{abcd} Within rows, means without a common superscript differ from one another at P<0.05

Table 43: Intake, *in-vivo* digestibility, nitrogen partitioning and retention of cattle fed grass diets supplemented with increasing amounts of WSC

	Level of added WSC (g/kg grass DM)					s.e.d.	Sig	L	Q	C
	0	30	60	90	120					
DMI (kg/d)	7.23	7.44	7.33	7.91	7.94	0.373	NS	0.03	NS	NS
N intake (g/day)	166	166.5	162.5	170.1	160.4	11.04	NS	NS	NS	NS
Daily N intake (g/kg BW)	0.42	0.42	0.42	0.42	0.41	0.025	NS	NS	NS	NS
Digestibility co-efficients (g/kg)										
Dry matter	626	627	660	634	636	18.3	NS	NS	NS	NS
Organic matter	720	723	744	729	732	13.1	NS	NS	NS	NS
Nitrogen	576 ^{ab}	557 ^{abc}	580 ^a	551 ^{abc}	516 ^c	21.4	*	0.012	NS	NS
ADF	656	645	669	638	631	24.2	NS	NS	NS	NS
NDF	733	723	739	716	707	19.3	NS	NS	NS	NS
DOMD (g/kg DM)	581	593	612	602	601	17.0	NS	NS	NS	NS
N loss (g/day)										
Faeces	70.1 ^b	73.4 ^{ab}	67.7 ^b	76.8 ^a	78.2 ^a	2.98	**	0.006	NS	NS
Urine	76.8 ^a	68.9 ^{ab}	61.6 ^{bc}	59.2 ^{bc}	52.7 ^c	6.57	**	<0.001	NS	NS
Total	146.9	142.3	129.3	136	130.8	8.5	NS	0.052	NS	NS
N retained	19.1	24.2	33.2	34.1	29.6	7.78	NS	0.085	NS	NS
Daily N retained (g/kg BW)	0.05	0.06	0.08	0.08	0.07	0.019	NS	0.074	NS	NS
Faecal N : urinary N	0.94 ^a	1.09 ^{ab}	1.15 ^{bc}	1.34 ^{cd}	1.51 ^d	0.100	***	<0.001	NS	NS
N loss(g/kg N intake)										
Faeces	427	447	427	453	491	26.8	NS	0.03	NS	NS
Urine	460 ^c	417 ^{bc}	380 ^{ab}	347 ^a	330 ^a	29.1	***	<0.001	NS	NS
Retained	113	137	193	201	179	44.1	NS	0.054	NS	NS

Table 43 shows the intake, *in-vivo* digestibility, nitrogen partitioning and retention of the cattle when fed grass diets supplemented with increasing amounts of WSC. There were no significant effects ($P>0.05$) of supplementary WSC on DMI or nitrogen intake in the digestibility study. However, there was a significant ($P=0.03$) positive linear trend in DMI as level of WSC increased. Addition of increasing increments of supplementary WSC had no effect on the digestibility of DM, OM, NDF, ADF or DOMD. Increasing levels of WSC did reduce ($P<0.05$) N digestibility and did so in a linear trend ($P=0.012$). As WSC addition increased there was a significant increase ($P<0.05$) in the faecal excretion of nitrogen and this was coupled with an inverse significant reduction of urinary nitrogen excretion. As a consequence, there were no significant ($P>0.05$) effects of WSC level on total loss of nitrogen in grams per day, or in grams of nitrogen retained per day. However, there was a decreasing linear trend ($P=0.052$) of total N loss as level of WSC increased, and an increasing linear trend ($P=0.085$) in nitrogen retained in grams per day, as level of WSC increased. Total faecal N output by cattle supplemented with 120 and 90 g of WSC per kg of grass DM was higher ($P<0.05$) than with the 0 and 60g. Faecal N output from cattle given the 30g WSC did not differ ($P>0.05$) from any of the other WSC levels. Faecal N output and increasing levels of WSC showed a significant positive linear relationship ($P=0.006$). The urinary N output by the cattle given 0g of WSC did not differ ($P>0.05$) from cattle given the 30g, but was significantly higher ($P<0.05$) than all other levels. Urinary N output for the 120g WSC level was lower ($P<0.05$) than 0 and 30 g, but did not differ ($P>0.05$) from 60 and 90 g. The fraction of N excreted in the urine decreased linearly ($P<0.001$) with increasing level of WSC. The partitioning of N excretion between the faeces and the urine (Y) (i.e. the ratio of faecal N excretion to urinary N excretion) (g/day) increased as WSC level (X) (g/kg grass DM) increased. The ratio increased in a linear fashion and the equation that represents this increase is as follows: $Y = 0.0046$ (s.e. 0.00072) $X + 0.927$ (s.e. 0.0532), $r = 0.7$, $P<0.001$. The proportion of N which was excreted in the faeces showed a significant ($P=0.03$) positive linear response to increasing WSC level. Urinary N excretion, as a proportion of N intake did differ significantly

($P < 0.05$) between the levels and decreased linearly ($P < 0.001$) as level of WSC increased. The amount of N excreted in the faeces in relation to N intake (Y) (g/kg N intake) and WSC (X) (g/kg grass DM) can be expressed as a linear equation of $Y = 0.45(\text{s.e. } 0.199)X + 421.7(\text{s.e. } 14.6)$, $r = 0.32$, $P = 0.03$. Nitrogen excreted in the urine as a proportion of nitrogen intake did not differ ($P > 0.05$) between levels 120, 90 and 60 g, but 120 and 90g were lower ($P < 0.05$) than 0 and 30 g, with 60 g being lower ($P < 0.05$) than 0 but not different ($P > 0.05$) from any other level. Nitrogen excreted in the urine as a proportion of nitrogen intake (Y) (g/kg N intake) and WSC level (X) (g/kg grass DM) can be expressed as a linear equation of : $Y = -1.105(\text{s.e. } 0.210)X + 452.9(\text{s.e. } 15.4)$, $r = -0.64$, $P < 0.001$. There was no significant ($P > 0.05$) treatment effect on total nitrogen retained, when expressed as a proportion of total N intake (Y) (g/kg N intake), but a positive linear trend ($P = 0.054$) existed as level of WSC (X) (g/kg grass DM) increased, and this can be described by the equation: $Y = 0.654(\text{s.e. } 0.324)X + 125.4(\text{s.e. } 23.8)$, $r = 0.26$, $P = 0.054$. The proportion of N which was excreted in the faeces showed a significant ($P = 0.03$) positive linear response to increasing WSC level. Urinary N excretion, as a proportion of N intake did differ significantly ($P < 0.05$) between the levels and decreased linearly ($P < 0.001$) as level of WSC increased. The amount of N excreted in the faeces in relation to N intake (Y) (g/kg N intake) and WSC (X) (g/kg grass DM) can be expressed as a linear equation of : $Y = 0.45(\text{s.e. } 0.199)X + 421.7(\text{s.e. } 14.6)$, $r = 0.32$, $P = 0.03$. Nitrogen excreted in the urine as a proportion of nitrogen intake did not differ ($P > 0.05$) between levels 120, 90 and 60 g, but 120 and 90g were lower ($P < 0.05$) than 0 and 30 g, with 60 g being lower ($P < 0.05$) than 0 but not different ($P > 0.05$) from any other level. Nitrogen excreted in the urine as a proportion of nitrogen intake (Y) (g/kg N intake) and WSC level (X) (g/kg grass DM) can be expressed as a linear equation of : $Y = -1.105(\text{s.e. } 0.210)X + 452.9(\text{s.e. } 15.4)$, $r = -0.64$, $P < 0.001$. There was no significant ($P > 0.05$) treatment effect on total N retained, when expressed as a proportion of total N intake (Y) (g/kg N intake), but a positive linear trend ($P = 0.054$) existed as level of WSC (X) (g/kg grass DM) increased, and this can be described by the equation: $Y = 0.654(\text{s.e. } 0.324)X + 125.4(\text{s.e. } 23.8)$, $r = 0.26$, $P = 0.054$.

Table 44: Purine derivative (PD) excretion and estimated microbial N supply in cattle fed grass diets supplemented with increasing amounts of WSC

	Level of added sucrose (g/kgDM)					s.e.d.	Sig	Orthogonal contrasts		
	0	30	60	90	120			L	Q	C
PD excretion (mmol/day)										
Allantoin	192.0	218.7	208.0	209.3	216.9	19.72	NS	NS	NS	NS
Uric acid	4.7	4.9	4.0	4.2	3.9	0.63	NS	NS	NS	NS
Total	196.7	223.5	212.0	213.5	220.8	19.80	NS	NS	NS	NS
Microbial N (g/day)	139.1	162.1	152.4	152.6	159.6	16.68	NS	NS	NS	NS
Microbial N (g/kg DM intake)	19.24	21.96	21.00	19.38	20.33	2.386	NS	NS	NS	NS
Microbial N (g/kg DOMI)	33.2	37.2	34.4	32.3	34.0	3.029	NS	NS	NS	NS

The urinary excretion of purine derivatives and estimated microbial nitrogen supply for the five treatments are presented in Table 44. Increasing levels of WSC did not alter ($P > 0.05$) total purine derivative excretion (Mmol/day), nor the individual components of the purine derivatives. Estimates using purine derivative excretion values indicate that microbial N synthesis was not significantly ($P > 0.05$) influenced by level of added WSC.

Thus, in conclusion, a sequential increase in the WSC content of a grass-based diet fed to growing cattle resulted in (1) a positive linear increase in dry matter intake, (2) a tendency to linearly increase liveweight gain, (3) no change in the digestibility of DM, OM, NDF or ADF but a decrease in the digestibility of nitrogen, (4) a shift in the site of nitrogen excretion from urine to

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faeces, thereby decreasing urinary N excretion and linearly increasing faecal nitrogen excretion and (5) a non-significant improvement in the quantity of nitrogen being retained by the animal, and (6) the synthesis of microbial nitrogen remaining unaffected by the addition of WSC to the grass diet at any level. The results of this study therefore suggest that if WSC levels in grass were increased sufficiently that animal productivity would increase concomitant with an improvement in the utilisation of the forage nitrogen.

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RMIS No. 5002

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Performance of steers grazing ryegrass genotypes of normal or elevated concentrations of water soluble carbohydrates

Grasses differing in their concentration of water soluble carbohydrates (WSC) could differ in the voluntary intakes they stimulate, in the growth and productivity of the animals grazing them and in the losses of nitrogenous materials via urine. A previous experiment with zero-grazed cattle indicated that increasing the concentration of WSC (as added sucrose) in grass could deliver the above benefits. This experiment aimed to assess the performance of steers grazing either 'Fennema' (normal WSC) or 'Aberdart' (bred for elevated concentration of WSC) perennial ryegrass. Swards were sown in September 2001. Thirty 2-year-old continental steers were rotationally grazed on each grass from 28 April to 29 September, 2004. There were no differences between 'Fennema' and 'Aberdart' ryegrasses in terms of steer live-weight gain (0.98 vs. 1.01 kg/day), carcass weight (336 vs. 337 kg) and kill-out fraction (0.527 vs. 0.528). Further analyses will quantify the nutritive value of the two grasses throughout the growing season.

This experiment was partially funded by the European Union under the Fifth Framework Programme (QLK5-CT-2001-04980).

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RMIS No. 5002

The extent and identity of visible fungi on baled grass silage at feed-out on farms in the Midlands

A pilot survey on 35 farms in March 2003 recorded visible fungal growth present on 58/64 (0.9) bales examined. The objective of the present survey was to investigate the extent and identity of visible fungi growing on the surface of baled silage throughout the winter-feeding season on farms in the Midlands.

Between mid-November 2003 and mid-March 2004, baled silage was surveyed on 50 farms along a route between Cavan town and Thurles (*ca.* 150 km). This route was sub-divided into five sections with two different farms per quintile visited once a month for five months (i.e. 10 farms/month). A detailed questionnaire was completed on each farm visited - records were taken of the number of bales present, harvesting and storage characteristics of the bales, type and extent of damage to the plastic wrap and the means used to prevent and remedy damage to the plastic wrap on the bales. Two bales in readiness for feeding were examined in detail on each farm (total of 100 bales examined). Protocols were as described in the previous pilot study. Briefly, the plastic wrap on each bale was examined for damage. On removal of the plastic wrap, all visible mould and yeast colonies were located and sampled. Samples were collected by removing a

small fragment of foliage colonised with fungal material into an individual sterile container. The surface area of each colony was determined by placing a plastic grid over the colony and visually estimating its area. Dry matter (DM) and pH were determined using clean silage from each bale. The direct plating method was used to isolate the moulds and yeasts; malt extract agar (MEA, Oxoid), and dicloran rose bengal agar (DRBA, Oxoid) containing antibiotics were the isolation media used. Fungi were identified to genera/species by their macro- and micromorphology features. Statistical analysis was performed using the GLM procedures of SAS, Version 8.2.

Bales had 336 (s.d. 105.4) g DM/kg and a pH of 4.5 (s.d. 0.29). Fungal growth was observed on 90/100 (0.9) bales. The proportion of bales affected with fungal growth (n = 20 bales/month) ranged from 0.75 (January) to 1.0 (March). Throughout the winter feeding season, the mean proportion of the bale surface area affected ranged from 0.04 in November to 0.09 in January, with mean proportion coverage of 0.07 over the five months (Table 45). On average, there were 6 visible fungal colonies on each affected bale and this ranged from one to 21 colonies per bale. A total of 582 fungal colonies were sampled, resulting in 862 isolates. The most frequently isolated fungus was *Penicillium roqueforti* (0.41) (Table 46) and this was present on 78/100 (0.78) bales examined. Other frequently isolated fungi included yeasts, *Schizophyllum commune* and mucoraceous moulds. Visible damage to the plastic film (including repaired damage) was recorded on 49/100 (0.49) bales. Of the 90 bales that were affected with fungal growth, those that had damaged film had proportionally higher mean fungal coverage (0.10) than where the film appeared undamaged (0.04) and this difference was statistically significant (P<0.05).

Table 45: Prevalence of visible fungal growth on baled grass silage from November 2003 to March 2004 on 50 Midland farms

Month of survey	Proportion of bales contaminated (20 bales/month)	Proportion of the bale surface area affected Mean±(s.d.)
Nov	0.90	0.04 (0.036)
Dec	0.90	0.07 (0.132)
Jan	0.75	0.09 (0.080)
Feb	0.95	0.08 (0.086)
Mar	1.00	0.08 (0.086)
Mean	0.90	0.07 (0.089)

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Table 46: Fungi isolated from baled grass silage on farms from November 2003 to March 2004

Fungal genera/species and the proportion of fungal isolates (n=862) from bales (n=90)	
<i>Penicillium roqueforti</i>	0.41
Yeasts	0.15
<i>Schizophyllum commune</i>	0.11
Mucoraceous moulds	0.08
<i>Penicillium panuem</i>	0.04
<i>Geotrichum</i>	0.04
<i>Fusarium</i>	0.02
Other <i>Penicillium</i> spp.	0.02
<i>Trichoderma</i>	0.01
Other unidentified moulds	0.11

It is concluded that a high incidence of visible fungal growth was recorded on bales at feed-out throughout the winter season. Air ingress *via* holes/damaged plastic film surrounding bales is a major factor permitting fungal growth in bales. *Penicillium roqueforti* and *Penicillium paneum* are two species that can produce harmful mycotoxins under certain conditions. Further research is needed to identify the mycotoxins routinely present in *Pencillium*-contaminated silage, with special emphasis on mycotoxins that could pose a threat to livestock health.

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National survey to establish the extent of visible mould on baled grass silage in Ireland and the identity of the predominant fungal species

On an annual basis, some 12 million bales of grass silage are made on Irish farms. A pilot survey of 35 farms in March 2003 recorded 90% of bales with some fungal growth present. The predominant fungus identified was *Penicillium roqueforti*, a species that can produce harmful mycotoxins under certain conditions. Evidence is increasing that mycotoxins are formed regularly under ensiling conditions. The objectives were to determine (i) the proportion of bales with visible fungal growth on farms in Ireland, (ii) the extent of fungal contamination on the surface of bales and (iii) the identity of the largest fungal colony present on bales.

In February 2004, 180 farms were visited on six separate routes throughout Ireland. Each route was sub-divided into five sections with six farms per quintile visited. All routes were 100-150 km in length and were representative of geographical locations and farm enterprises. Two bales in readiness for feeding were examined on each farm (total of 360 bales). Protocols were as in previous experiments with the exception that in this survey only the largest fungal colony on each bale was sampled.

Table 47: Prevalence of visible fungal growth on baled grass silage on 180 farms in Ireland (60 bales/route)

Route location	Proportion of bales contaminated	Proportion of bale surface area affected Mean \pm (SD)
North west	0.77	0.09 (0.12)
North midlands	0.98	0.04 (0.03)
West midlands	0.98	0.09 (0.11)
Midlands	0.90	0.08 (0.08)
South west	0.98	0.07 (0.08)
South east	0.91	0.02 (0.04)
Overall mean	0.92	0.06 (0.06)

Table 48: Predominant fungi on baled silage in Ireland

Fungal genera/species	No. of bales on which predominant	Proportion of bales
<i>Penicillium roqueforti</i>	146	0.42
<i>Schizophyllum commune</i>	71	0.20
Yeasts	45	0.13
<i>Penicillium paneum</i>	15	0.04
<i>Geotrichum</i>	12	0.03
<i>Fusarium</i>	4	0.01
<i>Trichoderma</i>	3	0.01
Mixed mycobiota ^a	35	0.10
Unidentified moulds	13	0.03

^a>1 fungus was isolated from the predominant fungal colony

The majority of bales examined were harvested between June and August 2003. Bales had a dry matter (DM) concentration of 309 (standard deviation (SD) 111.4) g/kg and a pH of 4.5 (SD 0.43). Fungal growth was visibly present on 334/360 (0.92) bales examined. A total of 334 mould and yeast colonies were sampled and 344 fungi were isolated and identified as being the contaminant visible fungal growth on the bales. The extent of fungal growth on bales ranged from 0 to 0.82 surface coverage, with mean proportion coverage of 0.06 (Table 47). The fungus affecting the largest surface area on bales was *P. roqueforti* (Table 48).

It is concluded that the results of this study reflect the findings made in the previous pilot survey. Visible fungal growth on baled silage is extensive throughout Ireland, with some differences in the extent of contamination on bales between different regions. As a consequence, some baled silage will have an inferior feeding value, which in turn will negatively influence farm profits. Although a relatively small number of fungal species was responsible for most of the contamination, at least two of these fungi (*P. roqueforti* and *P. paneum*) could potentially cause health problems in livestock by their known ability to produce harmful mycotoxins.

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Schizophyllum on baled grass silage in Ireland: national farm survey 2004

Since the early 1990's, a mushroom-type growth has been appearing with increasing frequency protruding through the plastic film on baled grass silage in Ireland. The fungus was identified as *Schizophyllum commune*, a gilled bracket fungus that it is known primarily as a white rot fungus, found worldwide on fallen branches in woodlands. A countrywide survey conducted in 1999 recorded the presence of *Schizophyllum* on 0.53 farms in Ireland. The authors of that survey concluded that *Schizophyllum* was widely distributed throughout Ireland and had the potential to cause considerable loss of feedstuff on farms. The objective of this survey was to assess the current prevalence of *Schizophyllum* on baled silage.

In February 2004, collections of baled silage were inspected along six routes. These routes represented different geographical locations and farm enterprises in Ireland. Thirty farms were visited along each route and bale collections were examined for the presence and extent of the visible protrusion of *Schizophyllum* basidiomes through the plastic stretch film, in addition to other bale parameters. Silage samples were collected from two bales on each farm and silage dry matter (DM) and pH was determined.

Bales had a DM concentration of 309 (standard deviation (SD) 111.4) g/kg and a pH of 4.5 (0.43). Bales examined along Route 6 had lower silage DM concentrations relative to the other five routes. *Schizophyllum* was visible on 106/180 (0.58) farms surveyed, ranging from 0.13 (Route 6) to 0.76 (Route 1) (Table 49). Proportionally 0.36 farms had less than 0.1 bales affected by this fungus and 0.2 farms had between 0.1 and 0.5 bales of their bales affected.

Table 49: Occurrence and extent of *Schizophyllum commune* on Irish farms (N=30 farms/route, 180 in total) in 2004

Occurrence & incidence of <i>Schizophyllum</i>	Number (and proportion) of farms on which <i>Schizophyllum commune</i> was observed on bales						
	Route 1 (Midlands)	Route 2 (West midlands)	Route 3 (South-west)	Route 4 (South-east)	Route 5 (North midlands)	Route 6 (North-west)	Overall
Present	23 (0.76)	21 (0.7)	21 (0.7)	20 (0.66)	17 (0.57)	4 (0.13)	106 (0.58)
<0.1 bales affected	10 (0.33)	10 (0.33)	14 (0.47)	15 (0.5)	12 (0.40)	4 (0.13)	65 (0.36)
0.1 – 0.5 bales affected	12 (0.40)	11 (0.37)	4 (0.13)	4 (0.13)	5 (0.17)	0 (0)	36 (0.20)
>0.5 bales affected	1 (0.03)	0 (0)	3 (0.10)	1 (0.03)	0 (0)	0 (0)	5 (0.02)
DM (SD) g/kg	335 (118.7)	334 (105.4)	312 (113.6)	346 (114.5)	296 (94.2)	236 (83.8)	309 (111.4)
pH (SD)	4.4 (0.32)	4.5 (0.35)	4.6 (0.57)	4.6 (0.49)	4.4 (0.31)	4.5 (0.45)	4.5 (0.43)

It is concluded that *Schizophyllum* is still widespread on baled grass silage in Ireland and on a slightly higher proportion of farms (0.58) than recorded five years previously. In that study, a lower incidence of *Schizophyllum* was also recorded on farms in the north-west region (Route 6). A low grass DM at ensiling, as recorded for bales on Route 6, may be a factor that prevents bales being successfully colonized with *Schizophyllum*.

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Numbers of mould and yeast propagules in baled grass silage devoid of visible fungal growth

From previous survey work, the occurrence of visible fungal growth on baled silage is widespread. Fungal spores/mycelium fragments present on the grass crop prior to baling and wrapping are permitted to grow if the integrity of the plastic film is damaged during storage. This growth will result in quantitative and/or qualitative nutrient losses in silage, as well as possible exposure of livestock to fungal mycotoxins. The objective of this experiment was to determine the number of yeast and mould propagules in baled grass silage devoid of visible fungal growth.

In July-August 2004, a single bale (1.2 m diameter x 1.2 m high) of grass was obtained from each of 15 crops on eight farms in Co. Meath (no more than two crops (or bales) per farm). Bales were transported to Grange, weighed and immediately wrapped (McHale™ 991 BE) in six layers of black plastic film (Volac Silawrap®); the interval between baling and wrapping was < 2 h. A questionnaire was completed relating to each bale – a record of the duration of wilting, type and age of the grass crop and weather conditions from cutting to baling. Additional questions related to grass chopping and the use of additives. Each bale was sampled in eight positions after *ca* 42 d storage using a sharpened sterile cylindrical steel corer (length, 22 cm; inner diameter, 3.5 cm and thickness, 1 mm) powered by an electric drill. Sampling points were at 1400, 1600, 2000 and 2200 h clock positions on the bale barrel, *ca* 40 cm from each end. Cores were taken to a depth of *ca* 20 cm. The eight sub-samples per bale were composited and stored at 4 °C prior to microbiological analysis. Mould and yeast counts were determined using the spread plate method, with the following exceptions. Each silage sample was thoroughly hand mixed under aseptic conditions and three 30 g sub-samples were removed for microbiological analysis. The mould and yeast colony forming units (cfu) were enumerated separately and the mean of the three sub-samples was reported for both. The mould and yeast count refers to the number of cfu/g wet weight of sample. The remaining silage was used to determine the dry matter (DM) and pH of each bale. Correlation coefficients between the different bale characteristics were determined using Unistat 5.5 for Microsoft Excel.

Table 50: Mould and yeast colony forming units (cfu) in 15 bales of grass silage

Bale no.	Wt. at ensiling (kg)	DM (g/kg)	pH	Mould (log ₁₀ cfu/g)	Yeast (log ₁₀ cfu/g)
1	740	355	4.3	1.1	1.6
2	410	353	4.4	1.7	0.5
3	710	337	4.3	0.7	2.0
4	800	343	3.8	0.8	3.1
5	670	334	5.1	0.9	1.5
6	640	388	4.7	0.3 ^a	2.1
7	600	459	5.0	0.5	2.6
8	570	431	4.8	1.7	2.9
9	800	193	3.6	nd ^b	2.1
10	955	182	3.5	nd	3.4
11	600	373	4.8	0.6	2.2
12	920	282	3.9	nd	2.6
13	770	197	4.2	nd	5.1
14	780	228	3.7	nd	4.3
15	810	188	3.6	nd	2.0
Mean (s.d.)	718 (141.2)	309 (92.0)	4.2 (0.54)	0.6 (0.60)	2.5 (1.13)

^a lowest limit of detection; ^bnd, none detected.

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The 15 bales represented a wide range of crops and harvesting conditions. The weather between mowing and baling ranged between fine to unsettled, bales were harvested from both stemmy and leafy grass, and pastures were described as never having been reseeded, reseeded < 10 years or reseeded > 10 years ago. The duration of wilting ranged from 3 h to 3 d. Bales were predominantly chopped, tied with netting and additives were not applied. There was no visible damage to the plastic film on any bale throughout the storage period and all were free of visible fungal growth on the day of sampling. Moulds were cultured from 9/15 (0.6) bales, while yeasts were cultured from all bales (Table 50). Overall, yeast numbers were higher in the 15 bales than mould numbers (2.5 vs 0.6 log₁₀ cfu/g). Correlation coefficients values for various bale characteristics were as follows: bale DM vs pH (r=0.81), DM vs bale weight at ensiling (r=-0.70), pH vs bale weight at ensiling (r=-0.71), bale weight at ensiling vs mould (r=-0.75), bale weight at ensiling vs yeast (r=0.48), DM vs mould (r=0.70), DM vs yeast (r=-0.39), pH vs mould (r=0.55), pH vs yeast (r=-0.32) and mould vs yeast (r=-0.52).

It is concluded that yeast propagule numbers were greater than mould numbers per g of silage. Although mould propagule numbers were low and sometimes absent, these propagules have the potential to grow extensively in bales if enough oxygen is available. No one crop type predisposed bales to higher numbers of moulds and yeasts. However, DM was positively correlated to increasing numbers of mould and negatively to yeast. pH was also positively correlated to mould numbers and negatively to yeast, but to a lesser extent than DM.

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***Penicillium* found in mouldy silage - its metabolites and sensitivity to oxygen**

There were three components to this work:

Firstly, to analyse 78 presumptive isolates of *Penicillium roqueforti* and 82 presumptive isolates of *Penicillium paneum* (the first and fourth most common visible moulds on baled silage in Ireland) isolated from baled silage in Ireland, for their secondary metabolites to (1) confirm their identification and (2) determine what mycotoxins they can produce *in vitro*. The identity of *P. roqueforti* and *P. paneum* isolates were confirmed by their secondary metabolite profiles. *P. roqueforti* isolates were found to produce the mycotoxins mycophenolic acid, PR toxin or its analogues and roquefortine C. All *P. paneum* isolates produced marcfortine and 50% of these isolates were able to produce patulin. This fungus is the only known producer of marcfortine in the fungal kingdom.

Secondly, to screen a small number of silages for the presence of a range of mycotoxins. Fifteen grass and 20 maize silage samples that exhibited visible mould or no visible mould were obtained in Ireland and Denmark respectively. These were analysed using liquid chromatography- diode array detection-electrospray-high resolution mass spectrometry (LC-UV-HR-MS), LC-MS/MS and cytotoxicity by using the MTT-cell culture assay after Solid Phase Extraction purification on C18, and/or polymeric cat- and an-ion columns. The results show that mycophenolic acid (MPA) and roquefortine C (RC) were the most prevalent fungal metabolites in the most visibly mouldy samples. Besides these metabolites, roquefortines A, B and D, festuclavine, 2,5-dihydroxybenzoic acid, citreoisocumarin, marcfortines A and B, and several andrastins were also detected in the samples. Penitrem A, penicillic acid, cyclopiazonic acid and patulin were not detected in any sample, and PR-toxin was found only in trace quantities in one sample. This was backed up by the bioassay results, which did not indicate cytotoxic compounds in any of the samples. One reddish maize sample infected with *Monascus* sp. also contained mevinolin and several *Monascus*

pigments but not citrinin. Samples with no visible fungal contamination contained low quantities of RC and MPA, the presence of which however indicated low levels of fungal contamination. The presence of marfortines shows that growth of *P. paneum* had occurred, although it was difficult to subculture from the samples.

Thirdly, to quantify the extent to which *P. roqueforti* can grow in a low-oxygen environment, *P. roqueforti* isolates obtained from baled silage were incubated in a modified atmosphere (i.e. 1 % O₂ and 50 % CO₂) for 5 days to establish their ability to germinate and grow under these conditions – these gaseous concentrations are typical of the baled silage environment during storage. The growth rate of *P. roqueforti* isolates was reduced by 20% over a period of 5 days at 1% O₂ and 50% CO₂ compared to control plates in normal atmosphere. Spores were also able to germinate in these conditions, but growth was reduced by 22% compared to control plates. This experiment needs to be repeated using a longer duration than 5 days.

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Bacteria and yeast in round bale silage on a sample of farms in County Meath, Ireland

Baled silage is made on almost three-quarters of all Irish farms. As in conventional clamp silage, a dominant lactic acid fermentation is required to reduce pH and minimise both quantitative and qualitative losses caused by undesirable microorganisms (e.g. yeast, *Clostridia*, *Bacilli* and *Enterobacteria*). However, the higher pH achieved in baled silage compared with conventional silage is less inhibitory to these microorganisms. The thickness of the plastic barrier used to seal ensiled forage from air is thinner with conventionally wrapped bales (70 µm for 4 layers) than clamp silos (250 µm for 2 layers). Furthermore, with baled silage, ~0.5 of the silage is within 12 cm of the plastic wrap compared to less than 0.1 with conventional clamp silage. This may create a more aerobic environment at the bale surface reducing the hygienic quality of the silage. The microbial composition within baled silage in Ireland has not been quantified. This study enumerated the predominant bacteria and yeast in the outer and inner layers of baled silage at feedout.

Two bales of silage were sampled on each of 10 farms from February to April 2004, using an electrically powered aseptic cylindrical core bit (length, 65 cm; internal diameter, 3.5cm) at eight points around the bale. Subsampling points were at 1500, 1800, 2100 and 0000 h clock positions on the bale barrel, *ca* 40 cm from each end. There was no visible evidence of aerobic deterioration at these points. At each point subsamples were taken from both the outer 20 cm and through to the centre of the bale. The outer and inner core subsamples were each composited per bale, mixed and stored at 4°C for microbiological analysis. MRS nystatin agar, malt extract agar (MEA) containing streptomycin (pH 3.5), violet red bile glucose (VRBG) agar, nutrient agar (NA) and reinforced clostridial agar (RCA) with neutral red were used to enumerate for lactic acid bacteria (LAB), yeast, *Enterobacteria*, *Bacillus* spores and clostridial spores, respectively (Seale *et al.*, 1990). VRBG was incubated at 37°C for 2 days. RCA was incubated anaerobically

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in a GasPak 150 jar for 7 days at 37°C while all other media were incubated for 3 days at 30°C. The number of colony forming units (cfu) on each plate was enumerated and the number of microorganisms/g silage calculated.

The bales sampled had a mean dry matter of 354 (s.d. 118.5) g/kg and pH of 4.5 (s.d. 0.50). These are typical of values found on bales throughout the country, and indicate that the preservation of the wilted silage was satisfactory (Table 51). Type of microorganism had a major impact on the mean counts, with LAB > yeast > *Clostridia* > *Bacilli* > *Enterobacteria*. In contrast, the standard errors suggest the range in counts was in the order yeast > *Enterobacteria* > *Bacilli* > *Clostridia* > LAB. Yeast, LAB and *Enterobacteria* numbers were higher in the outer layer. There was no significant difference ($P > 0.1$) in pH, DM, *Bacillus* and *Clostridia* numbers between the outer and inner bale layers.

Table 51: Composition of outer layer compared with inner layer of baled silage

	Outer	Inner	s.e.m.	Sig.
Dry matter (g/kg)	352	355	8.5	NS
pH	4.51	4.53	0.034	NS
LAB (log ₁₀ cfu/g silage)	5.83	5.53	0.093	*
Yeast (log ₁₀ cfu/g silage)	5.24	4.37	0.197	**
<i>Enterobacteria</i> (log ₁₀ cfu/g silage)	1.41	0.78	0.186	*
<i>Bacilli</i> (log ₁₀ cfu/g silage)	2.49	2.33	0.138	NS
<i>Clostridia</i> (log ₁₀ cfu/g silage)	3.75	3.87	0.096	NS

* = $P < 0.1$, ** = $P < 0.01$, NS = not significant

It is concluded that the numbers of yeast, LAB and *Enterobacteria* were higher in the outer layer than the inner layer, but the preservation appeared similar in both parts of the bales. Higher yeast and *Enterobacteria* numbers may reflect more aerobic conditions at the bale surface. This may be due to an imperfect seal by the plastic wrap. The data provides a benchmark for future research on the fermentation kinetics of baled silage.

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A 16S rDNA-based quantitative assay for monitoring *Lactobacillus plantarum* in silage

Ensilage of herbaceous biomass can be enhanced by applying pre-selected fermentative bacteria. However, insufficient is known about the population dynamics of such starter cultures under a range of ensiling conditions. Classical approaches for species-specific quantification of bacteria are labour intensive. An alternative approach is the detection of bacteria based on molecular markers for species-specific regions within their genomic DNA (e.g. the 16S rDNA sequence). In this study, a quantitative marker assay using the real-time PCR technique (Q-PCR) is described for *Lactobacillus plantarum*, a bacteria often used for silage starter cultures.

Based on a variable region within the 16S rDNA of *L. plantarum* published by Chagnaud *et al.* (2001), the following PCR-primers were developed: forward primer Lplan-vreg1-F 5'-TTACATTTGAGTGAGTGGCGAACT, reverse primer Lplan-vreg1-R 5'-AGGTGTTATCCCCGCTTCT, TaqMan[®] probe Lplrh-vreg1-T 5'-VIC[®]-GTGAGTAACACGTGGWAACCTGCC-TAMRA[®].

The Q-PCR reactions were carried out on an ABI 7000 system applying following optimised conditions: reaction mixture 1x JumpStart™ Taq ReadyMix™, 900nM forward primer, 300nM reverse primer, 200nM TaqMan® probe, 0.25µl internal dye ROX®, ad 25 µl H₂O_{dd}; cycle regime 1: 120s 94°C, 2: 15s 94°C, 3: 60s 59°C, 4: 60s 72°C, 2-4 were repeated 40x. Generally, every Q-PCR was set up in triplicates. Plasmid pATB875 containing a 1500 bp fragment of *L. plantarum* 16S rDNA was used as a standard. Grass was inoculated with equal concentrations of *L. plantarum* ATB-8 and *L. rhamnosus* ATB-14 and sampled on days 2, 13, 20 and 40 of ensilage. The genomic DNA was prepared from padded samples (Rheims & Stackebrandt 1999).

Using the pATB875 plasmid as a standard, an optimised Q-PCR protocol was developed. Out of triplicates of a dilution series an equation for the species-specific estimation of the copy number of *L. plantarum* 16S rDNA sequences in unknown samples was calculated (Fig. 1 A, B). Applied to grass silage samples, the assay monitored the raise of the *L. plantarum* population during the first two weeks of ensiling. Due to the increased acidification, the population decreased during prolonged ensiling (Fig. 1 C).

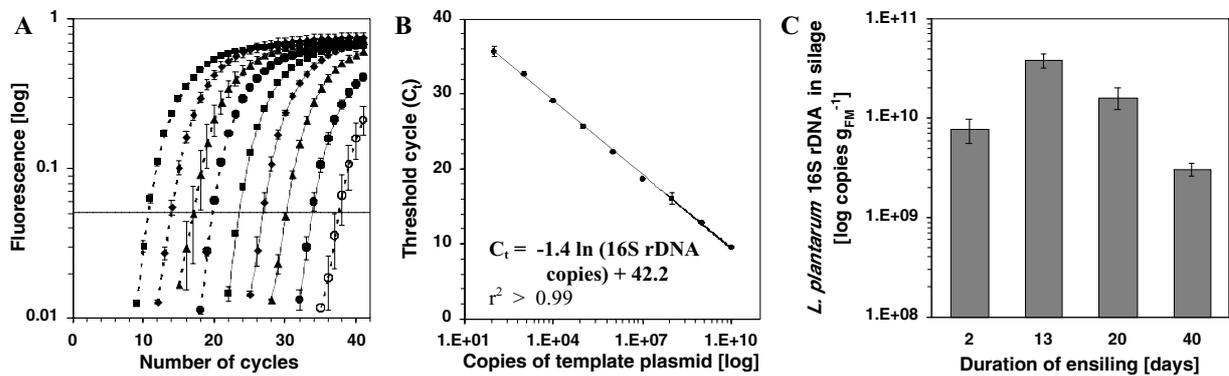


Figure 1 (A) Representative Q-PCR amplification curves of pATB875 10 fold dilution series (10^{10} (■) to 10^3 (●) and no-template control (○)). (B) The mean C_t values of triplicates were plotted against the copy number of pATB875. (C) Determination of *L. plantarum* 16S rDNA copy number within grass silage samples.

In conclusion, this Q-PCR assay is a first attempt at a direct DNA-based quantification of *L. plantarum* within silages. The Q-PCR assay enables the analyses of population dynamics of starter cultures containing *L. plantarum*. Similar approaches should also be applicable for monitoring other fermentative bacteria species.

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Microbiological composition of silage: the effects of wilting, chopping, compaction, and anaerobiosis

The fermentation profile of baled silage appears different to that of comparable conventional precision-chop silage ensiled in a horizontal bunker silo. The difference is manifested in a higher pH for baled silage. This creates conditions more conducive to the activities of undesirable microorganisms (e.g. yeast and *Enterobacteria*) that cause qualitative and quantitative loss of feedstuff. Numerous physical differences between the two systems can potentially explain the fermentation differences. This study sought to determine the relative effects of forage wilting, chopping and compaction, and air infiltration, on the microbial population at the end of ensiling.

Grass (0.8 kg dry matter (DM)/silo) was ensiled on three consecutive days (unwilted or after 24 or 48 h wilting) in a total of 72 laboratory silos. Each grass was ensiled either unchopped or precision-chopped, with or without air infiltration and with or without compaction. Within the factorial arrangement of treatments, individual treatment combinations were repeated in triplicate. Air infiltration was achieved by both the lid and the base of these silos being finger-tightened, so that over the course of the experiment air could filter into the silo and through the forage. The base and lids of the remaining silos were fully sealed creating an anaerobic environment. Compaction was achieved by steel weights (21 kg) being placed in the silo to exert vertical pressure on the forage. The remaining silos had no weights on the forage. The silos were stored at 15°C for 100 days before being opened and aseptically sampled. MRS nystatin agar, malt extract agar (MEA) containing streptomycin (pH 3.5), violet red bile glucose (VRBG) agar and nutrient agar (NA) were used to enumerate for lactic acid bacteria (LAB), yeast, *Enterobacteria* and total viable counts (TVC), respectively (Seale *et al.*, 1990). The number of colony forming units (cfu) on each plate was enumerated and the number of microorganisms/g silage calculated. Data was analysed as a 3 x 2 x 2 x 2 factorial arrangement of treatments using the GLM procedures of SAS, Version 8.2.

Extensive wilting tended to reduce the numbers of LAB, *Enterobacteria* and yeast on grass (Table 52), whereas each day of wilting increased ($P < 0.001$) yeast counts on silage (Table 53). The highest ($P < 0.01$) LAB and *Enterobacteria* counts on silage were with a 24 h wilt, and wilting increased ($P < 0.001$) TVC numbers. Higher numbers of LAB and yeast occurred on silage than grass. Air infiltration increased ($P < 0.001$) the numbers of each type of microorganism on silage whereas neither chopping nor compaction altered ($P > 0.05$) final counts.

Table 52: Mean (s.d.) microbial counts (\log_{10} cfu/g) on fresh grass

Wilt (h)	DM g/kg	LAB	<i>Enterobac.</i>	Yeast
0	193	4.2 (0.09)	4.1 (0.19)	2.5 (0.23)
24	497	5.4 (0.13)	4.7 (0.21)	3.3 (0.23)
48	670	4.1 (0.39)	3.6 (0.59)	2.0 (0.55)

Table 53: Effects of wilting, chopping, air infiltration and compaction on microbial counts (\log_{10} cfu/g silage)

	LAB	<i>Enterobacteria</i>	Yeast	TVC
Wilt				
0	5.61	1.37	3.65	5.64
24	6.18	2.63	4.33	6.4
48	5.83	1.39	5.42	6.36
s.e.m.	0.067	0.261	0.132	0.091
Sig.	***	**	***	***
Chopping				
Unchopped	5.92	1.76	4.39	6.16
Chopped	5.82	1.84	4.52	6.10
s.e.m.	0.054	0.213	0.108	0.074
Sig.	NS	NS	NS	NS
Air				
Anaerobic	5.25	0.73	3.93	5.39
Aerobic	6.51	2.88	5.0	6.89
s.e.m.	0.054	0.213	0.108	0.074
Sig.	***	***	***	***
Compaction				
No weight	5.92	1.91	4.55	6.25
Weight	5.82	1.69	4.36	6.03
s.e.m.	0.054	0.213	0.108	0.074
Sig.	NS	NS	NS	NS

** = $P < 0.01$, *** = $P < 0.001$, NS = not significant

Interactions were not significant between most factors ($P > 0.05$). However, infiltration of air increased ($P < 0.001$) both LAB and TVC numbers more as the duration of wilting was extended. Chopping forage increased ($P < 0.001$) LAB numbers on silage when air was present during ensilage but led to a reduction under anaerobic conditions. Air infiltration increased ($P < 0.001$) yeast numbers with wilted but not unwilted forage and it increased ($P < 0.05$) *Enterobacteria* numbers more after a 24 h wilt compared to unwilted or a 48 h wilt.

In conclusion, the data suggest that where similar grass is conventionally ensiled as baled or precision-chop silage, the extent of anaerobiosis is the major factor influencing silage microbial composition. However, further work on the fermentation variables of this silage along with enumeration of *Clostridia* and *Bacilli* needs to be undertaken.

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Grange Research Centre

Uni-axial stretching of baled silage wrap films: gas permeation properties

The most common methods of manufacture of silage stretch film are blown and cast extrusion, using low density polyethylene (LDPE) and linear low density polyethylene (LLDPE). Film is pre-stretched during application to ensure a tight film seal is maintained. Generally films are stretched to 1.7 times their original length. This experiment investigated the effects of uni-axial stretching on the gas permeation properties of films used in baled silage wrapping.

Three films were manufactured from LDPE-LLDPE blends (70/30: w/w), using LLDPEs of different densities and melt flow indices (MFIs). These are denoted films A, B and C. The films were manufactured using a cast film extrusion system. Films were stretched to 1.0, 1.2, 1.4, 1.6, 1.8, 2.0, 2.4, 2.7 and 3.0 times original length using a specially designed clamping system on an Instron 4411 Universal tensile tester. This also measured the forces generated while stretching. A manometric gas permeability measuring apparatus was used to determine the permeation coefficient (PC) of film samples. A lower permeation coefficient indicates less gas transmission per unit thickness of film. The apparatus was designed for direct measurement of the gas transmission rate (GTR) through plastic films in accordance with B.S.2782, method 821A, ASTM D. 1434 and similar methods. The test gas was CO₂ (99.8% purity). Five samples were tested for each film at each stretch level. The crystallinity of the various films was determined using a Perkin Elmer DSC-6, and the samples were heated from 30 to 140°C at a rate of 10 deg C/min. The latent heat (ΔH J/g) was calculated for each sample, which is a measure of crystallinity. Three samples were tested for each film. Infra red spectra of the films were obtained using a Perkin-Elmer FTIR spectrometer (spectrum 1000) fitted with a 0.12 μ m zinc selenide grid polariser. The instrument operated with a resolution of 1cm⁻¹ and 40 scans were obtained for each sample. The IR absorbance scans were analysed between 700cm⁻¹ and 750cm⁻¹, for changes in the intensity associated with the *a* and *b*-axes (730cm⁻¹ and 720cm⁻¹) in both the machine (MD) and transverse (TD) direction, which give an indication of the molecular orientation of the films. Data were subjected to two-way analysis of variance appropriate for a factorial (film and stretch) arrangement of treatments.

Table 54: Effect of uni-axial stretching on the permeation coefficient, crystallinity and orientation of films

Film	Polymer			Stretch								
	Density (g/cm ³)	MFI (g/10 min.)		1.0	1.2	1.4	1.6	1.8	2.0	2.4	2.7	3.0
A	0.918	4.5	PC ⁺	37.8	31.6	26.2	22.8	19.1	20.4	19.6	16.6	10.9
			Force (mN)	0.0	32.8	36.7	38.7	39.4	38.6	38.4	38.8	38.9
			ΔH	64.3	70.9	67.8	61.0	72.0	64.0	67.0	78.0	85.0
			<i>b/a</i> ratio MD	1.2	1.9	2.3	2.7	2.6	2.4	2.3	2.3	2.3
			<i>b/a</i> ratio TD	1.3	1.4	0.0	1.6	1.7	1.7	1.8	2.2	2.0
			B	0.903	1.5	PC ⁺	63.8	52.1	41.4	33.7	30.1	32.2
Force (mN)	0.0	32.0	22.4			24.6	25.9	26.3	26.5	27.0	27.7	
ΔH	16.3	39.8	33.6			37.1	42.8	36.1	39.6	49.7	51.1	
<i>b/a</i> ratio MD	1.2	1.6	1.8			2.1	2.5	2.8	2.6	2.7	2.7	
<i>b/a</i> ratio TD	1.3	1.3	1.2			1.2	1.3	1.7	1.6	1.8	2.0	
C	0.917	4.0	PC ⁺			34.6	25.3	25.9	24.0	22.1	17.9	22.2
			Force (mN)	0.0	35.0	33.3	33.7	33.6	33.2	32.6	32.8	33.3
			ΔH	51.0	61.8	56.4	56.1	59.6	59.9	67.2	67.0	60.2
			<i>b/a</i> ratio MD	1.3	1.7	2.3	2.7	2.8	2.5	2.9	2.9	2.7
			<i>b/a</i> ratio TD	1.4	1.3	1.2	1.4	1.4	1.5	1.7	1.8	1.8

⁺PC = CO₂ permeation coefficient (cm³(STP)cm⁻¹cm²cm(10⁻⁶)). PC SEM = 0.76, ΔH SEM = 0.75. Stretch (S), Polymer (P) and SxP were each significant at P<0.001 for PC and ΔH .

The gas permeability of films have been reported to be dependant on the film crystallinity and molecular orientation (Laffin, 2004). Crystallinity of polyethylene films is dependant on the density and molecular orientation will be affected by the amount of film stretch. It was shown that the CO₂ permeation coefficient of all films decreased (P<0.001) when stretched, with the biggest reduction exhibited by the film of lowest density (Table 54). The crystallinity of all films increased (P<0.001) with increasing stretch. All stretched films exhibited an increase in orientation in the MD as indicated by FTIR analysis (*b/a* ratio) in Table 54, and the results also show a change in crystalline structure at around 1.8 to 2.0 times stretch.

