



Annual Report 2015

NEW ZEALAND
AGRICULTURAL GREENHOUSE GAS
Research Centre

Leading Partners in Science



Cover image: Crystals of an essential methanogen marker protein from the inhibitor programme

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PROGRESS TOWARDS SOLUTIONS

Identifying mitigation solutions is a key component of the New Zealand Agricultural Greenhouse Gas Research Centre's (NZAGRC) Vision and Mission. The complexity of the problem means that identifying solutions is a long term goal. Successfully reducing greenhouse gas (GHG) emissions below a 1990 baseline within the New Zealand context of an expanding agricultural sector will require progress in both direct and indirect mitigation options. Direct mitigations are those solutions that reduce absolute emissions per unit of substrate (e.g. feed, nitrogen). Indirect mitigations are those that arise as a result of general improvements in the efficiency of production (for example by improved animal genetics and feeding practices which will reduce emissions per unit of product but not necessarily reduce absolute emissions).

It is important that the new knowledge developed in NZAGRC funded/co-funded research programmes is utilised in order to have a practical impact on the greenhouse gas emissions emitted from New Zealand agriculture. The table below highlights key some key outputs from 2014/15 and their envisaged impacts.

NZAGRC/PGgRc output	Expected impact
<p>The breeding work has found:</p> <ul style="list-style-type: none"> Differences in methane emissions between lines persist at pasture. Rumen microbial profiles are highly correlated with methane yield and contain a heritable and repeatable component suggesting that these profiles could be highly predictive of the methane status of an animal. Initial results indicating that moles of total gas produced may be a useful proxy for feed intake. 	<p>These results give important confirmation that methane differences are maintained at pasture and suggest that there could be an opportunity for more cost-effective identification and testing of large numbers of animals needed for the successful breeding of low emitting phenotypes.</p> <p>If moles of gas produced does turn out to be a good proxy for intake then the important question of whether there are differences between selection lines in the quantity of feed eaten at grazing can then be addressed.</p>
Feeding fodder beet at high levels of inclusion (90%) reduces methane emissions by 50%	Although preliminary this does indicate that fodder beet, which is an increasingly popular crop, could be a potent methane inhibitor if the effect is maintained over time and across a broad range of feeding situations.
Results from animal trials show that prototype vaccinations can produce high levels of methanogen-specific antibody in the saliva and rumen. Completion of methanogen genome sequencing projects in this and allied programmes allows better identification of universal vaccine targets.	More rapid identification of potent antigens for animal testing.
Five inhibitory compounds have been able to reduce methane production <i>in vivo</i> by up to 90% in two day animal trials. One compound reduced emissions by 26-16% in a sixteen day trial.	Passed proof of concept of the screening pipeline with promising compounds identified that can be developed further. Provides a good foundation for PGgRc commercialisation discussions with international partners.
A preliminary study in sheep indicating that if methanogenesis is inhibited, homoacetogenesis (the production of acetate from hydrogen and carbon dioxide) is unlikely to be a major sink for the hydrogen not converted to methane.	This result brings into question the idea that the suppression of methane formation would result in its replacement by acetate formation by different hydrogen-using microbes.

Plant genotype can influence nitrous oxide emissions with emissions varying between -58 and 120% of the median value. The lowest value came from the Italian ryegrass cultivar Moata and the highest from the brome cultivar Gala.	Provides important confirmation of laboratory studies indicating the role that plants could play in mitigating emissions from urine deposited onto pastures by grazing animals.
Soil carbon losses following cultivation at a Waikato site were not greater than losses following no-till and direct drilling treatments to re-establish the pasture sward. Rather, the soil water content conditions at the time of pasture renewal and duration between spraying and seedling emergence are more important drivers of changes in carbon storage.	The main implications for the farming community are that (a) the site preparation for pasture renewal is considerably less important than rapid re-establishment of the new sward and (b) selecting a critical time for the conversion when soil water content allows good pasture growth but when weather conditions minimise carbon losses.
Early results from the two-year comparison of changes in soil carbon storage under conventional ryegrass/clover and a mixed sward incorporating deep-rooting species show that the potential for net carbon uptake into the soil for the mixed sward is greater because of increased carbon inputs from roots. Preliminary findings on the water use efficiency of high and low diversity swards also show that the high diversity swards can produce equivalent amounts of biomass while using less water than a conventional ryegrass-clover sward.	Provides some encouragement for the idea that it may be possible to increase the quantity of carbon stored under New Zealand pastoral soils, in a win-win with more water and nutrient efficient pasture systems.

CHAIR'S REPORT

At the end of 2015, approximately 40,000 delegates will meet in Paris with the hope of finalising a new global agreement that will put in place actions to stabilise atmospheric concentrations of GHG. The international response to climate change began at the Rio Earth Summit in 1992. In 2015 COP21, also known as the 2015 Paris Climate Conference, will now aim to agree a universal approach to tackling climate change, with the broad aim of keeping global warming below 2°C above pre-industrial levels.

There is growing political and public pressure to develop ambitious targets for country-level GHG reductions with some concerns as to whether current pledges are consistent with the 2°C target. It is therefore important to put long term strategies to reduce GHGs into place now. New mitigation technologies will be valuable tools in the medium to long term.

In the case of New Zealand, the agriculture sector contributes 48% to the country's GHG emissions. Therefore, the sector will need to continue to play its part to help New Zealand meet any internationally agreed emissions reduction target. The emissions intensity of New Zealand agriculture, that is the gases generated per unit of meat or milk produced on farms, has declined on average by about 1% since at least 1990. However, the reduced emissions intensity has been more than offset by the increased overall product generated by the sector. As a result, New Zealand's total agricultural emissions have risen by 14%. Without the efficiency gains on farms, emissions would have grown much more, by almost 40%. So, while New Zealand farmers are already making a contribution and their efficiency gains are addressing a large portion of the problem, they are not enough to counter the extra GHGs being produced overall.

By continuing to improve on-farm efficiency and productivity, there is the opportunity to further reduce the intensity of emissions per unit product. However, this is unlikely to stop the country's total agricultural emissions from rising in the future given the growing global demand for high-value protein-rich food. Practical and cost-effective new and enhanced approaches to reducing agricultural GHG emissions are required to help meet environmental, social and international aspirations and obligations as well as economic growth targets. This is the role of the NZAGRC alongside the jointly industry/government-backed PGgRc. Our efforts are a prime example of Government, industry and researchers working together, combining resources to identify and develop additional interventions that will provide effective and practical results by 2020 and beyond.

Over the past two years the NZAGRC has critically evaluated all of its research programmes. The joint NZAGRC-PGgRc Methane programme was formally reviewed by an international science panel in April 2015 and has subsequently been updated. The NZAGRC Nitrous Oxide, Soil Carbon and Integrated Farm Systems programmes have been updated and contracted until June 2017, following extensive stakeholder consultation in 2013/14. A new three year programme that aims to develop lower emission systems for Māori farms started in July 2014.

The governance bodies of the NZAGRC and PGgRc continue to meet quarterly and there is now a strong drive towards engaging commercial partners for new methane mitigation technologies, the PGgRc will take the lead in this. A number of key science results in 2014/15 demonstrate that the science teams are getting closer to viable solutions to reduce agricultural GHGs.

Through its national and international roles and responsibilities, particularly through its active involvement in the Global Research Alliance on Agricultural Greenhouse Gases (GRA), the Centre continues to build on its reputation as an important source of clear and unbiased advice on the science behind agricultural greenhouse gases and their mitigation options.

Professor Warren McNabb
Chair of NZAGRC Steering Group
August 2015

NZAGRC DIRECTOR'S REPORT

It is hard to believe that in March 2015 it was five years since the NZAGRC was officially opened. In April 2015 we held our first joint conference with the PGgRc in Palmerston North. This was attended by one hundred and thirty GHG mitigation focussed scientists, policy makers and industry representatives. The day proved an excellent opportunity to reflect on the past five years and the roles that both science and policy play in the drive to reduce agriculture's environmental footprint in the face of climate change.

