

## **Joint RuminOmics/Rumen Microbial Genomics Network Workshop, 22 June 2013**

### *Harmonization of techniques associated with ruminal genome, microbiome and metagenome analysis*

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The analysis of how the ruminal microbial community influences methanogenesis, one of the primary objectives of the EC Framework 7 consortium, RuminOmics, depends on a set of techniques that are used to characterize the genome, microbiome and metagenome. The outcome of the analysis often depends on the details of the techniques used. How should samples of ruminal digesta be taken? And when? How should they be stored? How should DNA be extracted? Following extraction, several methods are available for analysing the community, but are they all valid? How best can we use NGS to achieve our objectives in this respect? And how does metagenomic sequence data from massively parallel sequencing help us to analyse the structure and activity of the community? This workshop was organised as a satellite meeting to the main Greenhouse Gases in Animal Agriculture meeting being held in Dublin at the Bellfield campus of University College Dublin, and was sponsored by RuminOmics and the Rumen Microbial Genetics Network. Its aim was to explore the various techniques that are being used and, if possible, to make them consistent, in order to facilitate comparisons between the results of experiments carried out by the many international groups working on rumen microbial ecology as it relates to methane and N emissions from ruminant livestock production.

In the first section of the workshop, eleven of the most influential workers in the field addressed the audience of >70, provoking a lively discussion – perhaps not the 'harmonization' mentioned in the title, but at least information exchange and therefore a first step towards that goal. The speakers kindly agreed to submit summaries of their talks, which follow this introductory article.

The second phase of the workshop comprised three discussion groups, which were asked to discuss

- Cultivation methodologies
- Taxonomy
- The future role of the Rumen Microbial Genomics Network

The discussion leaders, John Wallace, Tim McAllister and Jamie Newbold, reported back. The conclusions were rather similar from each of the three break-out groups. All lamented that relatively few labs were actually carrying out cultivation and isolation work. They

welcomed, however, the Hungate 1000 initiative, and the strategies that had been initiated by Janssen and his colleagues in terms of the design of growth media, which have scarcely changed since the days of Hungate and Bryant, and the principle of 'dilution to extinction'. The need for phenotypic characterisation of the large number of bacteria that were known from community 16S rRNA genetic analysis was uppermost in people's minds. For example, which species dealt with xenobiotics and plant secondary compounds and how? Culture of methanogens was tricky, with few labs even attempting it, but was vital if the correct live species were to be used for vaccine development. Phenotypic measurements were vital to back up and to understand the large volume of sequence information that is now available. The discussion leaders concluded that most participants considered taxonomy of ruminal microorganisms to be a mess, partly because of the lack of good phenotypic information. They also found confusing the different depths of taxonomic analysis being presented in different studies. Very often it is difficult to compare different studies directly because their data are presented so differently. Could RuminOmics or the RMG take the lead in standardising methodologies or presentation standards? – which then led into conclusions about the broader role of the RMG and whether it should continue. Harmonisation of methods across the globe was probably unattainable (and perhaps not even desirable); a single repository for descriptions of the methods that are used in different labs would be useful, however - a living Wiki as it was described by one delegate. Could that be hosted by RMG? Or RuminOmics? Probably not, because of the limited funding span of both projects. So, therefore, where? was the unanswered question. Discussion concluded that the RMG had certainly proved itself internationally useful for its role in the Hungate 1000 and Global Rumen Census initiatives.

The last session was devoted to the work of the RMG. Once again, the speakers kindly agreed to provide summaries of their talks, which appear in the current volume. The day closed with a dinner hosted by RuminOmics at a local hotel.