Project number: 6378
Funding source: Teagasc and seed industry
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Project dates: Jan. 2013 – Dec. 2019

Grass and Clover Breeding

Key external stakeholders:

Grassland farmers, seed industry, forage breeders, advisors and trials evaluators.

Practical implications for stakeholders:

- Eleven new perennial ryegrass varieties, five new white clover varieties and the first ever Irish red clover variety were bred and commercialised.
- Genomic selection was incorporated in the breeding methodology to advance the rate and reduce the cost of genetic gain.
- Recommendations were made on the optimum experimental design, statistical analyses and resource allocation for perennial ryegrass variety yield trials to increase the efficiency and reduce the cost of yield estimates.

Main results:

Seventeen new varieties were awarded Recommended/National Listing in Ireland, UK and/or other countries. The new varieties consisted of: 11 perennial ryegrass (*Lolium perenne* L.) varieties including diploids and tetraploids, and intermediate and late maturity groups; 5 white clover (*Trifolium repens*) varieties ranging from small to medium to large leaf size; and the first ever Irish red clover (*Trifolium pratense*) variety.

Genomic selection was incorporated in the breeding programme for the first time. Three cycles of genomic selection in a tetraploid perennial ryegrass population were completed. The resulting varieties are currently under evaluation.

The breeding programme entered into a new partnership with Goldcrop Ltd., an Irish seeds and inputs company, to support the programme and ensure the timely commercialisation and release of new varieties.

An extensive data set of historical perennial ryegrass variety trials conducted over 11 years and five locations in Ireland was analysed, and recommendations made on the optimum experimental design, statistical analyses and resource allocation for perennial ryegrass variety yield trials.

Opportunity / Benefit:

This project provides farmers with new varieties of perennial ryegrass, white clover and red clover that offer greater genetic merit and improved agronomic performance thereby increasing the productivity, profitability and sustainability of grassland farms. A commercial agreement between Teagasc and the seed industry ensures the timely production and availability of seed for farmers, and financial support for the breeding programme. The research findings on the optimum methodology for the experimental design, statistical analyses and resource allocation for perennial ryegrass yield trials will increase the efficiency and reduce the cost of perennial ryegrass variety trials in Ireland.

Collaborating Institutions:

Department of Agriculture, Food and the Marine
US Department of Agriculture
Goldcrop Ltd.
DLF Seeds and Science
Germinal Holdings Ltd.

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1. Project background:

Grassland is Ireland's greatest renewable feed resource accounting for >90% of agricultural land and providing the main feed for ruminant livestock. Plant breeding is man-directed evolution. It is powerful, precise and predictable. Forage grass and clover have been subjected to very little formal breeding. Genetic variation and potential breeding gain remains high.

The Teagasc grass and clover breeding programme can positively influence the 3.9 million ha of grassland in Ireland by breeding improved varieties. A new, improved variety provides farmers with the raw material to improve the productivity, quality and sustainability and reduce the environmental footprint of Irish grassland.

Teagasc has a long and successful history of forage breeding. The commercial breeding programme was established in Oak Park, Carlow in the mid 1980's. This project continues to build on this work through advancements in methodology and technology.

2. Questions addressed by the project:

- How successful is forage breeding in producing improved varieties of perennial ryegrass, white clover and red clover?
- Can genomic selection be successfully incorporated into the commercial breeding programme?
- What is the optimum experimental design, statistical analyses and resource allocation for perennial ryegrass yield trials?

3. The experimental studies:

The breeding process consisted of a multistep and cyclic process where the best plants (genotypes) were evaluated, selected and intercrossed to produce a new variety. Plants were selected based on their individual performance, progeny performance or DNA (genomic selection). Traits measured and selected for included: seasonal and annual yield, nutritional value, leafiness, disease resistance, ground cover, persistency, and palatability and grass utilization under cattle grazing.

A new variety was produced by crossing in all possible combinations a number of selected plants. The new variety was then independently tested by the government agencies in the relevant country. Varieties with value for cultivation and use (VCU) and that were distinct from other varieties, uniform and stable (DUS) were added to the Recommended/National List. Commercial seed of Teagasc bred varieties are produced and sold under license by Goldcrop Ltd., DLF Seeds and Science and Germinal Holdings Ltd.

Resources are always limited in plant breeding programmes, limiting the number of plots or plants that can be evaluated. Therefore, the available resources must be used in the most efficient manner. An extensive data set of historical perennial ryegrass variety trials conducted over 11 years and five locations in Ireland by the Department of Agriculture, Food and the Marine was analysed. The information provided by these past trials was used to gain a greater understanding of the effects of trial design and analyses on the accuracy, precision and cost of the yield estimates, and identify opportunities and improvements for the design of future perennial ryegrass yield trials.

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4. Main results:

Seventeen new varieties were awarded Recommended/National Listing in Ireland, UK and/or other countries† and commercialised since 2013.