Working alongside MPI and the PGgRc, usable results, outputs and publications continue to emerge from our research. We keep a close eye on ensuring that the outcomes of our funding can be translated into practical solutions; in some areas, results have already reached the stage where engaging with potential commercial partners is a priority. In 2014/15 it was formally agreed that the PGgRc will lead the interaction with potential commercial partners with strong support from the NZAGRC. A commercialisation strategy is now in place for emerging new technologies in particular from the methane programme.

A key focus this year has been to re-evaluate and update the Centre and PGgRc's co-investment in the methane mitigation space for the period 1 July 2015 to 30 June 2019. This has involved significant input from NZAGRC Principal Investigators, science teams, the International Science Advisory Panel (ISAG) and the NZAGRC Steering Group and PGgRc Board who provided review and feedback. I would like to thank everyone that has contributed to this process. I am happy that the new work plans are both scientifically rigorous and highly targeted towards solutions and look forward to continued outputs emerging from these updated programmes.

A three year NZAGRC-funded project that aims to assist the Māori pastoral sector to improve its collective capacity to increase resource efficiency and farm productivity while lowering emissions started on 1 July 2014. This project allows us to ensure that our research is applicable to all sectors of New Zealand society and will also provide tangible knowledge transfer materials that can be used by our Member organisations and industry partners. The project is progressing well with 29 farms engaged and a research paper detailing the first stages of the project ready for submission.

We continue to work collaboratively with the PGgRc, MPI and a wide range of national and international organisations. The Centre's role in administering GRA funding on behalf of MPI ensures excellent coordination of the New Zealand research programme with international efforts.

Highlights for the Centre staff this year include the development of a collaborative project with the Food and Agricultural Organisation of the United Nations (FAO) and the formal conclusion of the most recent assessment by the Intergovernmental Panel on Climate Change (IPCC) in which both myself and NZAGRC Deputy Director Andy Reisinger have played significant roles. At an operational level, we ended 2014/15 with the Centre Administrator role vacant and have recruited a new person to start in September 2015. Victoria Hatton, the Operations Manager (International) has been based in Rome since January 2015, working as the project coordinator of the joint project we have with the FAO.

I would like to express my thanks to all of our Advisory Groups, especially the International Science Advisory Group for their input into the methane review, conference and associated workshops. The Steering Group continue to be exceptionally dedicated to the Centre and have provided valuable and knowledgeable advice throughout the last year.

Dr Harry Clark
NZAGRC Director
August 2015

THE NEW ZEALAND AGRICULTURAL GREENHOUSE GAS RESEARCH CENTRE

The NZAGRC is 100% government-funded by the Ministry for Primary Industries through its Primary Growth Partnership Fund. It is a core component of the New Zealand Government's approach for addressing the reduction of greenhouse gas emissions from agriculture. This includes New Zealand becoming: (a) a major investor in agricultural GHG mitigation research; (b) a world leader in finding solutions to agricultural GHG emissions via its domestic investment programme; and (c) a leader in international initiatives to advance the search for mitigation solutions and help ensure international treaties address agricultural GHG emissions in an appropriate manner. The Centre is a science funder, has additional responsibilities for strategic research coordination, capacity building and leads New Zealand science input into international activities and policy processes in the agricultural GHG area.

The NZAGRC is a partnership between the leading New Zealand research providers working in the agricultural GHG area and the PGgRc. About NZ\$48.5 million is being invested by the NZAGRC into research and development activities over ten years. The NZAGRC is a "virtual" Centre and the research that it funds is carried out by researchers working in their own organisations and collaborating across organisations.

NZAGRC is not the only significant investor into agricultural GHG mitigation research in New Zealand. Much of NZAGRC methane research builds on research investments made by the PGgRc, and since 2013 the NZAGRC and PGgRc investments have been formally aligned. This involves a single research strategy with shared advisory groups and administrative processes. Targeted mitigation research and proof-of-concept trials are also carried out under the Sustainable Land Management and Adaptation to Climate Change (SLMACC) programme coordinated by MPI. In addition, the New Zealand government provides funding for projects that support the goals and objectives of the Global Research Alliance, which build on and extend New Zealand-based research through international collaboration and data sharing. Various investments by industry into on-farm tools and trials and extension complete the picture. Research investment by NZAGRC within this funding landscape is based on an assessment of national needs and priorities, existing knowledge and expertise, and major gaps.

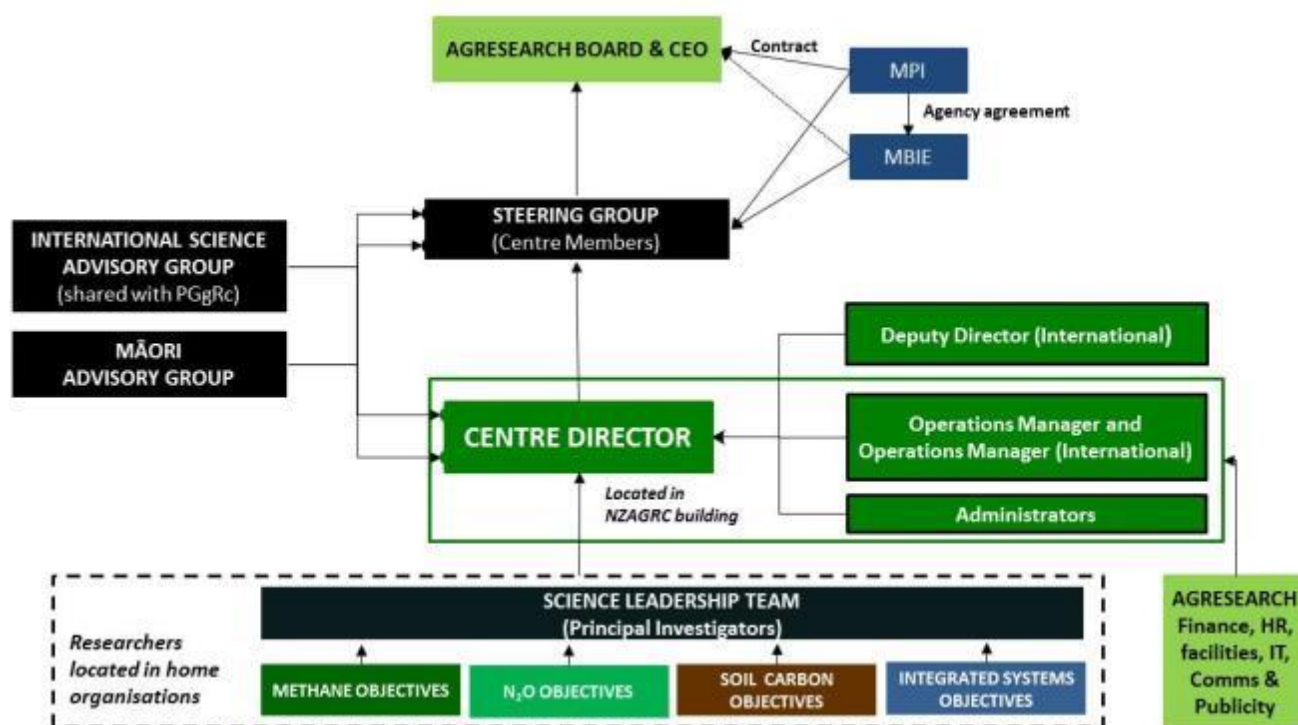
The NZAGRC is physically located on the AgResearch Grasslands Campus in Palmerston North. The Director, Operations Manager, Operations Manager (International), Project Analyst and Administrator are employed by AgResearch on behalf of the NZAGRC and are based in this building. The Deputy Director (International), also employed by AgResearch, is located in Wellington. The Operations Manager (International) has been sharing her time between the NZAGRC and the Food and Agriculture Organization of the United Nations (FAO) in Rome since January 2015, taking up a full-time secondment from July 2015 until at least March 2016.