In conclusion, the reduction in permeation coefficient recorded indicates the scope for working with more layers of highly stretched films. This may give improved barrier properties with less film applied. However the DSC and FTIR analysis results indicate a change in crystal structure that would result in disimproved mechanical properties.

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Carbon dioxide permeation properties of polyethylene films used to wrap baled silage

Polyethylene film wrapping for baled silage was introduced in the mid-1980s. This system is now used on 73% of farms in Ireland, accounting for one-third of the total national silage tonnage, or in excess of 9 million bales annually. The film must possess good gas barrier and mechanical properties to ensure satisfactory levels of anaerobiosis are achieved and maintained in the bale. This investigation determined the CO₂ barrier properties of a number of commercially available films.

Five commercial films (designated A to E) were investigated. Film C was reported to have been pre-stretched during manufacture. In each case a single layer of black film was tested, with 5 samples being obtained from the top, middle and bottom sections of a roll of film. A Davenport gas permeability measuring apparatus was used to determine the gas transmission rate (GTR) and permeation coefficient (PC) of film samples to CO₂ (99.8% purity). The GTR was measured according to B.S.2782, method 821A, ASTM D. 1434 and indicates the volume of gas transmission, with PC indicating the permeation per unit thickness Youngs modulus was measured from tensile analysis of the films in both the machine and transverse directions. The comonomer type of the films was predicted from differential scanning calorimetry (DSC) analysis. Each variable was subjected to two-way analysis of variance appropriate for a 5 x 3 factorial arrangement of treatments.

Table 55 shows that the gas transmission rates (GTR) for films A, B, D and E were in good agreement with previous reports for 25µm polyethylene film (Briston, 1983). The GTR is influenced by film thickness, with Film C (thickness 12µm) showing the highest GTR (49x10³ ml/m²/day/atm). Film C also exhibited the lowest permeation coefficient (24x10⁻⁶ml at standard temperature and pressure (STP)cm² versus 31 – 36x10⁻⁶ml(STP)cm²). The higher GTR of film C at least partially reflected its lower thickness (12µm), but the lower permeation coefficient appeared to be as a result of pre-stretching during manufacture leading to greater crystallinity and molecular orientation, which would improve the barrier properties of the film (Laffin, C. et al, 2004). Mechanical analysis also showed significant variation (P<0.001) in the Youngs modulus across the width of the roll, with the largest variation being recorded for film D (108.3 – 146.5MPa in the machine direction).

Table 55: CO₂ permeation and mechanical properties of five commercial films tested

Film(F)	Co-monomer	Thickness (µm)		Location in roll of film (L)			s.e.m ¹	Significance		
				Top	Middle	Bottom		Location	Film	L. x F.
A	Octene	25	GTR [#]	27.7	27.2	38.5	1.5	***	***	***
			PC ⁺	27.7	28.6	37.6	1.1	***	***	***
			YMm ^a	103.6	125.0	116.6	2.5	***	***	***
			YMt ^b	129.3	135.5	140.5	4.0	***	***	***
B	Octene	25	GTR [#]	35.3	33.6	45.1	1.5	***	***	***
			PC ⁺	32.5	33.0	42.9	1.1	***	***	***
			YMm ^a	133.2	117.7	131.8	2.5	***	***	***
			YMt ^b	151.2	150.7	132.5	4.0	***	***	***
C	Hexene	12	GTR [#]	49.8	47.4	49.1	1.5	***	***	***
			PC ⁺	25.8	23.5	21.7	1.1	***	***	***
			YMm ^a	98.9	111.1	115.1	2.5	***	***	***
			YMt ^b	109.6	128.4	136.5	4.0	***	***	***
D	Octene	25	GTR [#]	34.3	37.6	41.3	1.5	***	***	***
			PC ⁺	33.4	34.0	40.5	1.1	***	***	***
			YMm ^a	108.3	120.3	146.5	2.5	***	***	***
			YMt ^b	135.5	140.2	172.8	4.0	***	***	***
E	Octene	25	GTR [#]	34.9	31.6	34.5	1.5	***	***	***
			PC ⁺	38.4	33.4	35.6	1.1	***	***	***
			YMm ^a	115.9	117.8	124.8	2.5	***	***	***
			YMt ^b	117.8	127.3	125.3	4.0	***	***	***

[#]GTR = CO₂ gas transmission rate ((ml/m²/day/atm)(10³)) ⁺PC = CO₂ permeation coefficient (ml(STP)cm²(10⁻⁶))

^aYMm = Youngs modulus machine direction (MPa) ^bYMt = Youngs modulus transverse direction (MPa) ¹s.e.m = L. x F.

Film C exhibited the lowest Youngs modulus of all five films given its different co-monomer type. All films showed significant variation (P<0.001) in mechanical and gas permeation properties across the width of the roll, indicating non-perfect manufacture.

In conclusion, all films had similar permeation coefficient and Youngs modulus values, except for Film C which was pre-stretch and has a different co-monomer. The considerable variation in permeation and mechanical properties across the width of the roll for all films indicates an aspect of manufacture that needs improvement.

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Plastic film use on Irish farms

European Union policy strongly encourages a sustainable and multifunctional agriculture. Therefore, in addition to providing European consumers with quality food produced within approved systems, agriculture must also contribute positively to the conservation of natural resources and the upkeep of the rural landscape. Plastics are widely used in agriculture and their post-use fate on farms must not harm the environment - they must be managed to support the enduring sustainability of farming systems. This survey estimated the quantities of flexible plastic film used on Irish farms and their destinations post-primary use.

The survey was a supplement to the Teagasc National Farm Survey. The 1,167 farms involved represented 120,678 farms nationwide. The 961 farms responding to the plastics survey questionnaire were a stratified sample representative of farm enterprise, hectareage and geographic location. The survey quantified the primary and post primary use of polyethylene- and polypropylene-based products used to package fertiliser, mulch maize, tie silage bales and seal silage. It related to the crop growing season of 2001 and the following winter. Data were classified into two- or three-way tables using Statistical Analysis Systems and, where zeros and skewed distributions were absent, subjected to analysis of variance when appropriate.

Inorganic fertiliser was used on 97% of farms, averaging 14.8 t/farm (0.46 t/ha). Delivery was in plastic bags containing 50 or 500 kg or in bulk (i.e. no plastic) (Table 56). The mean weight was 2.4 and 2.2 kg plastic per tonne fertiliser packaged in bags containing 50 and 500 kg, respectively. Farm size and enterprise influenced ($P < 0.05$) the quantity of fertiliser and form of delivery. Maize was grown on 19700 ha, mainly on larger sized farms and dairy farms. Plastic mulch was used on 20% of this area (18% of maize farms). Punch plastic (*ca.* 12 μ m thick; 88 kg/ha; 55 t) and complete cover plastic (*ca.* 7 μ m thick; 51 kg/ha; 166 t) accounted for 16% and 84% of the area mulched, respectively. They were left to photo-degrade in the field. Silage was made on 87% of farms. Pit silage (68% silage area) was generally sealed beneath 2 sheets (57% new and 43% used once previously) of 125 μ m thick plastic and baled silage (32% silage area) was usually sealed under 4 layers of 25 μ m thick (before on-farm stretching) plastic (Table 57). Farm size and enterprise influenced ($P < 0.05$) plastic use per ha, partly due to changing the pit:baled silage ratio. There was 21 kg plastic used per ha baled silage (725 g/bale; 29 bales/ha) compared to 4.7 kg new plastic/ha pit silage. Moreover, use of new plastic decreased from 7.2 to 3.7 kg/ha pit silage as farm size increased from < 20 to ≥ 100 ha due to silos on larger farms having a greater area and height. About 1059 t plastic twine (*ca.* 58 g/bale) and netting (*ca.* 114 g/bale) were used in tying silage bales. Farms participating in the Rural Environment Protection Scheme (REPS) had a higher rate of recycling than other farms (Table 58).

Table 56: Plastic used for fertiliser delivery

Delivery unit size	50 kg	500 kg	Bulk
Fert. delivered (t)	964562	541039	205773
% of farms	89	17	8
% of fertiliser	56	32	12
Plastic used (t)	2315	1190	0

Table 57: Plastic (t) used for covering silage

Plastic sheets for silage pits - new	4,242
- old	3,222
Plastic stretch film for baled silage	8,617
Total plastic used (i.e. old + new)	16,081
Total plastic sold (i.e. new used)	12,859

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Table 58: Post primary use (%) of plastic

	REPS	Non-REPS	All
Silage sheets			
- recycled	49	27	33
- municipal dump	9	3	5
- on-farm disposal	36	65	57
- other	6	5	5
Bale stretch-film			
- recycled	54	33	40
- municipal dump	9	5	6
- on-farm disposal	32	60	51
- other	5	2	3
Twine and netting (for baled silage)			
- recycled	14	4	7
- municipal dump	10	3	5
- on-farm disposal	73	92	86
- other	3	1	2
Bags for 50 kg fertiliser			
- recycled	7	6	7
- municipal dump	6	2	3
- on-farm disposal	37	51	46
- other	50	41	44
Bags for 500 kg fertiliser			
- recycled	8	8	7
- municipal dump	11	2	4
- on-farm disposal	67	81	79
- other	14	9	10

Nationally, estimated total (mean) plastic use was 4,242 t (4.7 kg/ha) for new pit silage sheets, with corresponding values of 8,617 t (21 kg/ha) for baled silage stretch-film, 1,059 t (2.5 kg/ha) for baled silage twine/netting, 221 t (56 kg/ha) for maize mulch and 3,505 t (1.1 kg/ha) for fertiliser bags. A range of post primary uses of these plastics occurred which differed depending on the primary use of the plastic and the type of plastic.

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Genatex® as a protective sheeting for silage

Forage ensiled in bunker or clamp silos is conventionally sealed beneath two sheets of black 0.125 mm polythene (IS 246 1989). These are overlaid with a layer of touching tyres. The latter prevent movement of the polythene sheets that could draw air across the top surface of the silage. They also provide limited protection from some vertebrates. This system can produce silage free of visible surface waste and involves relatively modest material costs (tyres normally obtained free-of-charge). However, labour input in placing and removing the tyres is an additional cost. Some countries, due to perceived environmental risks, require farmers to have a licence to place tyres over silage. Tyres are susceptible to degradation over time and reports in the Veterinary Record indicate the diagnosis of pericarditis and traumatic reticulitis in cows which was attributed

to ingestion of tyre wire fragments with silage. Genatex® (Westfalia Farm Ireland Ltd., Coolroe, Ballincollig, Cork) is a polyethylene woven black mesh sheet made from a combination of tape and monofilament. Its efficacy as a protective sheeting for silage was evaluated in terms of its effects on silage surface waste compared to the standard system of using a complete layer of tyres.

Two outdoor, walled, concrete silos (each 24.2m long x 6.5m wide) were filled with grass (16 June and 5 August, 2003) to a mean height of 2.4m and sealed beneath two sheets of black polythene. Polythene was not placed along the inner surface of the silo walls before filling with grass. The two sheets were securely weighted around their edges with a continuous layer of silt. Their top surface (excluding ramp) was divided into four bands (6.5m wide x 5.0m long), and alternate bands were covered with car tyres or Genatex®. The latter had a row of weights placed around its edges and along its centre. The silos were opened in early November and early January and the silage removed during a 65 and 61 day duration, respectively. During feedout, polythene was maintained tightly in place on top of the silage, but was kept off the silage face. Three silage samples per silo were chemically analysed. On two faces per band, the pH of the top 1m of silage was profiled (in 0.1m horizons and 0.5 to 1.0 m widths). Because of the probable non-independence of the values for corresponding grid positions in the two faces per band, these values were treated as duplicate estimates and averaged. Data for each grid position were analysed for treatment and block (i.e. silo) effects using a General Linear Model with four estimates/treatment. A similar procedure was followed when obtaining one value per band for determining treatment effects within horizon.

The mean (s.d.) composition of the silage in each silo was dry matter (DM) 185 (2.6) and 239 (4.7) g/kg, pH 3.9 (0.06) and 3.9 (0.15), crude protein 133 (7.9) and 152 (4.6) g/kgDM and *in vitro* DM digestibility 640 (4.2) and 618 (38.5) g/kg. The pH profile of the top 0.8m silage is summarised in Table 59. The value for each position on the 0.9 and 1.0 m horizons was 3.9 (s.e. 0.01). Elevated pH likely reflects aerobic metabolism of fermentation acids and breakdown of N products. In general, higher pH values were found towards the top and sides of the silage reflecting the greater access by oxygen in these locations. Higher variances (s.e.) were also found towards the sides and top of the silages. Treatment effects were not significant ($P>0.05$; Table 59) at any of the 110 grid positions examined in the top 1m of silage. Similarly, when mean values for each horizon were compared, there was not a significant ($P>0.05$) treatment effect.

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Table 59: pH profile of top 0.8m of silage feed face

H ¹	Incremental distances from left-hand side of feed face (m) ²									
	0.5	1.0	1.5	2.0	3.0	4.0	5.0	5.5	6.0	6.5
Tyres - mean value for each grid position on feed face										
0	6.0	4.9	4.4	4.3	4.2	4.0	4.2	4.2	4.9	6.0
0.1	6.0	4.5	4.2	4.3	4.2	3.9	4.2	5.2	4.7	5.8
0.2	5.4	4.7	4.2	3.9	3.9	3.9	4.2	3.9	4.5	6.0
0.3	5.2	4.0	3.9	3.9	3.9	3.9	3.9	3.9	4.2	5.0
0.4	5.0	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	4.8
0.5	4.5	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	4.4
0.6	4.1	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
0.7	4.0	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
0.8	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
Genatex® - mean value for each grid position on feed face										
0	6.0	4.6	4.5	3.9	4.1	3.9	4.0	4.3	4.8	6.0
0.1	6.0	4.5	4.0	3.9	3.9	3.9	3.9	5.2	4.5	5.6
0.2	5.3	4.4	4.2	3.9	3.9	3.9	4.2	3.9	4.5	5.6
0.3	4.2	4.1	3.9	3.9	3.9	3.9	3.9	3.9	3.9	4.5
0.4	4.2	3.9	3.9	3.9	3.9	3.9	3.9	3.9	4.2	5.0
0.5	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	4.0	3.9
0.6	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
0.7	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
0.8	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
Standard error of mean value for each grid position on feed face										
0	³	.38	.43	.15	.20	.05	.18	.29	.30	³
0.1	³	.43	.18	.15	.19	.01	.19	.23	.39	.27
0.2	.32	.44	.19	.01	.01	.01	.19	.01	.32	.32
0.3	.50	.13	.01	.01	.01	.01	.01	.01	.19	.26
0.4	.32	.01	.01	.01	.01	.01	.01	.01	.19	.28
0.5	.21	.01	.01	.01	.01	.01	.01	.01	.08	.16
0.6	.11	.01	.01	.01	.01	.01	.01	.01	.01	.01
0.7	.05	.01	.01	.01	.01	.01	.01	.01	.01	.01
0.8	.01	.01	.01	.01	.01	.01	.01	.01	.01	.01

H¹: horizon of feed face (m; relative to top surface); ²i.e. 0.5 = 0-0.5, 1.0 = 0.5-1.0, etc.; ³mean = 6.0 or higher and no estimate of variance.

Genatex® was as effective as the standard system involving tyres for restricting aerobic deterioration on the top surface of silage. It was observed that polythene placed between the silo wall and the forage at ensiling, and folded onto the top of the forage, was required to limit aerobic losses below the top corners of the silage.

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RMIS No. 5137

Investigating development options for Irish suckler beef producers using mathematical programming

The changes in European agricultural policy brought about by the Luxembourg Agreement (LA) of 2003 have led to uncertainty with regards the future profitability of beef production in Ireland. A mathematical programming model, the Grange Beef Model, has been used to investigate the

impact of farm developments on the profitability of beef farms in Ireland within the policy environment of the Luxembourg Agreement.

Three development and expansion options open to beef farmers were examined in addition to one scenario where de-stocking occurs and a scenario where the farmer opts to remain static. A feature of all scenarios is the importance of grazed grass in the annual feed budget. In addition, rental land is crucial in the expansion scenarios. Whilst providing access to supplementary forage hectares, this land is also necessary to maintain total N usage below 260kg./ha. If land is not available to rent, expansion is not possible within the REPS compatible systems simulated in this study.

In general the financial results indicate a difficult period of adjustment for suckler beef farmers from 2005 to 2012. System 1 may be used as a benchmark since in this scenario the production system does not change. Results indicate little nominal change in farm margins for System 1 up to 2008. Following 2008, the increase in beef price results in improvements in profitability. By 2012 the farm margin is 135% of the farm margin in 2005.

It has been suggested that farmers may partially or fully de-stock following the implementation of LA since the requirement to stock animals for the purposes of collecting premia no longer exists. System 2 represents a partial de-stocking scenario. Whilst margins in 2012 are the lowest of the five scenarios, this may remain a potentially attractive option on some farms given the reduced labour requirements associated with this scenario. Many beef farmers have off-farm jobs with 51% of total beef farmers reported to have off-farm jobs in 2003. It is possible that some current full-time farmers may also opt for this scenario and take up off-farm employment.

The high cost associated with the expansion scenarios result in negative farm margin in 2006 and low margins in 2007 through to 2009. Although the margins for these scenarios recover to around €35,000 in 2012, it is unlikely farmers would accept these scenarios given the margins in 2006 to 2009. The cost of expansion is clearly an issue and if these costs can be reduced expansion of the scale modelled in this study may become more attractive to beef farmers. It is likely that as farmers de-stock, opportunities will arise for farmers wishing to expand to source suckler cows at a lower cost than the cost of replacement heifers as used in this study. Alternatively, farmers may elect to expand at a slower rate than that used in the scenarios investigated in this case using replacements sourced from within the herd.

Closer investigation of System 4 and System 5 indicates that the form of capital development has a relatively modest impact on the overall financial performance of the enterprise relative to the operating costs of herd expansion. As expected, the low cost option modelled in System 5 results in higher margins for this scenario when compared to System 4. Expansion at a greater scale than modelled here would further exaggerate the differences in the two scenarios. For the purposes of comparison in this study, no grant aid was assumed. The provision of grant aid would improve the attractiveness of these scenarios to farmers.

Thus, it is concluded that farm margins will come under increasing pressure in the years 2005 to 2012 and that expansion may be hindered by high costs associated with these scenarios.

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The impact of concentrate price on the utilisation of grazed and conserved grass

A linear programming model was designed and constructed to facilitate the identification of optimal beef production systems under varying technical and policy scenarios. The model operates at a systems level and most activities that could occur in Irish spring-calving, suckler beef production systems are included. In this summary, the components of the model are described together with a simple application of the model involving changing concentrate prices.

The model was developed using a mathematical programming methodology which encompasses the following characteristics: 1) a range of possible activities, 2) various constraints to prevent free selection from the range of activities, and 3) an objective which can be quantified. It is a single year steady-state design. The fundamental unit on which the model is based is the cow unit. Due to the predominance of pasture-based systems in Ireland a detailed set of grazing options that are typical of those available to Irish cattle farmers is specified. Model details are specified on a monthly basis. This enables it to respond to monthly fluctuations in feed supply and animal requirements. Financial budgets assign a cost or revenue to each activity and thus the program identifies the optimal net farm margin. Nutritional specifications are described in terms of net energy (NE) requirements subject to a maximum intake capacity.

A scenario investigating the impact of a change in concentrate price on optimal systems is presented. Concentrates are generally regarded as the most expensive feedstuff and farm margin and system operated are influenced to a large extent by the cost of concentrates. The influence of concentrate price on the optimal system is presented below (Table 60) together with the resulting impact on net margin (Figure 2). Above €140/tDM it was found that there was no response to further increases in concentrate price. Therefore, results are presented for the range €100/tDM to €140/tDM.

Table 60: Production results for concentrate price change scenario

Concentrate price (€/tDM)	100	105	110	115	120	125	130	135	140
Area for grazing (ha)	60.0	60.0	60.0	50.0	50.0	49.0	45.3	35.7	35.7
Area for grass silage (ha)	0.0	0.0	0.0	0.0	0.0	1.0	4.7	14.3	14.3
Land rented (ha)	10.0	10.0	10.0	0.0	0.0	0.0	0.0	0.0	0.0
Total N applied (kg/ha)	199.4	196.3	196.3	196.3	196.3	202.4	217.7	243.4	243.4
Concentrates fed (t)	94.5	84.5	84.5	70.4	70.4	58.8	39.1	6.0	6.0
Suckler cow numbers	61.3	61.2	61.2	51.0	51.0	53.7	56.4	56.2	56.2

Up to around €120/tDM all animals are finished on concentrate based diets but above this steer progeny are finished off grass and from about €130/tDM all progeny are finished off grass. Stock numbers initially decrease with an increase in concentrate price but recover somewhat following the change to grass-based finishing.

It is concluded that the price of concentrates has a crucial impact on optimal systems driving both the finishing system operated and grass silage requirements. The model can also be used to analyse current or prospective scenarios of interest. Future changes in agricultural policy can be routinely investigated. Whilst much of the production data is based on performances obtained at Grange Research Centre, the parameters can be modified to reflect other situations.

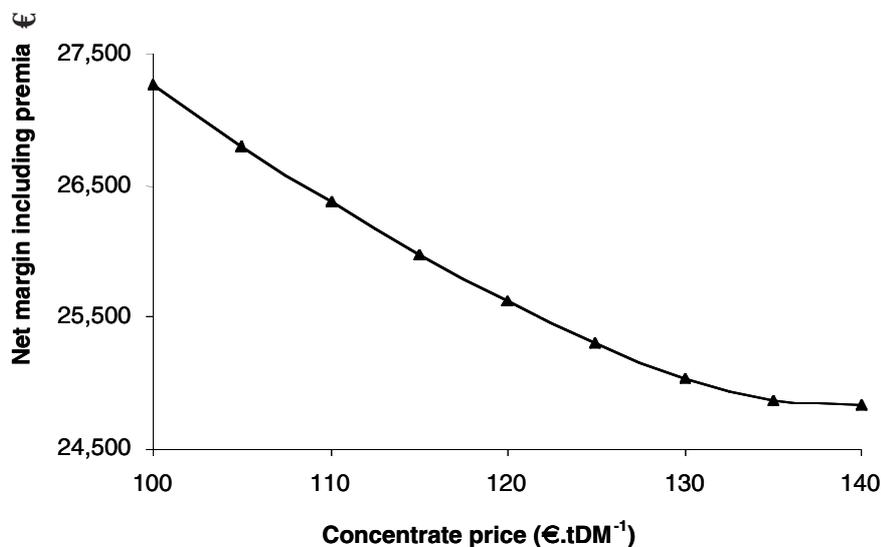


Figure 2. Change in net margin with increasing concentrate price

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RMIS No. 5137

Grass and maize silages in Ireland

This survey identified the types of farm making grass silage, characterised how it is made and highlighted the interactions between these factors. It also categorised the relationship between maize silage and both farm enterprise and farm size. The results are based on data collected in the Teagasc National Farm Survey. The latter was conducted during 2002 and obtained answers to questions relating to the year 2001. It involved 1,167 farms that represented a total population of 120,678 farms nation-wide.

The popularity of silage was influenced both by the dominant enterprise on farms (Table 61) and by farm size (Table 62). At least twice as much land was used for silage-making on farms involved in dairying (20 ha or more) compared to other enterprises, with the smallest areas being on cattle rearing (7.9 ha) or sheep (7.1 ha) farms. Farms of less than 20 ha were less likely to make silage (78%) than farms larger than 50 ha (90-95%). As might be expected, the area of land used for making silage increased as farms size gets larger. The average proportions of the total area of silage harvested for first, second and subsequent cuts were 78, 21 and 1% respectively. The high proportion of the total silage area harvested for first cut was a recent significant increase from the values recorded in 1996 (72%) and 1999 (71%). The recent proportions vary among enterprise, with dairy farms placing the highest emphasis on taking a second cut (69:30:1) and sheep farms the least (92:8:0).

The average area of grass silage made using precision-chop harvesters, single- or double-chop harvesters or round balers was 7.3, 1.0 and 3.9 ha/farm respectively, when expressed relative to the total number of farms making silage (Tables 61 and 62; e.g. on average, 7.3 ha/farm were harvested using a precision-chop harvester on farms where some silage was made). If the areas harvested by each individual system were expressed relative to the number of farms using that

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system, the corresponding values were 18.1, 9.9 and 5.3 ha/farm (e.g. on average, 18.1 ha/farm were harvested using a precision-chop harvester on farms where the precision-chop system was used). Farms involved in dairying (68-71%; Table 61) and larger sized farms (68-71%; Table 62) were the most likely to make precision-chop silage and to have a greater area of grassland allocated to this system of harvesting. This resulted in approximately three-quarters of the silage area on dairy farms and on larger sized farms being made with a precision-chop harvester. Single- and double-chop harvesters were much less common than heretofore, being made on 11% of farms. They were not particularly popular within any enterprise or farm size category. Most baled silage was made in large round bales, with large square bales accounting for only 1.2% of all baled silage (Table 63). Almost no silage was made using small bales. Round big bale silage was made on 74% of all farms and although it was popular across all enterprises (Table 61) and farm sizes (Table 62), it was particularly common on cattle rearing (84%) and sheep (81%) farms and smaller sized (82%) farms. Round bale silage accounted for the smallest proportion of land area allocated to silage on farms involved in dairying (14-18%) and those of 50 ha or more (18%), whereas farms majoring in cattle rearing (58%) or sheep (55%) or with a total area below 20 ha (63%) had the largest proportion of their silage area harvested as baled silage. Thus, baled silage was the primary silage-making system on beef farms and smaller sized farms, and was the secondary system on dairy farms and larger sized farms. In many cases where it was a secondary system, it was being used tactically to remove grass from paddocks that were surplus to the short-term needs of the herd (thereby facilitating improved grazing management and overall animal nutrition) or to remove small yields that would be difficult or expensive to successfully ensile in conventional silos. An estimate based on the data from this survey is that 12.3m bales (round + square) were made in 2001, with an average of 153 bales per farm making baled silage and an average of 29 bales per ha. The latter value does not seem to vary strongly with farm enterprise or farm size. More bales were made on larger than on smaller farms (Table 64), and farms with cattle or cows generally had more bales than sheep farms which in turn had more bales than tillage farms (Table 63). The pick-up wagon has made a re-entrance to the Irish market in recent years. It accounted for approximately 1% of the silage area, being more prevalent on farms in the 20-100 ha size category and on those with dairying or cattle rearing.

Most grass silage (89%) was made without the use of an additive and this characteristic held across enterprise (Table 61) and farm size (Table 62) categories. It continues a trend evident from values of 58% in 1991/92, 73% in 1996 and 84% in 1999. Farms classified as being mainly dairying or in the size category 50-<100 ha were more likely to have additives used during silage-making than other farm categories. Almost no additives were used in making baled silage, which means that where additives were used it was with precision-, single- or double-chop silages. Where additives were used, the general trend was for biological additives to be used the most, followed by molasses or (beet or citrus) pulp and with the area of grassland treated with acid additives being quite small. The use of additives other than these was uncommon.

The presence of a highly specialised and commercial agri-contractor service was vital to the continued viability of current Irish ruminant production systems. This was particularly evident on dairy and beef farms, but was also important on most sheep farms. Overall, 86% of all grass silage was harvested by contractors, with the remaining 14% harvested by farmers using their own machinery. Where only conventional (single-, double- and precision-chop combined) silage was made, contractors were particularly important in making silage on beef (92-95%), sheep (87%) and dairy (77-84%) farms but less significant on tillage (43%) farms (Table 63). Their importance was more evident on small (98%) compared to large (50%) farms (Table 64). On farms where only baled silage was made, an average of 89% of silage was made by contractors, with a higher proportion than this on smaller sized farms.

Maize silage was estimated to have been grown on 19,700 ha in 2001 according to CSO data. The largest area was on farms involved in dairying, followed by tillage and finally by cattle (Table 65). It was absent on small farms and the area per farm increased as farm size got larger. Despite the significant increase in the area of forage maize in recent years, it still accounted for a relatively modest proportion of the total area of forage conserved as silage. Thus, forage maize accounted for about 1.5% of the total area harvested as silage (i.e. grass + maize) and about 1.9% of the total area allocated for silage. These proportions likely underestimate its contribution since a much higher yield of dry matter per ha should be expected with forage maize than from single cuts of grass silage. In addition, maize is concentrated more on dairy farms (and in particular on those involved in winter milk production) and should therefore be making a larger contribution to overall feed supply in those circumstances.

Table 61: Scale and characteristics of grass silage production within different farming enterprises

	Dairying	Dairying/ cattle	Cattle rearing	Cattle fattening	Mainly sheep	Tillage	All systems
Total no. of farms represented	19516	13819	33887	28681	17109	7667	120678
No. of farms making silage	19272	13573	30413	23586	12093	5756	104693
- % farms making silage	99	98	90	82	71	75	87
Area of silage (ha)							
- first cut	282239	195517	211266	200813	78306	45978	1014119
- second cut	120590	74359	28438	33040	6891	9694	273012
- later cut	7756	1715	0	0	0	625	10096
- total	410585	271591	239704	233853	85197	56298	1297227
% of national silage area							
- precision-chop	74	77	30	49	30	61	59
- single/double chop	7	7	11	8	15	6	8
- round baler	18	14	58	42	55	31	32
- other	1	2	1	1	0	2	1
% silage area treated with additive							
- none	81.3	88.0	96.5	94.1	98.4	89.9	89.3
- acid	3.3	0.9	0	1.2	0.1	0.3	1.5
- molasses/pulp	4.6	5.9	2.8	2.5	0	1.4	3.7
- biological	10.2	5.2	0	1.5	1.5	8.4	5.0
- other	0.2	0	0.7	0.7	0	0	0.3

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Table 62: Scale and characteristics of grass silage production within different farm size categories

Farm size (ha)	< 20	20 to <30	30 to <50	50 to <100	≥100	All sizes
Total no. of farms represented	43965	23853	29092	19148	4620	120678
No. of farms making silage	34051	21321	27015	18155	4152	104693
- % farms making silage	78	89	93	95	90	87
Area of silage (ha)						
- first cut	147165	151036	287495	314752	113670	1014119
- second cut	24825	28200	70108	114095	35785	273012
- later cut	292	0	2581	5215	2008	10096
- total	172282	179235	360185	434062	151464	1297227
% of national silage area						
- precision-chop	29	41	56	75	74	59
- single/double chop	6	16	8	7	8	8
- round baler	63	41	35	18	18	32
- other	1	2	1	1	0	1
% silage area treated with additive						
- none	96.5	92.9	92.5	82.8	87.7	89.3
- acid	1.6	2.7	1.3	0.8	2.3	1.5
- molasses/pulp	1.8	1.8	2.7	4.8	7.8	3.7
- biological	0	2.3	3.1	10.8	2.0	5.0
- other	0	0.3	0.5	0.5	0	0.3

Table 63: Contractor and baled silage characteristics within different farming enterprises

	Dairying	Dairying/c attle	Cattle rearing	Cattle fattening	Mainly sheep	Tillage	All systems
Farms with baled silage							
Mean bales per farm	164	140	160	156	144	115	153
Total no. bales	2233166	1198773	4133210	2800816	1420390	522908	12309263
% bales as big square bales	1.1	3.0	0.2	2.8	0	0	1.2
Farms with only baled silage							
Mean bales per farm	276	165	147	125	94	72	133
Farms with conventional silage							
% area as baled silage	11.0	9.4	14.0	14.9	9.8	13.5	11.6
% total silage area harvested by contractors	83	85	92	90	84	77	87
Farms with only conventional silage							
% of silage area harvested by contractors	77	84	92	95	87	42	84

Table 64: Contractor and baled silage characteristics within different farm size categories

	< 20	20 to <30	30 to <50	50 to <100	≥100	All sizes
Farms with baled silage						
Mean bales per farm	116	133	179	192	285	153
Total no. bales	3347417	2101104	3699220	2350816	810706	12309263
% bales as big square bales	2.2	2.5	0.7	1.7	0.3	1.2
Farms with only baled silage						
Mean bales per farm	88	128	216	247	284	133
% baled by contractor	92	93	85	81	62	89
Farms with conventional silage						
% area as baled silage	8.6	13.4	13.6	10.2	11.6	11.6
% total silage area harvested by contractors	97	92	91	81	70	86
Farms with only conventional silage						
% of silage area harvested by contractors	98	89	82	75	50	84

Table 65: Scale and characteristics of maize silage production within different enterprises and farm size categories

Enterprise	Variable	30-<50ha ¹	50-<100ha	100+ha	All sizes
Dairying	% farms in category	3	21	29	6
	ha maize per farm	3.5	5.7	13.6	6.3
Dairying and other	% farms in category	0	12	30	6
	ha maize per farm	0	6.0	10.1	7.4
Cattle and Other	% farms in category	3	3	0	1
	ha maize per farm	2.8	8.1	0	4.8
Mainly Tillage	% farms in category	0	12	33	8
	ha maize per farm	0	6.7	5.9	6.2
All systems	% farms in category	1	9	18	3
	ha maize per farm	3.2	6.0	9.1	6.5

¹No maize was recorded on farms of less than 30 ha

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Yield and composition of forage maize: interaction of harvest date, cultivar and plastic mulch

Forage maize is established as a crop with the potential to consistently supply high yields of quality forage on some Irish farms. Despite its success, considerable variability in crop yield, quality and maturity at harvest can exist from year to year. These reflect differing prevailing weather conditions, particularly temperature during May to September. The use of plastic mulch has increased the likelihood of achieving higher yields of high quality maize crops and has permitted its sowing into areas once considered unsuitable for the crop. In this experiment two cultivars of differing maturity were grown with or without plastic mulch to examine how yield and composition altered during the harvest window of early September to early November.

Two forage maize cultivars of differing maturity under Irish conditions (Tassilo: FAO 210(early) and Benicia: FAO 270(late)) were grown at Teagasc, Oak Park in 2002. Each plot consisted of 4 rows (70cm spacing) of 5m length sown in duplicate blocks either uncovered (NP) or under complete-cover clear polythene mulch (P; 6 micron; IP Europe Ltd) on 24 April using a Samco precision seed drill at a seed rate of 100,000 seeds/ha. Standard fertiliser (150kg N, 50kg P, 200kg K/ha) and weed control (4.5l atrazine/ha) were both applied pre-sowing. Crop samples (2x1m per plot) were taken every 10 days from 10 September to 9 November. Data were analysed as a repeated measures analysis of variance using Genstat 7th Edition.

Plastic mulch increased ($P<0.001$) crop DM yield, the proportion of cob in crop DM, crop starch content and cob DM content for both cultivars, as shown in Figures 3-6. The late cultivar Benicia demonstrated the greater increase ($P<0.05$) in cob proportion and starch content when sown under plastic cover. As harvest date was delayed an increase ($P<0.001$) in cob DM content, cob proportion in DM and starch content was observed (Figures 4-6). Yields of DM increased initially

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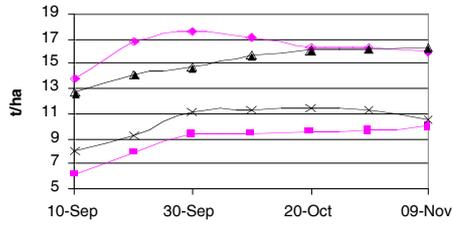


Figure 3. Crop DM yield over time

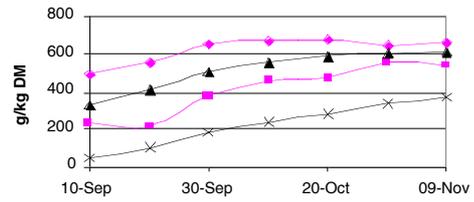


Figure 4. Cob DM in crop over time

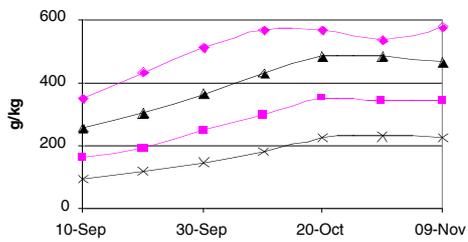


Figure 5. Cob DM content over time

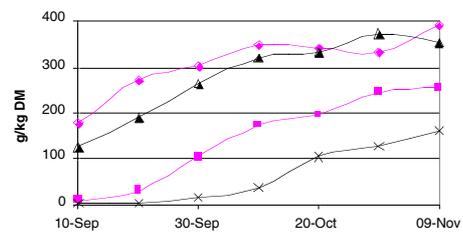


Figure 6. Crop starch content over time



but remained constant once peak yield was achieved which tended to be before mid-October (Fig. 3). Cultivar type did not influence ($P>0.05$) overall DM yield but the early cultivar Tassilo did have increased ($P<0.001$) cob DM, cob proportion and to a lesser extent increased ($P<0.05$) starch content compared to Benicia under both sowing regimes.

Plastic mulch increased crop DM yield, cob proportion and starch content and advanced cob ripeness (increased cob DM content) in both cultivars. Little yield benefit was obtained from prolonging harvest after 20 October, however, cob maturity (i.e. starch content) of the plants not grown under plastic (NP) continued to increase. Tassilo (early) was about three weeks more advanced in terms of cob ripeness than Benicia (late) when grown under plastic. Benicia grown without plastic mulch did not mature adequately and was unsuitable for growing without plastic mulch.

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Cob development in forage maize: influence of harvest date, cultivar and plastic mulch

Forage maize grown for silage tends to be a compromise between reproductive and vegetative yield, and the cob component is the main driver of feeding value (Keane et al., 2003). Thus the aim is to produce a well-developed crop of high dry (DM) matter and starch content reflecting large cobs of well-filled grains rather than crops with low DM and starch contents reflecting poorly developed (immature) cob components at harvest. The use of plastic mulch can increase total DM yields with the increase in cob yield accounting for 75% of the total yield increase (Easson & Fearnough, 1997). In this experiment the composition of cob components (i.e. rachis plus kernel) of two cultivars of different maturity under Irish conditions grown with or without plastic mulch were monitored between the harvest dates of 10 September to 9 November.

Two forage maize cultivars of different maturity under Irish conditions (Tassilo: FAO 210(early) and Benicia:FAO 270(late)) were grown at Teagasc, Oak Park in 2002. Each plot consisted of 4 rows (70cm spacing) of 5m length sown in duplicate blocks either uncovered (NP) or under complete-cover clear polythene mulch (P; 6 micron; IP Europe Ltd) on 24 April using a Samco precision seed drill at a seed rate of 100,000 seeds/ha. Standard fertiliser (150kg N, 50kg P, 200kg K/ha) and weed control (4.5l atrazine/ha) was applied pre-sowing. At 10-day intervals from 10 September to 9 November the cob component was removed from plants from a one metre length per plot. *In vitro* DM digestibility (DMD) was measured using the Tilley and Terry (1963) two-step method and acid detergent fibre (ADF) was measured using the Ankom fibre analyser. Data were analysed as a repeated measures analysis of variance using Genstat 7th Edition.

As harvest date was delayed cob starch content increased ($P<0.001$) and cob DMD and ADF generally decreased ($P<0.001$) (Figs 7-9). However, cob ADF did increase initially in uncovered plants before decreasing. Plastic mulch increased ($P<0.001$) cob starch content, this effect being most evident with Benicia ($P<0.01$) particularly in early September. It also reduced ADF ($P<0.01$) and DMD ($P<0.05$). Tassilo generally had a higher starch and lower DMD and a similar ADF to Benicia.

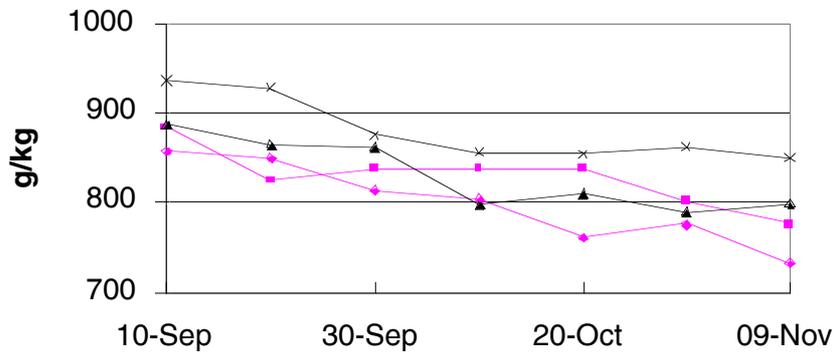


Figure 7. Cob *in vitro* DMD over time

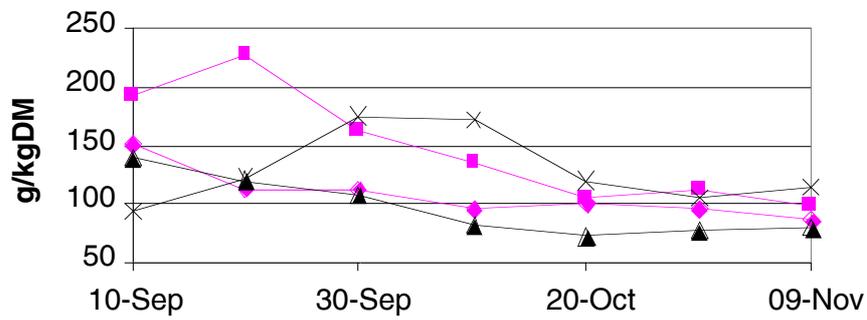


Figure 8. Cob ADF content over time

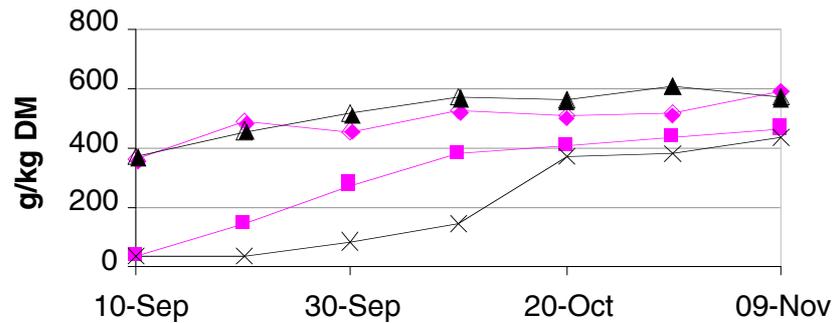
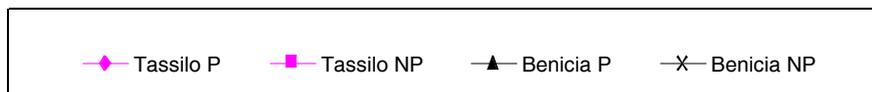


Figure 9. Cob starch content over time



In conclusion, a progressive rise in starch content was observed over time and was most evident in uncovered plants reflecting the greater maturity of covered plants. A corresponding decrease in ADF was observed as starch rose. The initial rise in cob ADF of uncovered plants could indicate the later, final stages of rachis development when compared to those under plastic cover. The temporal decline in cob *in vitro* DMD was quite large and may reflect decreasing degradability of starch and/or increasing indigestibility of the rachis as the cob matures.

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Composition of maize stover: interaction of harvest date, plastic mulch and cultivar

Currently just under 20,000 ha of forage maize is grown in Ireland with almost all of this conserved as silage. The grain portion is the main driver of feeding value in forage maize. Despite the considerable importance of the cob component the relevance of *ca.* 0.5 of crop dry matter (DM) present in the stover (stem and leaf fraction) must also be taken into consideration. The aim of this experiment was to evaluate the effects of harvest date, plastic mulch and cultivar on stover composition and proportion in the crop DM.

This experiment took place at Teagasc Oak Park during 2002 where two forage maize cultivars of different maturity under Irish conditions (Tassilo: FAO 210 (early) and Benicia: FAO 270 (late)) were sown uncovered (NP) or under complete cover clear polythene mulch (P; 6 micron; IP Europe Ltd). Each plot consisted of 4 rows (70cm spacing) of 5m length. Plots were sown in duplicate on 24 April using a Samco precision seed drill at 100,000 seeds /ha. Standard weed control (4.5l atrazine /ha) and fertiliser (150kg N, 50kg P, 200kg, K) was applied pre sowing to all plots. Samples of one metre length per plot were taken at ten day intervals from 10 September to 9 November. The cobs (grain and rachis) were removed and the remaining proportion of stover in the crop DM was calculated. Samples were dried, milled and assayed for *in vitro* DM digestibility (DMD; Tilley and Terry, 1963) and neutral detergent fibre (NDF; measured using the Ankom fibre analyser). Data were analysed by repeated measures analysis of variance using Genstat 7th Edition.

Delaying harvest date from early September until November resulted in a decrease ($P < 0.001$) in the proportion of stover in the crop DM and a reduction ($P < 0.001$) in stover DMD (Table 66). Stover NDF correspondingly increased ($P < 0.001$) over time. Stover DM increased in early November after air frost damage intensified leaf senescence. The use of plastic mulch did not influence ($P > 0.05$) stover DM content but it did reduce ($P < 0.001$) the proportion of stover in the DM. Plastic mulch use also affected quality, producing a higher ($P < 0.001$) NDF and a lower ($P < 0.01$) DMD than uncovered plants. Cultivar had no effect ($P > 0.05$) on stover DM or NDF concentration but the earlier cultivar Tassilo generally had a higher ($P < 0.01$) DMD and a lower ($P < 0.001$) stover proportion in the DM. The response of stover proportion, DMD and NDF to plastic mulch was not consistent over all harvest dates and in the case of NDF and DMD varied with cultivar.

In conclusion, delaying harvest date reduced the contribution of the stover to crop DM yield and considerably reduced its DMD and increased its NDF over the same period. Plastic mulch advanced the plant growth stage and level of cob development thus reducing the contribution of the stover to crop DM yield and reducing stover DMD on any given date. Stover in the later

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cultivar, Benicia, accounted for a greater proportion of crop DM yield, in particular when grown without plastic mulch, but its DMD was lower when compared to the earlier cultivar Tassilo.

Table 66: Stover composition over time

Harvest date	Cultivar	Plastic mulch	Stover in crop ¹	DM ²	DMD ²	NDF ³
(H)	(C)	(M)				
10-Sep	Tassilo	NP	765	183	703	559
	Tassilo	P	510	187	698	572
20-Sep	Benicia	NP	953	180	677	599
	Benicia	P	665	197	687	581
	Tassilo	NP	783	202	734	514
	Tassilo	P	446	190	683	589
30-Sep	Benicia	NP	896	196	691	566
	Benicia	P	587	195	679	591
	Tassilo	NP	620	185	707	554
	Tassilo	P	343	186	634	676
10-Oct	Benicia	NP	814	195	665	562
	Benicia	P	489	186	625	637
	Tassilo	NP	542	168	650	614
	Tassilo	P	329	179	611	712
20-Oct	Benicia	NP	754	191	629	628
	Benicia	P	439	172	569	710
	Tassilo	NP	526	198	629	651
	Tassilo	P	323	226	624	713
30-Oct	Benicia	NP	720	210	592	645
	Benicia	P	410	188	589	738
	Tassilo	NP	446	196	588	741
	Tassilo	P	352	197	536	769
9-Nov	Benicia	NP	657	196	548	688
	Benicia	P	394	202	531	772
	Tassilo	NP	460	240	534	774
	Tassilo	P	339	292	523	781
9-Nov	Benicia	NP	630	227	574	729
	Benicia	P	384	269	483	787
Sig	H		***	***	***	***
	C		***	NS	**	NS
	M		***	NS	**	***
	HxC		NS	NS	NS	NS
	HxM		*	NS	*	**
	CxM		**	NS	NS	NS
	HxCxM		NS	NS	**	*
s.e.m	HxCxM		19.0	7.5	9.8	9.3

¹gDM/kgDM; ²g/kg; ³g/kgDM

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The feeding value of conserved whole-crop wheat and forage maize relative to grass silage and *ad-libitum* concentrates for beef cattle

Grass is the predominant forage ensiled in Ireland. However, the relatively modest yields achieved in a single harvest allied to variability in digestibility and ensilability (and thus in intake and animal performance response) and the likelihood of effluent production create disadvantages for grass silage compared to the potential of some alternative forage crops. Thus, alternative forages are worthy of consideration on many farms. The objectives of this study were to quantify the relative intake, digestibility (results not presented) and performance of beef cattle offered grass silage, forage maize silage and whole-crop wheat (fermented or urea-treated), rank these relative to cattle offered an *ad libitum* concentrate based diet and compare the “alkalage” system of urea treated processed whole-crop wheat with whole-crop wheat silage.