New Perennial Ryegrass Varieties

Intermediate tetraploid

Carraig	Ireland 2012, Northern Ireland 2013, England/Wales 2013, Scotland 2013
Elysium	Ireland 2018
Chatsworth	England/Wales 2020

Late diploid

Glenroyal	Ireland 2015
Kerry	Ireland 2016
Oakpark	Ireland 2019, England/Wales 2018
Smile	Ireland 2019, England/Wales 2018, Scotland 2017
Gleneagle	England/Wales 2019
Glenrock	Scotland 2019

Late tetraploid

Solas	Ireland 2015, England/Wales 2015
Glenfield	Ireland 2021

New White Clover Varieties

Small leaf size

Galway	Ireland 2017, England/Wales 2005
Coolfin	Ireland 2017, England/Wales 2019, France 2015, Germany 2015

Medium leaf size

Iona	Ireland 2014, Northern Ireland 2013, England/Wales 2011, Scotland 2011
Buddy	Ireland 2015, Northern Ireland 2015, England/Wales 2013, Scotland 2013

Large leaf size

Dublin	Ireland 2018, England/Wales 2015, Scotland 2016, France 2016

New Red Clover Variety

Fearga	Ireland 2019, England/Wales 2019, Scotland 2019

†Northern Ireland Recommended List ceased in 2016.

Genomic selection was successfully incorporated in the breeding programme. Varieties are now being selected based on their DNA. Genomic selection will increase the rate and reduce the cost of genetic gain.

Analysis of historical perennial ryegrass variety trials indicated that future perennial ryegrass trials with > 20 varieties should be based on modern designs that are capable of blocking in two directions to control spatial variability within field sites. Trials should be based on incomplete block designs such as α-designs or resolvable row-column designs using block sizes ranging from 6 to 20, depending on block shape. Each replicate of the design should be as square as possible. Square replicates are more efficient with lower residual variances compared to long and rectangular replicates. Increasing the number of replicates decreased residual variances but this effect was considerably smaller than improving blocking effects including size (number of varieties per block) and shape. Increasing plot size was not effective in increasing trial efficiency.

Spatial analyses should also be used to supplement the experimental design. Spatial analyses allows adjustment of variety means for spatial variation, improving the statistical precision of estimates. Analyses without adjustment for spatial variation often resulted in erroneous variety rankings in the presence of spatial variation. The correlated errors model was the most effective for making spatial adjustments to variety yields.

Resources are always limiting in evaluation trials. The effects of reducing the number of harvest years, locations and

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replicates on the relative efficiency of the trials, based on the average variance of genotypic difference, was determined. Of the five test locations compared, no one location was identified as particularly unique. Reducing the number of replicates per trial had the smallest predicted impact on relative efficiency, followed by reducing the number of locations. Reducing the number of harvest years, even for one location, resulted in a large loss in relative efficiency.

5. Opportunity/Benefit:

This project highlights the benefit of forage grass and clover breeding to grassland agriculture and the significant improvements in sward productivity and quality that can be achieved through breeding. The project offers farmers new improved varieties of perennial ryegrass, white clover and red clover of greater genetic merit that when sown may increase the productivity, profitability and sustainability of Irish farm systems. A commercial agreement between Teagasc and the seed industry ensures the timely commercialisation and release of new varieties.

Research on the optimum testing methodology for grass yield estimates will increase the rate and reduce the cost of the genetic gain in perennial ryegrass breeding and variety evaluation programmes by improving the precision and accuracy of the yield estimates, and optimising the use of available resources.

6. Dissemination:

The characteristics and agronomic performance data of commercial Teagasc varieties are annually updated and published in the relevant national Recommended Lists of Grass and Clover Varieties for Ireland, Northern Ireland, Scotland, England/Wales, France and Germany. In addition, variety information has been widely disseminated to farmers via discussion groups, open days, farm walks, popular press and Teagasc publications. An overview of the project was given at the Moorepark, Grange and Clonakilty Open Days.

Main publications:

Sripathi, R., Conaghan, P., Grogan, D. and Casler, M.D. (2018) Modeling genotype x environment correlation structures in long-term multi-location forage yield trials. *Crop Science* 58:1447-1457.
Sripathi, R., Conaghan, P., Grogan, D. and M.D. Casler, M.D. (2017) Spatial variability effects on precision and power of forage yield estimation. *Crop Science* 3:1383-1393.
Sripathi, R., Conaghan, P., Grogan, D. and Casler, M.D. (2017) Field design factors affecting the precision of ryegrass forage yield estimation. *Agronomy Journal* 109:858-869.

Popular publications:

Byrne, S.L., Milbourne, D. and Conaghan, P. (2018). Turbocharging perennial ryegrass breeding. TResearch 13 (3): 30-31.
Arojju, S.K., Conaghan, P., Barth, S., Milbourne,D., Casler, M.D., Hodkinson, T.R., Thibauld, M. and Byrne, S.L. (2018) Genomic prediction of crown rust resistance in *Lolium perenne. BMC Genetics* 19:35.
Byrne, S.L., Conaghan, P., Barth, S., Arojju, S.K., Casler, M.D. and Milbourne, D. (2017) Using variable importance measures to identify a small set of SNPs capable of predicting heading date in perennial ryegrass. *Scientific Reports* 7:3566.
Arojju, S.K., Barth, S., Milbourne,D,. Conaghan, P., Velmurugan, J., Hodkinson, T.R. and Byrne, S.L. (2016) Genome wide association study of heading and re-heading in perennial ryegrass full-sib families. *BMC Plant Biology* 16:160.
Sripathi R. (2016) Improving precision and modeling genotype x environment interactions of forage yield trials: A case study. Ph.D. Thesis. University of Wisconsin-Madison. 117 pages.
Conaghan, P. Red clover – agronomy and management (2016) In: *Guidelines for Successful Organic Beef Production*, pp. 30-33. Teagasc, Ireland.
Conaghan, P. (2016) Seeds of progress: The journey of grass varieties. Irish Grassland Association Quarterly Newsletter, Winter 2016, issue 34, page 31.

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