NZAGRC GOVERNANCE

As the NZAGRC is set up as a unit operating within AgResearch, the Board and Chief Executive (CEO) of AgResearch have ultimate responsibility for the NZAGRC. However, a Steering Group (SG) comprising a representative of each NZAGRC Member provides advice and recommendations to the AgResearch CEO and Board on the operation of the NZAGRC. The NZAGRC Director reports to the AgResearch CEO and Board via the NZAGRC's SG. The International Science Advisory Group (ISAG) monitors, advises and reports on the NZAGRC's science quality and direction to the SG and NZAGRC Director. The NZAGRC SG meets formally with the PGgRc Board every six months and this provides guidance in relation to the needs of the industries that are intended to take up its research outcomes. The advisory roles of the ISAG and PGgRc Board are primarily in the areas of science quality, research direction and industry relevance.

A Māori Advisory Group (MAG) was established in 2011/12 to ensure that the research and development undertaken by the NZAGRC is relevant and accessible to all sectors of New Zealand society. The primary role of the MAG in 2014/15 has been to provide input and guidance into the early stages of a research programme focussing on low emission farm systems for the Māori sector. This project started in July 2014 and runs for three years.



NZAGRC Governance Structure

Role of the Steering Group (SG)

The NZAGRC Director reports to the Steering Group of the NZAGRC Members and via them to the AgResearch CEO and Board on the performance of the NZAGRC, including (with appropriate quantitative measures):

- Relevance of the NZAGRC's R&D to the agriculture sector and New Zealand
- Science quality
- Performance to contracted goals
- Human resource development and constraints
- Financial performance.

The main roles of the SG over the past financial year have been to ensure that the NZAGRC is operating effectively, funding decisions are made in a robust fashion and that the new science programme contracts are in line with the Centre strategy.

During 2014/15 the SG met quarterly, once in Wellington and three times in Palmerston North. They also provided comment and feedback on documents via video/teleconference and email as required. Quarterly face-to-face meetings were run in a similar fashion to Board meetings with papers circulated prior to, and detailed minutes signed off after, each meeting.

The compositions of the SG, ISAG and MAG and meeting dates during 2014/15 can be found in Appendix 1.

2014/15 SUMMARY OF ACTIVITIES AND ACHIEVEMENTS

The need for research to find cost-effective practices, tools and technologies to reduce agricultural GHG emissions that are consistent with New Zealand's pastoral farming base is as important as ever. Consequently, the Centre's vision and mission (see below) remain highly relevant in the changing context in which it operates. Progress towards achieving the specific goals set out under the vision and mission of the Centre is documented below.

The Vision

'To be an internationally renowned centre for research and development into agricultural greenhouse gas mitigation solutions'

The NZAGRC plans to be (i) a source of practical, cost effective technologies and/or practices that reduce emissions/increase sinks and clearly demonstrate that farm businesses can be both lower emitting and profitable; (ii) a focal point for New Zealand activities in agricultural greenhouse gas mitigation/soil carbon sink solutions; (iii) the key authoritative source of technical advice and support on agricultural greenhouse gas emissions and soil carbon sinks. Additionally, the NZAGRC will lead NZ's science input into the Global Research Alliance.

The Mission

'To provide knowledge, technologies and practices which grow agriculture's ability to create wealth for New Zealand in a carbon-constrained world'

The Goals

The NZAGRC has five major goals:

- 1: Advance knowledge and understanding***
- 2: Enhance awareness among stakeholders***
- 3: Contribute to policy***
- 4: Develop science capability***
- 5: Develop science and commercial partnerships***

These have been defined and quantified in order to be consistent, realistic and achievable and detailed targets are included in the NZAGRC Strategic Plan. The Centre has made substantial progress towards achieving its Vision and Mission through its on-going achievements in the five major business goal areas. Each goal is discussed in more detail in the following pages.

Centre progress towards achieving vision and mission

In 2014/15 particular high level achievements include:

- Continuing to act as a focal point for New Zealand research activities in agricultural GHG mitigation, building on international reputation for the quality of our research and progressing towards solutions. Our one day conference in April 2015 allowed us to showcase NZ work to date in this space.
- Running an efficient organisation with sound governance and financial control. The NZAGRC transferred all science contracts into the MPI contracts management system during 2014/15 and all contracting from Q4 2014/15 will be done via this system.

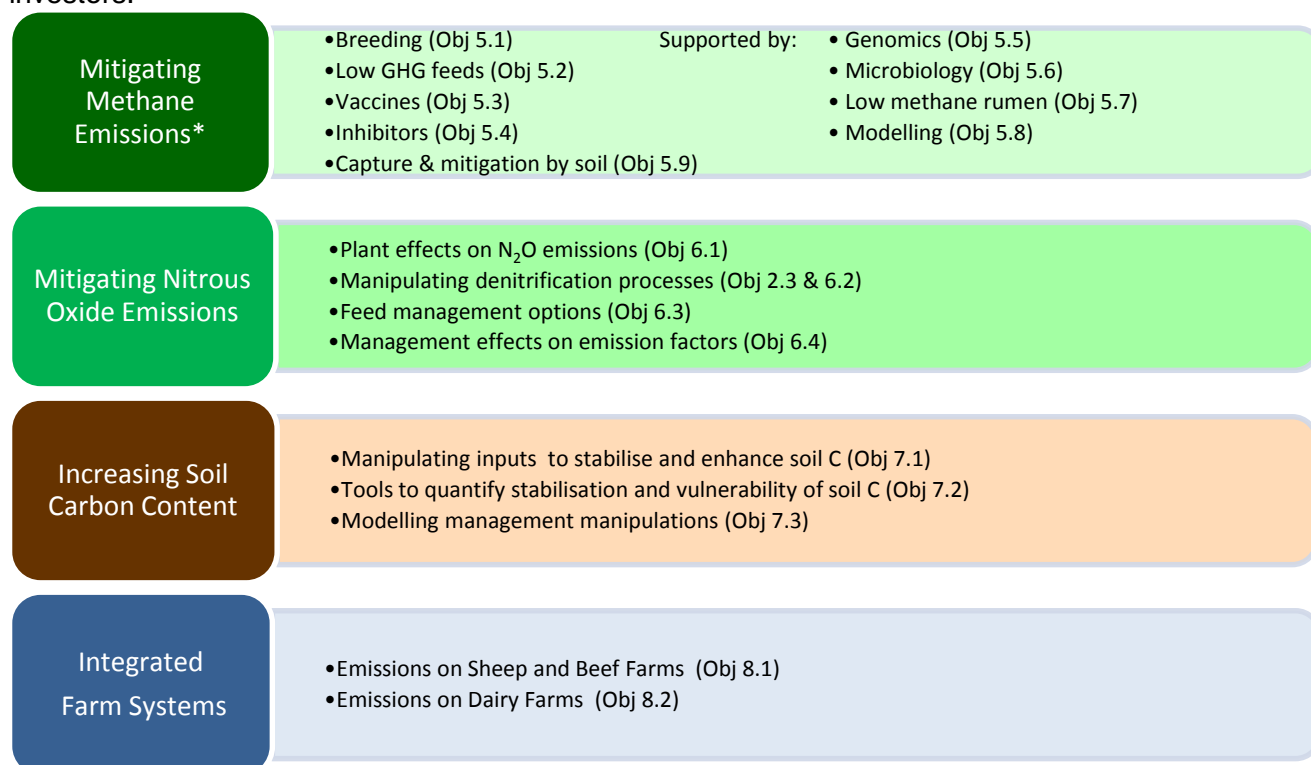
- On-going alignment with the PGgRc and, through this relationship, starting to actively engage with commercial entities to establish pathways to market for our technologies.
- Enhanced engagement with Māori, including starting a new three year Māori-focussed research programme.
- Increased efforts to communicate our science and how it fits into the bigger picture to stakeholders, media and the general public through the production of a series of fact sheets, press releases and a significantly increased media profile.
- Actively contributing to the success of the Global Research Alliance and coordinating New Zealand's science input to the Alliance and providing strategic advice to MPI, including leveraging substantial co-funding from the United Nations Environment Programme for an international project that is jointly led by FAO and NZAGRC.
- Active engagement with New Zealand policymakers through provision of emissions and mitigation scenarios to inform the development of New Zealand climate change policy, emissions targets and pathways.
- Contribution to, and in some cases coordination of, key science networks and funding mechanisms, including the Sustainable Land Management and Climate Change (SLMACC) fund and Methanet, and internationally, the Intergovernmental Panel on Climate Change (IPCC), the global Climate and Clean Air Coalition, and European FACCE-JPI and Horizon 2020 committees.
- Actively contributing to the development and retention of GHG-related scientific capability.