Seventy continental cross-bred beef steers, mean initial live weight 424 (sd 33.0) kg, were blocked for live weight and breed and allocated to one of 5 dietary treatments in a randomised complete block design. Treatments were grass silage (GS), maize silage (cv. Benecia) (MS), fermented whole-crop wheat (cv. Soissons) (FWCW), alkalage whole-crop wheat (cv. Soissons) (ALK) and *ad-libitum* concentrates (ALC). The four forages were precision-chop harvested. The ALK harvester was fitted with a grain processor and ensiled with 45kg Home ‘N’ Dry (Volac International Ltd.) /t DM. Forages were offered *ad-libitum* through individual Calan gates and supplemented with 3kg concentrates/head/day. The ALC treatment was supplemented with 5kg grass silage/head/day throughout the 160-day trial period. The mean DM (g/kg) (uncorrected for volatiles) of the GS, MS, FWCW and ALK were 161, 303, 391 and 705 respectively. *Ad-libitum* concentrate composition was 830g rolled barley, 100g soya-bean meal, 50g molasses and 20g minerals & vitamins /kg (DM 838 g/kg) and the concentrate supplement was 650g rolled barley, 280g soya-bean meal, 50g molasses and 20g minerals & vitamins /kg (DM 839 g/kg). Liveweight was recorded every 3 weeks and starting and finishing live weight calculated as the mean of two consecutive day’s weighings. Blood samples were taken from all animals mid-way through the experiment. The data were analysed using analysis of variance taking account of diet and block.

Total DM intake and carcass growth were lowest for GS ($p < 0.001$) (Table 67). Relative to ALC, GS, FWCW and ALK had a poorer ($p < 0.05$) FCE, lower liveweight ($p < 0.05$) and carcass ($p < 0.01$) gain and a poorer ($p < 0.05$) kill-out proportion. Despite ALK having the highest ($p < 0.05$) forage DM intake, kill-out proportion and rate of carcass gain were lower ($p < 0.05$) than MS. MS had a better FCE than the ALK ($p < 0.001$) or the FWCW ($p < 0.05$). Plasma urea concentration was lowest for MS and highest for ALK ($p < 0.001$).

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Table 67: Feed DM intake, growth, kill-out proportion, feed conversion efficiency (FCE) and plasma urea

	GS	MS	FWCW	ALK	ALC	S.E.M.
Forage DM intake (kg/d) 0.166	4.54 ^a	6.75 ^b	7.07 ^b	7.56 ^c	0.95 ^d	0.966
Total DM intake (kg/d) 0.194	7.07 ^a	9.27 ^c	9.59 ^{bc}	10.06 ^{bd}	9.86 ^b	.194
Liveweight gain (g/d) 48.3	802 ^a	1120 ^{bc}	1149 ^c	1132 ^c	1302 ^b	483
Carcass gain (g/d) 30.9	479 ^a	776 ^{bc}	723 ^{cd}	686 ^d	851 ^b	309
Carcass weight (kg)	290 ^a	335 ^{bc}	329 ^c	321 ^c	348 ^b	5.2
Kill out (g/kg)	523 ^a	547 ^{bc}	539 ^{cd}	532 ^{ad}	551 ^b	4.2
FCE (kg DM intake/kg carcass gain) 0.50	15.2 ^a	12.1 ^b	13.5 ^c	14.8 ^{ac}	11.9 ^b	
Plasma urea (mmol/l)	4.6 ^a	2.7 ^b	5.0 ^{ac}	6.8 ^d	4.9 ^a	0.21

Within row, means with the same superscripts are not significantly different ($p > 0.05$)

In conclusion, forage maize and whole crop wheat silages supported superior levels of growth by cattle compared to grass silage (*in-vitro* DMD 698g/kg), and the FCE with maize silage and *ad-libitum* concentrates did not differ. Fermented whole-crop wheat gave a similar level of animal performance to maize silage but both forms of whole-crop wheat had a poorer FCE than maize silage. There was no animal productivity advantage with alkalage compared to fermented whole-crop wheat.

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Behaviour and meat colour of beef cattle offered conserved whole-crop wheat and forage maize relative to grass silage and *ad-libitum* concentrates and an assessment of the aerobic stability of these forages

Forages are conserved from contrasting plant species and using different techniques. They consequently can differ in conservation efficiency and nutritive value. This experiment compared the aerobic stability of grass silage, forage maize silage and whole-crop wheat (fermented or urea-treated) and compared the behaviour and meat colour of cattle offered these forages or concentrates *ad-libitum*.

Seventy continental cross-bred beef steers, mean initial live weight 424 (sd 33.0) kg, were blocked for live weight and breed and allocated to one of 5 dietary treatments in a randomised complete block design. Treatments were grass silage (GS), maize silage (cv. Benecia) (MS), fermented whole-crop wheat (cv. Soissons) (FWCW), alkalage whole-crop wheat (cv. Soissons) (ALK) and *ad-libitum* concentrates (ALC). The ALK harvester was fitted with a grain processor and ensiled with 45kg Home 'N' Dry (Volac International Ltd.) /t DM (dry matter). Forages were offered *ad-libitum* through individual Calan gates and supplemented with 3kg concentrates/head/day. The ALC treatment was supplemented with 5kg grass silage/head/day throughout the 160-day trial period. Aerobic stability of the forages was assessed in triplicate on five occasions for 8 days at 20^oC using an automated temperature recording system.

Table 68: Forage aerobic stability, animal behaviour and meat colour

	GS	MS	FWCW	ALK	ALC	s.e.m.
Forage aerobic stability						
Duration to temp. rise ¹	131 (45)	39 (13)	180 (18)	149 (58)	-	
ATR to day 5 ²	7 (7)	53 (26)	2 (1)	8 (8)	-	
Animal behaviour						
Eating ³	13.8 ^a	8.0 ^c	11.1 ^d	8.8 ^c	4.9 ^b	0.28
Ruminating ³	37 ^a	30.1 ^c	33.5 ^{ac}	25.6 ^d	19.9 ^b	0.66
Drinking ³	0.7 ^a	0.8 ^a	1.6 ^b	1.1 ^{ac}	1.5 ^{bc}	0.07
Idle ³	48.8 ^a	61.6 ^c	54.4 ^d	64.5 ^c	73.9 ^b	0.79
Meat colour						
Muscle 'I' value	35.7 ^a	36.4 ^a	35.4 ^a	37.2 ^a	36.7 ^a	0.92
Muscle 'a' value	18.4 ^{ac}	19.3 ^{ac}	17.3 ^c	18.4 ^{ac}	20.9 ^{ab}	0.91
Fat 'b' value	12.7 ^a	9.4 ^b	11.3 ^c	7.6 ^d	9.9 ^b	0.47

¹Hours, s.d. in brackets; ²accumulated temperature rise to day 5 (⁰C), s.d. in brackets; ³percentage of occasions observed.

Within a row, means with the same superscripts are not significantly different (p>0.05)

Animal behaviour, recorded as either eating, ruminating, drinking or idle, was assessed every 15 minutes over a 24-hour period. Meat colour was measured the day after slaughter with a 'Minolta ChromaMeter CR100' instrument. Subcutaneous fat colour ('b' value) was measured at the same position on each carcass. Muscle samples taken from between ribs 5 to 7 of the *M. longissimus dorsi* were overwrapped with Clingfilm and left to bloom for 2 hours before measuring redness ('a' value) and lightness ('I' value). Animal behaviour data were analysed using Chi-square analysis and treatment comparisons of LSMEANS made using the PROC GLM procedure in SAS. Meat colour was analysed using analysis of variance taking account of diet and block.

The mean DM (g/kg) (uncorrected for volatiles) (s.d.) and NDF (neutral detergent fibre) concentration (g/kg DM) (s.d.) of the GS, MS, FWCW and ALK were 161 (10.4) and 611 (8.9), 303 (4.3) and 256(15.2), 391 (4.2) and 367 (18.8) and 705 (6.4) and 575 (35.2), respectively. The stability of MS on exposure to air was poorer than the other three forages which were very stable (Table 68). The percentage of time for which animals were observed eating was in the order GS>FWCW>ALK=MS>ALC. Animals offered GS spent the greatest percentage of time ruminating while those offered ALC spent the least. Animals were adjudged to be idle in the order ALC>ALK=MS>FWCW>GS. FWCW had a less red muscle (lower (p<0.01) 'a' value) than ALC. GS had the most yellow fat (higher (p<0.05) 'b' value) and ALK had the most white fat (lower (p<0.01) 'b' value). Animals on MS and ALC had a similar fat colour.

In conclusion, MS was the only forage that was aerobically unstable during feedout. Animals offered GS tended to spend the highest percentage of time eating and ruminating, with those on ALC spending the least time. Of the four forages, ALK resulted in animals spending the least percentage of time ruminating. Forage type influenced muscle and fat colour with GS and ALK producing the yellowest and whitest fat, respectively.

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Alternative conservation strategies for high moisture wheat grains

Cereal grains are important sources of nutrients for ruminants in many livestock production systems, and may need to be stored for up to twelve months before being fed. During this time both quantitative and qualitative losses must be minimised, with fungal growth in particular needing to be prevented. Grain with a dry matter (DM) content ≥ 860 g/kg can be stored for extended durations. However, the duration of safe storage progressively decreases and/or the requirement for additional protective treatments increases as the DM concentration at harvesting decreases. This experiment quantified the conservation characteristics of high moisture wheat grains stored anaerobically following contrasting processing and additive treatments.

Wheat grown at Teagasc, Oak Park in 2001 (cv. Madrigal) and 2002 (cv. Falstaff) was combine-harvested and the grain was either not processed (whole: W) or was rolled (R) using a crimper-roller (Murska 350S) prior to additive treatment. The additive treatments applied in 2001 (Experiment 1) were: (1) none (NA), (2) Crimpstore 2000 (A1; Kemira Chemicals (UK) Ltd.) at 6 ml/kg, (3) Graintona (A2; FSL Bells Ltd., UK) at 8 ml/kg, (4) NuGrain urea (U; Hydro Nutrition, Hydro Agri (UK) Ltd.) at 50 ml/kg or (5) Biograin (B1; Biotal Ltd., Wales) at 100 ml (10 ml additive + 90 ml additional water)/kg. The additive treatments applied in 2002 (Experiment 2) were: (1) NA, (2) A2, (3) U, (4) B1 at 50 ml (10 ml + 40 ml additional water)/kg or (5) Siloking (B2; Agri-King, Inc., USA) at 500 mg (50 mg + 450 mg dry carrier)/kg. Approximately 4 kg grain DM was ensiled in each of triplicate laboratory silos per treatment. All additives were intimately mixed with the grains prior to ensiling. Silos were stored at 15°C for >100 days. Sampling procedures and analyses were carried out as described by Stacey *et al.* (2003). Data for each year were subjected to analysis of variance for a 2 (processing) x 5 (additive) factorial arrangement of treatments within a completely randomised design using Genstat 6.0.

Mean (s.d.) DM concentration of grain at harvesting was 616 (1.2) and 657 (3.8) g/kg in 2001 and 2002, respectively. Across additives, rolling wheat grains before ensiling resulted in lower ($P<0.001$) DM, starch and organic matter digestibility (OMD) values but higher ($P<0.001$) levels of fermentation products (Table 69). The latter indicated that a more extensive fermentation had occurred. Regardless of grain being rolled, B1 in particular and U to a lesser extent reduced ($P<0.001$) DM content. This reflects the volume of additional water applied with these specific additives. The content of crude protein (CP) was increased ($P<0.001$) considerably by U, particularly when applied to rolled grain. This increase reflected the large amount of N applied in this additive. In 2001, A2 and U had a lower ($P<0.001$) starch content than NA, especially with rolled grain, while these effects were less evident in 2002. High moisture wheat conserved without an additive underwent sufficient natural fermentation to reduce pH to 3.9 in 2001. In 2002, a similar fermentation was achieved with rolled grain while the more restricted fermentation with whole grain resulted in a final pH of 4.4. Acid additives (A1 and A2) restricted fermentation ($P<0.05$) with A2 resulting in elevated final pH values. U stimulated ($P<0.05$) fermentation in 2001 but restricted ($P<0.05$) it with the slightly drier crop in 2002. In both years U resulted in a high, alkaline final pH. B1 had little effect on pH or lactic acid, but generally increased acetic acid. The latter likely reflected the effects of *Lactobacillus buchneri* in this additive. B2 (2002) had relatively minor effects on the fermentation indices.

Table 69: Treatment effects of processing (P) and additives (A) on wheat composition at silo opening

	DM ¹	CP ²	Dig ³	Star ⁴	pH	LA ⁵	AA ⁶
2001							
W-NA	638	118	819	659	3.9	11	3.1
W-A1	649	117	816	656	4.2	6	0.8
W-A2	645	112	864	638	5.4	4	1.3
W-U	623	167	861	633	7.0	11	7.6
W-B1	577	118	851	640	4.2	11	15.0
R-NA	632	120	816	644	3.9	23	6.2
R-A1	637	120	809	625	4.2	12	2.8
R-A2	633	116	835	589	4.7	7	5.4
R-U	618	250	857	589	9.0	37	7.8
R-B1	560	128	841	648	4.1	24	22.4
s.e.m. ⁷	1.2	5.0	9.6	7.6	0.28	1.5	0.82
Sig. P	***	***	NS	***	***	***	***
A	***	***	***	***	***	***	***
PxA	***	***	NS	**	***	***	**
2002							
W-NA	672	126	897	652	4.4	6	4.3
W-A2	684	121	892	620	5.4	1	2.6
W-U	654	153	913	648	9.0	1	3.0
W-B1	638	128	891	678	4.3	4	20.8
W-B2	680	125	882	637	4.2	9	3.5
R-NA	661	125	888	628	3.9	30	10.7
R-A2	661	122	893	611	4.8	6	7.6
R-U	633	181	898	571	8.7	5	7.6
R-B1	631	127	877	660	3.9	30	11.6
R-B2	661	124	868	625	4.0	25	7.9
s.e.m. ⁷	2.0	1.4	3.5	10.9	0.03	0.6	0.77
Sig. P	***	***	***	***	***	***	***
A	***	***	***	***	***	***	***
PxA	**	***	NS	*	***	***	***

¹g/kg; ²g crude protein/kgDM; ³in-vitro organic matter digestibility (g/kg);
⁴g starch/kgDM; ⁵g lactic acid/kgDM; ⁶g acetic acid/kgDM; ⁷PxA

In conclusion, high moisture wheat stored anaerobically preserved successfully, with rolled grain fermenting more extensively than whole grain but at the cost of a decrease in starch content and OMD. Acid additives, urea or bacterial inoculants containing *Lactobacillus buchneri* each altered the characteristics of the conserved grain.

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Nutritive value for finishing beef steers of wheat grain conserved by different techniques

Wheat grain harvested at dry matter (DM) concentrations above 860g/kg is slow to deteriorate during long-term storage. However, high moisture grain (HMG) ranging from below 600 to 750 gDM/kg is conserved on some farms in the form of anaerobic storage of acid-treated, rolled wheat (AR) and urea-treated whole-wheat (UN). This experiment quantified the nutritive value for beef cattle of standard wheat grain (propionic acid-treated and rolled:PR) compared to AR and UN at different levels of intake.

The experiment was a 3 (forms of wheat: AR, UN, PR) x 3 (levels of wheat offered: low (L), medium (M), high (H)) factorial arrangement of treatments, and with a control group of animals on grass silage only (GS). Friesian steers (n=120) were allocated to 10 treatments in a randomised complete block design. For 144 days, all animals were offered grass silage *ad-libitum* together with (i) no wheat (GS only); (ii) - (iv) PR at 3 kg (L) or 6 kg/head daily (M), or *ad libitum* (H); (v) - (vii) UN at equivalent DM allowances to (ii) and (iii), or *ad libitum*; (viii) - (x) AR at equivalent DM allowances to (ii) and (iii), or *ad libitum*. Total faecal collections were made on all animals over a 24 h duration between days 102 and 109, and assessed for DM and starch concentration. Carcass weight (hot carcass x 0.98) was recorded after slaughter and carcass weight gain was estimated as the difference between final carcass weight and 0.48 of initial live weight. Samples of *M. longissimus dorsi* were taken 24 h post-mortem from between ribs 5 to 7 and stored at 3°C for a further 24 h. Colour measurements (lightness (l), redness (a) and yellowness (b) of the muscle and subcutaneous fat) were made using a Minolta ChromaMeter CR 100. Animal data were analysed as a factorial arrangement of nine treatments (3 wheat forms x 3 wheat levels) or as 10 treatments within a randomised complete block design using Genstat 5.0.

The mean (s.d) DM, pH, crude protein (CP) and organic matter digestibility (OMD) values for GS at feedout were 226(9.7) g/kg, 3.9(0.11), 152(4.6) g/kg DM and 679(14.1) g/kg, respectively. The mean (s.d.) DM (g/kg), pH, CP (g/kg DM), starch (g/kg DM) and OMD (g/kg) values at feed out for AR were 693(10.1), 4.3(0.15), 116(2.4), 671(18.5) and 925(7.4). The corresponding values for UN were 738(9.1), 9.3(0.07), 145(3.9), 664(39.0) and 934(9.7) and for PR were 827(8.1), 4.8(0.26), 111(4.8), 655(23.4) and 933(9.4), respectively. GS had the highest (P<0.001) silage DM intake (SDMI) but the lowest (P<0.001) daily live weight (DLG) and daily carcass weight (DCG) gains (Table 70). Increasing levels of wheat consumption progressively reduced SDMI and increased DLG and DCG. SDMI was equally lower (P<0.001) with AR and PR compared to UN whereas DLG and DCG were equally higher (P<0.05) with AR and PR compared to UN. For steers offered wheat *ad libitum*, wheat DM intake was lower (P<0.001) with AR than UN or PR, while DLG and DCG were lower (P<0.001) with UN than AR or PR. UN had the highest (P<0.001) amount of starch in the faeces indicating considerable loss of undigested grains. Muscle redness ('a value') was not influenced by method of wheat management but was higher at M compared to L level of supplementation. Fat yellowness ('b value') was higher (P<0.01) with UN than AR, while M>L>H.

Table 70: Performance, DM intakes, faecal results and meat data from 144 day feeding trial

Diet	GS	AR			UN			PR			s.e. ¹	Significance (3 x 3)			
		L	M	H	L	M	H	L	M	H		W _F ²	W _L	W _F x W _L	
Wheat level (W _L)	0														
SDMI ³ (kg/day)	7.4	5.4	3.7	1.3	5.9	4.6	1.5	5.8	3.9	1.2	0.15	***	***	NS	
WDMI ⁴ (kg/day)	0	2.5	4.9	7.8	2.4	4.8	8.3	2.4	4.9	8.2	0.11	NS	***	*	
DLG ⁵ (g)	100	719	887	983	612	724	843	622	870	1043	64.1	*	***	NS	
KO ⁶ (g/kg)	484	503	502	516	495	502	501	497	511	520	4.1	**	***	NS	
DCG ⁷ (g)	64	421	517	629	351	433	491	362	545	676	35.1	***	***	NS	
Faecal DM (g/kg)	143	158	160	184	155	162	204	147	152	175	5.9	**	***	NS	
Starch ⁸ (g/kg DM)	8	9	15	31	51	99	118	9	14	20	10.4	***	**	**	
Muscle 'a' value	13.0	13.1	13.4	14.3	13.1	14.0	13.5	12.9	14.2	13.3	0.38	NS	*	NS	
Fat 'b' value	13.7	12.6	13.2	11.4	13.6	14.5	12.4	13.2	14.3	11.5	0.39	**	***	NS	

¹ W_F x W_L; ² Wheat form; ³ silage DM intake; ⁴ wheat DM intake; ⁵ daily liveweight gain; ⁶ killout; ⁷ daily carcass gain; ⁸ in faeces

In conclusion, AR can replace PR in finishing beef rations without compromising performance or meat colour, provided excellent management at the silo substantially restricts qualitative losses. In contrast, the severe faecal losses of undigested grains with UN resulted in inferior growth rates compared to AR or PR. The relative magnitude of the decrease in performance appeared greater as the level of wheat ingestion increased.

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Red clover grown for silage

The year 2003 represented the second growth season of an experiment to evaluate factors effecting the yield and quality of red clover grown for silage. Four replicate blocks, each with 24 treatments, allocated randomly among plots (2m x 10m) were sown in autumn 2001. Grass (cv. Greengold) plots received 0, 50, 100 or 150 kg inorganic N/ha; red clover (Merviot or Ruttinova) was sown alone or in mixture with grass, with or without a spring application of inorganic N, with first-cut harvest dates in late May or early June. Subsequent harvests were taken in July, August and October.

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RMIS No. 5137

ANIMAL NUTRITION

Determination of nutrient supply to the omasum from conserved grass, maize and whole-crop wheat

As a ruminant feedstuff, grass silage can be less attractive than alternative forage crops due to relatively modest yields obtained in a single harvest, variability in digestibility and ensilability and therefore nutrient supply, coupled with ever-increasing costs of production and effluent management. The composition of feeds consumed by ruminants differs markedly from the nutrients available for metabolism due to the modifications caused by microbial fermentation in the rumen. Consequently, to be able to predict the supply of energy and other nutrients to ruminants, processes occurring in the rumen have to be assessed. The objective of the experiment was to quantify the nutrient supply to the omasum from grass silage, maize silage and whole-crop wheat-based diets. Four ruminally fistulated Holstein-Friesian steers were assigned to a 4 x 4 Latin-square design experiment with 21-day experimental periods (12-day diet adaptation and 9-day sampling). The dietary treatments were the following four forages offered *ad libitum* and supplemented with 3 kg of concentrates /head/day: (1) grass silage, (2) maize silage, (3) fermented whole-crop wheat silage (45% DM) harvested at normal cutting height and (4) alkalage (urea-treated whole-crop wheat; 70% DM) harvested at normal cutting height. The diet was offered twice daily @8.30 and 20.30h. During the sampling period the diet was offered at 0.90 *ad libitum* intake. Rumen fluid samples were obtained at 0, 2, 4, 6, 8 and 10 hours post-feeding. Digesta markers (Co-EDTA, Yb acetate) were continuously administered into the rumen using a multi-channel peristaltic pump. Spot samples of digesta flow leaving the rumen were obtained *via* a tube passed through the rumen cannula and positioned in the omasal canal. Samples were collected using an alternating vacuum and pressure system twice daily at 6 hr intervals with the time of sampling advancing 90 minutes daily over 4 days so as to represent a 12-hr period, considered representative of the entire feeding cycle. Grab samples of faeces were obtained *via* rectal palpation at the same times as the omasal canal samples. Reticulo-rumen samples were obtained once daily for microbial protein determination. Two blood samples were obtained by jugular venipuncture from each animal immediately before the morning feeding and subsequently 3 and 6 hours after feeding for plasma metabolite analysis. Sample processing and laboratory analysis is in progress.

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Investigating the effects of supplementing grass silage with sucrose or starch-based concentrates on nutrient flow from the rumen

The asynchronous release of energy and nitrogenous components from grass silage has often been considered an important reason for low efficiency of rumen microbial growth on silage diets and sugar supplements have been suggested as a way to improve rumen ammonia utilisation. This study aimed at simulating the sugar concentrations that could occur in silage if grasses of very contrasting sugar concentrations were preserved optimally. Concentrate supplementation of the grass silage was also compared so that observations would be comparable to commercial practice. Four ruminally fistulated Holstein-Friesian steers were assigned to a 4 x 4 Latin-square design experiment with a 2 x 2 factorial arrangement of treatments. Experimental periods were 21-days consisting of 10-day diet adaptation and 11-day sampling time. The four diets were; (1) grass silage, (2) grass silage + 9% sucrose, (3) grass silage and 3 kg concentrates and (4) grass silage + 9% sucrose and 3 kg concentrates.

The sugar was mixed with the silage prior to feeding using a feeder wagon. The diet was offered twice daily at 8.30 and 20.30h. During the sampling period the diet was offered at 0.90 *ad libitum* intake. Two blood samples were obtained by jugular venipuncture from each

animal immediately before the morning feeding and subsequently 3 and 6 hours after feeding for plasma metabolite analysis. Rumen fluid samples were obtained at 0, 1.5, 3.0, 4.5, 6.0, 7.5, 9.0 and 10.5 hours post-feeding. Digesta markers (i.e. Co-EDTA, Yb acetate) were continuously administered into the rumen using a multi-channel peristaltic pump. Spot samples of digesta flow leaving the rumen were obtained via a tube passed through the rumen cannula and positioned in the omasal canal. Samples were collected using an alternating vacuum and pressure system twice daily at 6 hr intervals with the time of sampling advancing 90 minutes daily over 4 days so as to represent a 12-hr period, considered representative of the entire feeding cycle. Grab samples of faeces were obtained via rectal palpation at the same times as the omasal canal samples. Reticulorumen samples were obtained once daily for microbial protein determination. To determine rumen passage rates, marker infusion was ceased pre-feeding and grab samples of rumen contents were obtained at 0, 1.5, 3.0, 4.5, 6.0, 7.5, 9.0, 10.5, 12.0, 24.0 and 36.0 hours post-feeding. Sample processing and laboratory analysis is on-going.

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Effect of offering two levels of crude protein and two levels of milk replacer on calf performance

Recent reports from the United States propose new nutritional guidelines for calves (VanAmburgh 2003). They suggest an accelerated growth programme where calves are offered a high protein milk replacer (CMR) at a level sufficient to achieve a liveweight gain of 1000 g/d up to 8 weeks of age. This is in contrast to many European feeding programmes where calves are offered 25 kg of CMR over 42 days with an expected liveweight gain of 500 to 600 g/day. The objective of this experiment was to evaluate the accelerated growth system under Irish conditions.

In experiment 1, sixty-four 2 to 3 week-old Holstein/Friesian calves with an initial weight of 50 kg (+/- 1.8 kg) were allocated immediately following purchase to the following treatments: (1) 23% crude protein CMR at 600 g/d (LL), (2) 23% crude protein CMR at 1200 g/d (LH), (3) 30% crude protein CMR at 600 g/d (HL) and (4) 30% crude protein CMR at 1200 g/d (HH). The milk replacer was offered warm by bucket with the daily allowance reduced to encourage solid food intake in the period 42 to 56 days. All calves had *ad libitum* access to a concentrate diet throughout the 16-week experimental period. In the initial 56-day period all calves were individually penned on straw with a pen area of 1.5 m² per calf. Thereafter, groups were penned according to treatment on concrete slats. In Experiment 2, thirty-six 2 week old Holstein/Friesian calves with an initial weight of 45 kg (+/- 1.6 kg) were allocated on purchase to the following treatments: (1) 23% crude protein CMR at 750 g/d (LM) and (2) 30% crude protein CMR at 1200 g/d (HM). Feeding and management of the animals was similar to Experiment 1 for the first 112 days of the experiment. Thereafter, all calves had *ad libitum* access to a 16% crude protein concentrate ration. The calves were accommodated according to treatment in four pen on concrete slats. Grass silage was offered daily as a roughage source (approximately 10% of total dry matter intake). The animals were slaughtered after 388 days on experiment. In Experiment 1 the data was subjected to a two-way analysis of variance. In Experiment 2 data was subjected to analysis of variance.

The higher level of crude protein in the milk replacer (23 v 30%) did not affect feed intake or liveweight gain. In Experiment 1, the level of feeding significantly decreased concentrate intake in the period 1 to 56 days and increased liveweight gain in the period 1 to 56 days, however the high level of milk replacer did not significantly affect liveweight gain in the period 1 to 112 days (Table 71). In Experiment 2, the higher level of milk replacer feeding significantly reduced concentrate intake in the period 1 to 56 days but had no significant effect on liveweight gain (Table 72). After 204 days the difference in daily liveweight was 6 kg between the two treatments and after 388 the difference was 12 kg. These differences were not significant in Experiment 2 and feeding additional milk replacer with a high protein in the first 56 day did not effect carcass weight, conformation or fat score.

Table 71: Effect of level of feeding and level of protein in the CMR on calf performance (Experiment 1)

	<u>LL</u>	<u>LH</u>	<u>HL</u>	<u>HH</u>	<u>Sem</u>	<u>Sig</u>		
						<u>L</u>	<u>P</u>	<u>L x P</u>
Liveweight gain g/d								
1 - 56 days	690	760	720	870	48	*	-	-
57 - 112 days	1020	1140	1070	1110	67	-	-	-
1 - 112 days	860	950	900	990	48	-	-	-
Concentrate intake (kg/DM)								
1 - 56 days	34.0	28.2	41.1	23.2	4.12	**	-	-
57 - 112 days	180.5	189.6	168.2	211.9				
1 - 112 days	214.5	217.8	209.3	234.1				
CMR intake 1 - 56 days (kg/DM)	30.3	57.4	29.9	56.7				

Table 72: Effect of CMR programme on calf performance (Experiment 2)

	<u>LM</u>	<u>HM</u>	<u>sem</u>	<u>Sig</u>
Liveweight gain g/d				
1 - 56 days	550	640	45	-
57 - 112 days	1030	1070	85	-
1 - 112 days	790	860	55	-
1 - 204 days	1070	1100	23	
205 - 388 days	1060	1120	43	
1 - 388 days	1070	1110	33	
Final Carcass (kg)	457	475	12.9	-
Cold Carcass (kg)	230.5	239.7	6.65	-
Conformation	3.82	3.88	0.04	-
Fat score	2.64	2.31	0.24	-
K O%	50.4	50.4	0.01	-
Concentrate intake (kg/DM)				
1 - 56 days	29.0	17.4	2.75	**
57 - 112 days	150.8	150.1		
1 - 112 days	179.8	167.5		
CMR intake 1 - 56 days (kg/DM)	29.6	46.3		

In conclusion, increasing the daily CMR allowances from 600 g to 1200 g increased LWG in the period 1 to 56 day and there was no response to increasing the level of crude protein from 23 to 30%.

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MEAT QUALITY

Growth and rumen fermentation in steers offered a sunflower oil-enriched supplement to conserved forage

Addition of sunflower oil (SO) to cattle rations has been reported to enhance the concentration of conjugated linoleic acid (CLA) in meat in a dose-dependent manner. However, to avoid impairment of rumen function it is advised to limit the total supply of lipid in forage-based diets to 50-60 g/kg dry matter (DM). In production systems where concentrates are offered in discrete meal(s), separately to forage, optimisation of CLA production would require the concentration of additional oil in the concentrates to exceed this limit. Little information is available on the effects of an acute addition of oil to the rumen. The objective of this experiment was to determine the effect of SO offered in one meal on growth and rumen fermentation in beef cattle.

Growth study: Twenty crossbred heifers (mean initial weight = 426kg, s.d. 34.33 kg) were offered either grass silage (G) or one third G and two-thirds (DM basis) whole crop wheat (W) (519 gDM/kg + 30 kg urea/t) *ad libitum*. Three kg of isonitrogenous and isoenergetic concentrates that contained 0g or 110g SO/kg were offered once daily, before silage was offered. Animals were slaughtered after 150 days on the dietary treatments.

Rumen study: The four rations were offered to ruminally fistulated steers in a 4 (rations) by 4 (periods) Latin Square design. Ration allowances were on a bodyweight basis based on the intake and associated ration forage to concentrate ratio in the growth study. On day 14 of each period, rumen fluid samples were collected before and 1, 4, 8 and 12 hours after feeding. Intake and growth data were subjected to analysis of variance according to a split-plot design with silage in the main plot and oil concentration in the subplot. Rumen fermentation data were analysed according to a Latin Square factorial, split-plot (time) design.

There was no effect of ration composition on silage consumption but carcass weight tended to be higher for animals offered W (Table 73). Volatile fatty acid proportions were similar for all treatments, but total VFA concentration was decreased by SO only when included with W (Table 1). Inclusion of SO increased ruminal ammonia concentration in the first 4 h after feeding when included with W, but later in the day when included with G (Figure 10).

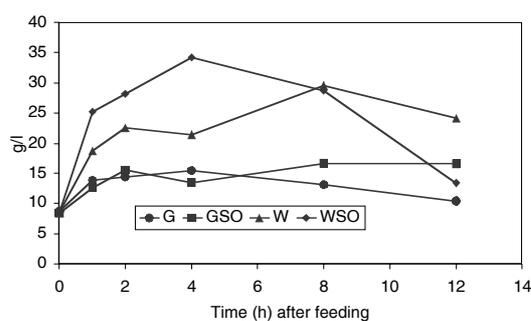


Figure 10. Ammonia concentration in rumen fluid (sed for forage type*SO*hours = 5.24)

Feeding a concentrate containing 110g SO/kg in one meal had no effect on rumen fermentation of a grass silage diet but altered rumen fermentation of a urea-treated whole crop wheat diet. These alterations did not influence forage consumption, FCE or animal growth.

Table 73: Feed intake, growth and rumen fermentation

	G	GSO	W	WSO	SED ^a	Significance		
						F	SO	F*SO
<i>Performance</i>								
Silage intake (kg dry matter/day)	5.34	5.36	5.13	5.84	0.496	NS	NS	NS
Carcass weight (kg)	284	289	300	303	11.4	0.08	NS	NS
KCF ¹ (g/kg carcass)	37.5	48.5	27.6	31.6	11.74	NS	NS	NS
FCE ² (gDM intake/kg carcass gain)	12.7	13.4	12.6	13.1	1.20	NS	NS	NS
<i>Rumen fermentation</i>								
PH	6.55	6.54	6.60	6.64	0.059	NS	NS	NS
Acetate (mol/100mol)	68.4	68.9	68.8	69.1	1.49	NS	NS	NS
Propionate (mol/100mol)	17.3	17.6	17.1	16.9	0.67	NS	NS	NS
Butyrate (mol/100mol)	11.3	10.6	11.31	11.4	0.76	NS	NS	NS
Volatile fatty acids (mmol/l)	78.7	78.8	87.2	79.8	1.94	*	*	*

^aFor interaction between effects of forage type (F) and sunflower oil (SO). ¹Kidney/channel fat; ²Feed conversion efficiency

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RMIS No. 4788

Quantification of maize silage in a beef ration using carbon stable isotope ratio analysis of meat

Stable isotope ratio analysis is a potentially useful technique for investigating the authenticity of animal-derived food products, including the authenticity of the geographic origin and feeding history of beef (Grange Research Centre, 2003 Annual Report p 74). The carbon stable isotope composition (expressed as $\delta^{13}\text{C}$) of beef tissue primarily reflects the proportion of C₃ and C₄ photosynthetic plants consumed by cattle. Confirmation that particular feeds were used might allow identification of the production origin of beef, i.e. whether the beef was organically or conventionally produced. The objective of this experiment was to establish a quantitative relationship between dietary C₄ plant intake and bovine muscle $\delta^{13}\text{C}$.

Forty-five continental crossbred heifers were fed with either grass silage (GS), an equal mixture (dry matter basis) of grass silage and maize silage (GMS), or maize silage (MS) *ad libitum* in a randomised block design (N=15). All animals also received 3 kg concentrates (composition per kg: 310 g citrus pulp, 460 g barley, 160 g soybean, 50 g molasses and 20 g mineral/vitamin mixture) daily. The *Longissimus thoracis et lumborum* muscles taken at 24 h post-mortem were freeze-dried and pulverised in a ball mill. Lipid was extracted from the milled samples using hexane: isopropanol (3:2 v/v) and the solvent was removed under a continuous flow of N₂. Lipid free muscle (0.9-1.1 mg) and lipid samples (0.2-0.9 mg C) were analysed by continuous flow isotope ratio mass spectrometry. The values of the isotope ratios were calculated and expressed in delta-notation [δ per mille (‰)] according to the formula: δ (‰)=[(R_{sample}/R_{reference})-1] × 1000, where, R is the ratio of the heavy to light stable isotope in a sample (R_{sample}) and a reference standard (R_{reference}). Results are referenced to an international standard (V-PDB). Data were subjected to analysis of variance (ANOVA) and regression analysis using SAS v.8.2 (SAS Institute).

The $\delta^{13}\text{C}$ of lipid free muscle from GS fed cattle (-25.1‰) was more negative than that from GMS (-22.1‰) and MS fed cattle (-18.1‰) (SED±0.1). This reflected the $\delta^{13}\text{C}$ values of dietary grass silage (-29.6±0.3‰), maize silage (-11.8±0.1‰) and the mixture of grass silage and maize silage (calculated $\delta^{13}\text{C}$, -21.0‰). A similar trend was evident in the lipid fraction, for which the

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$\delta^{13}\text{C}$ values were -29.2 , -25.8 and -21.1‰ ($SED\pm 0.3$) for GS, GMS and MS fed beef, respectively. Compared to lipid free muscle, the $\delta^{13}\text{C}$ of the lipid fraction was more negative. This depletion in ^{13}C likely reflected discrimination against ^{13}C during the process of lipid synthesis (DeNiro & Epstein, 1977).

There was a linear relationship ($P<0.001$) between the proportion of maize carbon in the diets and $\delta^{13}\text{C}$ of lipid free muscle ($r^2=0.98$) and lipid ($r^2=0.93$) (Fig. 11). The regression analysis indicated that, with 95% confidence, each 10% change in the dietary maize carbon corresponds to a 0.9 to 1.0‰ shift of $\delta^{13}\text{C}$ in lipid free muscle and a 1.0 to 1.2‰ shift of $\delta^{13}\text{C}$ in lipid. The regression lines for lipid free muscle ($\delta^{13}\text{C}=0.0958$ maize carbon % - 25.215) and lipid ($\delta^{13}\text{C}=0.1117$ maize carbon % - 29.338) had significantly different intercepts (differences 4.123‰, $P<0.001$) and slopes (differences 0.015, $P<0.01$), the latter possibly indicating an increasing tendency for maize carbon to be assimilated into beef intramuscular lipid during the fattening phase.

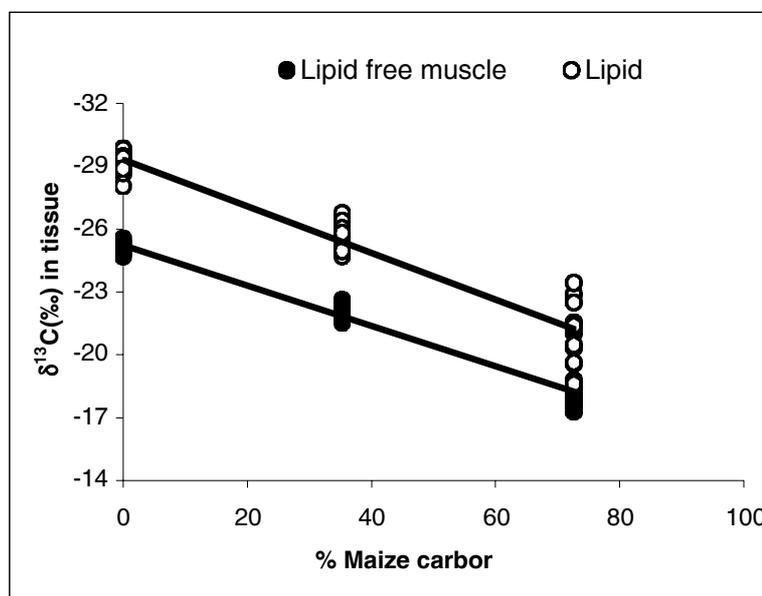


Figure 11. Relationship between maize-derived carbon in the diet of beef cattle and carbon stable isotope composition ($\delta^{13}\text{C}$) of lipid free muscle and lipid.

Maize silage used in the finishing diet of beef cattle could be detected and quantified by stable carbon isotope analysis of lipid free beef muscle or lipid. Thus, $\delta^{13}\text{C}$ has potential as a marker for authenticating the origin of beef from production systems using different levels of maize and other C_4 plant products.

RMIS No. 5214

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The composition and oxidative stability of lipids in *longissimus* muscle from grazing cattle supplemented with sunflower oil or linseed oil

Manipulation of ruminant ration composition has been employed to enhance the concentrations of conjugated linoleic acid (CLA) and omega-3 polyunsaturated fatty acids (PUFA) in milk and meat. Compared with conventional indoor rations, consumption of grass, rich in n-3 PUFA, led to an improvement in the fatty acid profile of beef, by increasing PUFA and CLA concentrations, decreasing the n-6:n-3 PUFA ratio and decreasing the saturated fatty acid concentration. Enrichment of concentrate rations with plant oils, such as linseed oil or sunflower oil, also resulted in an increase in the concentration of CLA in bovine muscle. The first objective of this study was to investigate the effect of plant oil supplementation of grazing cattle on the fatty acid profile of muscle, in particular the n-3 PUFA and CLA concentrations.

Strategies that improve the fatty acid composition must not impair other quality characteristics of beef. Appearance, specifically colour, is an important quality attribute influencing the consumer's decision to purchase. Increasing the PUFA concentration *per se* and/or increasing the concentration of longer carbon chain PUFA, predispose lipids to oxidation. The second objective was to determine the effect of alterations in the fatty acid composition on colour and lipid stability of beef.

Forty-five Charolais crossbred heifers (mean initial bodyweight = 330 kg, s.d. 39.90 kg) were blocked by initial bodyweight and, within block, randomly assigned to one of three dietary treatments (N = 15): unsupplemented grazing (GO); restricted grazing plus 2 kg/head/day of linseed oil-enriched meal (LO) or restricted grazing plus 2 kg/head/day of sunflower oil-enriched meal (SO). Concentrate and grass allowances were monitored at three-week intervals during a 5-month experimental period to achieve similar carcass weights across the treatments. Animals were slaughtered at a commercial facility, carcasses were chilled for 48 h at 4°C, and the *M. Longissimus dorsi* (LD) was excised from each carcass. Intramuscular fat was extracted from muscle samples using chloroform and methanol (2:1 v/v), methylated at 50°C for 20 minutes in alkaline and then acidic conditions and the fatty acid methyl esters obtained were analysed by gas chromatography (Supelcowax 100 m CP-Sil 88, Varian 3800).

Samples of LD collected 48h post-mortem were vacuum packaged and stored at 4°C for a further 24h prior to analysis. Samples were cut into steaks (25.4 mm thickness) and placed in retail display trays. Trays were over-wrapped with oxygen permeable film for aerobic storage or flushed with 80% O₂: 20% CO₂ for storage under modified atmosphere conditions. All samples were stored for up to 10 days at 4°C under simulated retailed display conditions (616 lux fluorescent lighting). Colour measurements were made at 2 day intervals using a Cr-300 Chromameter (Minolta Co. Ltd., Japan) set on the CIE colour scale and reported as the 'a' (redness value). Lipid oxidation was measured by distillation and results were expressed as 2-thiobarbituric acid reactive substances (TBARS) in mg malondialdehyde/kg muscle. The data were analysed as a randomized block design using Genstat 6.0.

Fatty acid data are summarised in Table 74. In general, the fatty acid composition of GO-fed cattle was similar to that previously reported. Compared to GO, SO-fed cattle had a higher concentration of C18:1 trans-11, C18:2, cis 9, trans-11 CLA, C20:4, total PUFA and n-6 PUFA but a lower concentration of C12:0, C18:3 and C22:5 and higher P:S and n-6:n-3 PUFA ratios. Compared to GO, LO-fed cattle had a higher concentration of C18:1 trans-11, cis 9, trans-11 CLA and n-3 PUFA and n-6:n-3 ratio but a lower concentration of C12:0, C20:4, C22:5 and C22:6. Compared to LO, SO-fed cattle had a higher concentration of C18:1 trans-11, C18:2, cis

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9, trans 11 CLA, C20:4, C22:6 and n-6 PUFA, a lower concentration of C18:3 and a higher n-6:n-3 ratio.

Table 74: Fatty acid and Vitamin E concentrations in *M. longissimus dorsi* from grazing cattle, either unsupplemented (GO) or supplemented with sunflower oil (SO) or linseed oil (LO)

Fatty acids (mg/100 g muscle)	GO	SO	LO	s.e.d.	Significance ¹
C12:0	1.52 ^b	0.88 ^a	0.88 ^a	0.157	***
C14:0	53.36	47.77	49.81	6.926	NS
C16:0	542.2	520.1	525.6	69.23	NS
C18:0	435.6	431.6	406.1	56.45	NS
C18:1 cis-9	843.0	847.1	780.1	125.3	NS
C18:1 trans-11	76.63 ^a	227.0 ^c	157.8 ^b	24.60	***
C18:2n6 cis	58.80 ^a	78.39 ^b	62.50 ^a	4.816	***
CLA c9,t11	18.37 ^a	47.43 ^c	32.00 ^b	5.976	***
CLA t10, c12	1.73	0.93	1.51	0.479	NS
C18:3n3	34.34 ^b	22.14 ^a	31.72 ^b	2.929	***
C20:4n6	11.75 ^a	12.47 ^c	9.57 ^b	0.803	***
C20:5n3	7.63	6.40	6.40	0.561	0.06
C22:5n3	12.69 ^b	10.40 ^a	9.69 ^a	0.688	***
C22:6n3	2.71 ^b	2.34 ^b	1.65 ^a	0.269	**
SFA ²	1089	1058	1037	134.5	NS
MUFA ²	1032	1186	1037	143.3	NS
PUFA ²	158.0 ^a	203.0 ^b	181.1 ^{ab}	14.93	*
P:S Ratio	0.15 ^a	0.21 ^b	0.18 ^{ab}	0.015	**
n-6 PUFA ²	86.92 ^a	106.5 ^c	92.51 ^c	6.405	***
n-3 PUFA ²	59.49	48.18	55.03	4.725	0.07
n-6:n-3 Ratio	1.46 ^a	2.24 ^c	1.72 ^b	0.096	***
Total fatty acids	2513	2688	2513	329.1	NS
Vitamin E (ug/g)	2.70 ^a	3.16 ^b	1.99 ^c	0.224	**

¹NS = not significant; *, ** and *** = P<0.05, P<0.01 and P<0.001, respectively; ²SFA = total saturated fatty acids; MUFA = total monounsaturated fatty acids; PUFA = total polyunsaturated fatty acids; n-6 PUFA = sum of C18:2, C18:3n-6, C20:2, C20:3n-6, C20:4 and C22:2; n-3 PUFA = sum of C18:3n-3, C20:3n-3, C20:5, C22:5 and C22:6.

Vitamin E concentration was lowest in LO-fed cattle and highest in SO-fed cattle (Table 74). Since similar amounts were supplied by the concentrates this suggests greater metabolism and a possible greater requirement for Vitamin E in the diet that supplied the greatest amount of n-3 PUFA.

There was no effect of diet on colour stability of beef (Table 75). Muscle lipids tended to be more susceptible to oxidation in MAP (higher TBARS) than in aerobic packaging with muscle from SO-fed animals more stable than LO-fed animals (Table 75). Muscles from LO-fed animals had lower lipid stability compared to GO-fed animals on day 2 and 6 of display in MAP.

