

**Project number:** 5972  
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## Probiotic lactobacilli survival and impact in the animal gut



### Key external stakeholders:

Animal feed manufacturers; thoroughbred racehorse industry, veterinary health professionals

### Practical implications for stakeholders:

- This project provides first time information on the microbial ecology of the equine, and other mammalian species gut.
- This project also provides information on commensal lactobacilli found in the gut microbiota of humans and animals.

### Main results:

- The project provided definitive genome-based evidence to support the fermentation patterns of sixteen strains of *Lactobacillus ruminis*, and has identified prebiotic carbohydrates with the potential to promote *L. ruminis* growth *in vivo*.
- This project identified the core faecal microbiota of ruminants, hindgut fermenters and mono-gastric animals co-localised to a single farm in Ireland.
- The project provided details for the first time, on the faecal microbiota of thoroughbred racehorses, both active and at rest.
- Analysis of the thoroughbred horse microbiota has revealed *Lactobacillus equi* to be a predominant *Lactobacillus* species in the hindgut. Genome analysis identified genes and enzymes highlighting *L. equi* adaptations to the herbivorous gastrointestinal tract of the horse, including fructan hydrolases.
- Having sequenced the genome of *Lactobacillus equi*, will help to further understand the microbial ecology of the equine hindgut and the influence lactobacilli have on it.

### Opportunity / Benefit:

The outcomes of this project is of relevance for the basic understanding of commensals/probiotics, potential mammalian applications, and potential alternatives to in-feed antibiotics for the animal production industry and generation of information of direct relevance for human probiotic consumption.

### Collaborating Institutions:

University College Cork

**Teagasc project team:** Prof. Paul Ross  
Dr. Michelle O' Donnell

**External collaborators:** Prof. Paul O'Toole, UCC  
Dr. Bridget Younge, UL

### 1. Project background:

Lactobacilli constitute a diverse genus of the Gram Positive bacteria found ubiquitously in nature and also within the gastrointestinal tract of humans and animals. At least twenty species appear to be the dominant Lactobacillus species in human faeces or the GI tract. They represent a wealth of potentially useful organisms and products for exploitation. Lactobacilli are often the target of dietary treatments like prebiotics as they are considered beneficial, commensal bacteria. Therefore, identification of the pathways involved in prebiotic utilization in an autochthonous member of the mammalian microbiota (*L. ruminis*) would greatly benefit the functional food sector.

The microbiota of both humans and animals in conjunction with dietary intake can have an effect on the health of the host. Many diseases including IBD, IBS and laminitis are thought to be caused by microbial dysbiosis within the gut. Elucidation of the herbivore microbiota would therefore be of benefit for animal husbandry and the economy.

### 2. Questions addressed by the project:

- What are the key properties of lactobacilli that promote survival in the gut, and what is the effect of introducing Lactobacillus on the gut microbiota?
- Can a combination of genome sequencing, comparative and functional genomics, and animal feeding trials identify gut survival and host interaction mechanisms?

### 3. The experimental studies:

In order to identify the factors involved in motility, prebiotic utilization and growth within *L. ruminis* we sequenced the genome of six strains from a variety of human and animal sources. The genomes of each were analysed and metabolic pathway maps were developed. A number of genes and operons were identified as being key to the utilisation of prebiotics (fructooligosaccharides, galactooligosaccharides and soybean-oligosaccharides).

This project was also interested in the effect that digestion type had on the microbiota of domesticated herbivores. In order to research this, fecal samples were collected from a commercial mini-farm which housed 10 different animal species. These 10 animal species could be divided into three digestion types – ruminant, hindgut fermenter or mono-gastric fermenter. Bacterial DNA was extracted from each sample and sent for pyrosequencing. Following bioinformatics analysis the resulting taxa were compared, in order to identify a core microbiota present in domesticated herbivores, regardless of digestion type.

### 4. Main results:

- Identification of the genetic basis for motility and carbohydrate utilization in *L. ruminis*.
- The first study to investigate the microbiota of the Irish Thoroughbred racehorse using next generation sequencing techniques.
- We determined the genome sequences of six strains of a commensal lactobacillus spp. *L. ruminis*.
- Analysis of the microbiota of Thoroughbred racehorses, miniature ponies, deer, donkeys, Kune-kune pigs, pygmy goats, llamas, alpaca, rabbits and chinchillas.

### 5. Opportunity/Benefit:

This project has highlighted the need for further research on the effect of diet on performance animals. It also forms a knowledge base for future studies into the microbiota of domesticated herbivores and its influence on health status.

## 6. Dissemination:

### Main publications:

- O'Donnell MM, Harris HMB, O'Toole PW, Ross RP. (2014) The Genome of the Predominant Equine Lactobacillus Species, *Lactobacillus equi*, Is Reflective of Its Lifestyle Adaptations to an Herbivorous Host. Genome Announc. 2014 Jan 16;2(1).
- O'Donnell MM, Harris HMB, Jeffreys IB, Claesson MJ, Younge B, O'Toole PW, Ross RP. (2013) Hindgut microbiome of the Irish Thoroughbred racehorses. Letters in Applied Microbiology, 57, 6, 2013, 492-501.
- O'Donnell MM, O'Toole PW, Ross RP. (2013) Catabolic flexibility of mammalian-associated lactobacilli. Microb Cell Fact. 2013 May 16;12:48.
- Riboulet-Bisson E, Sturme MH, Jeffery IB, O'Donnell MM, Neville BA, Forde BM, Claesson MJ, Harris H, Gardiner GE, Casey PG, Lawlor PG, O'Toole PW, Ross RP. (2012) Effect of Lactobacillus salivarius bacteriocin Abp118 on the mouse and pig intestinal microbiota. PLoS One. 2012;7(2).
- Neville, B. A., Forde, B. M., Claesson, M. J., Darby, T., Coghlan, A., Nally, K., Ross, R. P. & O' Toole, P. W. (2012). Characterization of pro-inflammatory flagellin proteins produced by Lactobacillus ruminis and related motile lactobacilli. PLOS One 7, e40592.
- Forde BM, Neville BA, O'Donnell MM, Riboulet-Bisson E, Claesson MJ, Coghlan A, Ross RP, O'Toole PW. (2011) Genome sequences and comparative genomics of two Lactobacillus ruminis strains from the bovine and human intestinal tracts. Microb Cell Fact. 2011 Aug 30;10 Suppl 1:S13.

PhD awarded to Michelle O'Donnell

### Popular publications:

#### *Betting on Bacteria*

Bernard Dixon, Microbes Magazine Animalcules section, March 2014

<https://www.microbemagazine.org/images/New%20Folder/Mar2014/znw00314000090.pdf>

## 7. Compiled by: Dr. Michelle O' Donnell