Goal 1: Advance knowledge and understanding

The NZAGRC will be the most important and trusted NZ source of scientific knowledge in the field of agricultural GHG emission mitigation.

Since its establishment in 2010, the NZAGRC has endeavoured to fund scientifically robust research and provide reliable new knowledge to its stakeholders, the wider scientific community and the general public.

The NZAGRC supports four Science Programmes in alignment with other agencies and private investors.



*Joint programme with the PGgRc, with the exception of Obj 5.9 which is solely NZAGRC funded.

Formal alignment with the PGgRc led to a joint science plan and subsequent joint contracting in the Methane programme being implemented from 1 July 2013. Initial contracts covered the period 1 July 2013 – 30 June 2015 and had an annual review clause in them to ensure that the research remained solution-focussed. These contracts have now come to an end and new work will be contracted from 1 July 2015 – 30 June 2019. Though the contracts will run until 2019, work plans and associated funding will be agreed annually and there will be clear stop/go points in all contracts.

In the N₂O, Soil Carbon and Integrated Farm Systems programmes a number of new contracts were established in 2014/15. A significant amount of time and effort was spent formulating new work programmes for these during 2013/14. Descriptions of the Objectives outlined above, and their progress during 2014/15, are contained in Appendix 2.

In 2014/15, key science achievements included:

- Five inhibitor compounds successfully reduced methane emissions in two day animal trials. One of these compounds has progressed to a 16 day animal trial and the level of methane inhibition in the first two days of dosing was circa 26.5% and after a further 14 days the inhibition of methane yield was still 16.6%. No toxic effects were observed for either compound.

- A number of vaccine candidates have been tested in animal trials. These can successfully raise antibodies against rumen methanogens but so far have not consistently changed rumen microbial populations.
- Results from experiments feeding fodder beet to sheep suggest that the response of methane emissions to different levels of fodder beet inclusion is more like grain (either curvilinear or with an inflexion point), rather than the linear decrease in methane yield that we observed when feeding increasing amounts of forage rape in mixes with ryegrass.
- A manuscript describing a model of the interaction of methanogens and hydrogen concentrations is ready to be submitted to the Journal of Theoretical Biology. By providing a representation of the methanogens and their interaction with hydrogen in the rumen, this model enables mechanistic predictions of mitigation options, currently under development, that target rumen methanogens directly such as chemical inhibitors and vaccines.
- N₂O emissions from the urine treated field plots of 18 plant species over a six week measurement period were markedly different among plant genotypes with the lowest emitter being the Italian ryegrass cv. Moata and the highest being upland brome. This represented a range of about -58 to +120% of the median value for the dataset.
- In Canterbury, measurements have been completed on the effect of Italian ryegrass/white clover, lucerne, and perennial ryegrass/WC (all with or without gibberellic acid (GA)) on N₂O emission factors (EF) from urine. GA had no direct effect on from the urine EF, but may allow reduction of fertiliser use without yield penalty - thus reducing N₂O from fertiliser. The lucerne EF was lower than that from the other species.
- A laboratory based rapid screening N₂O methodology has been developed and in initial tests it has identified two compounds that reduced emissions by about 50%. The impact of these products and their rates on N₂O emissions will be further investigated.
- Early results suggest that deep rooted species may lead to increased levels of soil carbon and in addition may have higher water use efficiency.
- Measurements of carbon exchange and carbon balance at Troughton Farm are ongoing. Preliminary data analysis suggests carbon losses following cultivation were not greater than losses following no-till and direct drilling treatments to re-establish the pasture sward. Rather, the soil water content conditions at the time of pasture renewal and duration between spraying and seedling emergence are more important drivers of changes in carbon storage.
- The CenW model was included in an international model intercomparison project where 28 models were applied using the same data sets to estimate carbon exchange at five international grassland sites. The analysis confirms that model is appropriate for analysing and forecasting changes in soil carbon storage for grazed grassland systems. Further activity is planned for model intercomparisons and the submission of research papers of the findings.

More detailed information regarding science progress during 2014/15 can be found in Appendix 2 which includes the submitted annual reports from all NZAGRC-funded Objectives.

Goal 1 metrics:

<i>Measure</i>	<i>Progress in 2014/15</i>	<i>Overall 2010-2015 vs target</i>
Peer-reviewed scientific journal papers	20 papers published plus 22 papers submitted	148 (published & submitted) Goal by 2015 = 100
Scientific conference papers	36	149 Goal by 2015 = 100
Patents relating to agricultural GHG emission mitigation technologies	Patenting decisions are the joint responsibility of MPI and PGgRc (not the Centre directly); new IP protected and managed as commercial (in confidence) IP or shared freely as public-good information.	Target not applicable to the Centre
Practical on-farm mitigation practices and technologies identified and being promoted	Promotion of improved efficiency as the immediate action farmers can take to help reduce emissions	Target undefined

Goal 2: Enhance awareness among stakeholders

The NZAGRC will be the most important and trusted source of information for New Zealand agricultural stakeholders on agricultural GHG emission mitigation.

PGgRc Alignment

From 2002-2012, the PGgRc invested more than \$37m into agricultural GHG mitigation research with equal shares from industry and government. During 2012/13, PGgRc successfully renewed its Partnership funding with MBIE for a further \$37m joint investment over seven years. This renewal triggered a move for the NZAGRC, which had always aligned its activities to the PGgRc, to develop a much closer formal working relationship with the PGgRc.

Close cooperation with the PGgRc is a key pathway for the Centre to interact with industry stakeholders, assist MPI to manage IP and enable knowledge transfer through commercialisation of new tools, technologies and practices. Current industry co-investors within PGgRc are: Fonterra, DairyNZ, Beef+Lamb NZ, Deer Research and AgResearch. Since February 2013, the Centre Steering Group members have been periodically meeting jointly with the PGgRc Board members to monitor progress on joint initiatives and co-funded R&D. The Centre Director is an observer on the PGgRc Board and the PGgRc Manager is a member of the NZAGRC Steering Group.

Key joint initiatives in 2014/15 with the PGgRc included:

- Organising an ISAG review of the joint Methane research programme and acting on the feedback received, and input from others, to establish new work plans in order to contract from 1 July 2015 – 30 June 2019.
- Continuing to develop and implement the joint communications strategy and plan. A range of joint communication activities have been conducted in the past year. These include a conference, science workshops, a range of co-branded factsheets, a major press release and managing the associated interest, relaunch of a co-branded e-newsletter, proactive media engagement.
- NZAGRC support for PGgRc-led engagement to find commercialisation partners.

Other Stakeholder Engagement

Although the PGgRc provides a robust pathway for the NZAGRC to link with industry stakeholders, the Centre continues to maintain direct links with a broad range of other stakeholders, including policy makers, end-users, the science community and the wider public.