Table 75: Surface redness ('a' value) and lipid oxidation (TBARS) in *M. longissimus dorsi* from grazing cattle either unsupplemented (GO) or supplemented with sunflower oil (SO) or linseed oil (LO) stored in aerobic or in modified atmosphere packs (MAP)

	Packaging	Storage Time (days)					
		0	2	4	6	8	10
<i>Redness</i>							
GO	Aerobic	17.99	13.46	9.36	8.10	8.83	8.74
SO	Aerobic	17.79	12.78	10.11	8.63	9.15	9.13
LO	Aerobic	18.53	13.23	9.87	7.61	7.97	8.79
s.e.d.		1.067	0.900	0.639	0.528	0.551	0.607
Significance ¹		NS	NS	NS	NS	NS	NS
GO	MAP	17.99	17.33	14.84	13.03	10.20	9.09
SO	MAP	17.79	17.45	15.41	12.56	10.63	8.52
LO	MAP	18.53	18.30	16.56	14.60	10.81	9.50
s.e.d.		1.067	0.703	0.960	1.351	0.973	1.004
Significance ¹		NS	NS	NS	NS	NS	NS
<i>Lipid oxidation</i>							
GO	Aerobic	0.83	0.82	0.46 ^b	0.39 ^a	0.65 ^b	0.86 ^b
SO	Aerobic	0.37	0.37	0.26 ^a	0.28 ^a	0.31 ^a	0.41 ^a
LO	Aerobic	0.52	0.67	0.27 ^a	0.62 ^b	0.63 ^b	1.01 ^b
s.e.d.		0.200	0.258	0.088	0.065	0.135	0.169
Significance ¹		NS	NS	*	**	*	**
GO	MAP	0.83	0.77 ^a	1.27 ^b	0.97 ^a	3.14	4.83 ^b
SO	MAP	0.37	0.58 ^a	0.53 ^a	0.80 ^a	2.37	3.05 ^a
LO	MAP	0.52	1.16 ^b	0.93 ^b	1.82 ^b	3.48	4.82 ^b
s.e.d.		0.200	0.122	0.215	0.430	0.571	0.654
Significance ¹		NS	**	**	*	NS	*

¹NS = not significant, * and ** = P<0.05 and P<0.01, respectively. Within packaging type and day, means with a common superscript do not differ significantly.

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It is concluded that supplementing grazing animals with plant oil-enriched concentrates resulted in a further beneficial effect on the fatty acid composition of muscle compared to grazing alone. Sunflower oil was more effective than linseed oil in increasing the concentration of CLA and TVA, but had a negative effect on the n-6:n-3 PUFA ratio. Linseed oil had a less pronounced effect on the CLA concentration than sunflower oil, but it also had a less negative effect on the n-6:n-3 PUFA ratio. While linseed oil supplementation caused a transient increase in lipid oxidation, this was not reflected in a loss in colour stability.

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Lipid oxidation and sensory characteristics of grass-fed beef: Effect of duration of grazing prior to slaughter

Beef from cattle produced from grass has a higher concentration of fatty acids considered to be beneficial to human health than beef produced from more intensive production systems and this increase in fatty acid concentration is dependant on the duration at pasture prior to slaughter. Improvements in the fatty acid composition of beef must not impair other quality characteristics of beef. Little information is available on the pattern of change of quality characteristics in grazing animals. The objective of this study was to determine the shelf-life and eating quality of beef from cattle produced from a standard Irish grass silage/concentrates finishing system but allowed to graze grass for different periods prior to slaughter.

Sixty Charolais crossbred heifers (BW = 338 kg) were used. One group was offered a silage/concentrates based diet indoors for 158 days (0 days at grass). One group grazed a predominantly perennial ryegrass pasture for 158 days. Two groups were initially offered a silage/concentrates diet, but grazed the above pasture for 40 and 99 days prior to slaughter after 158 days, respectively. Concentrate and grass allowances were adjusted periodically to achieve a similar mean carcass weight for all treatments. Carcasses were chilled for 48h at 4°C. A sample of *longissimus* muscle was aged for 14 days and stored frozen prior to lipid oxidation analysis (thiobarbituric acid reactive substances (TBARS)). A similarly-treated sample was used for sensory analysis by a 10 member trained taste panel. Panellists rated cooked steak using 0-100 line scales where low values are low ratings and high values higher ratings for a particular trait. Data were analysed according to a randomised block design.

Extending the grazing period increased the concentration of individual long chain n-3 polyunsaturated fatty acids (PUFA), total PUFA and conjugated linoleic acid (CLA) in muscle but did not affect lipid oxidation after 5 or 10 days of retail display (Table 76). This was likely due to vitamin E supplied by the grass. Beef from 99-day grass fed animals was tougher than beef from animals fed silage/concentrates or grass for 158 days. Extending the grazing period increased the scores for "greasy" and "fishy". Beef from animals fed silage/concentrates was preferred to beef from 40 or 99-day grass fed animals. There were no differences for juiciness, or for beef, abnormal, bloody, livery, metallic, bitter, sweet, rancid, acidic, cardboard, vegetable/grassy or dairy flavours.

Table 76: Fatty acid concentration (mg/100g tissue), TBARS (mg malonaldehyde/kg) and sensory perception of beef

	Days at grass				SED	P
	0	40	99	158		
C18:3	19.6 ^a	25.4 ^b	30.9 ^c	34.4 ^c	1.86	***
C20:5	5.6 ^a	5.5 ^a	6.4 ^a	7.7 ^b	0.50	***
C22:5	10.1 ^a	9.4 ^a	10.6 ^a	12.7 ^b	0.74	***
CLA	12.3 ^a	12.1 ^a	15.2 ^a	18.4 ^b	1.79	***
PUFA	129.4 ^a	136.7 ^{a,c}	145.6 ^{b,c}	158.4 ^d	6.93	***
Total	2641	2329	2754	2525	177.5	NS
TBARS-10 d	4.06	3.05	4.04	4.04	0.678	NS
Toughness	44.9 ^{ab}	48.8 ^{bc}	52.7 ^c	42.7 ^a	2.16	***
Greasy	10.4 ^a	12.7 ^{ab}	13.1 ^{ab}	14.4 ^b	1.44	*
Fishy	0.7 ^a	3.5 ^b	2.3 ^{ab}	3.3 ^b	1.07	*
Overall liking	22.5 ^b	18.1 ^a	18.8 ^a	19.9 ^{ab}	1.48	*

Figures with different superscripts differ significantly (P<0.05)

Increasing the duration of grazing prior to slaughter improved the fatty acid composition, did not affect lipid oxidative stability, had minor effects on flavour but inconsistently influenced the toughness of beef.

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The fatty acid composition of *Longissimus* muscle from grazing cattle supplemented with sunflower oil and fishoil

Effects of sunflower oil (S) and fish oil (F) supplementation of grazing cattle on the fatty acid profile of muscle, in particular the conjugated linoleic acid (CLA) and vaccenic acid (VA) concentrations were examined. Grazing Charolais crossbred heifers (initial bodyweight = 407 kg, s.d. 31.3) were offered (N = 12/treatment): grazing only(G), or an individual daily supplement of 2.5 kg concentrates that supplied 290 g S(S1), 415g S (S2), 290g S + 85g F (FS1) or 415 g S + 85 g F (FS2). Animals were slaughtered after 150 days and lipids from the *Longissimus* muscle were separated into neutral (N) and polar (P) fractions prior to methylation and separation by gas chromatography. Data were subjected to analysis of variance and "a priori" contrasts were used to test for effects of S level and F inclusion. Daily bodyweight gain and carcass weight averaged 834 g and 294 kg, respectively, and did not differ (P <0.05) between treatments. The N fraction of muscle from G, S1, S2, FS1 and FS2-fed cattle had 1.29, 1.80, 2.18, 2.32 and 2.44 (sem 0.132) g c9,t11CLA /100g fatty acids, respectively. The corresponding values were 2.78, 4.40, 4.96, 5.51 and 5.33 (sem 0.318) for VA, 1.29, 1.66, 1.49, 1.39 and 1.54 (sem 0.083) for linoleic acid and 0.66, 0.44, 0.42, 0.42 and 0.47 (sem 0.030) for linolenic acid. The P fraction of muscle from G, S1, S2, FS1 and FS2-fed cattle had 0.59, 0.88, 1.12, 1.35 and 1.20 (sem 0.101) g c9,t11CLA /100g fatty acids, respectively. The corresponding values were 1.10, 3.19, 3.77, 5.64 and 4.46 (sem 0.529) for VA, 12.0, 17.5, 17.5, 10.9 and 14.4 (sem 1.66) for linoleic acid, 4.61, 2.21, 2.35, 2.19 and 2.53 (sem 0.258) for linolenic acid, 3.76, 1.98, 1.75, 2.37 and 2.31 (sem 0.250) for eicosapentaenoic acid and 0.15, 0.06, 0.02, 0.35 and 0.61 (sem 0.053) for docosahexaenoic acid. It is concluded that (1) supplementing grazing cattle with S-enriched concentrates increased

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muscle CLA concentration in the N fraction in a dose-dependant pattern, and (2) F consumption increased the concentration of long-chain n-3 fatty acids in the P fraction but increased CLA concentration only when added to the lower S concentrate.

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RMIS No. 5213

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Colour stability and lipid oxidation in *M. Longissimus dorsi* from lambs fed oils or oilseeds rich in polyunsaturated fatty acids

Due to ruminal biohydrogenation, the increase in muscle polyunsaturated fatty acids (P) in ruminant tissue is small relative to dietary supply. Some biohydrogenation is desirable since conjugated linoleic acid (CLA), considered to have human health promoting properties, results from incomplete ruminal biohydrogenation of linoleic acid and by tissue desaturation of ruminally derived trans-vaccenic acid (TVA). It has been reported (Grange Research Report, 2003, p 66) that controlling the rate of oil release from linseed and camelina to the rumen increased the efficiency of transfer of dietary P to tissue while allowing the production of CLA. Increasing the P concentration, in particular the longer carbon chain P, predispose lipids to oxidation which is believed to be linked to muscle pigment oxidation and consequently to colour instability. The objective of this study was to compare the effects of the above strategies to protect dietary lipids from ruminal biohydrogenation, on the colour and lipid stability of muscle.

Sixty-six wether lambs (40 kg, s.d 4.69 kg) were offered, for 100 days prior to slaughter, one of six barley/beet pulp-based rations which contained 500 IU vitamin E/kg and which differed in fat source: i.e. Megalac (ML), Camelina (C) oil (C18:2:C18:3 = 0.68, CO), Linseed (L) oil (C18:2:C18:3 = 0.47, LO), C seed treated with 10gNaOH/kg (CS), L treated with 5gNaOH/kg (LS) and CO treated with ethanolamine (CA). Feed allowances were adjusted periodically to achieve similar carcass weights. Following slaughter, intramuscular fat was extracted from *M. longissimus dorsi* (LD), methylated and fatty acid methyl esters (FAME) analysed by gas-chromatography. Samples of thawed LD were placed in retail display trays and stored under an atmosphere of 80% O₂ : 20% CO₂. Colour (redness) and lipid oxidation (2-thiobarbituric acid reactive substances; TBARS) were measured at 3-day intervals during retail display. Data were analysed according to a randomised block design using Genstat 6.0. "A priori" contrasts were also used to compare oils and NaOH treatment *per se*.

Data are summarised in Table 77. LS was most effective in increasing the C18:3 and CLA concentration and decreasing the n-6:n-3 P ratio in muscle. Within C treatments, CA resulted in the highest C18:3 and lowest CLA concentration in muscle. On average, oil resulted in lower C18:3 concentration and n-6:n-3 ratio and higher TVA concentration and than seeds. There was no difference in colour stability during retail display (data not shown). Vitamin E concentration was lower for all treatments compared to ML. TBARS were highest for LS and lowest for ML in the early part of display. The trend was for seeds to result in higher TBARS than oil and for L to result in higher TBARS than C.

Table 77: Fatty acids concentration (mg/100g muscle), TBARS (mg malondialdehyde/kg muscle) and vitamin E (mg/g) in *M. longissimus dorsi*

Treatment	ML	CO	LO	CS	LS	CA	s.e.d.	Sig	Oil	Seed	Sig	C	L	P
Fatty acids														
C 18:2	123.4	139.8	130.9	126.0	123.5	144.3	11.63	NS	135.4	124.8	NS	132.9	127.2	NS
C 18:3	18.4 ^a	60.9 ^b	76.5 ^{bc}	68.4 ^{bc}	103.1 ^d	81.1 ^c	8.47	***	68.7	85.8	*	64.7	89.8	**
CLAc9t11	29.3 ^a	44.2 ^{bc}	42.2 ^{abc}	49.6 ^{cd}	58.5 ^d	33.5 ^{ab}	7.36	**	43.2	54.1	*	46.9	50.4	NS
TVA	99.1 ^a	217.1 ^c	223.8 ^c	164.9 ^b	155.1 ^b	137.3 ^{ab}	25.74	***	200.5	160.0	**	191.0	189.5	NS
P:S Ratio	0.15 ^a	0.17 ^b	0.18 ^b	0.19 ^b	0.22 ^c	0.18 ^b	0.010	***	0.18	0.2	**	0.18	0.20	**
n-6:n-3 Ratio	5.28 ^c	2.14 ^b	1.75 ^b	1.83 ^b	1.15 ^a	1.72 ^{ab}	0.248	***	1.95	1.49	*	1.99	1.45	**
Total FA	3524	4659	4177	4056	3980	4171	376.0	NS	4418	4018	NS	4358	4079	NS
TBARS														
- Day 3	0.23 ^a	0.66 ^{ab}	1.33 ^{bc}	1.04 ^{ac}	2.89 ^d	0.24 ^a	0.605	**	1.00	1.97	NS	0.85	2.11	*
- Day 6	0.60 ^a	1.47 ^{ac}	2.99 ^{bcd}	2.05 ^{ac}	4.56 ^d	2.15 ^{ac}	1.230	*	2.23	3.31	NS	1.76	3.77	*
- Day 9	1.92	2.65	4.87	3.05	6.89	4.06	2.375	NS	3.76	4.97	NS	2.85	5.88	NS
- Day 12	3.81	5.99	7.41	5.48	11.05	7.16	3.250	NS	6.70	8.27	NS	5.74	9.23	NS
Vitamin E	4.71 ^c	2.27 ^a	2.99 ^{a,b}	2.71 ^{a,b}	2.05 ^a	3.41 ^b	0.467	**	2.63	2.38	NS	2.49	2.52	NS

ML, CO, LO, CS, LS, CA, C and L = megalac, camelina oil, linseed oil, camelina seed + NaOH, linseed + NaOH, camelina amide, camelina and linseed, respectively. Means with similar superscripts do not differ significantly

Caustic treatment of oilseeds *per se* and chemical protection of C increased the concentration of C18:3 in muscle compared to oil. Higher muscle C18:3 concentration was associated with higher oxidation during retail display despite high and similar vitamin E consumption, but this was not reflected in a loss of colour stability.

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Inferring the origin of beef from C, N and S stable isotope ratio analysis

About 10% of meat produced globally for human consumption is traded internationally. Yet, existing livestock traceability schemes, based on tags, tattoos, life numbers and animal passports, depend ultimately on a paper trail. There is an urgent need for scientific, independent technologies for meat authentication to reassure consumers, project regional designations and ensure fair competition. The objective of this feasibility study reported here was to explore the usefulness of the C, N and S stable isotope composition as a potential marker of the international geographical origin of beef cattle.

Samples of Belgian (N = 2), Dutch (N = 3), French (N = 2), German (N = 5), Italian (N = 1) and Brazilian (N = 10) beef were obtained from two licensed Irish meat importers. These meat samples (about 200 g each) had no background information other than country of origin, but were stated to be from different animals. Spanish samples (N = 5) were sourced through the Universidad Complutense de Madrid from a local commercial supplier. US samples (two lots, N = 11 and 12) came from research herds (University of Nebraska) fed typical diets high in maize for 85 and 180 d prior to slaughter. Conventional Irish samples were also included in this study.

Lipids were extracted from 100 mg muscle powder. Natural abundance stable-isotope ratios of carbon (¹³C/¹²C), nitrogen (¹⁵N/¹⁴N) and sulphur (³⁴S/³²S) were measured on the de-fatted muscle residue by continuous flow isotope ratio mass spectrometry. Isotope ratios are expressed in delta (δ) notation in parts per thousand (‰).

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Data were analysed by single factor Multivariate Analysis of Variance (MANOVA) based on Pillai's test statistic, using Minitab R 13.20 (Minitab Inc., State College PA, USA).

European beef (including conventional Irish) was significantly different from American beef (MANOVA, $F_{265} = 327.3$, $P < 0.001$) based on C and N isotopic compositions. The observed large difference in $\delta^{13}\text{C}$ between European and American beef (Fig. 12) can only be explained by contrasting proportions of plants with C_3 and C_4 photosynthetic pathways. Mean $\delta^{13}\text{C}$ values for conventional Irish ($-24.5^{0/00} + 0.7^{0/00}$) and other European ($-21.6^{0/00} + 1.0^{0/00}$) samples suggest a predominance of C_3 plants. By contrast, considerably less negative $\delta^{13}\text{C}$ values for US ($-12.3^{0/00} + 0.1^{0/00}$) and Brazilian ($10.0^{0/00} + 0.6^{0/00}$) beef reflect the almost exclusive use of C_4 foodstuffs, likely maize or (sub) tropical C_4 pasture grasses.

These results identify $\delta^{13}\text{C}$ as a single marker that distinguishes American from European beef. This probably holds true for northern Europe, including Ireland and Britain, where pastoral beef production systems predominate and use of the only C_4 crop, maize, is marginal. Interestingly, Irish conventional beef was also significantly different from other European beef (MANOVA, $F_{2,32} = 14.8$, $P < 0.001$) based on $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$. This result suggests that isotopic origin authentication of beef may work on smaller geographic scales.

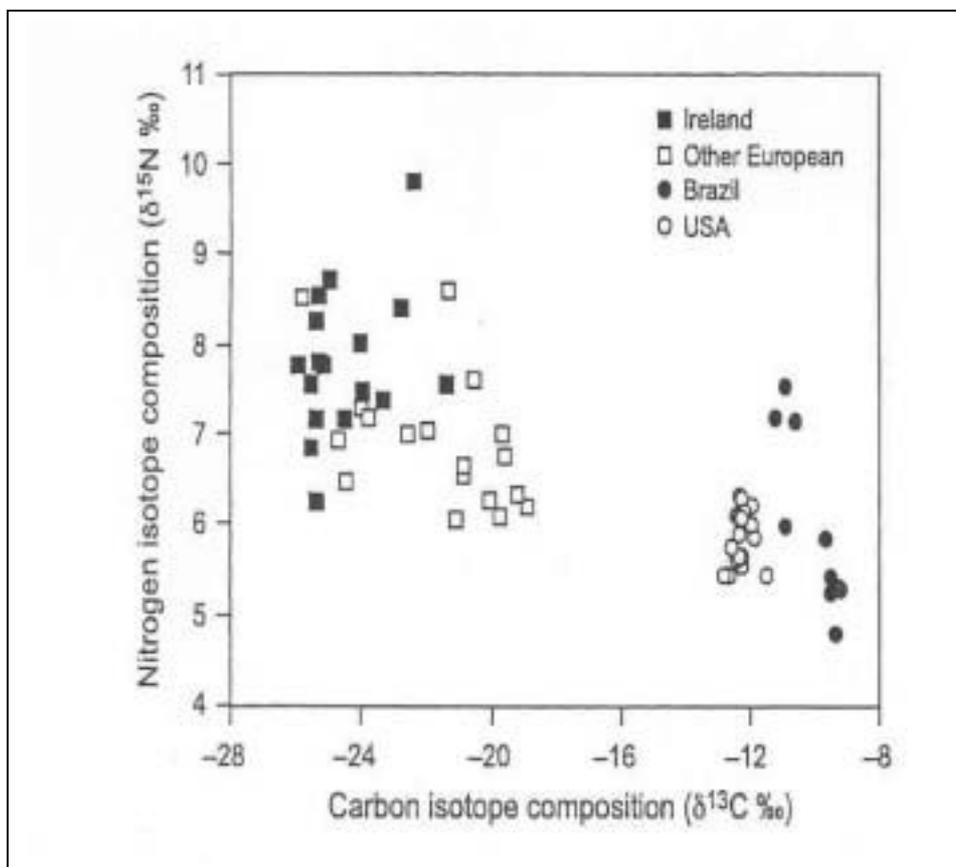


Figure 12. Nitrogen and Carbon isotope composition of meat samples from different countries.

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ANIMAL HEALTH AND WELFARE

Effect of 12-hour road transportation on adrenal, immune responses and body weight of bulls previously housed at different space allowances

Farm animals housed at different space allowances are routinely transported as a common management practice in the beef industry. Animals housed at reduced space allowance have been reported to show a degree of chronic stress and associated physiological changes. The objective of this study was to examine the effects of a 12 h transport of bulls by road which had previously been housed for 96 days at a range of space allowances, and to measure their adrenal, haematological, immune responses, and body weight (BW).

Seventy-two Holstein x Friesian bulls (mean BW = 403.4 kg, s. e. m. = 3.69 kg) were blocked by BW and randomly allocated to one of three (1.2, 2.7, 4.2 m²/bull) space allowances (N = 24/space allowance with 4 bulls/pen) and housed for 96 days. On day 97, bulls were assigned to two treatments, transport (T) and control (C). On days 97, 98 and 99 forty-eight bulls (N = 16/day) were transported (T) and twenty-four bulls were not transported and served as controls (C). Bulls were given access to grass silage (DM = 190.2 g/kg) and supplemented with a barley plus soybean mix daily and had free access to water. Indwelling jugular catheters for blood collections were fitted the day before each transportation. Blood samples, for cortisol determination were collected before transportation in the home pen (HP1), pre-loading crush facility (CR1), after loading in the transporter (load), after 12h transport in the transporter (unload), after unloading in the crush (CR2), in the home pens (HP2). Blood samples were also collected for interferon (IFN)- γ and Neutrophil % at HP1, C2, and after 24 and 168 h at HP2. C bulls were sampled on d 99 in parallel to T bulls for all the variables described above. The induced release of cortisol to a standardised dose of adrenocorticotrophic hormone (ACTH; 1.98 IU/kg metabolic BW) was tested in two bulls of T (after transport) and C treatment from each pen. Blood samples were collected, for a 4-h window at 30-min intervals following ACTH administration. T bulls were weighed before and after transport and C at time corresponding to T bulls. The data were analysed by two-way ANOVA. The model included the effect of treatment, space allowance and their interactions at each time point. Where appropriate the pre-transport treatment values were included as a covariate. The area under the cortisol vs. time curve (AUC) was calculated from the time of ACTH administration until the final blood sample was collected.

The basal median plasma cortisol concentrations in the C and T treatment bull were less than 3.28 ng/mL. The data for the cortisol concentrations are presented in the Table 78. There was no difference ($P > 0.05$) between treatment, space allowance and treatment x space allowance for the basal median plasma cortisol concentrations. Bulls assigned to the T had greater ($P = 0.001$) median plasma cortisol concentrations than C bulls. There was a treatment x space allowance effect ($P = 0.001$) at loading in the median plasma cortisol concentrations. The median plasma cortisol concentrations were greater ($P = 0.005$) in the bulls housed at 4.2 than 1.2 m² space allowance following loading in the transporter. The median plasma cortisol concentrations were greater ($P = 0.015$) in the bulls housed at 1.2 than 4.2 m² space allowance of the C treatment. The loading of the T bulls into the transporter previously housed at 1.2, 2.7 and 4.2 m² space allowance increased (for 1.2 m², $P = 0.007$; 2.7 m², $P = 0.001$ and for 4.2 m², $P = 0.001$, respectively) median plasma cortisol concentrations compared with C bulls housed in the corresponding space allowances. Following 12-h of transport, at unloading there was a treatment and space allowance effect. The median plasma cortisol concentrations were greater ($P = 0.006$) in the T than the C bulls and the bulls housed at 4.2 m² had greater ($P < 0.005$) median plasma cortisol concentrations than the bulls housed at 1.2 and 2.7 m² space allowance. Following unloading after the 12-hour road journey, there was a tendency ($P = 0.076$) for greater median plasma cortisol concentrations in the bulls housed at 4.2 m² than the bulls housed at 1.2 and 2.7 m² space allowance. The T bulls in the home pens housed at 1.2

m² space allowance had greater ($P = 0.053$) median plasma cortisol concentrations than the T bulls housed at 2.7 and 4.2 m² space allowances.

The adrenal glands of the T bulls responded acutely and showed an increased integrated cortisol response to the exogenous administration of ACTH than the C bulls housed at 1.2 ($P = 0.002$), 2.7 ($P = 0.044$) and 4.2 ($P = 0.026$) m² space allowance, respectively (Table 79). Following saline administration, no differences ($P > 0.05$) were observed in the T vs C bulls housed at 1.2, 2.7 and 4.2 m² space allowance, respectively. Bulls in the C treatment, housed at 2.7 m² space allowance and administered exogenous ACTH had greater ($P = 0.045$) integrated cortisol response than bulls housed at 1.2 m² space allowance. Integrated cortisol response in the T bulls administered with saline and housed at 1.2 m² was greater ($P = 0.046$) than the 4.2 m² space allowance. The integrated cortisol response was not different (T, $P = 0.974$; C, $P = 0.797$) in the bulls housed at 1.2, 2.7 and 4.2 m² space allowance for the T and C treatment, administered with ACTH and saline, respectively. The peak cortisol concentrations following the administration of ACTH in the bulls housed at 1.2 ($P = 0.007$) and 4.2 ($P = 0.047$) m² space allowance were greater in the T than the C bulls. The time to reach peak cortisol concentrations following administration of ACTH was greater in the T than C bulls housed at 1.2 m² space allowance. There was no difference in the time to reach peak cortisol concentrations in the T than C treatment bulls housed at 2.7 and 4.2 m² space allowance. There was a clear response (T, $P = 0.001$; C, $P = 0.001$) to exogenously administered ACTH when compared with the saline for the integrated mean plasma cortisol concentrations within the T and C treatments.

There was no ($P > 0.05$) effect of treatment, space allowance and treatment x space allowance before transport in the home pens, at 24 h and 168 h following transport on in vitro IFN- γ production (Table 80). Following transport, there was a treatment and space allowance main effect, indicated by the suppression ($P = 0.001$) of the mean Con A induced IFN- γ production in the T compared with C bulls and suppression ($P = 0.049$) of the mean Con A induced IFN- γ production in the bulls housed at 4.2 compared with those at 1.2 and 2.7 m² space allowance. Mean PHA induced IFN- γ production tended to be lower ($P = 0.067$) in the bulls housed at 4.2 than 1.2 and 2.7 m² space allowance following transport when animals were blood sampled in the crush holding facility.

There was a greater ($P = 0.001$) neutrophil % in the C compared with the T bulls in the home pens pre-transport. Following transport, the neutrophil % was increased in the T compared with the C treatment in the crush holding facility and at 24 h after transport (Table 81). Bulls in T treatment housed at 2.7 and 4.2 m² space allowance tended ($P = 0.089$) to have greater neutrophils % than the C treatment in the crush holding facility following transport. There was a main treatment effect on the lymphocyte %, described by the increased ($P = 0.018$) lymphocyte % before transport treatment in the T than in the C bulls. The lymphocyte % was not affected ($P = 0.014$) by transport, however, it tended ($P = 0.078$) to be greater in the bulls previously housed at 2.7 than 4.2 m² space allowance when sampled in the crush holding facility (crush2). Twenty-four hour following transport, the lymphocyte % was greater ($P = 0.002$) in the C compared with the T bulls. There was no ($P > 0.05$) treatment, space allowance and treatment x space allowance effect at 168 h. The eosinophil % was greater ($P < 0.05$) in the T than C bulls in the home pens prior to transport, in the crush holding facility following transport, and at 24 h following transport. No effects ($P > 0.05$) of the treatment, space allowance and treatment x space allowance were observed for the basophil and monocyte %. The band cells %, which are an immature form of neutrophils, were greater ($P < 0.05$) in the T bulls housed at 4.2 m² space allowance than C bulls, when sampled in the crush holding facility (crush2).

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The data for the blood cell constituents are presented in the Table 82. The main space allowance effect is indicated by greater ($P < 0.05$) red cell counts (RBC) numbers in the bulls housed at 1.2 and 2.7 than 4.2 m² space allowance before transport in the home pens, crush 2 and at 168 h after transport. However, at 24 h following transport, T bulls housed at 1.2 m² had a greater ($P = 0.012$) RBC count than the bulls housed at 2.7 and 4.2 m² space allowance. Packed cell volume tended to be greater ($P = 0.067$) in the T than in C bulls in the crush holding facility following transport. The treatment x space allowance effect at 24 h and 168 h post transport treatment is indicated by the increased PCV in the T than in C bulls housed at 4.2 m² space allowance. The Hb concentration was affected by main effect of treatment with greater ($P = 0.047$) Hb concentrations in the T than in C at crush 2. At 24 and 168 h post transport there was a treatment x space allowance effect. At 24 and 168 h, bulls in C housed at 1.2 m² space allowance had greater ($P < 0.05$) Hb concentrations than bulls housed at 2.7 and 4.2 m² space allowance and was greater (24 h, $P = 0.021$; 168 h, $P = 0.042$) in the T than in C bulls housed at 4.2 m² space allowance.

There was a greater ($P = 0.001$) loss of body weight following transport in the T than in C bulls (Table 83). Overall, the mean bodyweight loss was 2.5% in the C and 6% in the T bulls, respectively. Following transport, there was a greater tendency ($P = 0.095$) of weight loss in the bulls housed at 2.7 than 1.2 m² space allowance. At 168 h post transport, there was tendency towards increased weight gain ($P = 0.061$) in T than in C bulls. One week following transport, the weight loss had recovered in the T bulls and was only 0.5% different from their initial weight, while, C bulls had a 2% difference in the weight compared with initial bodyweight.

No ($P > 0.05$) effect of treatment, space allowance, or treatment x space allowance was observed in the rectal, shoulder, belly and rump temperature data (data not shown).

The mean daily air temperature (location 1: 11.5°C (range 7.9 to 14.9°C); location 2: 11.7°C (range 8.3 to 14.3°C) and relative humidity (location 1: 80.0% (range 57.9 to 90.8%); location 2: 77.9% (range 57.4 to 86.0) in the two housing locations were not different ($P > 0.05$). No obvious signs of injury or illness were detected during the experiment and the general health status of the animals was good.

In conclusion, housing bulls for 96 days in a range of space allowances did not affect basal cortisol response and immune function. Housing bulls at space allowance < 2.7 m² /bull produced a time-dependent acquired adaptation against the chronic low grade housing stress for cortisol release and is only apparent when bulls were administered with exogenous ACTH. ACTH administration may be a beneficial indicator for assessing the effects of chronic stress during the housing of cattle. ACTH challenge failed to differentiate between the increased adrenal response of cattle undergoing chronic stress and if subsequently exposed to the acute stress of transport. However, exogenous ACTH administration revealed that with the increase in the severity of the stressor from chronic to acute there is increased adrenal responsiveness and this warrants further investigation. The physiological data indicate that loading bulls on a transporter, transporting for 12-h and subsequently unloading reduced body weight, suppressed Con A and PHA induced INF- γ production, and produced neutrophilia, eosinophilia and lymphopenia. Transportation also affected the normal body homeostasis by increasing the packed cell volume, red blood cell and haemoglobin levels. The stress induced changes associated with transport were within normal physiological ranges and returned to basal levels within one week suggests that 12-h road transport does not adversely affect animal welfare.

Table 78: Effect of transporting bulls previously housed at 1.2, 2.7 and 4.2 m² on plasma cortisol (levels (ng/ml) are presented as median, lower (min) and upper (max) quartiles)

	Control						Transport				P ^a		
	1.2 m ²	2.7 m ²	4.2 m ²	1.2 m ²	2.7 m ²	4.2 m ²	Treat ^b	Space ^c	Treat x space				
Space allowance/bull	median	2.18	3.29	3.60	2.89	2.10	3.72	NS	NS	NS			
	min-max	(0.60 - 6.05)	(0.60-8.81)	(0.47-7.86)	(0.60-26.51)	(0.48-11.44)	(0.97-18.96)						
Home pen 1 ^d	median	2.18	3.29	3.60	2.89	2.10	3.72	NS	NS	NS			
	min-max	(0.60 - 6.05)	(0.60-8.81)	(0.47-7.86)	(0.60-26.51)	(0.48-11.44)	(0.97-18.96)						
Crush 1 ^e	median	1.55	1.18	1.13	5.12	4.67	5.85	0.0001	NS	NS			
	min-max	(1.15-2.76)	(0.60-2.99)	(0.60-1.48)	(0.60-17.50)	(0.60-19.04)	(2.57-19.71)						
Load	median	1.95	0.96	0.60	3.55	5.19	8.64	0.0001	NS	0.0019			
	min-max	(0.59-3.29)	(0.60-2.37)	(0.60-1.12)	(1.30-17.99)	(1.49-31.03)	(2.80-17.74)						
Unload	median	3.21	2.24	8.37	9.03	8.52	11.31	0.0006	0.0026	NS			
	min-max	(1.74-12.19)	(0.73-5.58)	(4.01-16.62)	(1.62-82.62)	(1.19-34.27)	(2.21-77.82)						
Crush 2 ^e	median	2.71	5.18	11.21	7.01	6.18	8.41	NS	0.0755	NS			
	min-max	(1.49-10.12)	(1.23-21.95)	(3.46-20.18)	(0.65-68.77)	(0.52-21.05)	(1.59-68.92)						
Home pen 2 ^f	Median	2.92	1.50	1.91	1.61	1.40	2.48	NS	0.0532	NS			
	min-max	(1.50-5.70)	(0.95-3.21)	(0.49-3.26)	(0.60-10.42)	(0.47-6.19)	(1.08-8.88)						

^aNon-significance, defined as $P \geq 0.10$, is denoted as NS

^bTreat = treatment; C-control; T-transport.

^cSpace = 1.2, 2.7, 4.2 m² average individual space allowance.

^dMean of cortisol concentrations at 1900 and 1915 GMT in the home pens before transport.

^eCrush holding facility before and after transport.

^fMean of the cortisol concentrations at 1000, 1024 and 1048 GMT in the home pens after transport.

Table 79: Effect of transporting bulls previously housed at 1.2, 2.7 and 4.2 m² on plasma cortisol following administration of ACTH (1.98 i.u. ACTH/kg metabolic body weight)

Variables ^a	Treatment	Space allowance/bull			Pooled s.e.	P
		1.2 m ²	2.7 m ²	4.2 m ²		
Post-ACTH cortisol AUC, ng/ml min	Control (N = 12)	5816 ^{b*}	9352 ^{c*}	7607 ^{bc*}	1246.2	0.05
	Transport (N = 20)	12423 *	12642 *	12301 *	788.4	NS
Post-saline cortisol AUC, ng/ml min	Control (N = 12)	920	820	833	109.1	NS
	Transport (N = 24)	989 ^b	816 ^{bc}	593 ^c	106.6	0.041
Post-ACTH peak ^d cortisol, ng/ml	Control (N = 12)	36.3 *	45.1	42.8 *	3.24	NS
	Transport (N = 20)	64.5 *	59.1	66.7 *	4.87	NS
Post-ACTH interval ^e to peak cortisol, min	Control (N = 12)	93 *	128	141	9.7	NS
	Transport (N = 20)	130 *	120	113	9.1	NS

^aCortisol concentrations at 1000,1024,1048 GMT in the home pens were averaged and included as a covariate.

^{bc}Means with different superscripts within variable and treatment differ, $P \leq 0.05$.

^dPeak cortisol is presented for the bulls administered with ACTH only.

^ePeak time to reach peak values following ACTH administration.

^{*}Means with in variables for treatment differ, $P \leq 0.05$.

The least square means are presented for the post ACTH and saline administration area under the cortisol vs. time curve (AUC), post-ACTH peak cortisol and interval to peak cortisol.

Table 80: The effect of transporting bulls previously housed at 1.2, 2.7 and 4.2 m² on slats, on mean concanavalin A (Con A) and phytohaemagglutinin (PHA) induced *in vitro* interferon- γ production (optical density @450nm) from cultured whole blood

Space allowance/bull	Control			Transport			Pooled s.e.	Treat ^b	P ^a	
	1.2 m ²	2.7 m ²	4.2 m ²	1.2 m ²	2.7 m ²	4.2 m ²			Space ^c	Treat x space
Con A										
Pre-transport, home pen	0.87	0.72	0.46	0.65	0.86	0.65	0.140	NS	NS	NS
Crush 2 ¹	0.48	0.67	0.48	0.32	0.39	0.10	0.092	0.001	0.049	NS
24 h post-transport	0.78	1.23	0.67	0.79	0.67	0.66	0.200	NS	NS	NS
168 h post-transport	0.57	0.54	0.47	0.72	0.77	0.70	0.155	NS	NS	NS
PHA										
Pre-transport, home pen	1.68	1.33	1.18	1.54	1.76	1.49	0.201	NS	NS	NS
Crush 2 ¹	1.34	1.52	1.19	1.12	1.42	0.81	0.185	NS	0.067	NS
24 h post-transport	1.30	1.62	1.18	1.22	1.32	1.09	0.246	NS	NS	NS
168 h post-transport	0.74	0.68	1.29	1.35	1.46	1.51	0.216	NS	NS	NS

¹Crush holding facility after transport.

Table 81: The effect^a of transporting bulls previously housed at 1.2, 2.7 and 4.2 m² on slats on differential white blood cell counts

Space allowance/bull	Control				Treatment				Pooled s. e.	P ^b		
	1.2 m ²	2.7 m ²	4.2 m ²	1.2 m ²	2.7 m ²	4.2 m ²	1.2 m ²	2.7 m ²		4.2 m ²	Treat ^c	Space ^d
Neutrophils, %	Pre-transport, home pen	40.3	41.8	40.8	36.0	36.3	34.5	1.04	0.0001	NS	NS	NS
	Crush 2 ¹	39.9	38.9	33.4	45.2	46.9	49.3	1.65	0.0001	NS	NS	0.0896
	24 h post-transport	39.6	39.7	39.1	47.9	39.8	44.5	1.26	0.004	NS	NS	NS
Lymphocytes, %	168 h post transport	42.7	40.1	40.5	40.1	40.2	41.0	1.00	NS	NS	NS	NS
	Pre-transport, home pen	43.0	44.1	48.4	47.1	47.1	44.6	1.20	0.0189	NS	NS	NS
	Crush 2 ¹	41.6	47.6	36.9	41.4	40.8	39.4	1.69	NS	0.0783	NS	NS
Eosinophils, %	24 h post-transport treatment	44.4	45.4	39.2	35.3	40.4	45.6	1.40	0.0002	NS	NS	NS
	168 h post transport treatment	39.9	43.8	43.5	44.1	44.2	39.9	1.03	NS	NS	NS	NS
	Pre-transport, home pen	8.3	6.8	9.9	10.5	8.8	6.8	0.83	0.0203	NS	NS	NS
Basophils, %	Crush 2 ¹	8.0	5.8	3.9	5.3	5.3	7.9	1.03	0.0312	NS	NS	NS
	24 h post-transport treatment	3.4	3.5	8.4	10.1	8.8	7.1	0.88	0.0001	NS	NS	NS
	168 h post transport treatment	6.8	5.6	7.6	8.8	7.8	7.5	0.64	NS	NS	NS	NS
Monocytes, %	Pre-transport, home pen	4.4	4.6	8.0	8.0	7.7	3.5	0.70	NS	NS	NS	NS
	Crush 2 ¹	6.5	4.0	4.2	6.7	4.8	6.2	0.80	NS	NS	NS	NS
	24 h post-transport treatment	3.2	4.6	4.8	4.5	6.4	4.9	0.75	NS	NS	NS	NS
Band, %	168 h post transport treatment	4.5	4.2	5.5	6.2	6.1	4.6	0.45	NS	NS	NS	NS
	Pre-transport, home pen	10.1	7.8	8.8	9.4	10.1	7.7	0.76	NS	NS	NS	NS
	Crush 2 ¹	8.4	7.8	6.7	8.0	4.3	6.6	0.98	NS	NS	NS	NS
Eosinophils, %	24 h post-transport treatment	8.8	10.6	8.5	9.1	8.0	10.2	0.88	NS	NS	NS	NS
	168 h post transport treatment	9.3	9.0	8.2	9.9	7.7	8.4	0.54	NS	NS	NS	NS
	Pre-transport, home pen	7.4	6.4	9.8	9.6	9.0	8.7	1.10	NS	NS	NS	NS
Monocytes, %	Crush 2 ¹	2.1	0.6	2.2	6.9	2.0	20.9	1.96	0.0013	0.0043	NS	0.0135
	24 h post-transport treatment	11.2	12.9	8.0	5.5	5.8	7.0	1.81	NS	NS	NS	NS
	168 h post transport treatment	8.6	7.3	12.9	5.7	4.2	5.8	0.89	NS	NS	NS	NS

^aData presented are angular transformed for individual percentages. ^bNon-significance, defined as $P \geq 0.10$.

^cTreat = treatment; C - control; T- transport. ^dSpace = 1.2, 2.7, 4.2 m² average space allowance. ¹Crush holding facility after transport.

Table 82: The effect of transporting bulls previously housed at 1.2, 2.7 and 4.2 m² on slats on haematological variables

Space allowance/bull	Control			Treatment			Pooled s.e.	<i>P</i> ^a		
	1.2 m ²	2.7 m ²	4.2 m ²	1.2 m ²	2.7 m ²	4.2 m ²		Treat ^b	Space ^c	Treat x space
Red blood cell, x 10 ¹² /l	Pre-transport, home pen	8.1	7.5	6.9	7.7	7.5	7.2	0.15	NS	NS
	Crush 2 ¹	8.4	7.9	7.5	8.2	8.1	7.8	0.17	NS	0.0358
	24 h post-transport	8.2	7.2	6.8	7.9	7.6	7.3	0.14	NS	0.0001
Packed cell volume %	168 h post transport	8.5	8.0	7.4	8.0	8.2	7.8	0.15	NS	0.0125
	Pre-transport, home pen	27.8	26.5	25.3	27.0	27.2	27.2	0.50	NS	NS
	Crush 2 ¹	28.7	28.0	27.5	29.1	29.1	29.3	0.49	0.0698	NS
Haemoglobin, x g/dl	24 h post-transport treatment	28.6	25.5	24.8	27.2	27.0	26.9	0.49	NS	0.0076
	168 h post transport treatment	29.4	29.0	27.2	28.5	29.9	29.8	0.5	NS	NS
	Pre-transport, home pen	10.2	9.9	9.3	10.2	10.2	10.1	0.17	NS	NS
Crush 2 ¹	Crush 2 ¹	10.7	10.5	10.4	10.9	11.0	11.0	0.18	0.0474	NS
	24 h post-transport treatment	10.8	9.7	9.4	10.3	10.2	10.2	0.17	NS	0.0076
	168 h post transport treatment	10.9	10.7	10.2	10.6	11.2	11.0	0.16	NS	0.0476

^aNon-significance, defined as $P \geq 0.10$, is denoted as NS.

^bTrans = treatment; C - control; T- transport.

^cSpace = 1.2, 2.7, 4.2 m² average space allowance.

¹Crush holding facility after transport.

Table 83: The effect of transporting bulls previously housed at 1.2, 2.7 and 4.2 m² on slats, on body weight^a

Space allowance/bull	Control			Treatment			Pooled s.e.	<i>P</i> ^b		
	1.2 m ²	2.7 m ²	4.2 m ²	1.2 m ²	2.7 m ²	4.2 m ²		Treat ^c	Space ^d	Trans x sp
Crush 2	-12.63	-16.00	-11.75	-25.00	-29.75	-29.93	1.226	0.0001	0.0852	NS
168 h post-transport	-11.38	-7.13	-7.25	-2.06	-5.19	-6.07	1.768	0.061	NS	NS

^aData are least square means and analysed by subtracting the values of post transport at crush holding facility (crush 2) and 168 h from the pre-transport values.

^bNon-significance, defined as $P \geq 0.10$, is denoted as NS.

^cTrans = treatment; C - control; T- transport.

^dSpace = 1.2, 2.7, 4.2 m² average space allowance.

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RMIS No. 5230

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Effect of transport for up to 24 hours followed by twenty-four hours recovery on liveweight, physiological and haematological responses of bulls

The objective of the study was to investigate the effect of transport on liveweight, physiological and haematological responses of bulls after road transport of 0, 6, 9, 12, 18 and 24h. Eighty-four continental x bulls (mean weight (s.d.) 367 (35) kg) were randomly assigned to one of six journey (J) times of 0, 6, 9, 12, 18 and 24 h transport at a stocking density of 1.02m²/bull. Blood samples were collected by jugular venipuncture before, immediately after and at 1, 2, 4, 6, 8, 12 and 24 h and bulls were weighed before, immediately after, and at 4, 12 and 24 h. Bulls travelling for 6, 9, 12, 18 and 24 h lost 4.7, 4.5, 5.7 ($P \leq 0.05$), 6.6 ($P \leq 0.05$) and 7.5 ($P \leq 0.05$) percentage liveweight compared with baseline. During the 24 h recovery period liveweight was regained to pre-transport levels. Lymphocyte percentages were lower ($P \leq 0.001$) and neutrophil percentages were higher ($P \leq 0.001$) in all T animals. Blood protein and creatine kinase concentrations were higher ($P \leq 0.001$) in the bulls following transport for 18 and 24h and returned to baseline within 24 h. In conclusion, liveweight, physiological and haematological responses of bulls returned to pre-transport levels within 24 h having had access to feed and water. Transport of bulls from 6 – 24 hours did not impact negatively on animal welfare. A more detailed report is in preparation.

Earley, B.

RMIS No. 5230

Effects of Carprofen administration on cortisol response and *in vitro* Interferon-production during banding and burdizzo castration of bulls

Castration of male cattle has been shown to elicit physiological stress, inflammatory reactions, pain associated behaviour, reduced performance (Fisher et al., 1996; Ting et al., 2003a,b) and reduced immune function (Fisher et al., 1997; Earley and Crowe, 2002; Ting et al., 2003a,b). Both Burdizzo and surgical castration have been reported to cause less chronic pain than banding castration (Molony et al. 1995). Carprofen, (6-chloro-alpha-methyl-9H-carbazole-2-acetic acid) is a non-steroidal anti-inflammatory medication (NSAID). Its elimination half-life is significantly longer than those of other NSAIDs used in veterinary medicine, which is in the range of 44.5 to 64.6 hours. The hypothesis of the study was that carprofen would suppress the rise in plasma cortisol and prevent immune suppression (*in vitro* IFN- production) without altering blood haematology following banding and burdizzo castration.

Fifty Holstein - Friesian bulls (5.5 mo old; 191 ± 3.7 kg) were assigned to one of five treatments: (1) untreated control (Con); (2) banding castration at 0 min (Band); (3) Band following an i.v. injection of 1.4 mg/kg of BW of carprofen at -20 min (Band+C); (4) Burdizzo castration at 0 min (Burd); or (5) Burd following 1.4 mg/kg of BW of carprofen at -20 min (Burd+C).

Bulls were housed in individual tie-stalls from d-14 (day of treatment = d0) to acclimate them to handling, restraint and their novel housing environment. Animals had *ad libitum* access to water and grass silage supplemented with 2.5 kg of barley/soybean mix concentrates per animal daily. Individual silage intakes were recorded daily from d -14 to 16 (from day 17, the animals were put to grass) to determine DMI. Animals were weighed on d -14 before assignment to treatment, and on d -1, 7, 14, 21, 28, and 35 to determine ADG.

Band and Band+C animals were castrated (time of treatment = 0 min) with latex bands applied to the neck of the scrotum using a banding instrument (Callicrate Smart Bander). The bands were applied following the protocol outlined in the instruction manual supplied with the instrument. Burdizzo castration (time of treatment = 0 min) was performed in the Burd, Burd+C bulls following the procedure of Fisher et al. (1996). As part of the castration procedure, gentle manual restraint of the bulls was used to facilitate the operator. Carprofen, a NSAID, was administered in Band+C and Burd+C bulls i.v. 20 min before treatment at the rate of 1.4 mg of carprofen/kg of BW (Rimadyl solution, 50mg/ml carprofen, Pfizer,) via an indwelling jugular catheter followed by 2 mL of 0.9% sterile saline to flush the catheter.

Animals in the control group were sham handled for a period equivalent to the time required to perform the castration procedure in the remaining treatment groups. Precise dose rates for C was determined based on the BW of each animal obtained on d -1 before treatments. Animals not receiving C in the S group were given an equivalent volume of sterile 0.9% saline solution via their jugular catheter.

To facilitate intensive blood sampling, indwelling jugular catheters were fitted aseptically on d -1 with a 12-gauge Anes spinal needle (Popper and Sons, Inc., New Hyde Park, NY) and vinyl tubing (approximately 1.47 mm o.d.; Ico-Rally Corp., Palo Alto, CA; catalogue No.SVL 105-18 CLR). All catheters were exteriorized on the neck of the animals, filled with sterile 3.5% sodium citrate solution, and plugged with a stopper. Catheters were secured in place in resealable patches with the aid of an adhesive cement (Big Bull Hip Tag Cement; Biguel Supply Co., Elysian, MN), Velcro, and zinc oxide wrapping bandages. After catheterization, animals were returned to their individual tie stalls. On d 0, blood samples (heparinized plasma) were collected at -2, -1.5, -1, -0.5, -0.25, 0, 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 6.5, 7, 7.5, 8, 10, 12, 24, 72 h relative to the time of treatment for each bull for subsequent cortisol assay. Heparinized blood samples for stimulated leukocyte production of interferon- γ (IFN- γ) and EDTA blood samples for routine hematology were collected before treatment on d 0 and on d 1 and 3. Further blood samples for hematology were collected on d 7, 14, 21, 28, and 35. Plasma samples were separated by centrifugation at 1,600 g at 8 °C for 15 min and subsequently stored at -20 °C until assayed. Rectal temperatures of each bull were monitored with a digital electronic thermometer (Jørgen Kruuse A/S model VT-801 BWC, Marslev, Denmark; catalogue No. 0701) on d -1, -2 h prior castration, 12 h after treatments and on d 1, 2, 3, 7, 14, 21, 28 and 35. All procedures were conducted under experimental license from the Irish Department of Health in accordance with the Cruelty to Animals Act 1876, and the European Communities (Amendment of Cruelty to Animals Act 1876) Regulations, 1994.

Plasma cortisol concentrations were determined using a commercially available RIA kit (Cortico-cote, ICN Pharmaceuticals, Orangeburg, NY; catalogue No. 06B-256440) adapted and validated for bovine plasma (Fisher et al., 1996). The intraassay CV (N = 6) for samples containing 4.9,

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17.3 and 51.7 ng of cortisol/mL were 15.3, 10.3 and 7.5%, respectively, and the interassay CV (N =10) for the same samples were 10.9, 8.6 and 5.9%. For each animal, the mean cortisol concentration was calculated for the periods -2 to 0, 0.25 to 1.5, 2 to 6, and 6.5 to 12 h relative to the time of treatment. The peak post-treatment cortisol concentration was recorded, and the area (ng·ml⁻¹·h) under the cortisol vs. time curve (AUC) was calculated for the pre-treatment period from -2h to 0 h, and for above the pre-treatment baseline from 0 to 12 h using the linear trapezoidal rule:

$$\sum \{[(C_t + C_{t+1}) \times 0.5] \times \Delta I\}$$

where C_t is the concentration of a plasma cortisol sample in ng/ml of an animal at time t , and for the next sample C_{t+1} with a time interval of ΔI in hours between them, and \sum is the sum of the responses from C_t to $n-1$ total number of concentration time points (Friend et al., 1977).

The stimulated lymphocyte production of IFN- γ was determined from harvested supernatant following a whole-blood culture (Wood et al., 1990). Duplicate 1.48-ml aliquots of heparinized blood were cultured in sterile 24-well flat culture plates (Sarstedt Ltd., Drinagh, Wexford, Ireland) with 20 μ l of PBS (GibcoBRL, Life Technologies Ltd., Paisley, Scotland; catalogue No. 14190-094) containing either 1 mg/ml of PHA (Sigma, product No. L-9132) or 1 mg/ml of Concanavalin A (Con A; Sigma-Aldrich, Inc., St. Louis, MO; product No. C 5275) or no additive for 24 h at 37 °C and in an atmosphere of 5% CO₂ in air. Aseptic techniques were practised during this procedure under laminar flow conditions. The culture plates were then centrifuged and the supernatant harvested and frozen at -20°C until it was assayed for IFN- γ using an ELISA procedure (Rothe et al., 1990; Bovigam, CSL Biosciences, Parkville, Victoria, Australia; catalogue No. 03000201). The *in vitro* PHA- and Con A-stimulated IFN- γ production was calculated by subtracting the absorbance at 450 nm of wells that received PBS alone from the absorbance of wells that received either PHA or Con A.

Red blood cell (RBC) number, white blood cell (WBC) number, differential WBC (percentage granulocyte, percentage monocyte, and percentage lymphocyte), packed cell volume (PCV), hemoglobin concentration, mean corpuscular volume (MCV), and platelet numbers were determined for unclotted (EDTA) whole-blood samples with an automated cell counter (Celltac MEK-6108K; Nihon-Kohdon, Tokyo, Japan) within 1 to 2 h of blood sampling. Thin blood smears were also prepared and stained using the haematology three-step stain for differential WBC counts.

All statistical analyses were performed using SAS V8.2. Data that departed from the assumptions of normality and/or homogeneity of variance were subjected to suitable transformations before ANOVA. Data relating to the mean plasma cortisol by period, peak cortisol, interval to peak cortisol, plasma haptoglobin and log₁₀ of plasma fibrinogen, haematological variables (RBC number, log₁₀ of WBC number, angular transformed ($x = (180/\pi) \times \arcsin(\sqrt{p/100})$), where p is a percentage ($0 < p < 100$; $\pi = 3.1416$)) differential WBC subpopulations, PCV, haemoglobin concentration, MCV, and platelet number), and ADG were analysed by ANOVA using a randomised complete-block design for the main effect of treatments at each individual time point (Steel and Torrie, 1960). The AUC for cortisol from 0 to 12 h, log₁₀ ($x + 1$) of IFN- γ production, total antioxidant status, rectal temperature, and DMI data were similarly analyzed by ANOVA with the pre-treatment values (from -2 to 0 h of the AUC for cortisol, d 0 for INF- γ and total antioxidant status, d-1 for rectal temperature and d 9 to d-4 for DMI) included as significant covariates.

Following an F -test, Fisher's least significant difference test was applied to determine statistical differences between treatments (Steel and Torrie, 1960). A probability of $P < 0.05$ was chosen as the level of significance for the statistical tests.

Mean plasma cortisol concentrations in the control animals from -2 to 12 h was less than 5.7 ng/ml, and there were no differences ($P=0.68$) among treatment groups in plasma cortisol concentrations from -2 to 0h before treatment. From 0.25 to 1.5 h following castration, plasma cortisol concentrations in the castrated animals increased acutely (Table 84) and were greater ($P < 0.001$) than in Con; Band animals had higher ($P<0.05$) cortisol concentration compared with Burd animals. The administration of C during the same time period failed to prevent the elevation of cortisol in both the Band+C ($P=0.18$) and Burd+C ($P=0.72$) animals. From 2 to 6 h post castration, only Band animals had higher ($P<0.05$) cortisol concentration than Con animals, though the administration of C had no effect at the same time period. Peak cortisol concentrations were greater ($P < 0.05$) in all castration treatments than in Con, C administration had no effect in this parameter; the intervals to peak cortisol concentrations in Band castrated animals were longer than ($P < 0.001$) in Burd castrated animals. Mean cortisol concentrations of Band+C animals from 6.5 to 12 h after castration was lower compared with Band animals ($P =0.04$); Burd animals had higher value compared with Con animals ($P=0.03$) and with no difference compared with Burd+C animals in the same period of time (Table 84). On d 1, mean cortisol concentration of Band and Burd animals were still high ($P<0.05$) than Con. C was not effective in reducing cortisol concentration during this same period, though the C treated groups had no differences compared with Con. On d3, mean cortisol concentration of Burd animals was high ($P<0.05$) compared with Con; in contrast, Burd+C animals had reduced ($P<0.05$) cortisol concentration compared with Burd animals.

Overall, the integrated cortisol response (i.e., AUC) was greater ($P < 0.05$) in all the castrated animals except Burd+C animals compared with Con animals. In contrast, the provision of C failed to reduce the integrated cortisol response in Band+C ($P=0.08$) and Burd+C ($P=0.07$) animals compared with Band and Burd animals respectively.

Table 84: The effect of treatment on mean plasma cortisol concentration by period, peak cortisol concentration, interval to peak cortisol, and area under the cortisol time curves (AUC)

Item	Treatment			
	Con	Band	Band+C	Burd
Burd+C				
Time	<u>Mean plasma cortisol, ng/ml</u>			
-2 to 0 h	6.7±0.27	6.3±0.26	6.2±0.28	6.4±0.28
0.25 to 1.5 h	8.4±2.63 ^X	32.7±2.54 ^Y	27.7±2.71 ^{YZ}	23.3±2.73 ^Z
2 to 6 h	5.5±1.21 ^X	10.0±1.17 ^{YZ}	8.5±1.25 ^{XZ}	7.7±1.25 ^{XZ}
6.5 to 12 h	5.2±0.84 ^{XZ}	7.1±0.81 ^{W X}	4.6±0.87 ^{YZ}	7.9±0.87 ^W
1 d	3.9±1.34 ^X	8.1±1.29 ^{YZ}	4.9±1.38 ^{XZ}	8.7±1.39 ^{YZ}
2 d	4.4±1.29	4.5±1.24	5.4±1.33	6.7±1.33
3 d	3.2±0.89 ^X	2.3±0.86 ^X	2.8±0.92 ^X	7.7±0.92 ^Y
Peak plasma cortisol, ng/ml	17.6±3.08 ^X	42.7±3.08 ^Y	34.7±3.25 ^{YZ}	34.4±3.08 ^{YZ}
Interval to peak cortisol, h ^a	-----	1.18±0.103 ^X	1.17±0.108 ^X	0.48±0.103 ^Y
AUC, (ng/ml)·h	59.9±12.27 ^X	138.3±12.27 ^Y	106.5±12.94 ^{YZ}	124.2±12.27 ^{YZ}

^a First peak reached within 2 h after treatment.

^{W,X,Y,Z} Means within a row that do not have common superscripts differ ($P<0.05$).

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There were no differences ($P=0.60$) among treatments in pre-treatment plasma fibrinogen concentrations (Figure 13a). On d1, Band+C animals had lower ($P<0.05$) fibrinogen concentration compared with Band and Burd animals. On d3, Burd animals had higher ($P<0.05$) fibrinogen compared with Burd+C, Band+C and Con animals. On d14, Band and Burd animals had elevated fibrinogen ($P<0.05$) compared with Band+C, Burd+C and Con animals. On d21 and 28, Burd+C had lower fibrinogen ($P<0.05$) compared with Con. There were no differences ($P=0.58$) among treatments in pre-treatment plasma haptoglobin concentrations (Figure 13b). On d1, there were no differences in haptoglobin concentration between castrates and Con animals. On d3, Band animals had elevated ($P<0.05$) haptoglobin compared with Con. No differences in plasma haptoglobin concentration found among treatments on d7, 14, 21 and 28. On d35, Band animals had higher ($P<0.05$) haptoglobin compared with animals in other 4 treatment.

Figure 13a and Figure 13b: The effect of treatment on fibrinogen (a) and haptoglobin (b) concentrates.

Figure 13a:
Fibrinogen

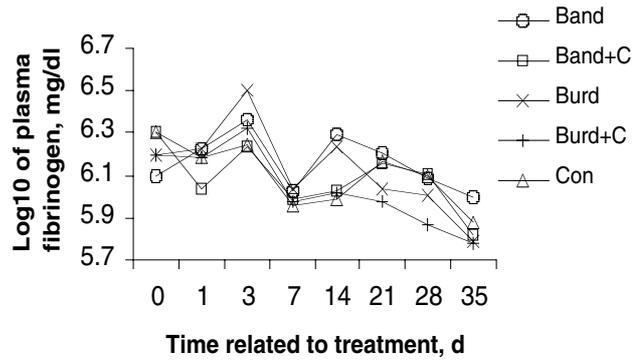
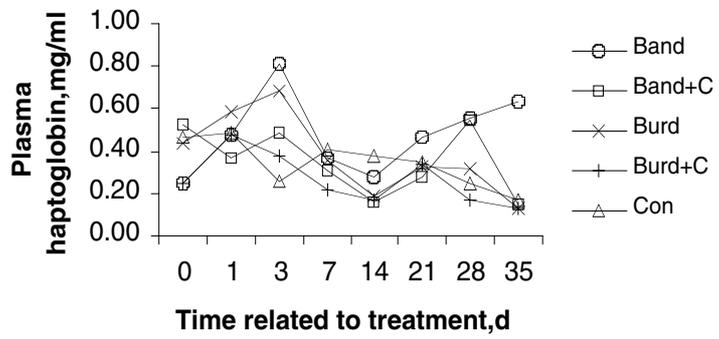


Figure 13b:
Haptoglobin



There were no differences ($P>0.05$) among treatments in IFN- γ production in response to Con A from leukocytes cultured in whole-blood samples collected on d0 before castration and on d2, neither in response to PHA on d0, 1 and 2. On d1, Burd animals had lower ($P<0.05$) IFN- δ production in response to Con A compared with Band (Figure 14a and 14b).

Figure 14a: Con A-induced Interferon-gamma

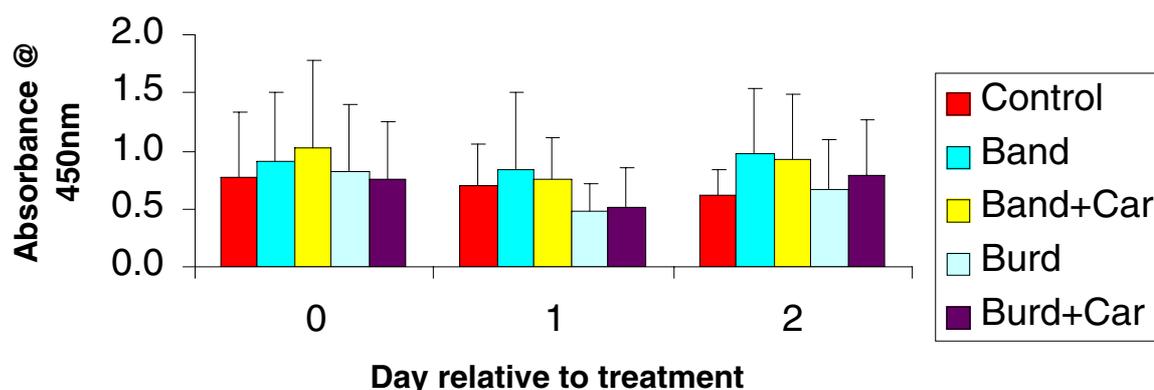
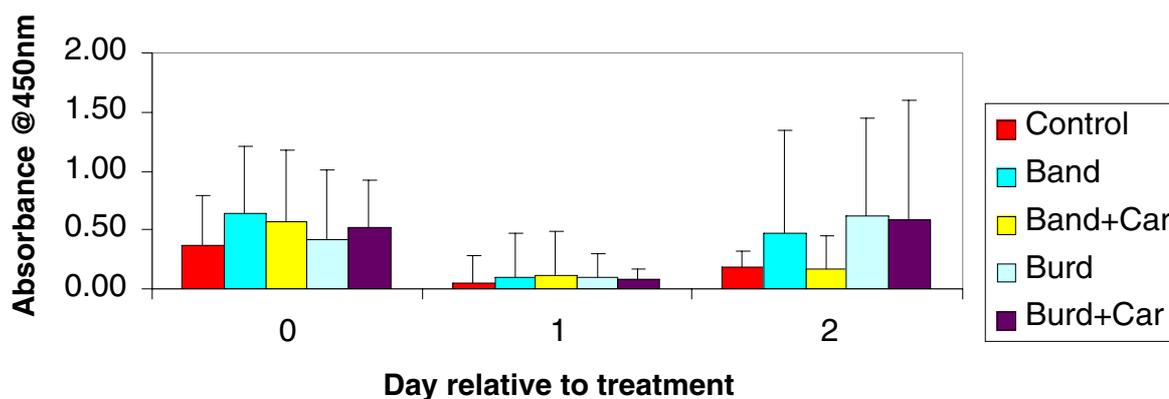


Figure 14b: PHA-induced Interferon-gamma



Generally, there were only limited effects of castration treatments on hematological parameters. There were no differences among treatments in the hematological variables on d0, except Band+C animals had higher ($P<0.05$) white blood cell (WBC) and lower ($P<0.05$) Red Blood Cell (RBC) counts compared with Con; Band+C animals had higher ($P<0.05$) percentage of granular cells (Gr%) compared with Burd, Burd+C and Con.

Band+C animals had lower ($P<0.05$) percentage of lymphocytes (Ly%) compared with Burd, Burd+C and Con; Band+C animals had lower ($P<0.05$) platelet number than Con.

Generally, there were no differences among treatments ($P>0.05$) on the changes in WBC numbers following castration, except on d1, Band+C animals had greater ($P=0.03$) number of

WBC than Con; on d7, Band animals had greater ($P<0.01$) WBC number than Burd and Con animals; and on d28, Band+C animals had greater ($P<0.05$) WBC number compared with Con. On d1, Burd+C animals had lower ($P<0.05$) RBC compared with Burd. On d7, Band animals had higher ($P<0.05$) RBC number compared with Con. On d14, Band+C and Burd+C animals had higher ($P<0.05$) RBC compared with Con. On d21, Burd+C animals had higher ($P<0.05$) RBC compared with Con. On d2, Band+C animals had lower ($P<0.05$) Gr% compared with Band and Burd+C. On d7, Band animals had higher ($P<0.05$) Gr% compared with Band+C, Burd and Con, with no difference between Band+C and Con. On d14, Burd+C animals had higher ($P<0.05$) Gr% compared with Con. On d21, Burd+C animals had higher ($P<0.05$) Gr% compared with Burd. On d2, Band+C animals had higher ($P<0.05$) Ly% compared with Band and Burd+C. On d7, Band animals had lower ($P<0.05$) Ly% compared with Band+C, Burd and Con, with no difference between Band+C and Con. On d14, Burd+C animals had lower ($P<0.05$) Ly% compared with Con. On d21, Burd+C animals had lower ($P<0.05$) Ly% compared with Burd. On d35, Band animals had lower ($P<0.05$) Ly% than Con animals. On d7, Band animals had higher ($P<0.05$) percentage of Monocyte compared with Band+C. On d7 and 14, Band animals had lower ($P<0.05$) platelet number compared with Band+C. On d21, Band+C animals had higher ($P<0.05$) platelet number compared with Burd and Burd+C. On d28, Burd had higher ($P<0.05$) platelet number than Burd+C. On d35, Band+C had higher ($P<0.05$) platelet number than Burd+C. There were no differences found in N:L ratios among treatments following castration.

There was no difference ($P=0.18$) in DMI among treatment groups from d-14 to d-1 before treatment. There were no differences ($P=0.70$ for d0 to d 6; $P=0.26$ for d7 to d16) in DMI among treatments before d 16, the day before the animals were put to grass. Overall from d 0 to 16, there were no difference ($P=0.45$) in DMI in all castrated groups compared with Con. There were no difference ($P=0.70, 0.36, 0.45, 0.14,$ and 0.53 for week 1, 2, 3, 4 and 5, respectively) in ADG among treatments (data not shown). Overall, from d -1 to 35, ADG was lower ($P < 0.05$) in Band compared with Burd animals.

Rectal Temperature

There were no rectal temperature (RT) differences ($P>0.84$) among treatment groups on d -1 and -2h before treatment (Table 85). At 12h following treatment, Burd animals had higher ($P<0.05$) RT compared with Con. At d1 post castration, Band and Burd animals had higher ($P<0.05$) RT than Con. At d2 following castration, all castrated animals had higher ($P<0.05$) RT compared with Con. There were no RT differences ($P>0.05$) among treatments on d 3, 7, 14, 21, 28 and 35 (data not shown).

Table 85: The effect of treatment on mean rectal temperature

RT	Treatment				
	Con	Band	Band+C	Burd	Burd+C
-1 d	38.3±0.22	38.2±0.22	38.5±0.23	38.5±0.22	38.4±0.22
-2 h	38.4±0.16	38.4±0.16	38.3±0.17	38.4±0.16	38.3±0.16
12 h	38.2±0.17 ^X	38.5±0.17 ^{XY}	38.5±0.18 ^{XY}	38.8±0.17 ^Y	38.6±0.17 ^{XY}
1 d	37.8±0.14 ^X	38.3±0.14 ^Y	38.1±0.15 ^{XY}	38.4±0.14 ^Y	38.2±0.14 ^{XY}
2 d	37.7±0.12 ^X	38.2±0.13 ^Y	38.4±0.13 ^Y	38.4±0.12 ^Y	38.3±0.12 ^Y
3 d	38.4±0.12	38.7±0.12	38.7±0.12	38.6±0.12	38.7±0.12

^{W,X,Y,Z} Means within a row that do not have common superscripts differ ($P<0.05$)

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Activation of the neuroendocrine-immune axis is a hallmark of stress, tissue injury, and infection. In the current study, the stresses induced by banding castration and burdizzo castration increased the integrated cortisol response (i.e., AUC) by 131% and 107%, respectively, compared with control animals. The pattern and duration of castration-induced plasma cortisol elevation measured in the burdizzo animals were in accord with previous experiments. There were no differences between Band and Burd animals, except Band animals had higher cortisol than Burd animals from 0.25 h to 2.5 h. However, on d 3, Burd animals had higher concentration of cortisol compared with animals in other 4 treatments. The Carprofen treatments failed to reduce AUC, peak plasma cortisol and the interval to peak cortisol in castrated animals, except that the mean cortisol on d 3 in Burd+C animal was reduced compared with Burd animals.

Following castrations on d1, 3, 7, 14, 21 and 28, the plasma haptoglobin concentration did not elevate in Band and Burd castrated animals compared with Con. However, On d35, Band animals had higher ($P<0.05$) haptoglobin compared with animals in other 4 treatment. The Band+C animals had no difference in haptoglobin with Con animals. Following castration on d3, fibrinogen concentration was elevated in Burd animals, and the C administration was effective in Burd+C animals in suppressing this elevation. On d14, Band and Burd animals had elevated fibrinogen ($P<0.05$) compared with Con animals. The C treatment were both effective in reducing this fibrinogen response in Band+C and Burd+C groups. In contrast, Ting et al. (2003b) reported that the burdizzo castration-induced increase in acute-phase proteins (plasma haptoglobin and fibrinogen) on d1 and 3 which were consistent with previously reported findings in surgical castrations. Haptoglobin production can be related to the magnitude of an inflammatory stimulus. Since on d35, Band animals showed higher haptoglobin compared with Burd animals, it would suggest that Band cause longer inflammatory response compared with Burd castration. Generally in this study, Band and Burd caused less acute-phase protein production than those reported in surgical castrations, this may also suggest that these two non-surgery castrations cause less inflammation compared with surgical method. The effect of C administration in reducing fibrinogen and haptoglobin would indicate C treatment is effective in reducing the magnitude of castration-induced inflammatory responses.

Immunological assessment is a useful indicator of cattle welfare, and the establishment of protective immunity depends critically on IFN- γ . IFN- γ is a cytokine produced by activated T lymphocytes and natural killer cells that helps to regulate immune responses to antigens. An increase in *in vitro* IFN- δ production correlates well with lymphocyte proliferation, and this has been used as a measure of immune responsiveness in calf castration studies. In the present study, there were no detectable differences in either Con A- or PHA-induced IFN- δ production in all the castration treatments compared with controls on d 0, 1, and 3. This would indicate the responsiveness of the lymphocytes of the Band and Burd castrated animals was not compromised. By contrast, Con A-induced IFN- γ production in the Burdizzo castrated animals was lower compared with Control animals on d 1.

Banding and Burdizzo castrations did not significantly affect WBC numbers except on d7, Band animals had greater ($P<0.01$) WBC number than Burd and Con animals; On d1 and d28, Band+C animals had greater ($P<0.05$) number of WBC than Con. However, the administration of Carprofen in Burdizzo-castrated calves did not affect WBC counts. On d2, Band+C animals had lower ($P<0.05$) Gr% compared with Band and Burd+C and on d7, Band animals had higher ($P<0.05$) Gr% compared with Band+C, Burd and Con, with no difference between Band+C and Con. This would suggest Carprofen administration could reduce Gr% in Banding castrated calves. However, on d14, Burd+C animals had higher ($P<0.05$) Gr%

compared with Con, and on d21, Burd+C animals had higher ($P < 0.05$) Gr% compared with Burd. This would suggest Carprofen was not effective in reducing Gr% in Burdizzo-castrated calves. Generally there was no difference ($P > 0.05$) found in neutrophil:lymphocyte ratio and other hematological parameters among treatments and would indicate the health of the animals was not compromised.

Overall from d 0 to 16, there were no differences ($P > 0.05$) in DMI and in ADG in all castrated groups compared with Con. By contrast, Ting et al. (2003) reported burdizzo-castration resulted in reduced animal growth rate but no feed intake differences. The ADG was lower ($P < 0.05$) in Band compared with Burd animals and this would be in agreement with the report that Burdizzo castration appeared to produce the least pain compared with surgical and banding castrations.

In conclusion, banding and burdizzo castrations acutely increased the secretion of cortisol with no suppression of Con A- and PHA-induced IFN- γ production from leukocytes in whole-blood cultures, and no reduction in animal feed intakes and growth rates. Carprofen treatments had no significant effects on the overall cortisol response and the initial peak response to castration. Castration caused an increase in acute-phase protein production and carprofen was effective in reducing this elevation.

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LABOUR USE ON BEEF FARMS

Agricultural contractors as a labour source on Irish suckler farms

Knowledge about agricultural contractors has not kept pace with their growing significance to agriculture. Indirect evidence shows that the number of contractors, as well as their degree of use on farm has increased substantially and is likely to continue to do so. As the cost of labour has risen, farm incomes have decreased, and fewer farm workers are seeking employment in agriculture. Farmers are increasingly looking for flexible, skilled labour sources at peak times of the year. Agricultural contractors can achieve economies of scale beyond the reach of individual farmers, through use of sophisticated and expensive agricultural machinery, as well as specialised knowledge and skills. Therefore, the objectives of this study were to 1) quantify the use of agricultural contractors on Irish suckler farms, and 2) identify the main tasks on Irish suckler farms for which contractors are employed.

Data were collected from 115 predominantly spring calving suckler farmers distributed evenly across the east and west of the country. Each farmer recorded all activities undertaken by an agricultural contractor on their farm for each month of the year. The task activity was recorded by the farmer, as was the length of time taken to complete the task. The participating farmers completed an accompanying questionnaire focusing on agricultural contractors. Uni-variate and bi-variate analysis was carried out on data using SPSS version 8.0.

From the data collected, it was found that 97% of the surveyed sample used an agricultural contractor on their farm for at least one task during the farming year. Figure 15 below shows the monthly fluctuations in the recorded use of agricultural contractors for all suckler farms surveyed. In the early part of the year the number of jobs carried out per month by agricultural contractors was low, and were chiefly concerned with farm maintenance and slurry and fertiliser spreading. Contractor use increased substantially in June, as sheep were sheared, silage was harvested, and slurry and fertiliser were spread. The number of contract jobs carried out per month remained high throughout the summer months, as a result of silage harvesting and slurry spreading, and towards the end of the summer, and early autumn as a result of cereal harvesting. Contract jobs began to decrease again into late autumn and winter.

Table 86 below shows that a total of 755 tasks were documented over the 12 months of the study. The average farmer involved in suckler beef alone was found to use the agricultural contractor for a total of 5.7 jobs per annum. The average suckler farmers who operated an arable enterprise, was found to be using an agricultural contractor for 7.2 jobs per annum. The average suckler farmer who had a mix of other enterprises in operation on the farm, used the agricultural contractor for 9.6 contract jobs per annum.

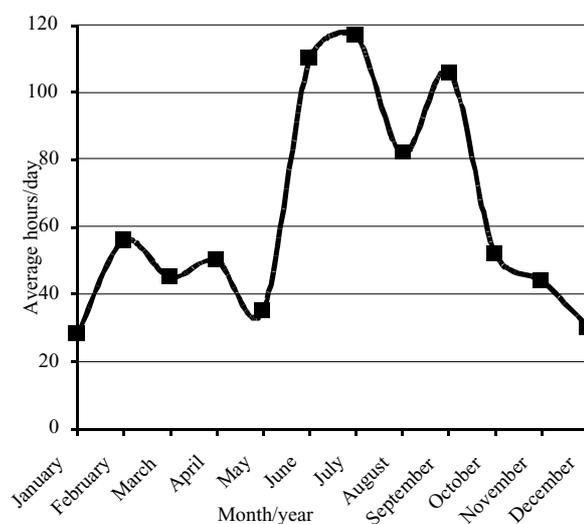


Figure 15. Monthly fluctuations in the recorded use of agricultural contractors for all tasks on farm.

From Table 86 it is clear that the majority of agricultural contractors activities were concerned with silage and hay making, slurry, fertiliser and farmyard manure spreading, and land and building maintenance.

Table 86: Suckler farm type and use of agricultural contractors by contract task (measured in tasks per farm per annum)

Farm type	Feeding Cleaning Land & Building Maint.	Spreading Slurry, Farmyard Manure & Fertiliser	Silage & Hay making	Other enterprises	Other tasks*	Total	Mean
(S) Only	58	74	62	0	24	218	5.7
(S) Sheep	70	60	66	39	15	250	6.4
(S) Dairy	4	7	14	1	4	30	6.0
(S) Arable	10	14	24	33	12	93	7.2
(S) Mixed	38	30	24	52	20	164	9.6
Total	180	185	190	125	75	755	6.7
% of total	24%	24%	25%	17%	10%	100%	

Further more a breakdown of the 11,788 hrs which contractors spend on suckler farms was assigned to various farm tasks as detached in Table 87.

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Table 87: Contractor tasks in hrs according to farm tasks

Task	Hrs	% of farms
Conservation	5,800	86
Slurry	1,720	57
Maintenance/fencing	3,377	44
Animal husbandry	301	40
Miscellaneous	<u>590</u>	
Total	11,788	

The tasks most assigned to contractors were conservation which accounted for 50% of contractor time, slurry handling for 15% and maintenance of 25% of contractor time with the final 10% allocated to fencing, animal husbandry and miscellaneous.

Contractors are widely used by suckler beef farmers. Contract work varies hugely across farm types, and with the time of year. Silage and hay making, slurry, fertiliser and farmyard manure spreading, and land and building maintenance appear to be the main tasks on suckler farms for which contractors are employed.

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RMIS No.5084

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ENVIRONMENT

Impact of cattle on nutrient losses in overland flow – Small plot study

The small plot study on the impact of cattle on nutrient losses in overland flow was set up in spring 2002. The rationale behind the work, the experimental set up and some of the methods employed are described in the Johnstown Castle Annual Research Reports 2002 and 2003.

The aims of the study were to measure the impact of cattle on soil physical properties relevant to the hydrological characteristics of a site, and to assess whether or not the presence of grazing animals is likely to influence the quality of the overland flow produced at a site.

The study was completed at the beginning of August 2004 and a detailed report on it was submitted to the EPA (Environmental Protection Agency), who co-financed the work with Teagasc, in early 2005.

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RMIS No. 5022

CATTLE REPRODUCTION

Progesterone regulation of uterine gene expression in cows

Previous studies at Athenry show that low systemic progesterone in the first week after AI is associated with significant embryo loss in dairy cows (see Research Report, 2003), however, the mechanism involved is not clear. One possibility is that progesterone may affect uterine function through alteration of uterine gene and ultimately uterine protein expression. There is evidence in other species that progesterone controls uterine retinol binding protein secretion which is essential for early embryo development. Retinol binding protein is essential for vitamin A transport to the embryo. The objective of this study was to establish the relationship between systemic progesterone during the first week after AI and the expression pattern of the retinol binding protein gene in the cow uterus.

Beef cross heifers (N=12) were blood sampled twice daily on days 0 to 6 (day of ovulation=day 0) and the systemic progesterone concentration measured by radioimmunoassay. Heifers were divided on the basis of their progesterone concentrations into high (H) or low (L) groups on day 4 after AI. Half of each group was in turn supplemented (S) with exogenous progesterone from day 4 to day 6 resulting in a total of four progesterone groups, low control (LC), high control (HC), low supplemented (LS) and high supplemented (HS). Uterine endometrial tissue was harvested *post mortem* on day 6 and snap frozen in liquid nitrogen. Retinol binding protein (RBP) cDNA probes were generated from total RNA using RT-PCR and a primer designed and based on published GenBank sequences. Total RNA from the endometrial tissue was separated by electrophoresis and, following blotting and hybridisation with the [α^{32} P]dCTP labelled cDNA probe, the blot was exposed to a phosphor imaging plate which was subsequently scanned using a phosphor imager. Following densitometric analysis of the image the data were normalised for loading and relative gene expression was calculated as the ratio between the calibrated volumes for the gene divided by the 28S rRNA volume. Data were analysed as a 2 x 2 factorial using analysis of variance (Proc GLM SAS 2001).

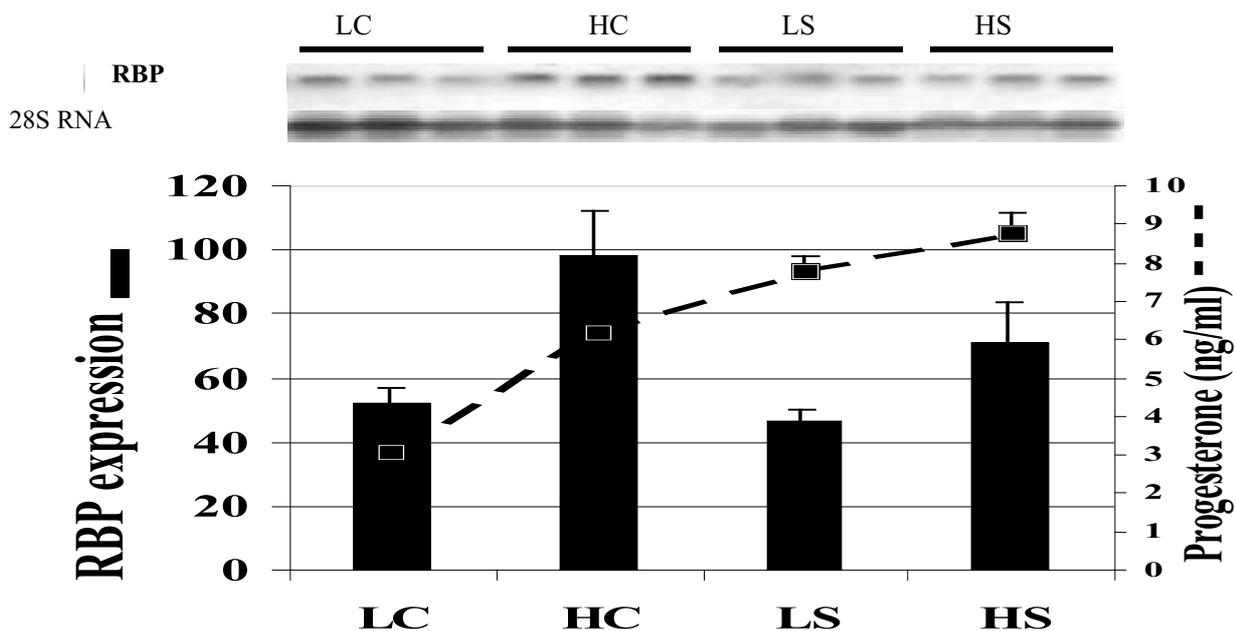


Figure 16. Northern blot and histogram showing progesterone concentration and relative expression of the retinol-binding protein gene in the uterus on day 6 after ovulation in the treatment groups

Expression of the retinol-binding protein (RBP) gene was detected in uterine endometrium from all 4 groups. High systemic progesterone concentration up to day 4 was associated with increased ($P < 0.01$)

RBP mRNA expression (Figure 16). In contrast, supplementation with exogenous progesterone between days 4 and 6 did not alter RBP gene expression ($P>0.05$). These results suggest that the concentration of progesterone that the uterine endometrium is exposed to during the first few days after ovulation is more important in terms of regulating RBP expression than subsequent exposure. It would seem that exposure of the uterus to optimal concentrations of progesterone is required prior to day 4 after ovulation in order to ensure adequate RBP support for the embryo which enters the uterus around day 4. It is known that RBP binds retinoids and retinoic acid and is vital for the transport of vitamin A, which is essential for regulating cellular differentiation and proliferation in the early developing embryo. Progesterone is known to affect early embryo survival in cattle and the progesterone induced changes in RBP gene expression coincide with the period of early embryo loss.

The expression of RBP mRNA is apparently very sensitive to the concentration of systemic progesterone during the first few days after AI. In this context inadequate progesterone support is likely to be associated with sub-optimal retinol-binding protein gene expression in cattle resulting in a reduction in embryo growth and increased embryo loss.

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RMIS No. 4675

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Maternal environment and embryo survival in cattle

Up to 40 percent of cattle embryos die within three weeks of fertilisation while they are nutritionally dependent on the fluids in the reproductive tract for their growth, development and survival. However, there is little published information on the composition of these fluids during the period of early embryo development. Previously we have reported on the pH, and on the concentrations of the main energy substrates in these fluids (see Research Report 2003). Despite the importance of amino acids as protein precursors and energy sources and the importance of ions as regulators of pH, fluid secretion, osmolarity and enzyme activity, little is known of their concentrations in cattle oviduct or uterine fluid. The objective of this study was to determine the concentrations of amino acids, anions and cations in cattle oviduct and uterine fluid on different days of the oestrous cycle.

Reproductively normal crossbred heifers ($N=59$), of similar age (average 20 ± 4 months), live weight (408 ± 4.6 kg) and body condition score (3.2 ± 0.1 units) were used for oviduct and uterine fluid collection. Oviduct fluid was collected on days 0, 2, 4 and 6 and uterine fluid on days 6, 8 and 14 after oestrus (day of oestrus = day 0). Amino acid concentrations were measured by high performance liquid chromatography and ion concentrations by ion chromatography. The results were analysed by analysis of variance (SAS v8.02). Model terms were tested using heifer, within-day mean square as the error term. Values are presented as least square means \pm SEM.

The comparative pattern of amino acid concentrations for oviduct and uterine fluid is shown in Figure 17. Glycine and alanine were the predominant amino acids in the oviduct while glycine and taurine were predominant in the uterus. The concentration of glycine was higher ($P<0.05$) and taurine was lower ($P<0.001$) in the oviduct than in the uterus. In general the amino acid concentrations were affected by day of cycle in the uterus ($P<0.05$) but not in the oviduct ($P>0.05$).

The comparative pattern of ion concentrations for oviduct and uterine fluid is shown in Figure 18. Sodium was up to 80-fold and chloride up to 40-fold higher than most of the other ions in both fluids. Phosphate, sulphate, magnesium and potassium were higher ($P<0.05$) in the uterus compared to the oviduct.

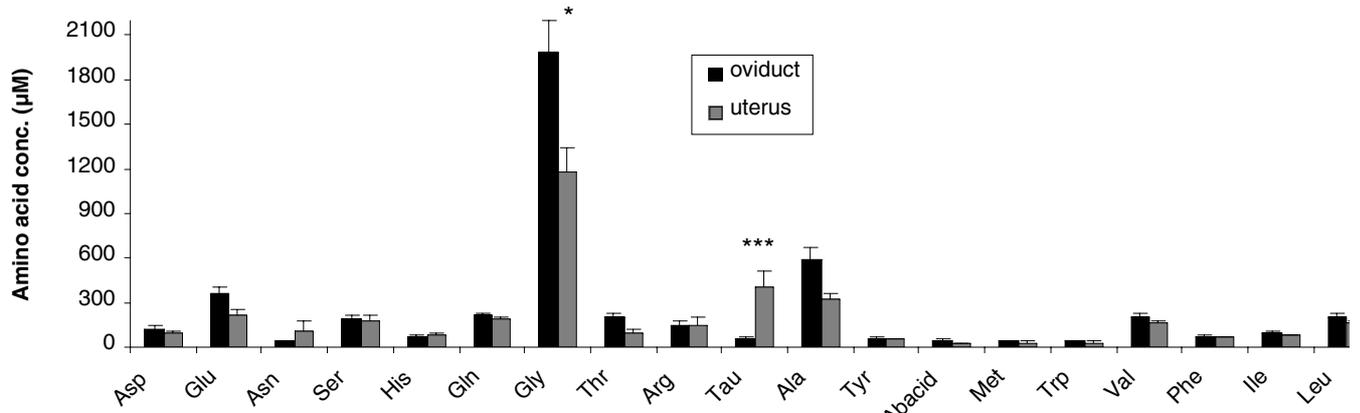


Figure 17. Concentrations (µM) of amino acids in bovine oviduct and uterine fluid on day 6 of the oestrous cycle. * = P<0.05; *** = P<0.001.

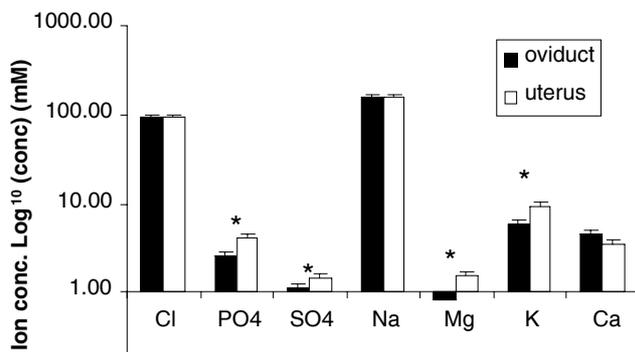


Figure 18. Concentrations (µM) of ions in bovine oviduct and uterine fluid on day 6 of the oestrous cycle. * = P<0.05.

The results provide novel baseline information describing the amino acid and ion concentrations in the maternal environment of the developing cattle embryo. The differential concentrations between the oviduct and uterus with respect to specific amino acids and ions and the differential effect of day of cycle suggest that the use of two sequential in vitro culture media, based on the composition of oviduct and uterine fluid, may enhance in vitro cattle embryo production. Furthermore, this information will help identify factors that may deleteriously affect the composition of oviduct and uterine fluid, and therefore reduce embryo survival.

RMIS No. 4951

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Negative energy balance and gene expression in dairy cow metabolic and reproductive tissues

Negative energy balance (NEB) is a severe metabolic disorder affecting high yielding dairy cows during the early *post partum* interval and this can limit peak milk production and impair cow fertility and animal health. The object of this study is to investigate the affects of NEB on global patterns of gene expression in liver and reproductive tissues collected during the period of greatest NEB between cows managed to achieve a high or low energy balance status. The biological material for this experiment was collected as part of a larger Moorepark energy balance study (RMIS 4997). Twenty-four multiparous dairy cows were blocked according to parity and BCS three weeks prior to expected calving date and were randomly allocated within block to one of two postpartum dietary treatments differing in energy density and designed to create two divergent groups of animals in terms of energy balance (mild versus severely negative) postpartum. From the day of calving onwards animals were individually fed and energy balance was calculated on a daily basis. Follicular dynamics were recorded on a daily basis from seven days postpartum. From the original 24 cows, 12 animals (6 per treatment) were selected for slaughter on the basis of energy balance and possession of healthy oestrogen active, dominant follicle. Animals were slaughtered on day 14 *post partum* (N=6 per treatment) and metabolic and reproductive tissues collected under RNase free conditions for subsequent molecular analysis. Mean energy balance on the day of slaughter for the mild and severe NEB cows was -1.4 UFL/day and -6.1 UFL/day, respectively. Glucose, metabolites (BHB, NEFA, urea) and systemic IGF-I concentrations were significantly different ($P<0.05$) between groups confirming the divergence in energy balance. Amyloid and PGFM and blood parameters including white blood cell count and percentage lymphocytes were also measured but showed no significant difference between groups ($P>0.05$). Total RNA was extracted from metabolic (liver) and reproductive (oviduct and uterine) tissues. All samples displayed intact 28S and 18S rRNA transcripts. The quality and quantity of the isolated RNA was also confirmed using automated capillary gel electrophoresis on a Bioanalyzer 2100 with RNA 6000 Nano Labchips. This RNA will be used for quantitative RT-PCR to examine the expression of IGFI, IGFII and their binding proteins. This RNA will also be used in year two to examine the affects of NEB on gene expression in liver and reproductive tissue using the recently constructed 23,000 Affymetrix bovine array.

RMIS No.5234

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Functional genomic analysis of the bovine corpus luteum

The lifespan of the bovine corpus luteum (CL) is an important factor in the control of normal ovarian cyclicity and the establishment and maintenance of pregnancy. Premature CL regression is associated with embryo loss while persistent CLs that fail to regress contribute to abnormal oestrous cycle duration. To gain further insights into the genes and mechanisms controlling the lifespan of the CL a 434 character customized ovarian cDNA array was constructed and used to analyse gene expression in non-regressed versus regressed CL tissue.

Corpora lutea deemed to be functional and non-regressed (N=6) generated a mean systemic progesterone concentration of 11.72 ± 1.17 ng/ml (mean \pm sem) within a range of 7.86 to 16.69 ng/ml and did not display DNA laddering Fig. 19. In contrast CLs deemed to be regressed (N=6) generated a mean systemic progesterone concentration of 0.84 ± 0.27 ng/ml (mean \pm sem) within a range of 0.24 to 1.76 ng/ml and exhibited the characteristic pattern of DNA laddering indicative of programmed cell death and structural regression. The occurrence of DNA laddering characteristic of apoptotic cell death and structural regression is detectable only in the regressed CLs. Lane 1 and 14 contain 100 bp molecular weight markers.

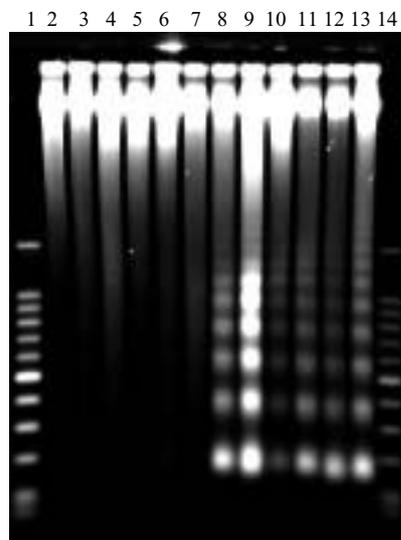


Figure 19. SYBR Green I stained DNA isolated from non-regressed (lane 2-7) and regressed CL tissue (lane 8-13).