In its on-going support of knowledge transfer the Centre was involved in key activities in 2014/15 that included:

- Hosting meetings with farmer groups and individual companies and organisations and giving presentations at farmer forums (e.g. NZAGRC conference, Fonterra, Royal Society).
- Presenting at conferences where industry is well represented (e.g. NZ Soil Science Society, NZ Society of Animal Production, Climate Smart Agriculture).
- Dedicated publications (e.g. annual Highlights document and regular e-newsletter) and articles in farming and general press and presenting on television and radio.
- Membership of MPI science-related advisory groups (e.g. SLMACC, Methanet, Agricultural Inventory Advisory Panel).
- Providing scientific information and expert advice to key stakeholders including government officials and industry.
- Hosting international visitors and showcasing New Zealand agricultural GHG science, including for ambassadors and high-ranking science delegations.

- Working directly with industry organisations and farmers as part of the Integrated Farm Systems programme (Pastoral 21 and B+LNZ) and Māori programme (29 farms to date).

Māori Engagement

During 2013/14 NZAGRC staff worked closely with the MAG to develop an RfP for a focussed Māori research project. A proposal for a three year programme of work was accepted from a cross-organisation team (AgFirst, AgResearch, Lincoln University and Scion) led by AgFirst, in late 2013/14 and work began on 1 July 2014.

Māori-focussed research

- Development of farm systems and farm typologies and selection of case study focus farms
- Mitigation modelling and scenario design
- Sector adoption and integration of project outcomes and practice change strategies

The “Low emission farm systems for the Māori sector” programme aims to assist the Māori pastoral sector to improve its collective capacity to increase resource efficiency and farm productivity while lowering greenhouse gas emissions. During 2014/15 the programme has developed a set of Māori farm typologies, which represent the predominant pastoral farming systems, identified key factors that underpin farm productivity, resource and emission efficiency and sustainable profitability, and started to identify and test a range of mitigation strategies. Farm typologies are important to avoid the problems of homogenizing a heterogeneous group that range from very small farms to large multi-enterprise corporates. These typologies have been compared against existing databases and used in the selection of four in-depth representative case study farms for scenarios of alternative farm system configurations that will evaluate mitigation options. At 30 June 2015, base GHG emission profiles of 29 Māori farms had been developed and the case study farm programme is underway. A paper has been written on the project to date which, following peer review, will be submitted to an international journal.

Communications and media

In early 2014 a joint communication strategy and action plan (CSAP) was approved by the NZAGRC SG and PGgRc Board. This plan was subsequently updated to provide twelve months of activities from July 2014 to June 2015. The key goal of this year’s plan has been to raise visibility, understanding and relevance of the work undertaken by the Centre and Consortium. Elements of the plan were to include changes in emission intensity in the overall scope of communications, engage more directly with our targeted audiences and build direct links to editors/journalists. The overall aim being to improve our target audiences’ understanding of where we and our work fit in the overall ‘NZ Inc’ approach to increasing agricultural production within environmental and GHG constraints.

Work to implement the action plan has progressed well, with highlights as follows:

- Production and active dissemination of three factsheets:
 - ‘Reducing New Zealand’s Agricultural Greenhouse Gases: What we are doing’
 - ‘Reducing New Zealand’s Agricultural Greenhouse Gases: How we measure emissions’
 - ‘Reducing New Zealand’s Agricultural Greenhouse Gases: Soil Carbon’
- Engagement with New Zealand journalists from a key contact list through email and familiarisation visits with interested journalists.
- Regular e-newsletter produced and distributed to approximately 300 recipients. Three newsletter science stories have been reproduced in print via the NZ Farmer (distribution ~76,000).

- Proactive media engagement following press release about the inhibitor programme at conference. Significant television (2), radio (4) and print/on-line (5) coverage of this release (major unique stories shown in brackets, plus additional syndicated repeats of these stories). Also on-going communication on the range of science covered by the Centre.

Additionally, during 2014/15 the NZAGRC has both hosted and attended a significant number of meetings and presentations with a diverse group of external parties, both in New Zealand and internationally. The NZAGRC has also actively promoted itself and its role in the media and to a scientific audience via conference papers and peer-reviewed publications. These are summarised below and detailed in Appendix 3.

Type of interaction/output	# in 2014/15
Meetings and Presentations (New Zealand)	64
Meetings and Presentations (International)	12
International Visitors and Groups	8
Global Research Alliance related interactions	73
Media interactions	7
Conference presentations	36
Journal articles in press	22
Journal articles published	20
Other interactions/publications	65

Goal 2 metrics:

<i>Measure</i>	<i>Progress in 2014/15</i>	<i>Overall 2010-2015 vs target</i>
Page views of Centre's website	35,039	111,045 views Target is more views than any other NZ agricultural GHG website
Senior Centre staff presentations to meetings of New Zealand industry and policy stakeholders	34	83 Goal to 2015 = 30
Centre funded scientist presentations to the farming community and general public	21	53 Goal to 2015 = 50

Goal 3: Contribute to policy

The NZAGRC will be the authoritative source of information for the New Zealand government on agricultural GHG emission mitigation.

Policy Advice

A key aim of the Centre is to be a trusted and independent source of knowledge - particularly to policy agencies – to enable sound, evidence-based policy development. The Centre's relationship with MPI (and other government departments in general) has continued to grow stronger and deeper in 2014/15, particularly as a result of its increased role in the GRA and other international initiatives, and the need for New Zealand to develop an emissions reduction target as part of the current UN process to develop a global climate change agreement. Likewise, MPI policy staff continue to appreciate the NZAGRC's robust scientific input and encourage and foster a culture of trust and open engagement.

The Centre's on-going inputs into the GRA and other international initiatives are prime examples of activities that the Centre engaged in during 2014/15 related to this goal.

Other activities by the Centre in 2014/15 include:

- Director and Deputy Director are members of MPI's Agricultural Inventory Advisory Board.
- Director is Chair of MPI Methanet (science grouping advising MPI on methane inventory development).
- Director is a member of the FACCE-JPI Science Advisory Board and Chair of the FACCE-JPI GHG Mitigation call International Advisory Committee plus Chair of science assessment committee on EU call on Climate Smart Agriculture.
- Deputy Director was a member of the core writing team for the IPCC's Synthesis Report, which was completed in November 2014 and concluded the IPCC's assessment of our current knowledge of climate change, impacts, and adaptation and mitigation options as input to climate change negotiations under the UN.
- Director and Deputy Director provided advice to policy agencies on an expert meeting and possible scope of a Special Report on Agriculture, Climate Change and Food Security, which may be produced during 2016-2018
- NZAGRC hosting international visitors (e.g. ambassador of USA and UK High Commissioner, delegation from China, amongst others).

Goal 3 metrics:

Measure	Progress in 2014/15	Overall 2010-2015 vs target
Senior Centre staff presentations to meetings of New Zealand government policy staff	9	42 Goal to 2015 = 10
Written reports prepared for government policy makers	2	7 Goal to 2015 = 10
Centre's science contributions directly influence and reflected in government policy	Range of technical advisory roles	Target undefined

Goal 4: Develop science capability

The NZAGRC will be a major source of new capability in the field of agricultural GHG emission mitigation.

Students and Post-doctoral fellows

Increasing the pool of researchers with skills in the agricultural greenhouse gas mitigation area is a major objective for the NZAGRC. To achieve this objective the NZAGRC is strategically directing funding to build capability for the future. Some of this funding is embedded within the funding of the core science programme, with additional funding being available on a discretionary basis when high quality students or projects are identified.