Two class analysis at a false discovery rate (FDR) of $<5\%$ revealed 15 differentially expressed genes of which seven increased and eight decreased expression in regressed relative to non-regressed CL tissue, Table 87. Representative autoradiograms obtained following complex cDNA probe hybridisation are shown in Fig. 20. The fold change in gene expression ranged from 1.74 to 2.37 for the up-regulated genes and 1.71 to 5.81 for the down regulated genes. Among the up-regulated genes, four encoded proteins associated with the extracellular matrix, and three had known functions relating to cell structure, oxygen radical metabolism and apoptosis. In contrast the eight down regulated genes encoded proteins mainly involved in steroid biosynthesis and are detailed in Table 87.

Table 87: Genes differentially regulated at luteolysis

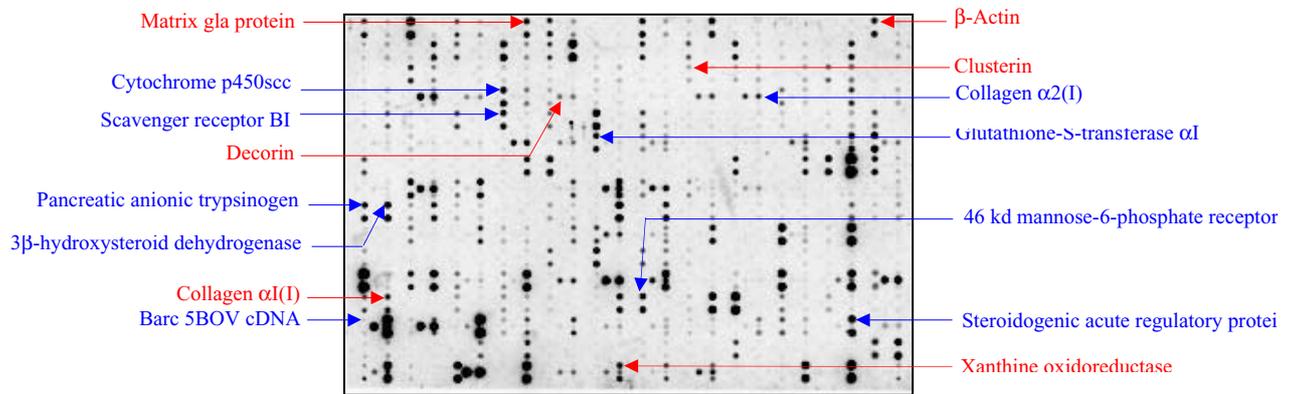
Biological Function	Gene name	Accession No.	Fold change	q-value (FDR) ^a
Extracellular matrix	Decorin	CV547971	2.37	1.22
	Matrix gla protein	BU917233	1.91	1.22
	Collagen α 1(I)	BU917189	1.85	3.96
	Collagen α 2(I)	CV547968	1.74	3.96
	Pancreatic anionic trypsinogen	BU917352	-3.95	1.22
Cell structure	β -Actin	BU917286	1.83	1.22
Oxygen metabolism	Xanthine oxidoreductase	BU917063	1.92	1.22
	Glutathione-S-transferase α I	BU917155	-3.82	1.22
Apoptosis	Clusterin	BU917282	1.76	3.96
Steroid biosynthesis	3 β -hydroxysteroid dehydrogenase	BU917207	-5.81	1.22
	Steroidogenic acute regulatory protein	BU917068	-4.69	1.22
	Scavenger receptor BI	BU917275	-1.96	2.04
	Cytochrome p450scc	BU917164	-1.85	2.84
Metabolism	46 kd mannose-6-phosphate receptor	BU917348	-3.98	1.22
Uncharacterised EST	BARC 5BOV cDNA	BU917392	-1.71	2.04

^aDifferentially expressed genes identified by SAM at a false discovery rate (FDR) of less than 5%.

To confirm the validity of the cDNA array results, five of the 15 differentially expressed genes including glutathione-S-transferase α I (GSTAI), scavenger receptor BI (SR-BI), collagen α 1(I), matrix gla protein (MGP), and clusterin were subjected to Northern blot analysis Figure 21. The Northern results indicated that the mRNA for GSTAI and SR-BI decreased in regressed relative to non-regressed CL tissue ($P < 0.01$) a result consistent with the array data. Genes shown to be up-regulated by array analysis (collagen α 1(I), MGP and clusterin) also increased expression by Northern blot analysis ($P < 0.01$). The mRNA encoding secreted protein acidic and rich in cysteine (SPARC) which did not change significantly by the array analysis also did not change significantly when analysed by Northern blot ($P > 0.05$) Figure 21. In addition the fold change in gene expression calculated for the Northern blots correlated closely with the array results ($R^2 = 0.992$). Collectively these findings demonstrate that the array experimentation in terms of screening and data analysis was accurate and reproducible.

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A. Non-regressed corpus luteum gene profile



B. Regressed corpus luteum gene profile

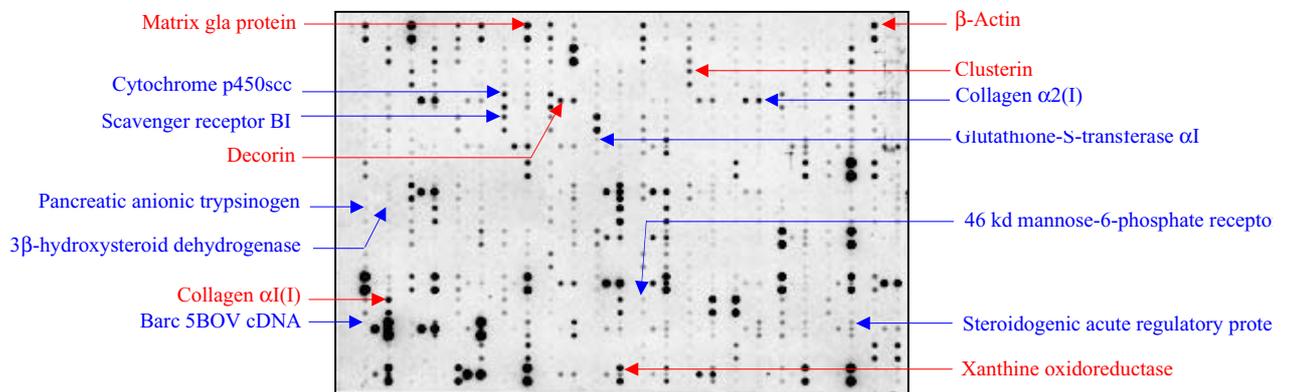


Figure 20. Representative gene expression profile at luteolysis. Hybridisation of ovarian cDNA array with $\alpha^{33}\text{P}$ labelled RNA derived from (A) non-regressed CL tissue and (B) regressed CL tissue. Clones up-regulated and down-regulated in regressed CL tissue compared to non-regressed CL tissue are indicated in red and blue respectively.

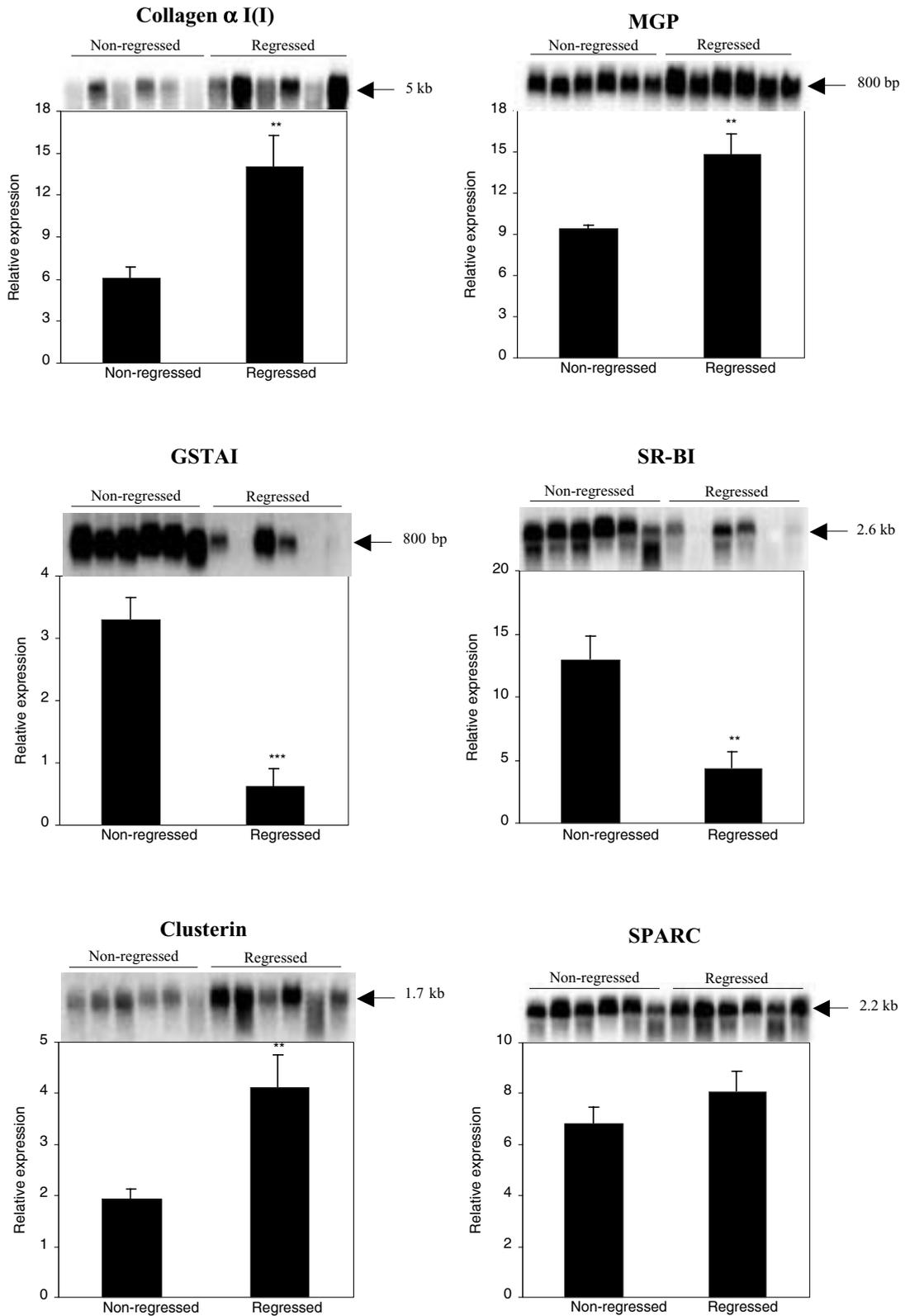


Figure 21. Northern blot analysis of collagen α I(I), MGP, GSTAI, SR-BI, clusterin and SPARC. The data are expressed as mean \pm S.E.M. of six CL tissues per group (** $P < 0.01$, *** $P < 0.001$).

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Using a customized 434 character ovarian specific cDNA array a total of 15 differentially expressed genes were identified in non-regressed compared to spontaneously regressed bovine CL tissue. The differentially expressed genes were grouped into functional families and the most interesting finds are summarized below.

Steroidogenesis

The decreased systemic progesterone concentrations in heifers with regressed CLs was accompanied by corresponding decreases in mRNA for genes involved in steroidogenesis including, 3 β -hydroxysteroid dehydrogenase (3 β -HSD), P450 cholesterol side-chain cleavage enzyme (p450scc) steroidogenic acute regulatory protein (StAR) and (SR-BI). The mRNA for 3 β -HSD and StAR were the most highly repressed and showed an almost 5-fold expression difference in non-regressed compared to regressed luteal tissue. Similarly, p450scc mRNA decreased 1.85 fold in regressed luteal tissue. These results confirm other studies linking the repression of these genes to cessation of progesterone biosynthesis during CL regression. The results of the cDNA array and Northern blot analysis demonstrated a greater than 1.9 fold decrease in expression for SR-BI mRNA. This receptor is involved in the uptake of (HDL) which is the major source of cholesterol for progesterone biosynthesis. The decreased expression of mRNA encoding SR-BI in regressed bovine CL tissue suggests that HDL uptake by SR-BI may play an important role in the control of progesterone biosynthesis. This suggestion is supported by the alternate regulation of SR-BI by the luteotrophic action of prolactin and the luteolytic action of PGF2 α in rat ovarian tissue.

Oxygen radical metabolism

Reactive oxygen species are recognised cellular signals implicated in the induction of apoptosis and are generated within the CL as a by-product of steroid hormone biosynthesis and as a consequence of PGF2 α induction at the end of the oestrous cycle. There is growing evidence that the balance between oxygen free radical generating systems and scavenging systems play a key role in regulating CL lifespan. In the current study mRNA encoding glutathione-S-transferase α I (GSTAI) was approximately 5-fold down regulated in regressed luteal tissue. One of the principal biological functions of GSTAI is to provide protection against oxidative stress and in particular the damaging effects of lipid peroxidation. Down regulation of GSTAI mRNA is therefore likely to increase luteal sensitivity to oxidative damage, hasten the onset of apoptosis and contribute to the demise of the CL.

Apoptosis

The mRNA encoding clusterin was elevated more than 2 fold in regressed relative to non-regressed CL tissue and was accompanied by the disappearance of systemic progesterone and the formation of DNA laddering, an indicator of apoptosis and structural regression. Levels of clusterin mRNA have been shown to increase in response to tissue regression, involution, damage and disease and functionally attributed to alterations in lipid transport, membrane remodeling and apoptosis. These observations suggest that clusterin may alter CL function at several levels. For example, it is possible that up-regulation of clusterin in regressed CL tissue reduces progesterone biosynthesis through alteration of cholesterol transport and distribution, contributing to a shortage of substrate for steroidogenesis. Although clusterin appears to be inherently a pro-survival protein, over-expression in neuron cells is known to induce mitochondria-dependent apoptosis. Increased clusterin expression has also been demonstrated in regressed rat and swine CL tissue. These observations, combined with the results found in the current study, implicate clusterin expression with yet another incidence of programme cell death and point towards a role for this glycoprotein protein in luteolysis.

Extracellular matrix

Structural regression of the CL is associated with extensive degradation and remodeling of the extracellular matrix (ECM) which can influence cell division, differentiation, migration and apoptosis. In the current study mRNA for collagen α 1(I) and collagen α 2(I), components of the triple helical structure of collagen type I, were greater than 1.7 fold up-regulated in regressed luteal tissue. The mRNA encoding pancreatic anionic trypsinogen also known as trypsinogen 2 was approximately four-

fold down regulated in regressed relative to non-regressed CL tissue. Trypsinogen 2 is known to degrade type I collagen and can also activate latent collagenases including matrix metalloproteinases MMPs -1, -8 and -13. Taken together these results indicate that collagen biosynthesis is active during the latter stages of CL regression and support the biochemical data showing that collagen type (I) is retained during luteolysis.

Decorin, an extracellular proteoglycan, was identified as the most highly induced mRNA (2.37 fold) represented on the cDNA array. Decorin has been implicated as a key regulator of matrix assembly through its ability to promote collagen fibril assembly. It has also been implicated in the negative control of cell proliferation by virtue of its ability to bind and sequester the bioactivity of transforming growth factor β . Up-regulation of decorin is therefore likely to promote CL regression by enhancing collagen stability and suppressing cellular growth.

The mRNA for MGP, a vitamin K dependent calcium binding protein, not previously identified in the CL was elevated 1.91 fold in regressed CL tissue. One of the principal roles of MGP is to promote the calcification, differentiation and migration of vascular cells. While these functions are cell type dependent and are also influenced by the presence or absence of particular growth factors, it is tempting to speculate that the role of MGP in CL regression is also mediated through alteration of the luteal vascular system.

This cDNA array study has resulted in the identification of genes with recognized roles in CL regression, genes with potential roles in this process and genes whose function have yet to be defined in this event. Overall our findings indicate that CL regression is a complex multi-mechanistic process that involves alterations in genes associated with extracellular matrix remodeling, oxygen radical metabolism, apoptosis and steroid biosynthesis. An improved understanding of the genes and mechanisms regulating CL function is central to the long term goal of developing new treatment strategies to overcome luteal inefficiency.

RMIS No.4953

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Repeatability of embryo survival beef heifers

Embryo survival rate is a major determinant of reproductive efficiency in dairy and beef herds. Data has shows that early embryo death, before about day 16 of gestation, is the major cause of low conception rates. There is some evidence of repeatable differences between animals in their ability to establish and sustain pregnancy as well as some genetic variability for heifer pregnancy rate and dairy cow sustainability. However, the endocrine, molecular and genetic bases for these apparent differences in embryo survival rate have not been established. The initial objective of this study was to establish repeatability estimates for embryo survival rate in heifers. A total of 69 reproductively normal heifers were used with each heifer artificially inseminated (AI) on 4 occasions over a 9-month period using the following regimen. Initially estrus was synchronised using a 2-injection prostaglandin (PG) regimen and heifers were inseminated at 6-12 hours after observed standing estrus (day 0). Heifers were ultrasonically scanned for pregnancy on day 28 and again on day 35. All pregnant heifers received a PG at day 35 to induce embryo loss. Six weeks after the induced embryo loss all heifers were re-programmed using a 2-injection PG-regimen, inseminated and scanned as described above. A similar schedule was followed for a further two rounds of AI. Semen from one high-fertility bull was used.

Conception rate was similar ($P>0.05$) for each round of insemination with an overall rate of 70%. After 3 rounds of insemination the fertility status of each heifers was categorised depending on the cumulative outcome to the first three rounds of insemination. Heifers that failed to become pregnant to

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any of the first 3 services were designated as 0, heifers that became pregnant once as 33, heifers that became pregnant twice as 67 and heifers that became pregnant to all 3 services as 100. The effect of

fertility status determined after three inseminations (independent variable) on pregnancy rate to the 4th insemination (dependent variable) was evaluated using PROC GENMOD (SAS). Pregnancy rate to the 4th insemination was affected ($P < 0.0056$) by fertility status and results are summarised in Figure 22.

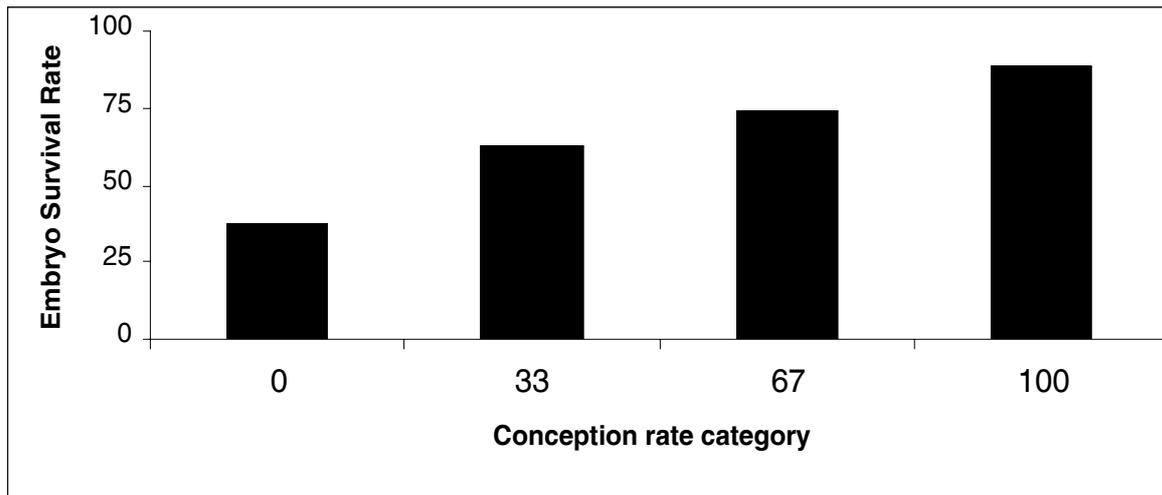


Figure 22. The outcome of the 4th insemination for each category of heifer.

Repeatability estimates were derived from an analysis of variance as the interclass correlation among records on the same individual in different rounds. Based on data to-date the repeatability of embryo survival is 0.18. This analysis indicates embryo survival has low to moderate repeatability.

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RMIS No.5235

**SHEEP PRODUCTION
GRASSLAND**

Grazing and territorial habits of hill sheep on the Teagasc hill sheep farm, Leenaun, using satellite tracking and field observations

The relevant objective of the sub-programme ‘Grassland and Grazing Management in Teagasc 2000 – Sheep Production’ was to: “Develop hill sheep production systems that ensure the continued viability of producers while reducing environmental impacts”. The latter part of this objective is addressed in the current project by monitoring the natural resources, particularly the unimproved hill and mountain vegetation, under a quantified, free-range grazing management system. The method of monitoring the hill vegetation is described in an earlier Research Report (1995). GPS technology and field observations are used to quantify the grazing and territorial habits of hill sheep on a seasonal basis. A survey of the frequency of vegetation species was repeated in 2004. The frequency of the group ‘no vegetation’ was almost halved compared with 1995 (Table 88).

Table 88: Species group frequency (%)

Group	1995	2004	Change
No Vegetation*	35.6	19.7	-15.9
Grasses	24.9	33.1	8.2
Sedges	22.2	24.0	1.8
Heathers	3.3	5.0	1.7
Rushes	0.7	0.7	0.0
Bryophytes	7.1	9.0	1.9
Other forbes	6.2	8.5	2.3

* Algae-covered soil and rock outcrop/boulder

All groups of vegetation, except rushes, contributed to the overall increase in vegetation. While the increases in vegetation frequency were significant the magnitude of the increases was not significantly affected by the variations in physiography. However, preferred grazing area (*Research Report 2002*) in the combined transportational mid-slope and colluvial foot-slope was significantly correlated with the increases in the frequency of the general vegetation and of *Molinia caerulea* (Table 89).

Table 89: Effect of Preferred Grazing Area (PGA) on changes in vegetation frequency (%) in two physiographic units in the period 1995-2004

PGA	All species	<i>Molinia caerulea</i>
<i>Inside</i> (N=35)		
mean (s.e.)	17.7 (1.2)	9.8 (1.29)
<i>Outside</i> (N=117)		
mean (s.e.)	10.8 (2.32)	4.5 (2.37)
Significance	<0.01	<0.05

Three season-based, sampling periods of flock and animal activities, using GPS collars in conjunction with field observations, have been completed. Preliminary analysis indicated a preference for territories with the steepest slopes and the lowest content of organic soils. Grazing preferences were concentrated in the *Molinia*-dominated and mixed *Molinia-Schoenus-Rhynchospora* communities. Greater numbers of sheep sought shelter in the dry and windy rather than in the wet and windy conditions.

Focal animal observation. One of the GPS-collared ewes was observed at intervals totalling almost 16.5 hours in July/August 2004. Grazing and moving activities accounted for 37 and 7% of the time, respectively. Being relatively motionless (standing/lying) accounted for

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approximately half that at just over 20 per cent of the time. The collared ewe was a member of a stable group of seven and associated with a clearly defined 'home range'. Grazing was not confined to the dominant vegetation available.

Sheep 'rests'. Rest areas ('camp' sites) are a feature of the unimproved hill vegetation under free-range grazing. They occupy 1.4% of the total area and may provide conditions that are favourable for the establishment of the unpalatable grass, *Nardus stricta* (*Research Report 2003*). The prominent position that is occupied by this plant is indicated in Table 90.

Table 90: Distribution of some plant species by dominance in sheep rest sites

Plant species	Per cent frequency		
	1st	2nd	3rd
<i>Molinia caerulea</i>	68.8	21.5	6.5
<i>Nardus stricta</i>	18.3	22.6	8.6
<i>Juncus spp</i>	6.4	11.8	11.8
<i>Schoenus nigricans</i>	1.1	26.9	16.1

A study of the vegetation and soils of a stratified random sample of the 'rests' based on physiography and size was completed in summer 2004. Two broad vegetation types (British National Vegetation Classification (NVC)) were observed after ordination of the field data: acid grassland (*Nardus stricta-Galium saxatile* grassland) and wet heath (*Scirpus caespitosus-Erica tetralix* wet heath) with some transitional types between the two main communities. Soil analysis confirmed the acidic nature of the peat substratum. *Nardus stricta* was a prominent component of the vegetation on the majority of rests but was rarely dominant. Colonisation by *Nardus* was common where bare peat aprons were present on or adjacent to sheep rests. Such areas were also strongly correlated with low herb cover and the presence of 'bog' or wet heath species. Examination of wool samples from 20 randomly selected ewes from the hill flock indicated no evidence for zoochory being responsible for the spread of *Nardus stricta* on the farm.

Overall vegetation cover of the unimproved hill, based on plant frequency study, increased from 64.4% in 1995 to 80.3% in 2004. The stocking density of the unimproved hill during this period ranged from 0.9 to 0.8 ewes per hectare. The quantification of the grazing and territorial preferences of hill sheep is essential to assessing their impact on the general vegetation cover and on the establishment and spread of unpalatable species.

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RMIS No. 5080

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Performance of ewes and lambs at Leenane hill farm

The flock at Leenane hill farm is self-contained (i.e. replacements are homebred) and consists of Scottish Blackface ewes most of which represent the local strain of the breed. The ewes are managed as two separate sub-flocks one of which gets most of its annual maintenance from the hill grazing while the other is largely confined to the Greenland. The sub-flocks are (1) Hill flock which is a purebreeding Scottish Blackface flock which is confined to the hill throughout the year except during the mating season and from just before lambing up to about 5 weeks post lambing when all ewes with single female lambs are put back to the hill; the remaining ewes remain on the greenland until weaning of their lambs (apart from short spells on the hill dictated by grass supply) and (2) Lowland flock which is a crossbreeding flock which consists of the older ewes (> 4 years old) which are crossbred with rams from lowland breeds to produce female replacements for lowland flocks. Wether lambs and surplus female lambs from the purebreeding system are sold for slaughter to the light lamb market

while surplus lambs (mostly wethers) from the crossbreeding system are finished for the French market.

The mating plan for the 2003 breeding season involve using five S Blackface rams on the Hill flock (single sire mating) while the ewes in the crossbreeding flock were joined with either Belclare (N=3) or Chamois (N=2) rams in single sire mating groups. The reproductive performance of the ewes is summarised in Table 91 and the values show that the performance was very satisfactory for both flocks and that the Lowland flock achieved a very good performance for number of lambs reared per ewe joined.

Table 91: Reproductive performance of the sheep flocks at Leenane

Item	Flock	
	Hill	Lowland
No. ewes joined	205	143
Liveweight (kg) at		
Mating	43.5 (0.27)	51.0 (0.59)
Lambing	39.7 (0.34)	44.8 (0.71)
Fertility	0.91	0.95
Litter size	1.15 (0.031)	1.71 (0.065)
Lambs reared/ewe joined	0.99 (0.040)	1.57 (0.088)

Aspects of lamb growth are summarised in Table 92. There is no basis for any direct comparison between the lambs in the Hill and Lowland flocks as they are managed in distinct systems. In the Lowland flock the performance of Chamois-sired lambs versus Belclare-sired lambs is of interest as the former are reputed to yield very good carcass conformation. Lambs sired by the Chamois were significantly lighter at birth (-0.9, s.e. 0.09, kg) and at weaning (-2.0, s.e. 0.50, kg). The differences among individual sires were quite small. In the case of the Hill lambs the growth rate up to 5 weeks was clearly less than for the crossbred lambs even though all were managed on greenland at that stage. The performance of the purebred lambs to weaning was relatively poor and there was no marked difference between the sexes even though the wether lambs remained on the greenland up to weaning at about 14 weeks of age.

The incidence of lamb mortality (i.e. dead born + died up to weaning) was extremely low in both systems at Leenane and this is consistent with results in previous years. The incidence of mortality in the two systems for single and multiple births is given in Table 93. Analysis of the mortality data showed that there was no difference between Belclare and Chamois lambs despite the significant difference in birth weight. Birth type had a significant influence on mortality, as expected but there was a significant interaction ($P < 0.04$) between the pattern of effect in the Hill and Lowland flocks. Purebred twin lambs (Hill flock) had a lower survival probability than purebred singles but the ranking of birth types was the opposite from the crossbred lambs (Lowland flock).

Table 92: Least squares means for effects of flock*breed and birth/rearing type on lamb growth traits at Leenane

Flock	Breed	Birth/rearing type	Growth trait					
			0 – 5* (g/day)	s.e.	0-14* (g/day)	s.e.	14 wk weaning weight (kg)	s.e.
Lowland	9209	Single	316	12.7	266	8.0	29.7	0.84
		Twin	248	8.4	236	5.3	26.7	0.55
	9220	Single	344	10.6	284	6.7	32.4	0.70
		Twin	256	7.9	248	4.9	28.0	0.52
Hill	9292	Single	266	4.5	190	2.8	22.8	0.30
		Twin	207	8.1	186	5.1	21.8	0.53

*growth rates from birth to five weeks

*growth rates from birth to fourteen weeks

Table 93: Incidence of total lamb mortality in Hill and Lowland flocks at Leenane

Flock	Birth type	Total lambs born	Mortality (%)
Hill	Single	160	4.4
	Twin	54	11.1
Lowland	Single	52	9.6
	Twin	164	3.3

The information on slaughter traits and carcass conformation (based on factory classification) is summarised in Table 94. The lambs sired by Chamois rams yielded significantly lighter carcasses and were 5 days older than Belclare-cross lambs at slaughter. The Chamois-crosses had significantly better conformation score which reflected the fact the 73% had a conformation score of R or better compared with 49% for Belclare crosses.

Table 94: Performance traits for lambs sold for slaughter

Trait	Sire breed			Difference B - C
	Scottish Blackface	Belclare(B)	Chamois (C)	
Age at slaughter (days)	171	196	205	NS
Weight at slaughter (kg)	28.3	39.5	39.1	NS.
Carcass weight (kg)	11.3	17.8	17.1	P<0.01
Conformation score [§]	1.4	2.7	2.9	P< 0.01

[§]On the EUROP scale 1 = P to 5 = E

Hanrahan, J.P. and O'Malley, L.

RMIS No. 5080

Observations on diurnal variation in incidence of lambing at Leenane

Supervision of the ewe flock during the lambing period represents one of the peak labour inputs in sheep production. If there was significant diurnal variation in the incidence of parturition this could allow the supervision input to be focussed on the period when incidence was high. In particular there is some evidence that under 'natural' conditions of outdoor lambing natural cues associated with daylight may act to minimise lambing during the night. This hypothesis was examined in the Scottish Blackface ewes at Leenane during the 2003 and 2004 seasons. The ewes at Leenane lamb outdoors and supervision are confined to daylight hours with the final check on ewes at about 9 PM and the next check at about 6.30 AM. This

allowed the ewes that lambed during the night time period to be identified. The observations were assessed separately for the two subflocks (i.e. Hill and Lowland) at Leenane. The median lambing date for the Low flock is in early April (5 April in 2004) while the Hill flock is joined to lamb about 2 weeks later (median 21 April in 2004). The incidence of night-time lambing is given in Table 95 for each flock by year combination.

Table 95: Observed and expected incidence of ewes lambing at night

Year	Flock	No. of ewes	No. lambing at night	
			Observed	Expected
2003	Hill	184	75 (40.8) ^a	73
	Lowland	124	52 (41.9)	49
2004	Hill	186	86 (46.2)	74
	Lowland	101	36 (35.6)	40

^aPercentage

The analysis of these data showed that there was no significant variation due to flock or year and the flock-by-year interaction was not significant. The overall percentage of ewes that lambed at night was 41.8 and the expected percentage, based on the assumption that there is no significant diurnal variation in time of parturition, is 39.6. The difference between the observed and expected number was associated with a chi-square value of 4.8 (exact probability = 0.031). Thus, the evidence suggests that a slightly greater proportion of ewes lambed during the night-time than was expected under the null hypothesis. Consequently, with the natural daylength conditions that the ewes at Leenane experience, there is no reduction in the incidence of night-time lambing and this is consistent with previous observations on a large commercial flock under indoor lambing conditions and all-night supervision (see Sheep Research Report 1999, p27).

Hanrahan, J.P. and O'Malley, L.

RMIS No. 5080

ANIMAL NUTRITION AND SUPPLEMENTATION

Effect of grass height and concentrate supplement on pre-weaning growth of lambs

This trial, which involved two contrasting sward heights combined with a daily concentrate allowance of either 0, 300 or 660 g/head of a commercial creep ration, was run on a farmlet basis as in previous years. The sward heights were established by using different stocking rates (20 or 24 ewes/ha) and the concentrate allowance treatment represented the maximum daily allowance. Ewes and their lambs (mostly twins) were turned out to the experimental plots shortly after indoor lambing and concentrate was offered to the lambs from around 3 weeks of age. The ewes received no concentrates after turnout. There were 10 farmlets representing two replicates of the two grass-only treatments and two replicates of the two grass heights combined with 300 g concentrate allowance. The two treatments involving 600 g of concentrates were not replicated. A total of 398 lambs were assigned to treatment and growth data to weaning ewes was available for 395 lambs.

The following growth traits were evaluated: growth rates from birth to 5 weeks (G05), birth to 14 weeks (G014), 5 weeks to 14 weeks (G514) and 10 weeks to 14 weeks (G1014). Weaning weight adjusted to 14 weeks of age (W14A) was calculated. The data were analysed by fitting a linear model with effects for dam age (2, 3, or 4 years), sex, rearing type (single or twin), grass height (low, high), concentrate allowance (0, 300, 600 g) and the interaction between grass height and concentrate allowance. The differences among the treatment combinations were partitioned to evaluate linear and quadratic responses to concentrates and how this interacted with sward height.

Table 96: Least squares means for growth rate and weaning weight for the different combinations of grass height and concentrate allowance

Grass height	Concentrate allowance (g/day)	Variable			
		G514	G1014	G014	W14A
Low	0	271 ^b	218 ^c	292 ^b	337 ^b
	300	332 ^a	316 ^a	340 ^a	38.6 ^a
	600	338 ^a	299 ^{ab}	333 ^a	37.7 ^a
High	0	321 ^a	279 ^b	325	36.6 ^a
	300	324 ^a	308 ^a	330 ^a	37.3 ^a
	600	334 ^a	336 ^a	346 ^a	38.9 ^a
Approx s.e.		9.5	12.4	7.5	0.82

^{abc}Means without a common superscript are significantly different (P<0.05)

The least squares means for the six treatment combinations are given in Table 96 for selected growth traits and weaning weight. Inspection of the result shows that there was very little response to concentration supplementation at the high grass allowance but significant response was evident on low grass. This was reflected in a highly significant interaction (P<0.01) between concentrate allowance and sward height. The growth rate between 5 weeks of age and weaning provides the most direct test of the effects of sward height and concentrate allowance on lamb growth. Thus, over this period there was a significant linear response to concentrate supplementation with the low grass treatment (P<0.001) and the response also had a significant negative quadratic component (P<0.01). However, there were no significant effects on the high grass treatment (P>0.2). Grass height had no significant effect on growth rate to 5 weeks of age. These results are consistent with corresponding trials

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in previous years in showing that there is little worthwhile response to concentration supplementation unless grass availability is low. Thus, if grassland management is such that guidelines for grass supply (sward height) are followed there is no worthwhile response from the addition of concentrate supplementation.

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RMIS No. 4929

SHEEP BREEDING AND REPRODUCTION

Comparison of pre-weaning growth of Suffolk and Texel lambs

The purebred flocks of Suffolk and Texel sheep maintained at Athenry are managed together throughout the year except during the mating. These flocks have been under selection for increased growth rate to weaning since their establishment in the mid 1970s (see Sheep Research Report 1999, 20-22). Selection has been limited in 2003 and 2004 due to a process of selection of rams for increased resistance to scrapie. In the case of the Suffolk flock ARQ/ARQ animals were not used while in the Texel this restriction was applied plus the carriers of the VRQ allele were not selected. All of the animals in the flock, including the 2004-born individuals, were genotyped during the year and all carriers of the VRQ allele were culled (Texel flock). The allele frequencies at the PrP locus are summarised in Table 97 for the two flocks. The frequencies are very close to the baseline frequencies obtained from a large number of sheep of these breeds from the national registered populations.

Table 97: Relative frequency of the alleles at the PrP locus in Suffolk and Texel flocks

PrP allele	Breed	
	Suffolk	Texel
ARR	0.47	0.33
ARH	0	0.33
ARQ	0.53	0.13
AHQ	0	0.13
VRQ	0	0.08

The average performance of these two flocks over the period 2002 to 2004 is summarised in Table 98. The only significant difference between the breeds was for birth weight which was greater in Texels. The difference in birth weight is consistent with the values reported previously. However the absence of any difference between these two breeds for growth rate to weaning is not consistent with earlier results and tends to confirm the suggestion that a greater response to selection has been obtained in the Texel population and this would be consistent with the higher estimates of heritability for pre-weaning growth in the Texel breed.

The proportion of total phenotypic variance due to maternal influence declined from 0.37 for birth weight and 0.45 for growth rate from birth to 5 weeks to 0.21 for growth between 5 weeks and 10 weeks of age and to 0.03 for growth between 10 weeks of age and weaning. Thus maternal influences on growth due to either prenatal factors or milk yield are relatively minor for the growth from around 8 weeks of age onwards which is consistent with the rapid decline in milk after about week 5 of lactation.

There was no evidence for any interaction between rearing type and breed for growth rate at any stage which conflicts with previous data from this study which showed that twin-reared lambs on Texel ewes had a significantly better growth rate between birth and 5 weeks than did twin-reared Suffolks whereas the opposite was the case for singletons (see Research Report 1999, 20-22). The reason for this change in maternal growth effects is unclear.

The fact that there was no evidence for any differential decline in the growth rate of lambs in the immediate pre-weaning period when it is known that parasite burdens, as revealed by faecal egg counts, diverge substantially between the breeds is surprising. It suggests that the Suffolk may have a high tolerance for gastro-intestinal parasites. It would be informative to compare these two breeds under 'clean grazing' conditions as well as under the current conditions where they are exposed to a natural mixed infection. The presence of a genotype-environment interaction would provide a valuable insight into the mechanisms involved in responding to parasite infection.

Table 98: Comparison of purebred Suffolk and Texel lambs for pre-weaning growth

Trait	Breed	
	Suffolk	Texel
Birth weight (kg)	4.6 (0.12) [§]	5.0 (0.11)
G_05 (g/day)	323 (11.2)	326 (9.8)
G_510 (g/day)	357 (7.4)	343 (8.0)
G_1014 (g/day)	223 (7.7)	219 (7.4)
G_014 (g/day)	301 (5.6)	302 (5.1)
Weight at weaning (kg)	34.6 (0.59)	35.2 (0.54)

[§] () = s.e.

Hanrahan, J.P.

RMIS No. 4462

Estimates of heritability for growth and carcass traits

Progeny performance data for the 57 sires (40 progeny per sire) involved in the study on effects of the PrP gene on lamb performance was used to estimate the heritability of growth and carcass traits. The resulting estimates, from the REML estimates of the sire and residual variances are presented in Table 99. All of the estimates were statistically significant different from zero except for the fat classification score. The confidence intervals on the estimates are reasonably narrow and indicate that for growth traits the heritability is generally at least 0.1 and probably not much greater than 0.2. The estimates for these traits are consistent with estimates from a previous study. The relative magnitudes of the heritability estimates for ultrasonic muscle and fat depths are reversed compared to estimates from a previous study (0.20 and 0.28, respectively), but those estimates are within the confidence interval estimates in the present study. Thus, it is suggested that the heritability of UMD can be taken as about 0.25 while that for UFD is about 0.2.

Table 99: Variance due to sires and residuals and estimated heritability

Trait	Sire variance	Residual variance	h ²	90% CI for h ²	
				Lower	Upper
Birth-weight	0.0313***	0.621	0.19±0.038	0.13	0.29
G_05	71.37***	3224.99	0.09±0.017	0.05	0.15
G_514	139.76***	2739.57	0.19±0.039	0.13	0.29
G_014	77.49***	1737.60	0.17±0.034	0.11	0.26
W5A	0.18***	5.48	0.13±0.025	0.08	0.20
W14A	0.89***	19.16	0.18±0.036	0.12	0.27
WT_120	1.70***	26.19	0.24±0.049	0.17	0.36
UMD	0.46***	5.43	0.31±0.062	0.22	0.45
UFD	0.0037***	0.09991	0.14±0.029	0.09	0.23
CARC_WT	0.06842**	2.5683	0.10±0.027	0.05	0.21
NCON	0.005822***	0.2221	0.10±0.026	0.05	0.21
NFAT	0.000871 ns	0.09395	0.04±0.009	0.00	0.11
SL_AGE	88.8758***	906.12	0.36±0.092	0.23	0.59

* P<0.05, ** P<0.01 and *** P<0.001

The finding of no evidence for genetic variation in carcass fat score is at odds with an estimate of 0.26 (s.e. 0.073) from a previous study while the heritability of conformation score in that study was 0.21 (s.e. 0.067) which is about twice the estimate in Table 99. The differences may be partly due to the fact that in the present study the carcass classification was done by abattoir personnel.

Hanrahan, J.P. and Casey, K.

RMIS No. 4462

Genetic basis for ovarian hypoplasia in Cambridge and F700 Belclare ewes

A high incidence of sterility has been noted in Cambridge and F700 ewe and Belclare sheep and studies of inheritance patterns here indicated that an autosomal recessive gene was responsible, see Research Reports 1999 (p26) and 2000 (p23-24). Analysis of DNA sequence for two candidate genes has shown that point mutations in each gene were directly associated with the sterile phenotype. Both candidate genes code for growth factors and are expressed by the oocyte within the growing follicle. These growth factors play key roles in the co-ordination of the development of the Graffian follicle. One of the genes (GDF9) is on sheep chromosome 5 while the other gene (BMP15) is on the X chromosome. The evidence for the role of these genes in the ovarian hypoplasia is as follows: DNA from a total of 36 F700 ewes with the sterile phenotype was available and 35 of these ewes were homozygous for point mutations in either GDF9 or BMP15 and among a large number of normal ewes that were genotyped none were found to be homozygous; analysis of DNA from 30 sterile Cambridge ewes showed that 26 of these were homozygous from wither the BMP15 mutation or the

GDF9 mutation and no fertile ewe was found that was homozygous from any of the mutations.

The DNA sequence for BMP15 revealed two separate mutations, both within exon 2, that were non-conservative and associated with the sterility phenotype. One of these mutations (named $FecX^G$) was found in both F700 Cambridge populations while the other mutation ($FecX^B$) was only found in the F700 flock. Individuals with the $FecX^G/FecX^B$ genotype also had the sterile phenotype as expected from the fact that individual homozygous for either mutant were sterile. The GDF9 mutation was the same in both populations and is likely to have originated from the same source.

The four unexplained Cambridge sterile cases had the typical ovarian hypoplasia and infantile uterus associated with the animals that were homozygous for the mutant genes and two of these individuals were wild type for both BMP15 and GDF9. Examination of the pedigree of these individuals showed that one sire was a common ancestor of both the paternal and maternal lineages. It is hypothesised that an unidentified third gene is responsible for the sterile phenotype exhibited by these animals. The unexplained F700 sterile case had an atypical phenotype in that the uterus was recorded as being of normal size and follicles were noted on the ovaries at a number of laparoscopic examinations. It is possible that this animal was misclassified.

Specific matings were arranged to generate further information on the inheritance of the unexplained sterile condition in Cambridge sheep.

Hanrahan, J.P.

RMIS No. 4785

Incidence of mutations in Mark 2 Belclare flock

A total of 191 ewes were tested for presence of the BMP15 and GDF9 mutations that have been shown to be responsible for the large effects on ovulation rate seen in F700 belclare and Cambridge flocks.

No cases of the GDF9 mutation, $FecG^H$ were detected and thus it can be concluded that this mutation has been excluded from the Mark 2 Belclare. A total of 12 ewes were found with the $FecX^G$ mutation and a set of 13 ewes were identified as carriers of the $FecX^B$ mutation. All carriers that were still in the flock at the time the test results were obtained were transferred to the F700 flock. Since this initial screening descendants of the identified carriers were tested and any carriers were also transferred to F700 flock. The objective of the testing programme is to eliminate the mutations from the Mark-2 Belclare flock at Athenry.

Hanrahan, J.P. and Mullen, M.

RMIS No. 4785

Presence of major genes for ovulation rate in the pedigree Belclare population

The establishment of the Belclare breed on farms in the beginning of the 1980s involved dispersal of ewes and rams from the research flock at that time. This dispersal phase preceded the recognition of the possibility that genes with large effects on ovulation rate were likely to be present in the foundation population. In later years, after the establishment of a sub-line of Belclare ewes with exceptionally high ovulation rate for research into the basis for the extreme ovulation, only animals from the Mark2 line of the Belclare were released for use on pedigree flocks.

Since the identification of the BMP15 and GDF9 mutations that are responsible for large effects on ovulation rate in the F700-Belclare line and in Cambridge sheep (see Sheep

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Research Report 2002/3) screening of the Mark2-Belclare flock at Athenry for the mutations involved has revealed that both *FecX^G* and *FecX^B* mutations were present at low frequency (see above). Thus it is likely that these mutations are present in the pedigree population. This hypothesis was supported by the belief of one breeder that an above average litter size performance was a feature of a significant number of ewes in the breeder's flock and there was conscious selection to use such animals for breeding. Since the flock in question (Flock A) was the largest flock of Belclare sheep in the breed society all of the ewes in the flock were blood sampled onto FTA paper and the DNA extracted to determine genotypes at the BMP15 and GDF9 loci.

Flock A: A total of 95 ewes were tested along with 5 rams (four of whom were bred in the flock). Four ewes were identified as carriers of the *FecX^G* mutation and five were carriers of the *FecX^B* mutation. One ewe carried both mutations. No carriers of the known GDF9 mutation (*FecG^H*) were detected. One of the rams also carried *FecX^B* and this ram was bred in the flock. Examination of the pedigree of the carriers showed that four of the *FecX^B* carriers were sired by one ram; the dam of this ram was also tested and was a carrier of *FecX^B* (as expected). The pedigree of the *FecX^G* mutation was more diverse but it was possible to conclude that the mutation was inherited from some of the foundation ewes that were released to breeders at the time the pedigree society was established and two ewes in the pedigrees were identified as likely carriers of the mutations inherited by these four cases. It was also possible to show that the *FecX^B* mutation in this flock originated from among the foundation ewes released to breeders. Thus both mutations present in this flock originated from among the foundation sheep released for the establishment of the breed on farms.

The litter size records for the set of ewes tested was examined and while two of the six carriers with records had produced quadruplets there was nothing exceptional about the remaining cases and quadruplets were also recorded for five ewes that were not carriers. Thus identification of carriers based on a limited number of litter size records is not likely to be very reliable.

Flock B: Another Belclare flock came to attention because of a high incidence of apparent barren hoggets in the autumn of 2003. All of these ewes was bred in the flock and were by the same sire which was also bred in the flock. Subsequent physical examination of the animals in question revealed that some of them had the typical phenotypic appearance of ewes that are sterile due to homozygosity for any of the BMP15 mutants or for the GDF9 mutant. Genotypes of these animals (N=8) were also determined and this revealed that all of them carried the *FecX^G* mutation but that 6 also carried the *FecX^B* mutation and were thus sterile. All of the 8 cases were sired by the same ram and thus this ram was classified as carrying *FecX^G*. Scrutiny of the pedigree of this ram showed that the mutation came from a ewe sold out of the Mark2-Belclare flock at Athenry in 1997. The ewe had produced two sets of triplets at Athenry and subsequently had a litter size record of 4,3,2,3 in Flock B. Among the six carriers of the *FecX^B* mutation five had the same maternal grandsire and thus it was concluded that this individual was a carrier. The sire in question was bred in Flock A. The same ram was also the maternal great grandsire of the remaining ewe.

Based on the above findings a selective genotyping programme of ewes in the pedigree Belclare population is planned. Ewes with exceptionally high litter size records based on at least three lambings will be tested and all stock rams that are to be used in the next breeding season will be tested.