1. The provision of short term scholarships to promising undergraduate students with the aim of encouraging them to undertake post graduate studies
2. The provision of well-funded PhD stipends to high quality undergraduates
3. Employing high quality post-doctoral fellows and early stage scientists on 2-3 year contracts

In 2014/15 the dedicated undergraduate “pipeline” scholarship scheme continued with Massey, Lincoln and Waikato Universities. The updated N₂O programme recruited two new PhD students and a new postdoctoral researcher to measure N₂O emissions alongside the soil carbon work at Troughton farm. The NZAGRC also contributed funding to a student gaining work experience in the methane inhibitor group who subsequently gained employment in the dairy industry.

Type of Capability Development	# active in 2014/15	Total funded to date*
Undergraduate - Summer student	3	20
Undergraduate - Honours student	0	4
Masters Project	0	3
Masters	2	4
PhD	13	15
Post-doctoral fellow	4	4
Early career scientist	1	2
	23	52

*Including active 14/15 numbers

The NZAGRC continues to be a major funder of PhD students in agricultural sciences related to nutrition, animal and plant performance and greenhouse gas emissions in New Zealand.

Funding for international students under the LEARN fellowship scheme (under separate contract with MPI; see below under Goal 5) provides an international dimension to NZAGRC's overall capacity building efforts.

Goal 4 metrics:

Measure	Progress in 2014/15	Overall 2010-2015 vs target
PhD students studying and graduated	13 active students	15 students funded to date Goal to 2015 = 15
Post-doctoral researchers completed 2-year projects	4 active post docs	4 post docs funded to date Goal to 2015 = 10
FTEs of professional researchers working on NZAGRC research programmes	>50 researchers (19.9 FTE*) contributing to the Centre's research programme	Average of 21.3 FTE per annum Goal to 2015 = 25

*Includes PhD and post doc FTE contribution to core programme

Goal 5: Develop science and commercial partnerships

The NZAGRC will be a key player in many research and commercial partnerships relating to agricultural GHG emission mitigation.

International

The Global Research Alliance on Agricultural Greenhouse Gases (GRA), initiated by the New Zealand Government, is a key pillar in New Zealand's international science and policy engagement in climate change and agriculture. Notably, the Centre is referred to explicitly in New Zealand's commitment to long-term emissions reductions under the current UN climate change negotiations, reflecting the importance of both developing domestic solutions but also fostering international collaboration to address a globally significant problem.

Leadership of New Zealand's engagement in the Alliance rests with MPI and the Centre plays key supporting roles by providing science leadership in the GRA's research groups and resulting activities, monitoring and administering research contracts in support of the GRA on behalf of MPI, and providing strategic advice to MPI on collaborative funding opportunities, capacity building initiatives and linking of research projects with existing international initiatives.

The Centre Director continues to co-chair GRA's Livestock Research Group (LRG) together with his colleague from Wageningen UR (Netherlands), and the Centre Deputy Director acts as New Zealand's representative on the LRG. The Deputy Director, Operations Manager (International) and Project Analyst support the co-chairs in developing and monitoring the LRG's work plan, ensuring appropriate LRG presence at international events, and identifying opportunities for further engagement with existing research programmes, science institutions, international organisations and the private sector.

To increase awareness of LRG activities and engage new members and stakeholders, NZAGRC communicate the work and scope of the LRG to a global audience via newsletters, a regularly updated website, and presentations at scientific conferences and expert meetings.

Key activities for 2014/15 included:

- Planning, organising and leading two major meetings of the LRG, in November 2014 in Yogyakarta, Indonesia, and in June 2015 in Lodi, Italy. The meetings further strengthened understanding and identification of collaboration and capacity building opportunities across LRG member countries, and significantly strengthened links with GRA partners and other international institutions.
- Coordinating and participating in a workshop on soil carbon in June 2015 in Lodi, Italy. The workshop sought to identify synergies between a range of international initiatives designed to enhance the storage of carbon in agricultural, particularly pastoral soils, and to ensure good links between different scientific organisations, stakeholders and policy initiatives in this area, including defining a clear role for the GRA.
- Contributing to and hosting a GRA workshop at the Climate-Smart Agriculture conference in Montpellier, France, in March 2015, to raise the profile of the GRA and the contributions it can make to an effective integration of mitigation options with increased food security and increased resilience of the food system.
- Completion and dissemination of a summary document of current best practices and emerging options to reduce GHG emissions intensity from livestock systems around the world. This document was requested by and produced jointly with the Sustainable Agriculture Initiative (SAI) Platform, which represents many of the world's largest food producing companies.

- Coordination and successful submission of a bid to the UNEP Climate and Clean Air Coalition initiative on enteric fermentation. This project, led jointly by FAO and NZAGRC, will seek to identify and test regionally appropriate intervention packages that can jointly increase productivity, contribute to livelihoods and food security, and reduce emissions intensity of livestock production. The NZAGRC Operations Manager (International) split her time between New Zealand and FAO headquarters in Rome, Italy, to initiate the project, and for 9 months beginning in July 2015 will act as full-time project coordinator on secondment to FAO to advance this important project.
- As contracting agent for MPI, NZAGRC oversaw the monitoring and, in some cases, completion of a wide range of research projects in support of the GRA. Examples include:
 - ✓ A project to develop and test common protocols for the identification of naturally low-emitting animals was completed successfully, resulting in the initiation of a technical working group under the International Committee for Animal Recording (ICAR) to make this knowledge accessible to the widest possible international audience.
 - ✓ Advanced screening high-throughput technologies for methane inhibitors were developed and successfully implemented in two GRA projects. This resulted in the identification of several promising candidate compounds that were subsequently tested in sheep in respiration chambers, with promising significant reductions of methane emissions in initial two-day trials. Further evaluations of those and other compounds will now be conducted as part of the NZAGRC-PGgRc inhibitor development pipeline.
 - ✓ Three new research contracts were signed worth \$3.3M from round 3 of the New Zealand Fund for Global Partnership in Livestock Emissions Research (GPLER). These contracts are between MPI and research providers, but NZAGRC manages these contracts on behalf of MPI and assisted in the scientific evaluation of proposals by an international Technical Advisory Panel. This was the last round of this fund.
 - ✓ A significant new project was commissioned, in collaboration with the US-based company C-Lock, to design and test a system to rapidly screen methane emissions from dairy bulls along with feed intake, to assist in the identification and selection of naturally lower-emitting dairy animals for New Zealand.
 - ✓ NZAGRC continued to commission and manage a suite of LRG priority research projects in the area of methane and nitrous oxide.
- Continued administration of the LEARN/GRASS fellowship scheme, with 9 fellows involved this year.
- International capacity building and engagement efforts:
 - ✓ Successfully held a 2 week technician training course on measurement of GHG emissions from livestock in South Africa, in collaboration with the European AnimalChange project and the University of Pretoria. The course was held in September 2014 with participants from 12 countries across Africa. NZAGRC organised the event funded by MPI, with financial support from the EU funded AnimalChange programme.
 - ✓ Organised and participated in an engagement workshop at the RUFORUM conference in Maputo, Mozambique. The main objective of this biennial conference is to provide a platform for agricultural research for development stakeholders in Africa and beyond to exchange findings and experiences, while at the same time learning lessons towards improving performance of the agricultural sector and ultimately people's livelihoods.
 - ✓ Arranged for a regional capability building workshop for south-east Asia, following the annual meeting of the LRG in Yogyakarta, Indonesia. The workshop involved participants from India, Bangladesh, Japan, the Philippines, Indonesia, Thailand, Malaysia, and Vietnam, and identify key next steps for the region to improve their

ability to estimate GHG emissions from livestock systems and identify options to reduce emissions while supporting agricultural development and rural livelihoods.

- ✓ Planning for training courses on improved emissions inventories for countries from Africa and south- and south-east Asia, to be held in July and September 2016, and for a technical training course on measuring GHG emissions from livestock for south and south-east Asia, to be held in November 2015 in Thailand.

IP and knowledge management

The Centre does not own IP generated from its science investments and patenting and commercialisation decisions are the direct responsibility of MPI and/or PGgRc. The Centre's role is simply advisory and administrative. An on-line Release of Information (ROI) system, established and maintained by the NZAGRC, is used to keep track of the number and type of publications/presentations generated under NZAGRC funding and ensures that new IP is appropriately identified, protected and managed. The system is also used for approval and tracking of PGgRc and GRA outputs.

During 2013/14, the possibility of establishing a NZAGRC on-line reporting and progress management system for science contracts was investigated. This was superseded by MPI establishing a new contract management system and the NZAGRC being invited to use this in early 2014/15. NZAGRC staff have worked closely with MPI to set up suitable sections of the contract management system for NZAGRC and GRA contract monitoring. This was completed and all current contracts were migrated to the system by 30 June 2015. Q4 reporting was conducted via the system and it will be used for all suitable NZAGRC contract monitoring and reporting going forwards.

Thus far, only the methane mitigation area has identified products (e.g. methanogen inhibitors, anti-methanogen vaccines and low emitting sheep), with clearly identified commercial potential. During 2014/15, the NZAGRC has actively supported the PGgRc in its efforts to engage with industry partners to move these research areas closer to commercial reality.

Goal 5 metrics:

<i>Measure</i>	<i>Progress in 2014/15</i>	<i>Overall 2010-2015 vs target</i>
Leadership of science input into Global Research Alliance and coordination of Livestock Research Group with the Netherlands	Proactive NZAGRC input into Alliance during 2014/15	No target
Visiting fellows from overseas research organisations hosted	2 exchanges funded by LEARN/GRASS Fellowships	9 exchanges to date Goal to 2015 = 5
Memoranda of understanding covering research collaborations agreed with research centres around the world	Agreements with national and international research centres on-going and productive	Target not applicable
Confidentiality agreements with companies to discuss information related to agricultural GHG mitigation technologies	Signing confidentiality agreements with interested companies is the joint responsibility of MPI and PGgRc. The PGgRc are taking a lead role with regards to adoption and commercialisation, on behalf of industry and MPI. The NZAGRC role is one of advice and support.	Target not applicable to Centre
Licenses to companies to sell agricultural GHG emission mitigation technologies that the NZAGRC or its partners have developed or imported and implemented to suit NZ requirements	Signing licensing arrangements with interested companies is the joint responsibility of MPI and PGgRc. The PGgRc are taking a lead role with regards to adoption and commercialisation, on behalf of industry and MPI. The NZAGRC role is one of advice and support.	Target not applicable to Centre

SCIENCE FUNDING REPORT

Funding

In accordance with the NZAGRC's Business, Strategy and Science Plans, and with the approval of the SG, \$5.18 million was allocated to research and ancillary activities in the 2014/15 financial year. The detailed funding allocated to the core scientific programmes is reported in detail later in this section. All figures are exclusive of GST.

Infrastructure Update 2014/15

A major spending initiative on infrastructure was completed in the 2010/11 financial year with the New Zealand Ruminant Methane Measurement Centre (at the AgResearch Grasslands campus in Palmerston North) and the New Zealand Nitrous Oxide Measurement Centre (situated at Lincoln University) becoming operational. In the current financial year a contribution of \$150,000 was made to Landcare Research for the purchase and installation of lysimeters at the newly developed dairy facility at Ashley Dene farm.

Capability Development Funding 2014/15

The NZAGRC's strategy in this area is outlined under Goal 4 (see previous section). A portion of the Centre funding for this is embedded within the core science programme, another portion is provided via the undergraduate "pipeline" scholarship schemes, with the remaining funding being available on a discretionary basis when high quality students are projects are identified. Additionally, the NZAGRC advises MPI with respect to international capability building efforts and assists with the administration of Alliance funds in this area (see Goal 5).

Research Programmes 2014/15

The current Science Plan consists of 19 Research Objectives which align under five key areas: (i) methane; (ii) nitrous oxide; (iii) soil carbon; (iv) integrated farm systems; and (v) Māori. Those programmes marked with a dagger ([†]) are co-funded with the PGgRc and/or PGgRc/MPI and those marked with a diamond ([◇]) are solely funded by the PGgRc. Those left unmarked are solely funded by the NZAGRC.

Area	#	Objective Title	Objective Leader	Objective Leader Organisation	2014/15 Research FTE**	2014/15 \$NZ NZAGRC (GST excl)*
Methane	5.1 [†]	Breed low methane ruminants	S Rowe & A Jonker	AgResearch	0.99	338,000
	5.2 [†]	Identifying low GHG feeds	D Pacheco	AgResearch	0.12	40,000
	5.3 [†]	Vaccine	N Wedlock	AgResearch	0.94	280,000
	5.4 [†]	Identify inhibitors that reduce ruminant methane emissions	R Ronimus	AgResearch	1.06	357,500
	5.5 [†]	Microbial genomics to underpin methane mitigation	E Altermann & S Leahy	AgResearch	1.16	230,000
	5.6 [◇]	Microbiology to underpin methane mitigation	S Kittelmann & P Janssen	AgResearch	0	0
	5.7 [†]	Understanding the low methane rumen	M Tavendale & G Henderson	AgResearch	0.69	89,000
	5.8	Modelling rumen methane production	D Pacheco	AgResearch	1.57	200,000
	5.9	Dairy housing methane capture and mitigation by soil	S Saggar	Landcare Research	0.16	55,000
Nitrous Oxide	6.1	Plant Effects on N ₂ O Emissions	S Bowatte	AgResearch	0.81	276,500
	6.2	Denitrification Processes	S Saggar	Landcare Research	0.96	90,000
	6.3	Feed management options for mitigating N ₂ O emissions from grazed systems	C de Klein	AgResearch	1.46	447,000
	6.4	Additional N ₂ O work	H Di, J Luo & T van der Weerden	Lincoln University, AgResearch	0.64	154,000
Soil Carbon	7.1	Manipulation of carbon inputs to stabilise and enhance soil carbon stocks	D Whitehead	Landcare Research	3.91	409,000
	7.2	Tools to quantify the stabilisation capacity and vulnerability of carbon in grassland soils	F Kelliher	AgResearch	1.63	276,000
	7.3	Modelling management manipulations using the HPM	J Rowarth	Waikato University	0.18	75,000
Integrated Farm Systems	8.1	GHG Emissions on Sheep and Beef Farms	R Dynes & K Hutchinson	AgResearch	0.44	124,025
	8.2	GHG Emissions from Dairy Systems	R Dynes & K Hutchinson	AgResearch	1.94	618,050
Māori	20.1	Low emissions for the Maori sector	P Journeaux	AgFirst	1.25	300,793
Total					19.90	\$4,359,868***

*N.B. 2014/15 funding includes personnel costs, consumables and in certain cases, items such as SNP chips or services such as DNA sequencing. **NZAGRC PhD students and post-doctoral researchers time is included. ***Other research costs of \$477,442 are not included in this table.

Methane Research Programme Report - 2014/15

**Principal Investigators: Dr Peter Janssen and
Dr Graeme Attwood**



The NZAGRC CH₄ programme is jointly funded with the PGgRc and aligns with existing MPI programmes. It aims to reduce emissions by directly targeting the CH₄-producing methanogens through small molecule inhibitors and vaccines and indirectly through feeding and changes in animal phenotype. The current objectives within the NZAGRC CH₄ programme have made significant progress this year and contracted milestones came to an end at 30 June 2015. New work plans have been developed and contracts will be entered into in early 2015/16. A high-level product development pipeline, involving the NZAGRC-PGgRc and potential activities by industry and government, has been developed to guide and summarise the overall research effort (see following page).