Hanrahan, J.P.

RMIS No. 4785

Performance characteristics of Belclare sheep in pedigree flocks

The actual lamb weight at around birth and subsequent growth rate for 2004, according to birth type, are in Table 100.

Table 100: Performance for purebred Belclare flocks

Birth type	Lamb mortality	Birth weight (kg)	Growth rate (g/day)	Age interval (days)
Single	4.1	4.4	346	62
Twin	11.1	3.8	308	63
Triplet	25.0	3.4	280	63
Other		2.9	270	63

The average litter size in 2004 was 2.05 and the distribution of litter size was as follows: ewes with singles (23%), twins 53%, triplets 20% and with quads or more 4%.

The average breeding values for lambs born each year since 1995 are given in Figure 23. The pattern shows that there has been no progress since 2001. This means that breeders are not using the EBV95 values when selecting flock replacements or stock rams. Inspection of the flock average EBVs shows that some flocks are using the EBV information to select breeding stock and have average EBVs between 10 and 25 while another group of flocks have average EBVs that are only around 5.

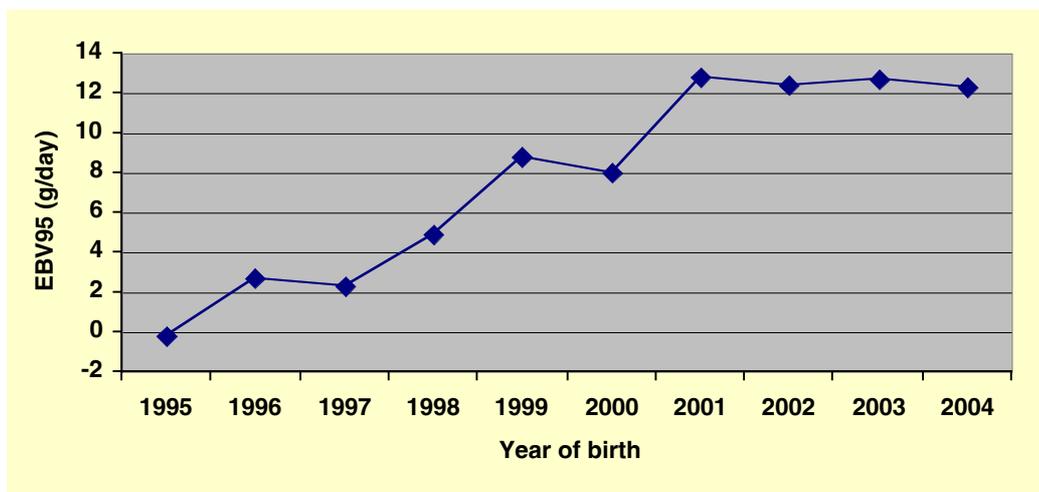


Figure 23. Estimated breeding values of pedigree Belclare flocks.

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RMIS No. 4453

Embryo survival in ewes heterozygous for mutations in BMP15 and GDF9 that have a large effect on ovulation rate

Growth factors produced by the oocyte, such as bone morphogenetic protein 15 (BMP15) and growth differentiation factor 9 (GDF9) are part of a complex system in the ovary out of which graffian follicles emerge that eventually ovulate oocytes capable of fertilisation and yielding a pregnancy. Both BMP15 and GDF9 are known to be involved in the orchestration of the process of follicle development and growth leading to the production of competent oocytes. Mutations in the BMP15 and GDF9 genes can have profound effects on this process. Thus, it

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has recently been shown (see above) that sterility due to ovarian hypoplasia in Cambridge and Belclare ewes is due to specific point mutations in either BMP15 or GDF9. It was also shown that heterozygous carriers of these mutations exhibit a much higher ovulation rate (+0.9 to +1.7 ova) than contemporary animals with the wild-type genotypes. A question that arises is whether these mutations compromise the competency of the resulting oocytes.

In the course of investigations on the genetic basis for exceptionally high ovulation rates in Cambridge ewes and in the F700-Belclare line, routine ovarian examinations to determine ovulation rate were carried out over a number of years. These examinations included a determination of ovulation rate at the cycle of conception. Litter size was recorded at birth. Following the discovery of mutations in the BMP15 gene (*FecX^G* and *FecX^B*) and the GDF9 gene (*FecG^H*), and their association with the sterility phenotype and increased ovulation rate, the genotype of all ewes, for whom DNA was available, was determined for these loci. The *FecX^G* mutation is common to both Cambridge and F700-Belclare populations while the *FecX^B* mutation is not found in Cambridge.

The data for all available records of litter size and the associated ovulation rate at conception for those ewes for whom the BMP15 and GDF9 genotypes were known was extracted from the data files on these genetic lines. A total of 73 Cambridge ewes and 84 F700 ewes (total records 136 and 180, respectively) contributed data. The data on ovulation rate and corresponding litter size were analysed using a linear mixed model with fixed effects for ewe age (2,3 or ≥ 4 years at lambing), number of BMP15 mutations (0 or 1) and number of GDF9 mutations (0 or 1) plus ewe identity as a random effect; ovulation was included as a covariate with linear and quadratic terms. The mixed model procedures of SAS were used for data analysis.

Table 101: Number of ewes by breed and genotype and the corresponding ovulation rate

Genotype status [†]		Breed	No. of records	Ovulation rate
BMP15	GDF9			
0	0	Cambridge	34	3.08
		F700	44	2.60
0	1	Cambridge	20	6.00
		F700	10	4.73
1	0	Cambridge	30	4.62
		F700	115	3.70
1	1	Cambridge	52	8.03
		F700	11	6.31

[†] 0 = wild type; 1 = heterozygous for mutant allele

The distribution of ewes according to genotype for BMP15 and GDF9 is given in Table 101 for both Cambridge and F700-Belclare along with the corresponding mean ovulation rate.

The average litter size adjusted for ovulation rate is shown in Table 102 for each breed-by-genotype combination. Genotype was a highly significant source of variation in litter size for F700 ewes ($P < 0.01$) but failed to reach statistical significance in the Cambridge ($P = 0.08$). The inclusion of ovulation rate in the model eliminated significant effects of ovulation rate on litter size in the F700 population but did not account for significant variation in the Cambridge. In the latter, ewes with only the BMP15 mutation tended to have a higher litter size than expected while the ewes with both mutations had a litter size that was lower than expected. This pattern was not evident in F700 and may be related to the fact the *FecX^G* mutation is a stop codon which eliminates gene product while the *FecX^B* mutation, which predominates in F700, is an amino acid substitution.

Table 102: Litter size adjusted for differences in ovulation rate

Genotype status [¶]		Breed	Litter size	s.e.
GMP15	GDF9			
0	0	Cambridge	3.0	0.51
		F700	2.8	0.22
0	1	Cambridge	2.5	0.31
		F700	2.4	0.38
1	0	Cambridge	3.5	0.29
		F700	2.5	0.11
1	1	Cambridge	2.2	0.41
		F700	2.9	0.52

[¶] 0 = wild type; 1= heterozygous for mutant allele

These results suggest that while in general these mutations do not impact on embryo survival there is an interaction between the GDF9 mutation (*FecG^H*) and one of the BMP15 mutations (*FecX^G*) which is detrimental to either fertilisation rate or embryo survival

Hanrahan, J.P.

RMIS No. 4785

Allele frequencies at the prion protein (PrP) locus in Suffolk, Texel and Charollais pedigree populations in Ireland

Susceptibility of sheep to scrapie and other transmissible spongiform encephalopathies (TSE) is strongly associated with polymorphisms of the prion protein (PrP) gene. Animals that are heterozygous for the ARR allele have a very low risk of developing scrapie while ARR homozygotes are essentially resistant to infection although a few cases have been detected worldwide. The European Commission has directed that a breeding programme for resistance to TSEs in sheep must be established in all member states by 2005 (Commission Decision 2003/100/EC). The objective of these programmes will be to increase resistance to scrapie by selecting for the ARR allele. In order to establish breeding programmes that are practicable for a particular breed it is necessary to know the frequency of the most resistant allele.

PrP genotype data were obtained from University College Dublin (animals born between 1998-2002) and the Irish Equine Centre (animals born in 2003). The final data set consisted of Suffolk (N=2868), Texel (N=1954) and Charollais (N=1033) sheep. These samples originated from two sources. The majority of blood samples were submitted by flock owners on a voluntary basis. A subset represented animals specifically chosen at random from flocks involved in the national sheep breed improvement programme for lean tissue growth rate. Information on all Suffolk, Texel and Charollais animals was extracted and screened so that only animals with correct pedigree registration details and complete PrP genotypes were retained. The pedigree information allowed the classification of individuals by year of birth. Because only small numbers of animals were born prior to 2000 and a PrP genotyping service was only introduced in 1998 all animals born prior to 2000 were classified as one group for data analysis purposes. The allele frequency data were analysed to evaluate the association with year of birth using Proc GENMOD of SAS, with alleles classified as either 1(=ARR) or 0 (not = ARR), and a logit link function. Orthogonal contrasts were used to evaluate time trends.

The observed frequencies of the five PrP alleles are shown in Table 103 for each of the breeds studied. The results show that the Suffolk had the highest frequency of the resistant ARR allele while the frequency of this allele was lowest in the Texel. The Texel had a high frequency of the ARH allele, which is essentially absent from the Suffolk and Charollais populations. The AHQ allele was essentially absent from the Suffolk and Charollais breeds.

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All five alleles were represented in the Texel population. The year of birth had a significant effect on allele frequency in all three populations ($P < 0.05$) and this reflected a definite increase in the frequency of the ARR allele in all breeds in the 2003-born cohort (see Figure 24). These changes are probably due to increased awareness among breeders of the need to increase scrapie resistance and the decisions of the EU in relation to scrapie.

Table 103: Overall estimates for allele frequencies at the PrP locus in Suffolk, Texel and Charollais sheep

PrP allele	Breed		
	Suffolk	Texel	Charollais
ARR	0.767	0.320	0.527
ARQ	0.226	0.245	0.398
ARH	0.006	0.360	<0.001
AHQ	<0.001	0.018	<0.001
VRQ	<0.001	0.057	0.074

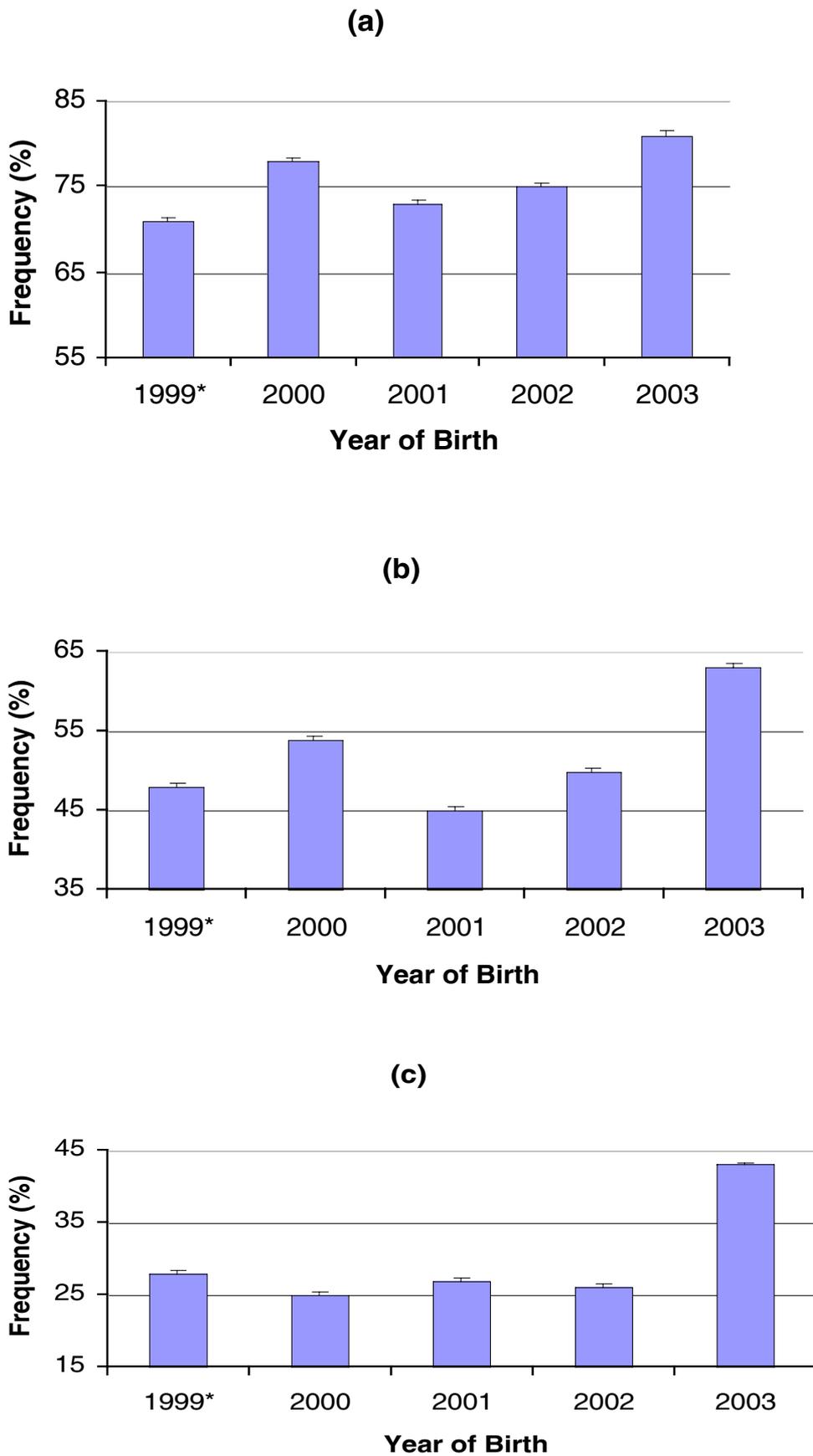


Figure 24. Frequency of ARR allele in Suffolk (a), Charollais (b) and Texel (c) sheep by year of birth (1999* = pooled data for all animals born prior to 2000).

The data were used to establish baseline frequencies for the ARR allele by regarding all animals born prior to 2003 as a representative of the breeds prior to any active breeding policies by breeders for resistance to scrapie. The resulting estimates are shown in Table 104.

Table 104: Baseline allele frequencies at the PrP locus in Suffolk, Texel and Charollais sheep

PrP allele	Breed		
	Suffolk	Texel	Charollais
ARR	0.743	0.263	0.493
ARQ	0.248	0.271	0.423
ARH	0.008	0.384	<0.001
AHQ	<0.001	0.015	0.001
VRQ	0.001	0.066	0.082

The difference in the frequency of the ARR allele between animals born in 2003 and the baseline values was 0.07, 0.17 and 0.15 for Suffolk, Texel and Charollais, respectively. In all cases the difference represented only about one half of that expected if all breeders had adopted a policy of only using stock rams that were homozygous ARR as sires of the 2003 lamb crop.

Estimates of the change in frequency of the ARR allele that would result from a breeding programme that involved using only ARR/ARR sires showed that after 5 years of such a policy the frequency of the ARR allele in lambs would be greater than 0.6 in all breeds and the corresponding frequency of ARR/ARR lambs would be 0.8, 0.5 and 0.6 for Suffolk, Texel and Charollais, respectively.

A selective breeding programme for increased resistance to scrapie would be easy to implement in the Suffolk and Charollais breeds but would seriously constrain Texel breeders in the scope they would have to select for other traits in the initial years of such a programme

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Effects of scrapie (PrP) genotype on lamb growth and on ultrasonic fat and muscle depths in commercial production systems

Susceptibility to scrapie in sheep is strongly associated with polymorphisms of the PrP gene. The ARR allele of this gene is associated with resistance while the ARQ allele is associated with increased susceptibility. Selection for the favourable ARR allele is desirable due to the growing concern about the possible relationship between scrapie in sheep, BSE in cattle and CJD/vCJD in humans. The EU Commission requires the initiation of a breeding programme for resistance to scrapie in all member states for the 2004-breeding season

The study was designed to provide direct estimates of the effects of substituting an ARQ allele with an ARR allele on lamb growth and carcass traits and was completed over two years. Pedigree rams (22 Suffolk, 18 Charollais and 18 Texels) were used for mating in autumn 2002 or 2003. The rams were purchased in pairs from pedigree breeders, with one member being ARR homozygous and the other being ARQ homozygous. The members of a pair were by the same sire, where possible, so as to minimise any confounding of PrP genotype and genetic merit for performance traits. Six flocks were used for progeny testing, four of which were commercial flocks. A minimum of 4 rams (2 pairs) was tested per flock

each year. In each flock ewes were assigned at random to individual mating groups (at least 40 ewes per group) and rams were randomly assigned to these groups.

All progeny were tagged and weighed at birth, weighed again at *ca.* 35, 98 and 130 days of age and fat and muscle depth over the loin was measured using ultrasonography in conjunction with the last two weights. These measurements were used to estimate live-weight (LWT), ultrasonic muscle depth (UMD) and ultrasonic fat depth (UFD) at 120 days of age for each lamb. These are the traits used in the Pedigree Sheep Breed Improvement Programme (PSBIP) operated by the Department of Agriculture and Food to calculate the Lean Meat Index.

The PSBIP traits, and growth rate to weaning (G014) and weaning weight (W14), were analysed using Proc MIXED of SAS to fit a mixed linear model with sire effects treated as random. The model included fixed effects for birth-type (1 to 4), rearing-type (1 to 3), sex, dam-age (2 to 4), flock, year-of-birth and sire breed. Interactions representing flock*sire-breed and flock*year-of-birth were also included. Other 2-factor interactions were examined in preliminary analyses and were not significant. The differences between ARR/ARR and ARQ/ARQ sires were obtained by appropriate linear contrasts within each breed of sire and sire was the experimental unit.

The average number of progeny with performance data at 120 days of age was 52, 59 and 60 per sire for Suffolk, Texel and Charollais, respectively. Sire effects were significant ($P < 0.001$) for all traits considered. The estimates of the effects of substituting an ARR allele for an ARQ on growth rate to 98 days (G014), weight at weaning (W14), LWT, UMD and UFD are shown in Table 105 and show that substituting an ARR allele for an ARQ allele had no effect on any of the growth traits measured, for any of the three breeds. There was no evidence for an effect on ultrasonic fat depth in any breed. In the case of muscle depth the substitution of ARR allele for an ARQ allele had a significant ($P < 0.01$) negative effect in the Suffolk breed and a significant ($P < 0.02$) positive effect in Charollais. There was no evidence for an effect of allele substitution on this trait in Texels. The finding of a significant association between PrP genotype and muscle depth along with previous reports for some breeds of an association between PrP genotype and conformation suggest that there is a gene linked with the PrP locus that has an effect on muscularity. The absolute magnitudes of the effect on muscle depth are not considered to be of any practical significance.

Table 105: Effect (\pm s.e.) of genotype of sire (ARR/ARR minus ARQ/ARQ) on lamb growth and ultrasonic measurements

Trait	Breed of sire		
	Suffolk	Charollais	Texel
G014 (g/day)	-2.0 \pm 2.11	-1.9 \pm 2.41	-0.5 \pm 2.17
W14 (kg)	0.15 \pm 0.223	-0.27 \pm 0.255	0.03 \pm 0.233
LWT (kg)	-0.31 \pm 0.273	-0.26 \pm 0.316	0.35 \pm 0.281
UFD (mm)	-0.00 \pm 0.016	0.02 \pm 0.018	-0.02 \pm 0.016
UMD (mm)	-0.33 \pm 0.130	0.36 \pm 0.153	0.12 \pm 0.133

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The effects of the PrP allele on carcass traits and slaughter age are given for each breed in Table 106. The ARR allele was associated with a small but significant ($P < 0.05$) increase in age at slaughter in Suffolks and in the Texel the ARR allele was associated with an increase of 0.2 kg in carcass weight ($P < 0.05$). None of the other effects approached statistical significance ($P \geq 0.2$).

Table 106: Effect (\pm s.e.) of genotype of sire (ARR/ARR minus ARQ/ARQ) on lamb age at slaughter and carcass traits

Trait	Breed of sire		
	Suffolk	Charollais	Texel
Slaughter age (days)	3.6 \pm 1.79	-2.4 \pm 1.87	-0.2 \pm 1.80
Carcass weight (kg)	-0.09 \pm 0.081	-0.01 \pm 0.083	0.20 \pm 0.082
Conformation score	-0.01 \pm 0.024	-0.02 \pm 0.024	0.00 \pm 0.023
Fatness score	-0.00 \pm 0.012	0.00 \pm 0.012	-0.00 \pm 0.012

The data support the hypothesis that substituting an ARR allele for an ARQ allele has no important negative effect on lamb growth or on the traits used in the PSBIP for any of the breeds examined. Likewise, there was no evidence for any important effects on carcass traits. Consequently it is concluded that the breeding programme required to be implemented by the EU, based on the selection for the ARR allele, will not affect any of the traits used in the PSBIP.

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Use of flow cytometric analysis to evaluate results from staining frozen-thawed semen to assess quality characteristics

On-going studies to identify an *in vitro* method that could be used to assess the ability of frozen-thawed semen to for use in cervical AI included use of specific stains for *in vitro* assessment. The staining methods used were:- (1) syber14 plus propidium iodide to identify live cells and the state of the acrosome and (2) merocyanine 540 plus Yo-Pro to identify live/dead status and whether the live cells were capacitated. Spermatozoa were also tested for osmotic resistance to provide an independent assessment of the proportion of live cells.

The results from these procedures were evaluated by processing the samples through a flow cytometer to count the number of cells in each possible category resulting from each test procedure. This allows a very large number of individual sperm cells to be evaluated per sample. The results were used to calculate the following variables for each straw (3 straws per ram): - (a) Live_PI = percentage live cells based on staining with syber/propidium iodide, (b) Dead_YPro = percentage dead cells based on Yo-Pro, (c) Live_Osm = percentage live based on osmotic resistance, (d) Capacitation = percent capacitated (among live cells) based on merocyanine stain, (e) Acrosome = percentage of live cells with damaged acrosome based on syber14 and (f) Damaged_Acrosome = percentage of cells with damaged acrosome based on syber14.

These variables were analysed to evaluate the effects of individual ram to ram variation and the estimates for individual rams were compared with *in vivo* fertility. Differences among rams were significant for Acrosome ($P < 0.05$), Capacitation ($P < 0.02$), Dead_Ypro ($P < 0.001$)

but were not significant for Live_PI. The individual ram effects approached significance for Damaged_Acrosome (P=0.07) and Live_Osm (P=0.095).

The relationships among these variables were examined and the correlation between the various estimates of the proportion of live cells was significant : Dead_Ypro and Live_Osm, r = -0.65; Dead_Ypro and Live_PI, r = -0.46; Live_PI and Live_Osm, r = 0.51.

The relationships between *in vivo* fertility and the variables defined above were all examined and in no case was a significant correlation detected. A sample of the pattern of relationships is shown in Figures 25 and 26.

The results from this study have failed to identify a laboratory test that could be used to select rams that would give high pregnancy rate following use in cervical AI with frozen-thawed semen.

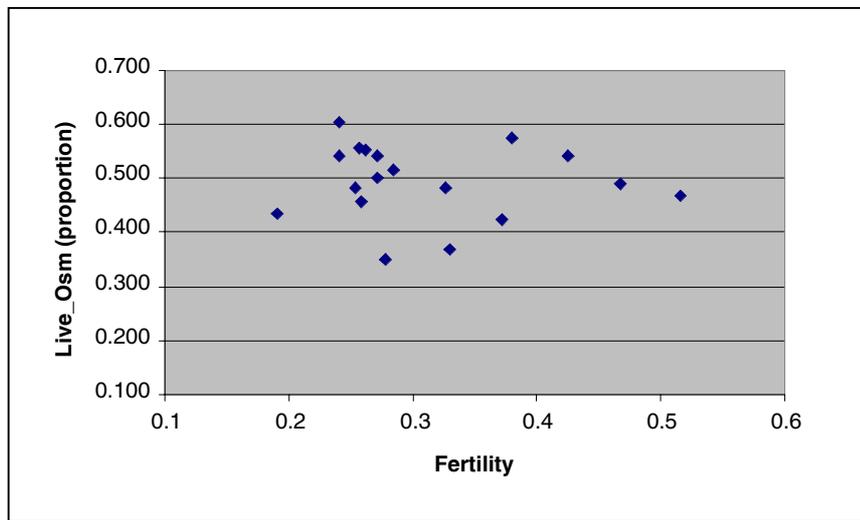


Figure 25. Relationship between *in vivo* fertility and the percentage of live cells based on osmotic resistance test.

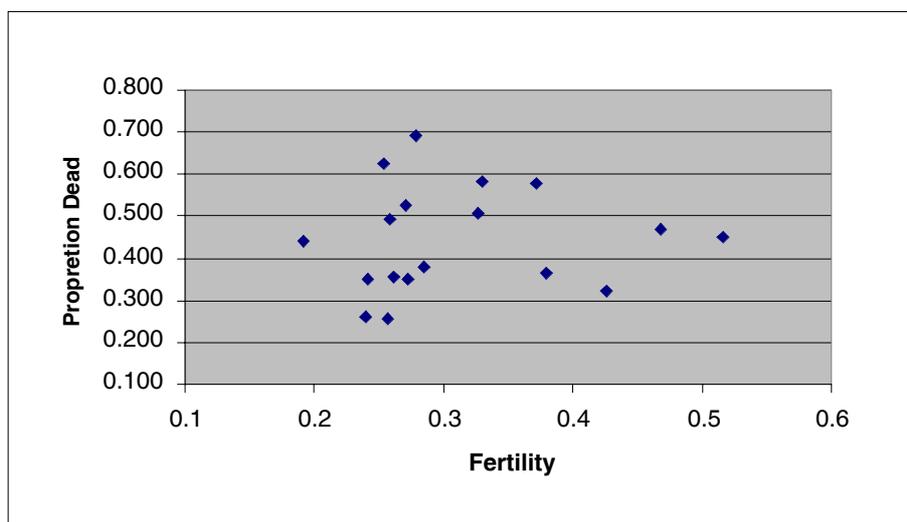


Figure 26. Relationship between *in vivo* fertility and the percentage of dead cells based on staining with Yo-Pro.

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Correlation of ram field fertility, following cervical insemination of ewes using frozen-thawed semen, with *in vitro* fertilisation of ewe oocytes

Differences in field fertility among rams using artificial insemination (AI) with frozen-thawed semen have been reported, however, no good laboratory test exists that can predict these differences. The objective of this study was to develop a suitable *in vitro* fertilisation (IVF) protocol capable of identifying differences among rams of differing field fertility after AI using frozen-thawed semen.

Experiment 1: In vivo fertility

Semen was collected from 18 rams and loaded in 0.25 ml straws (200 x 10⁶ sperm per straw). Straws were thawed at 70°C for 8 s, and synchronised ewes (N= 427) were cervically inseminated 56 to 58 h post-sponge withdrawal. On Day 28 after AI, ewes were slaughtered and evaluated for pregnancy. Average pregnancy rates for 18 rams tested, was 46.3%. Three high (50.3, 50.0 and 46.7%) and 3 low (30.4, 25.5 and 22.9%) fertility rams were identified for use in Experiment 2. These results are similar to those found previously, where pregnancy rate ranged from 35.1 to 78.9% between rams.

Experiment 2: In vitro fertilisation

The fertilising ability of 6 rams (from Experiment 1) was evaluated using IVF. Oocyte maturation, fertilisation and culture were standard. Either 2 x 10⁶ or 0.062 x 10⁶ sperm cells per ml were used for IVF of the ewe oocytes. There were 25 oocytes per well and 2 wells per ram per replicate with 6 replicates. Zygotes were cultured in 25- μ l droplets of SOF under mineral oil. Cleavage rate was monitored at 48 h post-insemination and blastocyst yield checked on Days 5 to 8.

When semen was added to oocytes at 2 x 10⁶ sperm per ml, no significant differences were found between high and low fertility rams for cleavage rate or blastocyst yield, in agreement with other studies. When the experiment was repeated using 0.062 x 10⁶ sperm per ml, no differences were found among rams for blastocyst yield; however, there was a significant difference among rams for percentage of oocytes cleaved (range from 41.4 to 69.4%; P=0.031). There was a significant (P<0.05) correlation between the field fertility of rams and the cleavage rate *in vitro*. To our knowledge, this is the first report of a correlation between IVF results and field fertility using frozen-thawed semen and cervical AI. The fact that previous studies have not used as few as 0.062 x 10⁶ sperm per ml for IVF may explain why a significant correlation was not found in previous studies.

It is concluded that there is a correlation between fertilisation rate *in vitro* and fertility *in vivo* when 0.062x10⁶ sperm cells were used for IVF. This may be a useful laboratory method for predicting field fertility of frozen-thawed ram semen.

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Use of computer-aided sperm analysis (CASA) to assess *in vivo* fertility of frozen-thawed semen

On-going studies to identify an *in vitro* method that could be used to assess the ability of frozen-thawed semen to for use in cervical AI was extended to include computerised image analysis of the activity of individual spermatozoa. Semen from 18 rams that have been evaluated for *in-vivo* fertility of their frozen-thawed semen was used in this study. Individual straws from a frozen semen bank established for such studies were assessed using three straws per ram. Typically about 40 individual spermatozoa were tracked per straw. The data set consisted of values for 2917 individual sperm. The variables measured were as follows :- Average velocity of motile sperm on curvilinear path in $\mu\text{m/s}$ (VCL), Average velocity of motile sperm on average path in $\mu\text{m/s}$ (VAP), Average velocity of motile sperm on straight line in $\mu\text{m/s}$ (VSL), Percentage of linearly motile sperm (Linearity) = ratio (VSL/VCL) x 100 (LIN), Percentage of straight motile sperm (Straightness) = ratio (VSL/VAP) x 100 (STR), Mean angular displacement in degrees along the trajectory (MAD), Beat cross frequency (Hz) (BCF), Amplitude of lateral head displacement (μm) (ALH).

The distributions of the variables was examined and based on this analysis log transformations were used prior to statistical analysis. The variables are clearly correlated given the definitions and in order to determine if any combination of the variables could effectively discriminate among the panel of rams with respect to *in vivo* fertility the transformed vales were subjected to cluster analysis using Proc CLUSTER of SAS. Two methods were examined – one is based on the average distance between pairs of observations (distance is a function of the values for the set of variables used as the basis for clustering) and the other is based on the Euclidean distance between cluster means (*centroid method*). There was good consistency between the results of the two methods as shown in Figure 27. A range of endpoints for the clustering process were examined and the outcomes from these and from two methods were compared. The results failed to reveal any critical difference between endpoints or the methods and the centroid method was used for final analyses with the clustering process stopped at the stage when the data was condensed to 6 clusters.

The percentage of sperm in each of the 6 clusters was as follows 24.3, 46.5, 28.8, 0.2, 0.1 and 0.07, for clusters 1 to 6, respectively. Cluster 1: contained poorly progressive and non-active sperm (24.3%); cluster 2: consisted of highly active but non-progressive sperm (46.5%); cluster 3: had highly progressive and motile sperm (28.8%). The other clusters were not relevant due to very low incidence. The percentage of spermatozoa that were each cluster was computed for individual rams and related to *in vivo* fertility. Examples of the results are in Figures 28, 29 & 30.

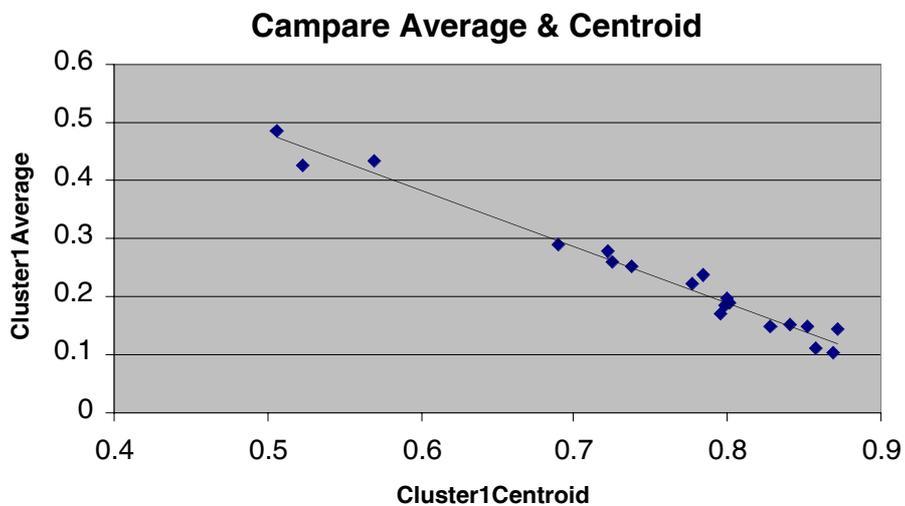


Figure 27. Average percentage of sperm in cluster 1 for each ram - comparison of two clustering methods.

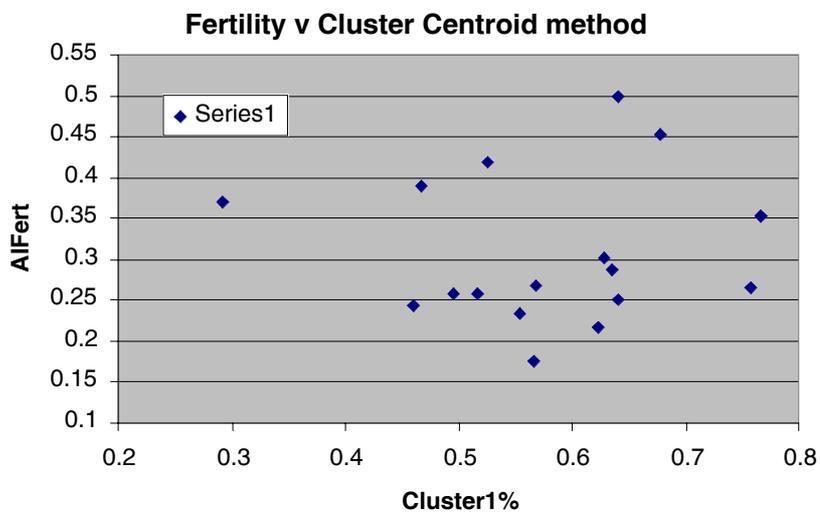


Figure 28. Relationship between in vivo Fertility and Cluster1%.

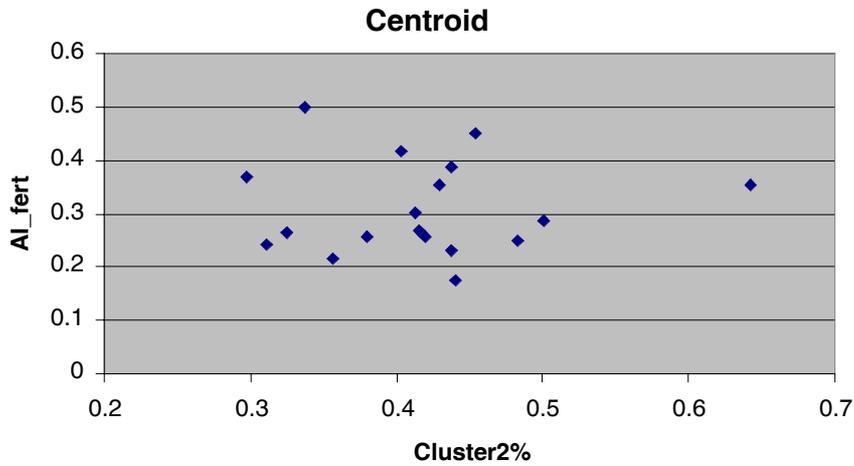


Figure 29. Fertility and percentage sperm in Cluster2.

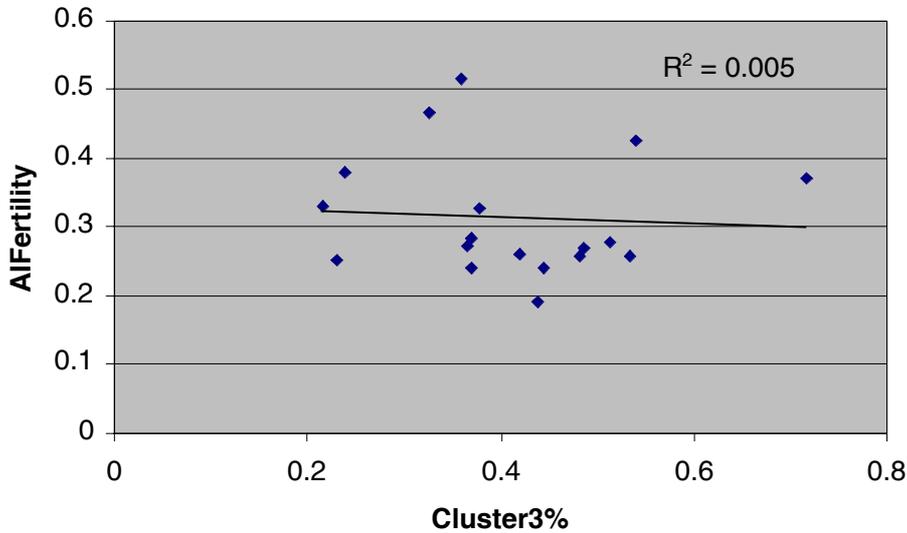


Figure 30. Fertility and percentage sperm in Cluster3.

From the results in the above figures it is clear that there is no evidence for a relationship between fertility and the incidence of specific sperm sub-populations. It is most unlikely that any other version of Cluster analysis will reveal a relationship.

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Effect on pregnancy rate of re-suspending frozen-thawed ram spermatozoa in seminal plasma prior to cervical AI

Impaired function of cryopreserved spermatozoa can be overcome by addition of seminal plasma (SP), resulting in normal fertility after cervical AI (Maxwell *et al. Reprod. Fertil. Dev.*, 1999, 11:123). The aim of this study was to evaluate the effect on pregnancy rate of adding SP to frozen-thawed semen prior to AI. Semen was collected from 6 rams (3 High and

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3 Low fertility from prior evaluation) and frozen in 0.25-ml straws (800×10^6 sperm/ml). SP was obtained by centrifugation (6500g for 15 min at 4°C) of pooled ejaculates from High (HSP) and Low (LSP) fertility rams and filtered (0.22 µm). Artificial SP (ASP) was made as per Mortimer and Maxwell (*Reprod.* 2004, 127:285). Prior to AI, frozen straws were thawed (70°C for 8 s), added to 62.5 µL of HSP, LSP or ASP (20% v/v), incubated at 33 °C for 5 min and loaded into 0.5-mL straws for AI. Ewes inseminated with untreated frozen-thawed semen from the same rams were controls (NSP). Ewes (N=295; 2 replicates) were synchronised (30-mg FGA pessaries), received 400 IU PMSG at pessary withdrawal and inseminated 57 h later (day 0). Pregnancy was determined at slaughter (day 29). Pregnancy rates were 28.5, 24.3, 24.6 and 15.0% for HSP, LSP, ASP and NSP, respectively (P>0.3). The relatively low value for NSP was due to one replicate. The difference between ram fertility types did not reach significance (P=0.1) and there was no interaction between SP treatment and ram type. The results do not support previous findings that SP has a beneficial effect on frozen-thawed spermatozoa.

RMIS No. 5081

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Is post-thaw visual assessment useful for quality control of frozen ram semen?

The quality of frozen ram semen is usually evaluated by visual assessment of the viability and motility of spermatozoa post-thawing. This evaluation typically leads to rejection of about 30% of samples. There is disagreement on the value of such *in vitro* assessment of semen as a predictor of fertility. In ongoing AI research in this laboratory, semen from individual rams is frozen in Minitüb[®] straws using standard procedures. Sample straws from daily collections from each ram are thawed and assessed visually (viability and motility) using phase-contrast microscopy (200X and 400X) and classified as:- accepted for use in AI, rejected or marginal. Parous ewes (N=148) were synchronised (30-mg FGA pessaries) and cervically inseminated, 57 h post-pessary removal, with frozen-thawed semen from a panel of 8 rams. Each ram was represented by semen from at least 2 (usually all 3) of the categories described above and representing the same collection period. Non-return rate was determined, by syndicate joining with entire rams, from day 14 post AI. Non-return rates were 24%, 20% and 28% for samples categorised as accepted, rejected or marginal, respectively - these differences were not significant ($\chi^2_{2 \text{ d.f.}} = 0.70$; P>0.7). Others have also demonstrated a lack of correlation between visual assessment and fertility. It is concluded that visual assessment was not related to *in vivo* fertility of cryopreserved ram semen and that the use such assessment is an unjustified overhead in routine semen freezing. An objective and practicable laboratory procedure that is reliably associated with fertility of frozen-thawed ram semen is required.

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RMIS No. 5081

Developmental competence of embryos from Belclare and Suffolk ewes

We have previously reported that the percentage of fertilised oocytes which reached the blastocyst stage by day 6 after AI with frozen-thawed semen was higher for Belclare (94%) than Suffolk (59%) ewes. This may reflect differences in oocyte quality or the timing of fertilisation. Mature Belclare (B; N=69) and Suffolk (S; N=71) ewes were synchronised (30-mg FGA pessary) in 4 replicates and laparoscopically inseminated 60 h post-pessary removal and slaughtered 1 or 2 days later. Presumptive zygotes were recovered and cultured *in vitro* (grouped by breed). Immature oocytes, aspirated from the recovered ovaries of the ewes, were matured, fertilised and cultured *in vitro* (by breed). Cleavage was determined 48 h post-insemination and blastocyst development was assessed on days 5-7. Data were analysed by

least squares with culture group as the unit. There was no significant ($P < 0.05$) ewe-breed effect on cleavage rate (%) of oocytes matured *in-vivo* (40 & 49, s.e. 9.5, for B & S) or *in-vitro* (49 & 54, s.e. 9.5, for B & S). Likewise, ewe breed had no significant effect on the percentage of cleaved oocytes developing to the blastocyst stage for *in-vivo* (29 & 19, s.e. 8.0, for B & S) or *in-vitro* matured oocytes (27 & 33, s.e. 7.5, for B & S). It is concluded that oocyte quality does not differ between the breeds and therefore previous differences in developmental competence of early embryos must reflect differences in the uterine milieu.

RMIS No. 5081

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Ewe breed differences in oestradiol and progesterone concentrations during the peri-ovulatory period

We have recently reported that the difference between Suffolk and Belclare ewes following cervical AI with frozen-thawed semen is due to (i) sperm traversing the cervix and uterus in a higher proportion of Belclare than Suffolk ewes, leading to a higher incidence of fertilization, and (ii) the lower developmental competence of fertilized oocytes from Suffolk ewes (Fair et al., 2005). This difference in sperm transport may be due to a difference in the finely tuned endocrine control of sperm transport mechanisms while the lower developmental competence in Suffolk than Belclare embryos may be due to a difference in the quality of the oocyte being ovulated or due to differences in the uterine environment.

The objective of this study was to examine differences in oestradiol- 17_β (E2) profiles in the pre-ovulatory period and progesterone (P4) levels from around the time of ovulation to Day 6 of the oestrous cycle in ewe breeds known to differ in pregnancy rate to cervical AI using frozen-thawed semen.

Multiparous purebred Suffolk (N=35) and Belclare (N=30) ewes were synchronised. Jugular venous samples were collected in EDTA coated vacutainer tubes from all animals at 3 h intervals between 18 and 54 h post pessary removal, then every 6 h until 83 h and then every 12 h until Day 6 of the cycle. Laparoscopy was performed on all ewes at midcycle. Two weeks prior to slaughter live weights were recorded from 73 purebred Suffolk and 72 purebred Belclare ewes. At slaughter their carcass weights were recorded and livers were isolated, examined and weighed. Only data from healthy livers were recorded.

Data from 62 Belclare and 58 Suffolk livers were included in the calculation of liver weights. There was no difference between Suffolk and Belclare ewes in live weight (75.47 ± 1.45 vs. 72.38 ± 1.56 kg respectively; $P=0.15$) or in carcass weights (33.78 ± 0.79 vs. 31.88 ± 0.85 kg respectively; $P=0.1$). Livers were heavier in Suffolk than in Belclare ewes (0.99 ± 0.04 vs. 0.88 ± 0.04 kg respectively; $P=0.04$).

At the time of laparoscopy, the view was impaired in one Suffolk and one Belclare ewe and thus these were excluded in the calculation of ovulation rate. Values from three Suffolk and two Belclare ewes were excluded from analysis due to abnormal hormone concentrations.

Belclare ewes had a higher ovulation rate than Suffolk ewes (3.31 ± 0.2 vs. 2.21 ± 0.18 respectively; $P < 0.001$). The interval from pessary removal to the LH surge was significantly shorter in Suffolk than in Belclare ewes (34.6 ± 0.8 vs. 37.95 ± 0.87 h, respectively; $P < 0.05$). E2 concentration rose up to -6 h and then fell sharply and this change was significant over

time ($P < 0.001$; Figure 31); however it was unaffected by breed. When an average E2 value was taken from all points prior to the LH surge (3.29 ± 0.18 and 2.96 ± 0.17 ng/ml for Suffolk and Belclare ewes respectively) there was a significant effect of ovulation rate on E2 concentration ($P < 0.05$) but no effect of breed or any breed by ovulation rate interaction ($P > 0.05$).

Ewe breed had no effect on P4 concentration ($P = 0.42$), however, P4 concentrations changed significantly over time ($P < 0.001$; Figure 32). There was a significant interaction between breed and time with the rate of increase of P4 occurring earlier in Belclare than Suffolk ewes ($P < 0.001$). There was a significant linear and quadratic relationship between ovulation rate and P4 concentration at all time points post Day 1 ($P < 0.05$). When an average value for P4 concentration was taken from points post Day 2 (2.01 ± 0.16 and 2.41 ± 0.18 ng/ml for Suffolk and Belclare ewes respectively) there was a quadratic relationship between P4 concentration and ovulation rate ($P < 0.01$; Figure 32). When a linear regression was performed on the relationship between P4 concentration, breed and ovulation rate, the difference in ovulation rate between breeds accounted for the differences in P4 concentration ($P < 0.05$). Liver weight was unrelated to P4 concentration ($P > 0.05$).

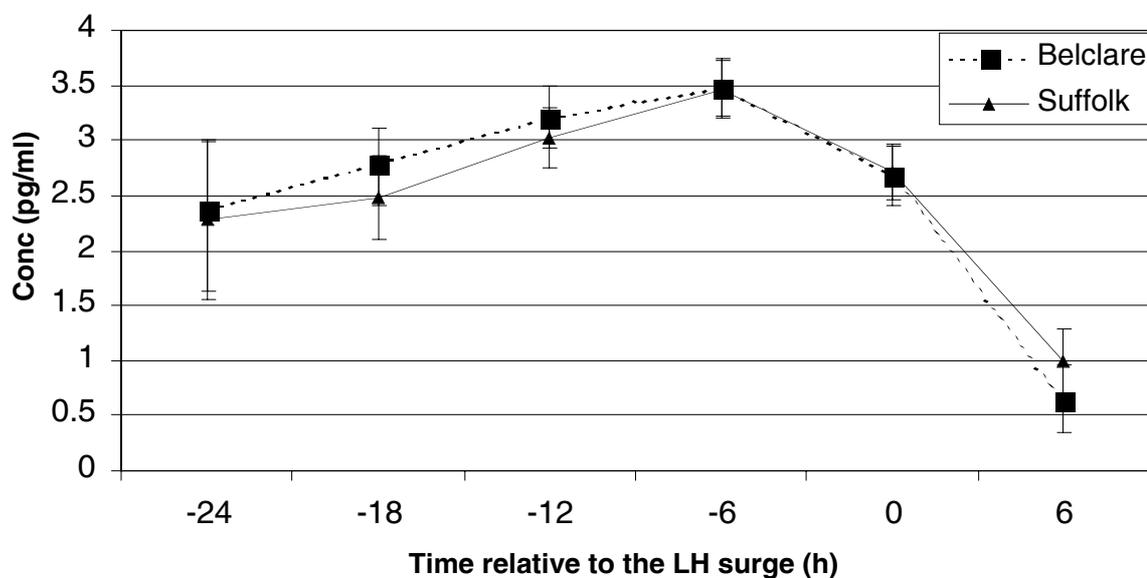


Figure 31: Oestradiol- 17_β concentrations (pg/ml) from -24 h prior the luteinising hormone surge to +6 h in Suffolk and Belclare ewes.

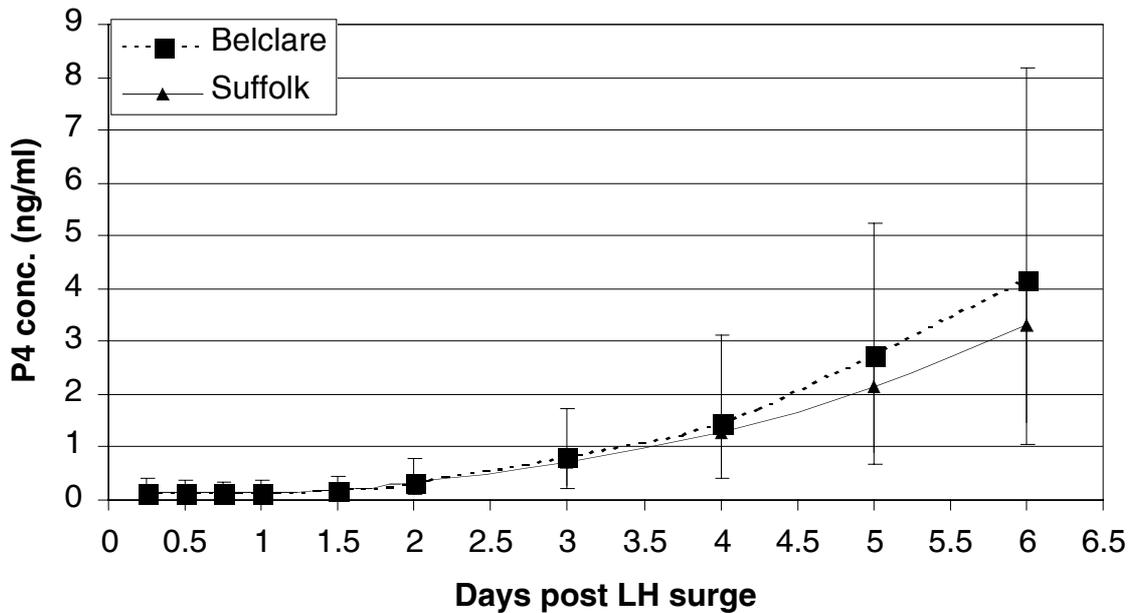


Figure 32: Progesterone concentration (ng/ml) from Day 0.25 to Day 6 post the luteinising hormone surge in Suffolk and Belclare ewes

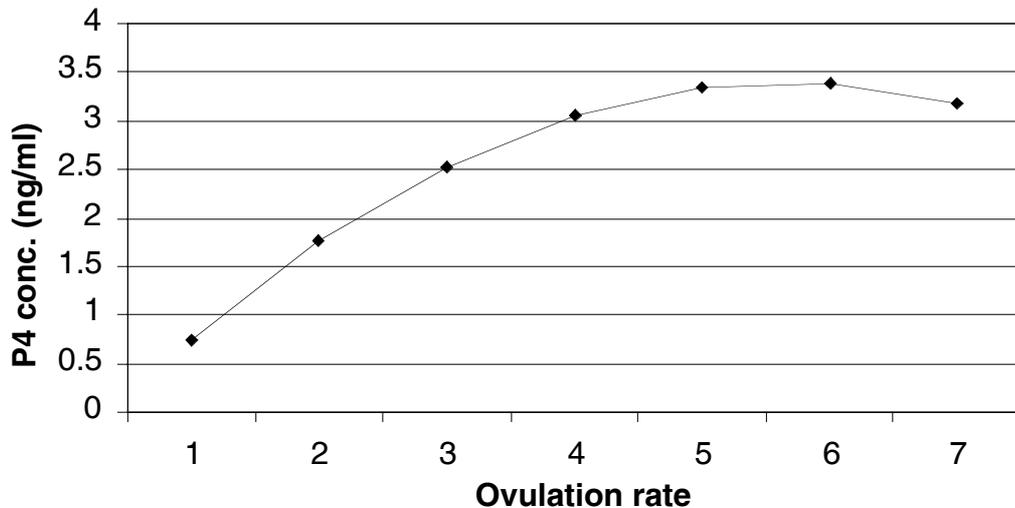


Figure 33: Quadratic relationship between ovulation rate and progesterone concentration in Suffolk and Belclare ewes (P4 concentration taken as an average value for each breed over a 3 day period from Day 3 to Day 6 - 2.01 ± 0.16 and 2.41 ± 0.18 ng/ml for Suffolk and Belclare ewes respectively)

RMIS No. 5081

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Performance summary for Galway pedigree flock

All flocks registered with the Galway Sheep Breeders Association Ltd. and located in the Republic of Ireland participate in the Galway Sheep Breed Improvement programme operated by the Department of Agriculture, Food and Rural Development. This programme involves the recording of litter size data on individual pedigree Galway ewes with the objective of calculating a merit index for prolificacy for individual ewes and their progeny. Breeders are encouraged to use this information in the selection of breeding stock so as to bring about genetic improvement in prolificacy. The data collected under the programme are entered, validated and processed by the Sheep Production Department of Teagasc at the Sheep Research Centre, Athenry.

A total of 376 ewes were recorded as having produced purebred Galway lambs and 5 of these were joined as ewe lambs. Only 73% of ewes on the original mating plan were recorded as having lambed to a purebred mating (lambing details for 5 ewes that were mated to Suffolk rams were also returned). The litter size data for all ewes, except yearling mothers, are summarised in Table 107 by age of ewe.

Table 107: Litter size in 2004 for pedigree Galway ewes in 21 flocks

Ewe age ⁺	No. of ewes		Litter size	
	Joined	Lambled	Total	Live
2	221	140	1.36	1.25
3	154	77	1.48	1.35
4	111	56	1.61	1.42
5	142	99	1.68	1.34
Overall	513	372	1.53	1.32

⁺5 ewe lambs were joined and all of these lambed singles

A total of 25 rams, from 13 different flocks, sired progeny in 2004. The number of sires represented among the lambs born in 2004 is detailed in Table 108 by flock in which the ram was born.

Table 108: Source of sires of 2004-born pedigree Galway lambs and number of progeny by sires from each source

Flock of origin of sire	No. of sires	No. of ewes lambed	No. of lambs
CD	1	7	13
DC	1	10	16
FC	1	19	27
GO	3	54	68
HM	1	25	21
JC	5	81	122
JM	2	27	31
MK	1	35	54
MM	3	25	23
MW	2	13	20
PS	3	46	57
TF	1	4	5
TK	1	39	45
Overall	25	372	500

While the number of rams used and the number of flocks in which these were bred is a positive indicator that the narrowing of the genetic base that was evident some years ago has been halted the failure of new entrants to remain in the Association is a cause for concern.

Conservation of semen from Galway rams

A programme was initiated to collect semen from a representative panel of pedigree Galway rams and freeze for long term storage as part of a project on the conservation programme for this native breed. Pedigree breeders provided access to stock rams for the project. The rams became available at two stages during the late autumn: group 1 (N=15 rams) was assembled in early November and group 2 (N=3) was assembled in mid December. All rams were housed on straw and fed silage and a high protein ration and allowed to acclimatise to their new environment. Rams were then trained to serve a teaser ewe that was restrained in a holding crate on a raised platform. Initially the rams were trained to go up the ramp and serve the oestrus ewe without any attempt to collect semen. This procedure was continued for a couple of days (depending on the temperament of the ram) with increased handling by the operator until the ram is fully accustomed to the handler. Then the ram was trained to serve into an artificial vagina.

The response to training, which was continued over several weeks was disappointing and unusually poor. Some rams exhibited slight interest in the oestrous ewe initially but interest then waned as time progressed. This is contrary to what would normally happen. Only 5 of the 18 rams were successfully trained to serve into the artificial vagina and of these only 4 rams gave satisfactory semen volume and thus contributed semen that could be frozen. Semen was frozen following standard dilution and processing procedures. The number of straws stored from these rams is summarised in Table 109.

Table 109: Summary of semen collection from Galway rams

Ram tattoo	No. of straws frozen
PS02/61	108
PS02/8	87
GO02/025	200
WD03/653	144

Because it was not possible to obtain the rams for collection purposes until after the joining period in the pedigree flocks it is possible that it was too late in the season. In addition 3 of the panel were rams lambs and while one of these was trained successfully the produced semen volume was insufficient for freezing purposes. It was concluded that to maximise the prospect of getting rams trained and yielding good semen volume it is probably necessary to collect from Galway rams during the early part of the normal breeding season. In addition, because it was more difficult to train and collect from ram lambs and the volume of semen was very low this age group should not be included in the semen collection programme in future years.

Genetic variation in the Galway sheep breed based on microsatellite markers

The Galway breed is the only native Irish sheep breed and is listed as a breed in danger of extinction. As part of the genetic conservation and overall study of the breed a DNA bank has been established. Blood samples were collected in 10-ml heparinized vacutainers from 163 pedigree Galway sheep. These animals were born in 15 different pedigree flocks and were

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the progeny of 44 sires. The set of sires originated from 20 different pedigree flocks. An aliquot (2 ml) of whole blood from each 10-ml sample was stored at -20°C and 180 μl was put on FTA paper. Genomic DNA from the remaining blood was extracted using a proteinase K extraction method. This bank is representative of the pedigree population in Ireland and will provide material for investigations on the level of genetic variation in the breed. The objective of this study is to describe the variability found in the breed and to compare these findings with variability studies on other sheep breeds.

A panel of 16 microsatellites has been selected and tested for use in the evaluation of the genetic variation in the Galway breed. This panel represents both arms of the three metacentric chromosomes and 10 other chromosomes. Linkage of the results with other studies on genetic variability was an important criterion in selecting the panel of microsatellites. A subset of 94 animals was chosen for use in the present study. These represented all of the sires in the DNA bank and generally only one offspring per dam was included. The DNA from this group has been genotyped using eight microsatellite markers out of the panel of 16. Polymerase chain reaction (PCR) was performed according to standard procedures. The PCR products were subjected to electrophoresis on an ABI3100 capillary sequencer. The resulting data on the length of PCR products were analysed using the GenemapperTM software package and genotypes were assigned to individual sheep based on this information. The variability at each marker locus was quantified by calculating the effective number of alleles, the expected heterozygosity and the polymorphism information content.

Table 110: Effective number of alleles, heterozygosity and polymorphism information content (PIC) for eight microsatellite loci in pedigree Galway sheep

Microsatellite	Chromosome	Effective no. of alleles	Gene diversity	Observed heterozygosity	PIC
BM1824	1	5.0	0.80	0.58	0.77
MAF64	1	3.2	0.68	0.64	0.63
OarFCB11	2	3.5 (4.1) ^a	0.81	0.59	0.79
OarFCB128	2	5.4 (5.3)	0.72	0.39	0.67
OarCP43	3	3.6	0.72	0.66	0.67
OarCP34	3	3.0 (3.7)	0.66	0.75	0.63
OarHH35	4	3.2	0.69	0.31	0.64
OarAE129	5	3.4	0.70	0.76	0.65

^a Average for 6 Spanish breeds .

The results on the variability at each microsatellite locus are presented in Table 110. All the microsatellite loci were highly polymorphic as indicated by the number of alleles present and were reasonably consistent with results from other studies using the same microsatellites. Available information on the effective number of alleles from other studies is included for comparison. The heterozygosity was around 60% or greater for 6 of the 8 microsatellite loci indicating considerable genetic variability. However, there is an indication that the effective number of alleles is somewhat less than found in other sheep breeds.

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RMIS No. 5254

ANIMAL HEALTH AND WELFARE

Survey of helminth control practices on lowland sheep farms in Ireland

Gastrointestinal parasites are an important cause of production loss in ruminants. Farmers have controlled the impact of parasites on performance by the intensive use of anthelmintics. Treatment frequency, the proportion of the parasite population exposed to the anthelmintic, optimum dose rates and movement of sheep containing drug resistant worm populations are considered important factors in influencing the rate of development and prevalence of drug resistance. The survey was conducted to determine parasite control practices and assess risk factors that may contribute to the development of anthelmintic resistance on lowland sheep farms.

A questionnaire relating to farm details, grazing and worm treatment practices was mailed to two groups of sheep farmers. The first group (N=128) was selected by Teagasc advisors based on the criteria that they were lowland producers with a history of a long-term sheep enterprise and with a ewe flock size ≥ 100 . The second group (N=38), were lowland producers already involved in a Teagasc Technology Evaluation Transfer (TET) study (RMIS 4813/4928).

69% non-TET and 74% of TET farmers returned questionnaires. A significant proportion of respondents had cattle /sheep enterprises (65% and 61% of non-TET and TET farms, respectively) while the remainder were sheep-only enterprises. The majority of farmers (99%) indicated they treated for roundworms with an anthelmintic. Some aspects relating to treatment practices are summarized in Table 111 and Figures 34 & 35.

Table 111: Summary of responses (% respondents) in relation to treatment strategy and dose calculation

	Sampling Group			
	<u>Lambs</u>		<u>Ewes</u>	
	Non-TET	TET	Non-TET	TET
Minimum number of respondents	86	27	80	24
<i>Treatment Strategy (%)</i> :				
Set dosing programme	91	74	95	88
Dose at sign of disease	6	22	5	12
<i>Weight basis for dosing (%)</i> :				
Heaviest actual	51	59	28	42
Heaviest guessed	31	30	46	39
Average guessed	17	11	24	19

Calculating the amount of dose administered based on either guessing the weight of the heaviest animal or on the average weight of group may lead to the administration of a sub-optimal dose, which enhances the selection pressure for resistant worms. Only 50% of farmers reported that they checked the accuracy of the dosing gun (65% non-TET, and 50% TET) prior to dosing.

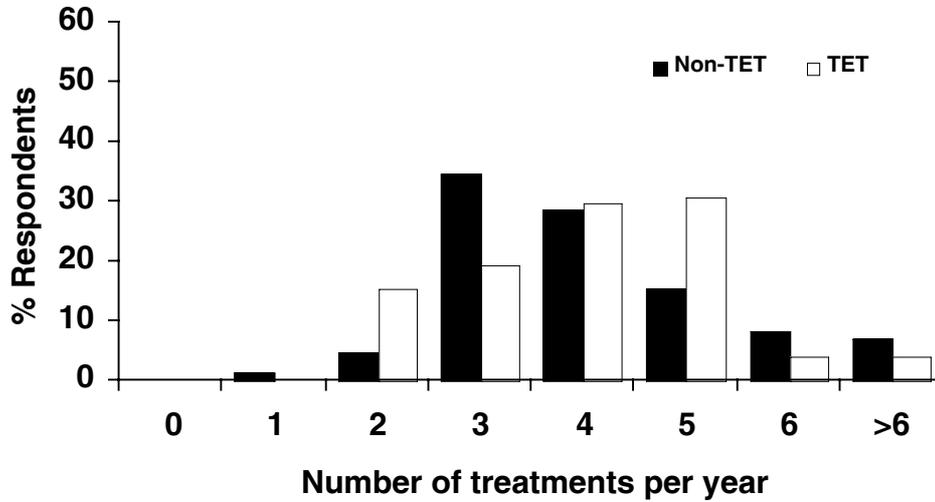


Figure 34. Frequency of treatment in lambs

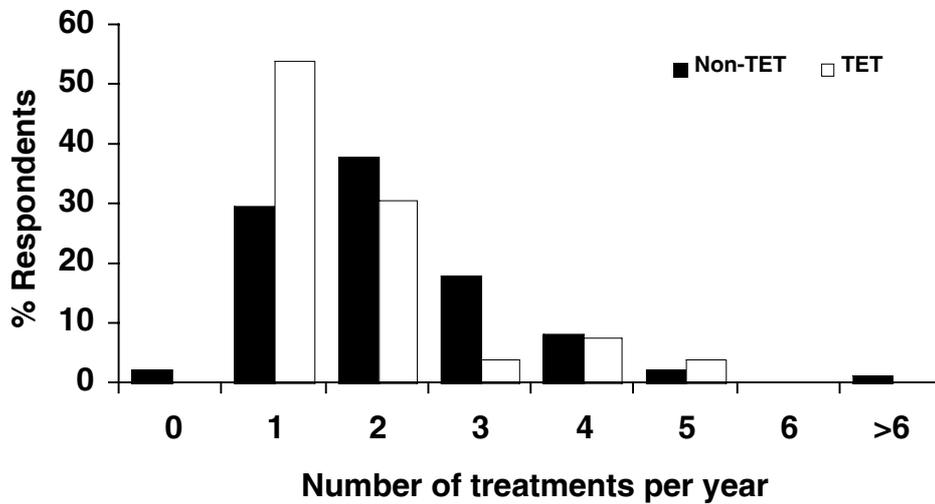


Figure 35. Frequency of treatment in ewes

34% of non-TET and 11% of TET farmers reported the practice of withholding food prior to dosing, which reduces rate of digesta passage and increases parasite exposure to the drug. In the majority of cases (>90%) the withholding period was <6 hours. For benzimidazoles and macrocyclic lactones, it has been shown that efficacy of these drugs is only enhanced when food is withheld for 12 to 24 hours prior to treatment.

Another important factor in increasing the risk of anthelmintic resistance on a farm is through purchased sheep that are host to drug-resistant worms. The dual practice of treating and quarantining purchased animals and delaying the move of treated stock to ‘clean’ pasture is now considered favourable practices in curbing the development and/or spread of resistance. Almost all farmers (93%) reported that incoming animals were treated with anthelmintic prior to mixing with the rest of the flock. Over 45% of farmers indicated that they usually moved lambs to ‘clean’ grazing after dosing (49% non-TET, 54% TET). This is now considered highly selective for resistance.

Clearly, anthelmintics play a dominant role in controlling parasites. The evidence indicates that there is a need for a greater appreciation of the principles that inform the sustainable use

of anthelmintics. A combination of practices such as excessive treatment frequency and sub-optimal dosing (as shown on some farms) does suggest there is a likelihood that a substantial proportion of lambs are not being dosed effectively. These departures from 'best' dosing practices will increase the risk of selecting worm populations that are resistant to anthelmintics. As successful worm treatment relies on effective on-farm management practices, it is vitally important that the basics in 'best' treatment practice are not compromised. It is imperative that practices that preserve anthelmintic efficacy are incorporated in helminth treatment programmes on each farm.

A follow-up study was undertaken to investigate the prevalence of anthelmintic resistance on these farms. Analyses of these data are ongoing, but preliminary results from the anthelmintic resistance tests (over 60 flocks) indicate there was a high incidence of resistance to benzimidazoles (70% of farms) while over 20% of farms had evidence of resistance to levamisole.

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RMIS No. 5253

Examination of the effects of DRB1 Alleles on FEC measurements

As shown in previous research reports Suffolk and Texels show a large difference in parasite resistance. The overall objective of this study was to examine the reasons why this disparity between breeds exists both at a genetic and immunological level. On the basis of previous analysis it was shown that in the Suffolk breed variation in exon 2 of the DRB1 gene in the major histocompatibility complex was significantly associated with faecal egg count (FEC). Detailed genetic analysis of the alleles at this locus showed that one allele (Ovar-DRB1*0203) was associated with reduced FEC while two other alleles (OAMHC213, Ovar-DRB10) were associated with increased FEC relative to the most frequent allele (DRB1*03411) in a purebred Suffolk population.

Crossbred progeny from Suffolk rams, carrying either the low FEC allele (Ovar-DRB33), or one of the high FEC alleles (OAMHC213) and the reference allele (DRB1*03411), were produced in 2004. Lambs were genotyped and lambs with the appropriate genotypes were selected for a post weaning study of FEC. The lambs were involved in a grazing trial at Athenry during the period from about 3 weeks of age until drafted for sale. As lambs from this trial, with suitable genotypes at DRB1 locus (DRB33, DRB3411, MHC213), were classified as fit for slaughter they were retained, amalgamated as one of two groups subject to the time of their availability. The first group were sampled and dosed according to manufacturer's recommendations (Oramec, Merial Animal Health Ltd) on 12th August. The second group were sampled and dosed (same product and procedures as earlier) on the 6th Sept and were placed in the same paddocks as the first group. Groups grazed for an 8-week period to provide a measure of FEC. The effectiveness of the anthelmintic treatment was tested on a composite faecal sample from the flock using FEC-PAC methodology at 10 to 12 days post-dosing. FEC was determined from individual lambs using McMaster methodology on 3 occasions, starting at a minimum of 35 days post-dosing. Results are presented in Table 112.

Results from the post-treatment efficacy check revealed that the anthelmintic was effective (both groups). The differences among grazing groups and dates were highly significant. The repeatability of FEC measurements was 0.7.

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Table 112: Allele effect on FEC

Allele from sire	Effect conferred by allele With respect to resistance shown in previous study	Number of lambs	Number of FEC measurements	Estimate of $\ln(\text{FEC}_{\text{OT}})$	s.e.
DRB33	Increased resistance: decreased FEC	27	80	6.55 (674)	0.189
DRB3411	Reference allele	48	136	6.84 (909)	0.145
MHC213	Decreased resistance: increased FEC	30	90	6.79 (864)	0.182

The test of difference among the DRB1 alleles was not significant ($P= 0.45$). The difference between DRB33 and the other two alleles was associated with a P-value of 0.23. Inclusion of the other allele (based on length only), inherited from the dam in the model did not change the estimates to any important degree. The results are not conclusive as the number of lambs per genotype was on the low side for the two most interesting genotypes (DRB33 & MHC213) The results confirmed the ranking of the alleles with respect to FEC in the purebred Suffolks but the numerical difference in FEC was not statistically significant, although it represented a 30% reduction in FEC. Further evaluation of the role of exon 2 of DRB1 gene is warranted to establish definitively the role of these alleles.

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RMIS No. 4927

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Treatment on the basis of various faecal egg count levels, examining the impact on lamb weight pre – and post- weaning

Faecal egg counts are an indirect measure of host response to parasite infection. The level of challenge combined with the host experience of infection will affect the number of worms that establish and the faecal egg count observed. The use of on-farm DIY technology (FECPAK) for determining faecal egg counts at farm level is an effective tool that can be used as an aid to sustainable parasite control management. However, this requires rules for relating observed faecal egg count to a decision to treat with anthelmintics. The aims of the following experiments were to examine the impact of operating within a range of FEC values on subsequent lamb performance i.e. weight gain, pre and post weaning.

Pre-weaning experiment

Twin lambs and their undosed ewes were co-grazed and set stocked on permanent pasture for the pre-weaning period. One lamb in each litter was randomly assigned to one of two treatment groups while the remaining twin was assigned to a ‘control’ group. Lambs in the control group were which suppressively treated to diminish the impact of parasitism. At 5 weeks the control lambs received an anthelmintic treatment that offered residual protection against *Teladorsagia* larval establishment (Moxidectin; Cydectin 0.1% Oral Drench for Sheep, Wlelehan Animal Health) and additional treatments (Levamisole; Chanaverm, Chanelle Pharmaceuticals Ltd) were given at approximately 2-week intervals to diminish the harmful effects of a potential *Nematodirus* challenge. The other twin was randomly assigned to either of 2 treatment groups. Following their anthelmintic treatment at 5 weeks (Chanaverm, Chanelle Pharmaceuticals Ltd), any subsequent treatment with Levamisole (Chanaverm, Chanelle Pharmaceuticals Ltd) was dependent on total faecal egg count (FEC) reaching either of 2 threshold levels ((900 e.p.g. or 1800 e.p.g). All drugs administered were given according to manufacturer’s instructions. Lamb weights were taken at 5, 10 and 14 weeks of age and FEC measurements monitored by pooling individual samples and using FECPAK methodology. FEC was monitored on a weekly basis except when a treatment

event occurred whereupon a period of 2 weeks was left before FEC was determined again. All lambs were treated with levamisole at weaning (14 weeks of age) and faecal sampled according to pre-treatment weaning group one month later (25/07/03).

The effect of group (control / '900' / '1800') and treatment frequency on lamb weight (adjusted) at 14 weeks and weight difference between 5 and 14 weeks of age were examined using GLM procedures of SAS.

Results are presented in Tables 113 and 114. The control group received 3 anthelmintic treatments while the 900 and 1800 group received 2 treatments preweaning. Neither treatment group nor treatment frequency effected lamb weight at 14 weeks or weight difference observed in lambs between 5 and 14 weeks of age. The higher FEC observed in the 'control' group compared to the treatment groups one month post-weaning may reflect a difference in immune status of this suppressively treated group compared to the other treatment groups (Table 115). However when the FEC was examined in a subgroup of these lambs, which remained untreated, this trend was not apparent (Table 116).

Table 113: Faecal egg count by age for each anthelmintic treatment group

Date	Age (wks)	Treatment Group			Action taken
		Control (n=54)	900 group (n=30)	1800 group (n=33)	
23/04	5		140 (90, 50)*		Control group treated: Moxidectin '900' group treated: Levamisole '1800' group treated: Levamisole
29/04	6	ND^		ND	
05/05	7	ND		ND	Controls treated: Levamisole
12/05	8	0 (0, 0)		270 (45, 225)	
19/05	9	0 (0, 0)		510 (75, 435)	
26/05	10	300 (300, 0)		975 (300, 675)	Control group treated: Moxidectin '900' group treated: Levamisole
02/06	11	ND	ND	1815 (1125, 690)	'1800' group treated: Levamisole
09/06	12	0 (0, 0)	0 (0, 0)	ND	
16/06	13	0 (0, 0)	45 (0, 45)	15 (0, 15)	
23/06	14	45 (0, 45)	285 (0, 285)	105 (15,90)	Control group treated: Levamisole '900' group treated: Levamisole '1800' group treated: Levamisole
25/07	18	735 (0, 735)	300 (0, 300)	255 (0, 255)	

* Figures in parenthesis correspond to Nematodirus FEC, other trichostrongyle FEC

^ ND = FEC not determined

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Table 114: Effect of pre-weaning treatment on live weight

	Treatment Group Weight (kg)			Significance (P value)
	Control (n=54)	900 (n=30)	1800 (n=33)	
Weight at 14 weeks of age (kg)	32.8 (0.50)*	31.7 (0.67)	32.7 (0.64)	NS
Weight change between 5 and 14 weeks of age (kg)	16.3 (0.48)	16.0 (0.64)	16.8 (0.61)	NS

* \pm s.e.

Table 115: Post-weaning FEC in lambs with different pre-weaning treatment history but who remained untreated post-weaning

Date	Age (wks)	Pre Weaning Treatment Group		
		Control (N=20)	900 group (N=10)	1800 group (N=8)
12/08	21	975 (60,915)*	750 (15,735)	1150 (20,1130)
03/09	24	1395 (30,1365)	1710 (90, 1620)	1140 (0, 1140)

* Figures in parenthesis correspond to *Nematodirus* FEC, other trichostrongyle FEC

Post-weaning experiment

One month post-weaning (and anthelmintic treatment) co-grazed lambs in two flocks (Flock A: cross-bred lambs, Flock B: pure-bred lambs) were randomly assigned to 3 treatment groups: control (supressively treated) '900' and '1800'. A pooled faecal sample for FEC determination was taken and all lambs were weighed. Lambs in the control group were administered with an oral macrocyclic lactone with residual activity (moxidectin; Cydectin, Wlelehan Animal Health) at the beginning of the trial. The '900' and '1800' groups were administered levamisole (Chanaverm, Chanelle Pharmaceuticals Ltd) when FEC was >900 e.p.g. and 1800 e.p.g, respectively. All drugs administered were given according to manufacturer's recommendations. The duration of the trial was 6 weeks and all lambs were weighed at the end of the trial.

The effect of group (control / '900' / '1800') and flock group (Flock A or flock B) on initial weight (late July) and final weight (early September) and the weight difference using GLM procedures on SAS.

Results are presented in Tables 117 to 118. Flock total FEC at the beginning of the trial in Flocks A and B was 430 and 240 e.p.g., respectively. *Nematodirus* was only observed in flock B (30 e.p.g.). In both flocks, the '900' group was treated once with levamisole. The '1800' group in both flocks remained untreated for the duration of the trial (total FEC remained below 1800 e.p.g.). The weight of the control animals at the beginning of the trial was significantly higher than the '900' group ($P < 0.05$) but not the '1800' group. The observed bias in the control weight was unexpected given the fact that animals were assigned randomly

to their groups *via* random numbers procedures. Neither flock group nor the interaction between flock group and treatment were significant on the initial weight. Treatment and flock group had a significant effect on lamb weight observed at the end of the trial. There was a significant difference between the control and the other two treatment groups ‘900’ / ‘1800’ group ($P < 0.05$, $P < 0.005$, respectively).

Treatment and flock group were significant effects on the weight change observed (final weight minus initial weight). The interaction (treatment* flock group) term was not significant. There was a significant difference between the control and ‘1800’ group ($P < 0.01$) but not between the control and ‘900’ group. The difference between the ‘900’ and ‘1800’ group approached significance ($P = 0.053$). These data indicated that post-weaning, one treatment is necessary and a FEC of 900 e.p.g. seems an appropriate threshold / trigger count to prompt an anthelmintic treatment action.

Table 116: Post-weaning lamb FEC (*Nematodirus*, other trichostrongyles) results and timing of anthelmintic treatments

Date	Flock A (N=110)			Date	Flock B (N=130)		
	Control (N=37)	900 group (N=34)	1800 group (N=39)		Control (N=43)	900 group (N=43)	1800 group (N=44)
30/07		570 (0, 570)	810 (0,810)	29/07	N.D	165 (0, 165)	90 (15, 75)
05/08	0	855 (0, 855)	1050 (15, 1035)	06/08	0 (0,0)	330 (45, 285)	300 (0, 300)
12/08	0	960 * (0, 960)	855 (0, 855)	13/08	0 (0,0)	675 (45, 630)	495 (0, 495)
19/08	15	N.D.	555 (30, 525)	21/08	225 (15, 210)	1485** (45, 1440)	1365 (135,
26/08	60 (40, 15)	15 (0, 15)	1110 (45, 1065)	27/08	345 (15, 330)	N.D	1440 (45, 1395)
03/09	255 (60, 195)	75 (0, 75)	1350 (60, 1290)	02/09	390 (15, 375)	75 (0,75)	1290 (15, 1275)
	N.D	N.D	N.D	09/09	510 (30, 480)	315 (0, 315)	1470 (60, 1410)

* ‘900’group, Flock A, dosed ** 900’group, Flock B, dosed ^ ND = FEC not determined

The effect of total number of treatments (pre- and post-weaning) administered to lambs in Flock A on final live weight (early September) was examined using GLM procedures on SAS. The number of treatments had no effect on final live weight (Table 119).

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Table 117: Post-weaning lamb weights by flock and treatment groups

	Flock A			Flock B		
	Control (N=37)	'900' group (N=34)	'1800' group (N=39)	Control (N=43)	'900' group (N=43)	'1800' group (N=44)
Initial Weight	36.9 (1.25)	34.9 (1.28)	35.9 (1.18)	36.1 (0.55)	35.1 (0.55)	34.8 (0.54)
Final Weight	42.9 (0.70)	40.2 (0.73)	40.4 (0.69)	43.7 (0.66)	43.0 (0.66)	41.8 (0.65)
Weight change (kg)	6.3 (0.36)	5.6 (0.38)	4.8 (0.36)	7.8 (0.34)	8.0 (0.34)	7.2 (0.33)

Table 118: Significant (probability) effects of treatment and flock on live weight

Variable	Treatment	Flock	Treatment *Flock
Initial weight	<0.05	NS	NS
Final weight	<0.01	<0.01	NS
Weight difference	<0.01	<0.0001	NS

Table 119: Effect of total number of treatments pre & post –weaning on final live weight

Flock ID	n	Total No. of Treatments	Final live weight (early Sept)
Flock A	18	3	40.5 (1.05)*
	60	4	41.0 (0.53)
	31	5	42.0 (0.74)

* ± s.e.

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RMIS No.4928

Effect of year-round grazing on grass supply and flock productivity

Previous results on extended grazing showed that a grazing season of 330 days was attainable by the application of grass budgeting principles (*see 2000 Research Report, Athenry Research Centre*). Following this development it was decided, in 2001, to investigate whether extended grazing could be applied also for the management of ewes in late pregnancy and during outdoor lambing. The objectives were:

1. To develop a feed demand and grass supply profile for the management of year-round grazing
2. To compare ewe productivity and lamb performance in flocks managed either on year-round grazing or on a standard system of grazing combined with silage feeding and housing
3. To determine whether extended grazing affects nitrate-N concentrations in ground water or has impact on soil structure or overland flow.

The pooled results for the years 2001 to 2004 are summarised in this report.

Two systems: A flock of 290 Belclare x Cheviot ewes was subdivided and managed in two self-contained farmlets as follows: (1) Grazing/silage/housing system, lambing date March 21, 9.6 ha Perennial ryegrass/clover pasture stocked at 14 ewes per ha, silage budget 0.6 per ewe (GS), (2) Year-round grazing system, lambing date April 1, 15.0 ha Perennial ryegrass pasture stocked at 10 ewes per ha, no silage budget (YRG)

Grassland management:

Belclare x Cheviot ewes were mated with Texel rams and the lambing dates chosen for the GS and YRG systems were March 21 and April 1, respectively. The ewes were joined with the rams in single sire mating groups for 6 weeks in October/November. The GS flock was housed on December 9 and offered silage *ad libitum* supplemented with concentrates in the final 6 weeks pre-lambing commencing at 200 g/ewe/day and increased to 600 g for the last 2 weeks. Surplus grass in autumn was one of the consequences of the low stocking rate in the YRG system and this surplus was manipulated for the practise of extended grazing. An area of 7.2 ha was closed in mid September, dressed with 33 kg N per ha and the resulting grass growth was reserved for winter grazing. Consequently, the stocking rate in this system during the breeding season was high, i.e. 20 ewes per ha. Extended grazing was commenced on December 7 and continued until March 7. A daily allowance of 1 kg grass DM per ewe was offered to the flock by block grazing in daily shifts. This allowance was increased to 1.3 kg DM on February 4 for late pregnancy and was supplemented with concentrates commencing at 250 g/ewe/day and increased to 400 g for 4 weeks pre-lambing.

The GS flock lambed indoor and the ewes plus lambs were turned out to pasture at 1 to 3 days post-lambing. To facilitate outdoor lambing in the YRG system, a six-paddock management plan was drawn up. The flock was scanned for litter size on day 80. In early February three paddocks including two used for extended grazing in December/January were dressed with 45 kg N per ha, closed and reserved for lambing. The YRG flock was removed off block grazing at 2 weeks pre-lambing, divided into single, twin and triplet bearing groups and transferred to the three lambing paddocks where they were set stocked and managed separately for lambing. The remaining three paddocks in this farmlet system were then dressed with 45 kg N per ha and reserved for grazing post-lambing. Daily supervision during lambing commenced at 6 am and continued until 10 pm indoor and 9 pm (nightfall) outdoor. Ewes considered by the stockman to require assistance at lambing were given assistance. Triplet bearing ewes lambing outdoor were housed for 2 to 3 days post-lambing for assessing their milking ability to rear triplet sets.

Following lambing, six paddock rotational grazing was practised on both farmlets. All lambs were offered creep feed, 250 g/lamb/day, from 10 weeks of age until slaughter. Lambs were weaned at 14 weeks of age and drafted for slaughter following weighing and handling for a standard degree of finish. Light lambs remaining in late September were removed off pasture and finished on a catch crop of rape supplemented with 220 g concentrates/lamb/day.

Fertilizer nitrogen:

Fertilizer N inputs were relatively low in both systems. The grass/clover pastures in the GS system had a high content of clover, e.g. 190 g per kg of pasture DM; hence no fertilizer N was applied during the main grazing season. The annual N input in this system averaged 79 kg N per ha applied in two dressings, i.e. for early grass and for silage. N applications in the YRG system consisted of strategic dressings for early grass and for autumn-saved pasture in September amounting to an annual input of 97 kg N per ha annually.

Measurements:

Feed demand was estimated from a review of published research results on grass intake by ewes and lambs together with published data on the feed requirements of ewes for target

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condition score at critical stages of the annual production cycle. Allowance was made in both systems for the following factors: flock turnout in the GS system was gradual and there was a time lag of 2 weeks post-lambing before the full stocking rate was completed; the number of lambs reared per ewe joined was 1.76 and the distribution of single, twin and triplet bearing ewes categorised by rearing type was 20%, 65% and 15% respectively. The data on grass intake were interpreted as indicators of feed demand by translating them into a practical feed demand profile that allowed for the changing feed requirements of ewes and lambs during the grazing season and expressed per ha for the stocking rate applicable to each system.

Grass supply was estimated by visual assessment of grass DM per ha made fortnightly by two experienced observers, supported periodically by grass clips taken at ground level from a 0.25 m² quadrat followed by drying and weighing. Sward height (SH) measurements were made fortnightly during the grazing season and used as a management tool to maintain pasture quality and to minimise stemminess. Flock performance information was collected on ewe liveweight, condition score, reproductive performance, lamb mortality, growth rate, carcass weight and age at slaughter.

Feed demand estimates are shown in Figure 36. GS feed demand increased rapidly to 80 kg DM/ha/day at 8 weeks post-lambing and to 95 kg ha/day prior to weaning on July 1. In contrast, YRG feed demand was relatively low due to the low stocking rate. Following weaning, there was a marked decline in feed demand in both systems as ewes were grazed tightly in order to facilitate the process of drying off. Thereafter, feed demand was reduced progressively as lambs were drafted for sale. Following mating, feed demand remained unchanged for the remainder of the grazing season.

Grass supply:

Results on the average supply of grass available for grazing during the four years under review are shown in Figure 37. Due to silage conservation, 2.6 ha on the GS farmlet were unavailable for grazing from April 1 to July 1 and, hence, the grass supply on this area was excluded from the results until grazing was resumed in July. The stocking rate on the grazing area during this period was 19.4 ewes per ha. Grass surpluses arose in the YRG system in June and July due to the low stocking rate and the absence of pasture closure for silage. On average over the 4 years of the trial these surpluses amounted to 25 t DM and were harvested for silage. Grass surpluses arose again in autumn and were used strategically for extended grazing as already stated. In early winter grass supply in the GS system declined rapidly as the farmlet was grazed off prior to housing while grass supply in the YRG system increased as a result of grass accumulation for extended grazing.

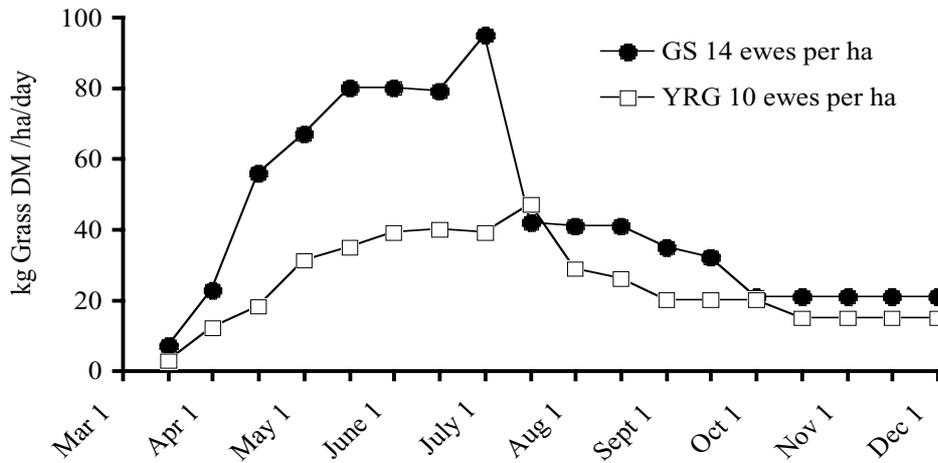


Figure 36. Feed demand (kgDM/ha/day) from March to December

Regular estimation of feed demand and grass supply and grazing capacity of the pastures facilitated flexibility in grazing management whereby adjustments were made on the basis of fortnightly assessments of the feed status of the farm. Application of grass budgeting principles proved effective for devising management strategies to cope with grass surpluses or deficits.

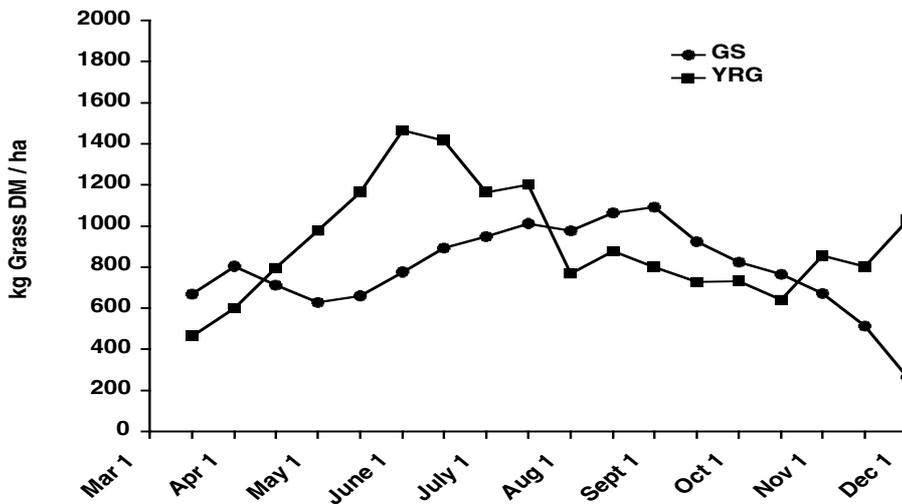


Figure 37. Grass supply on grazing area

Ewe liveweight:

Results on ewe liveweight and body condition score recorded during the critical stages of the annual production cycle are shown in Table 120. Although significant differences were recorded, in practice these differences were small. There were significant flock x year interactions.

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Table 120: Ewe liveweight and body condition: Pooled results 2001/02/03/04

	Liveweight (kg)			Body condition score		
	GS	YRG	Sig	GS	YRG	Sig
Pre-mating	68.0	69.9	***	3.43	3.47	NS
Post-mating	70.7	70.9	NS	3.59	3.61	NS
Mid pregnancy	74.2	70.7	***	3.47	3.51	NS
Lambing	-	-	-	3.18	3.41	***
5 wks post-lamb.	67.7	73.7	***	2.96	3.27	***
Weaning	69.1	71.2	***	3.23	3.28	NS

Ewe productivity:

Results on ewe reproductive performance and lamb birthweight are shown in Table 121. The data provide important assessments on the adequacy of feeding and management for lambing. Ewe productivity was high in both systems due to the high litter size of Belclare x Cheviot ewes. Lamb birth weight was significantly higher in the YRG system for all litter size classes.

Lamb survival rate:

Results on lamb survival rate are shown in Table 122. The results should be assessed in the context of the high litter size recorded in both systems. The distribution of birth types was 10% singles, 50% twins and 40% triplets/quads. The incidence of dead born lambs in the outdoor lambing system was relatively high, resulting in a lower survival rate compared with indoor lambing. Dead lambs were submitted for veterinary examination and causes of mortality were identified.

Table 121: Ewe reproductive performance and lamb birth weight (kg)

	System					
	GS			YRG		
No. ewes to ram 2001/02/03/04	541			629		
Ewes lambing (%)	93			94		
Litter size	2.13			2.18		
No. lambs reared/ewe joined	1.76			1.76		
Birth type:	No. lambs	Birth wt.		No. lambs	Birth wt.	Sig
Single	87	5.2		89	6.0	***
Twin	566	4.2		618	4.9	***
Triplets/quads	385	3.2		516	3.9	***

The results are shown in Table 123. It is evident that death in utero and difficult birth were the principal causes of dead-born lambs. Death in utero was associated with Toxoplasmosis infections in 2002 and 2003 identified at the Regional Veterinary Laboratory, Kilkenny.

Table 122: Lamb survival rate: Pooled results 2001/02/03/04

Lambing system	GS Indoor	YRG Outdoor
No. lambs born dead and alive	1075	1290
Lambs born dead (%)	5.5	11.0
Lambs died 0-10 weeks (%)	6.2	3.7
Lambs reared as per cent of all lambs born (%)	88.7	85.6

Table 123: Causes of lamb mortality

Lambing system	Indoor	Outdoor
No. lambs born dead	62	142
<u>Causes:</u>		
Abortions	6	12
Premature	3	1
Death in utero	29	51
Stillbirths	5	26
Difficult birth	10	43
Prolapse	3	2
Congenital deformity	2	2
Unknown	4	5
No. lambs died birth to 5 weeks	60	43
<u>Causes:</u>		
Congenital defect	1	2
Starvation/no suck	4	4
Hypothermia	3	8
Predation	1	5
Disease: E.coli	6	4
Gastritis	5	3
Pneumonia	7	6
Pulpy kidney	1	0
Accidents	27	10

Lamb performance:

Comparative results on lamb performance and output are shown in Table 124. With the exception of carcass weight, the differences in the components of lamb performance were highly significant. The lower growth rate recorded in the GS system at 5 weeks of age coincided with a rise in faecal egg count to 1850 epg. The effect of extensification on carcass output per ha was evident, i.e. almost 30% reduction in the YRG system compared with the GS system.

Table 124: Lamb performance and output

	System		Sig
	GS	YRG	
Ewes/ha	14	10	
No. lambs reared/ewe joined	1.76	1.76	
Growth rate(g/day):			
Birth to 5 weeks	267	291	P<0.001
5 to 10 weeks	241	278	P<0.001
10 to 14 weeks	231	214	P<0.001
Carcass wt. (kg)	18.9	18.8	NS
Age (days)	164	152	P<0.001
Lamb carcass output/ewe (kg)	33.3	34.1	
Lamb carcass output/ha (kg)	465	331	

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Environmental studies related to winter grazing

Questions were raised on whether extended grazing has environmental impacts, specifically on nitrate-N leaching, soil displacement and overland flow. Studies on these issues were commenced in 2001 by: (a) inserting ceramic cups at depths of 0.5 m, 1.0 m and 1.5 m in grazed and ungrazed plots for water sample extractions and measurements; (b) installing a soil detachment tray to collect splashed soil following rainfall; (c) using surface flow indicators to examine overland flow. Results on nitrate-N concentrations recorded in January and February 2004 are shown in Table 125.

The values were all lower than the EU Guideline of 5.65 mg/l. The EU Nitrate Directive threshold is 11.3 mg/l. Earlier results were presented in Research Report 2002, including results which showed no evidence of adverse impacts on soil structure and overland flow. Extended grazing did not pose a risk of sedimentation and enrichment of surface waters. Where a grazed paddock adjoins a watercourse, a simple precaution is to leave an ungrazed buffer zone between the grazed plot and the watercourse.

Table 125: Nitrate-N concentrations in 2004

Ceramic cup depth (m)	Mean NO ₃ -N concentrations (mg/l)		
	Plot 1 grazed	Plot 2 grazed	Plot 3 ungrazed
0.5	0	2.3	4.4
1.0	0	5.0	0.6
1.5	0.3	2.8	3.0

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RMIS No. 4925

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