Previous work in the breeding programme has shown that gross methane emissions and methane yield were heritable and repeatable traits when measured in respiration chambers. These results led to the development of selection lines of low and high methane yield animals. The work undertaken in the past year has focused on continuing this selection for divergent methane phenotypes. The current cohort differ in methane yield by ~7% in respiration chambers and by ~14% using a proxy for methane yield when measured in portable accumulation chambers (PAC) off pasture. Research in the current year also validates previous findings that, under the current SIL index, lower methane emitting animals having higher EBVs and the expectation of greater profit. This gain in profit comes from a tendency for low emitters to have a higher dressing out percentage and leaner growth. Other highlights this year include:

- SF₆ trial showing that differences in methane emissions between lines persist at pasture.
- Results showing that rumen microbial profiles can be reduced to 2 dimensions and were highly correlated with methane yield and contain a heritable and repeatable component suggesting that these profiles could be highly predictive of the methane status of an animal.
- Initial results indicate that moles of total gas produced may be a useful proxy for feed intake and this is a heritable trait even after adjustment for liveweight when used in PAC chambers on animals grazing pasture.

During 2014/15 the feeds programme continued to build onto the finding that feeding some brassicas (forage rape) results in lower methane emissions when compared with perennial ryegrass diets. We demonstrated that methane emissions from sheep linearly decreased as the levels of forage rape included in the diet increased. This suggests that reductions in methane emissions observed in brassicas occur through a different mechanism to those elicited by feeding grains. Future work will focus on looking at the whole system effects of increasing the quantity of forage rape fed. We have been able to translate knowledge generated as part of our brassica studies into identifying fodder beet as a potential low GHG feed. In an initial study with sheep, feeding a diet of 90% and 10% grass resulted in a methane emission reduction of 50%. We are currently following up and confirming results from this first study using a broader range of fodder beet inclusion rates. We conducted an initial assessment of the potential of near-infrared reflectance spectroscopy (NIRS) to predict methane yields from forages fed to ruminants. Although modest, the predictive ability of the NIRS calibration could be considered useful for screening purposes, particularly in terms of obtaining an initial, non-experimental prediction of the potential methane yield of a feed.

The vaccine programme has progressed in the past year by moving to large scale animal trials with two vaccine candidates. One of these has been extensively tested in sheep and cattle and the

other has commenced with a sheep trial. Two additional potential new targets have been tested in sheep with promising results. Antisera against some of these targets have shown inhibitory effects in *in vitro* methanogen culture assays and these targets will now be tested further using larger groups of animals.

Significant progress was made in the inhibitor area in 2014/15. Short-term sheep trials with animal-safe inhibitors identified five compounds showing significant (~20% or greater) methane inhibition, with brief levels of inhibition >90% just after feeding in three cases. In a 16 day animal trial one of these compounds showed an inhibition of 26.5% over the first two days, falling to 16.7% at day 16. The inhibitor programme is coming towards the end of its screening component now with *in silico* screening of >1,000,000 compounds, >20,000 against enzymes and >15,000 against a rumen methanogen having been conducted to date. This screening has identified numerous other potential inhibitors and a prioritised target list is being produced for the 2015/16 work programme which will focus strongly on animal testing and increasing the potency of identified inhibitors.

The study of microbial genomics continues to underpin the vaccine and inhibitor work. NZAGRC-PGgRc funded researchers have now created the largest known comparative genomic dataset on rumen methanogens in the world. These genomes are being mined to identify new targets for assessment as vaccine antigens.

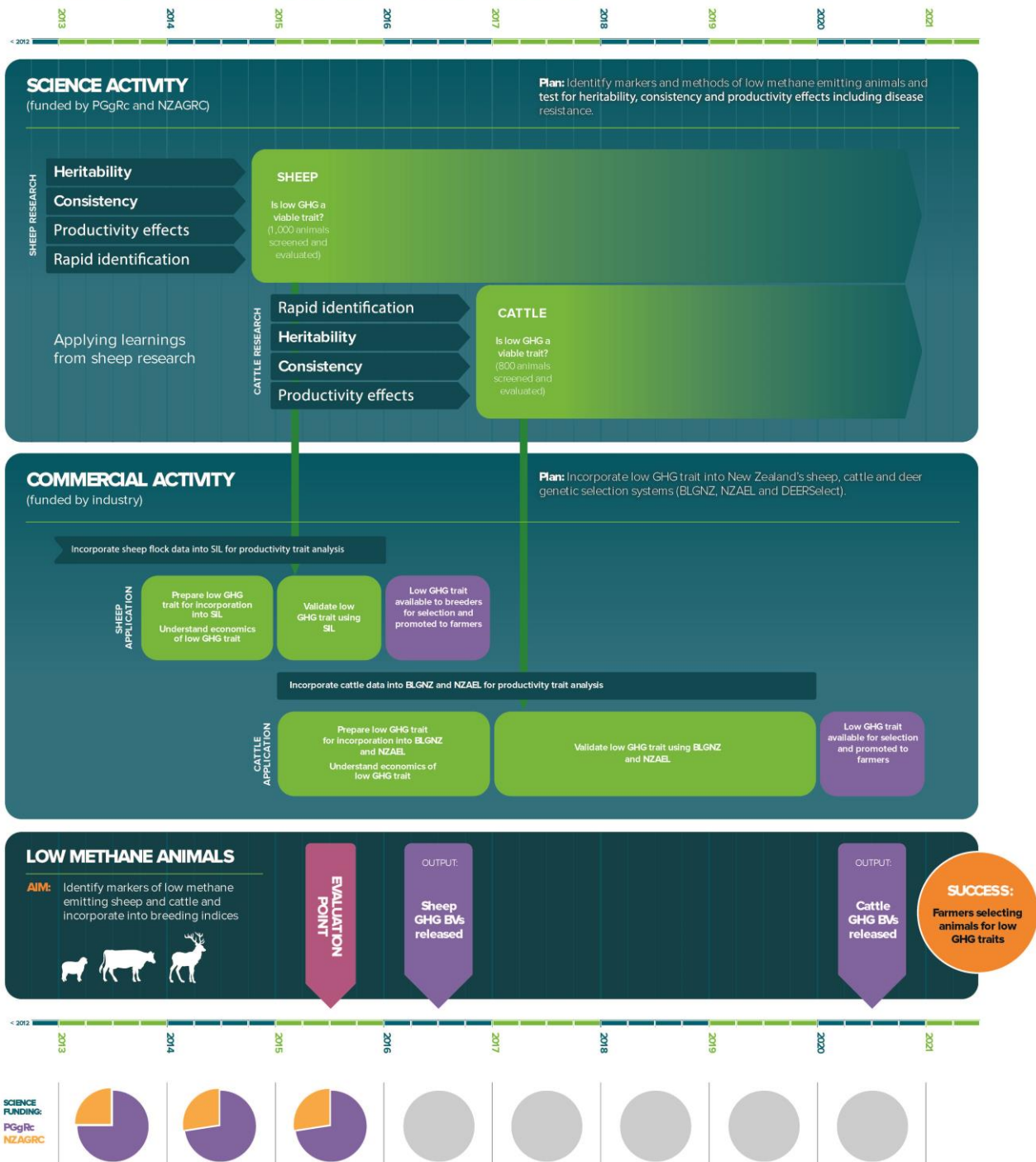
Rumen microbial ecology methods (high-throughput sequencing of microbial marker genes, enumeration of total methanogens by quantitative PCR) have been applied to the four research aims and the effects of different methane mitigation strategies have been evaluated. The microbiology team has also identified the dominant methanogens present in NZ ruminants at a species level which has helped to inform other parts of the programme. In order to ensure that the research utilises the most appropriate and effective tools, barcoded Illumina MiSeq amplicon sequencing and a validated buccal swab methodology have been established for microbial community analysis during 2014/15.

Work on understanding the potential impacts of a low methane rumen has continued. This has demonstrated that homoacetogenesis occurs in the ovine rumen, even when methanogenesis is not inhibited. Ruminal homoacetogenic activity increased when methanogenesis was inhibited with a model inhibitor compound. An increase in propionate, a further ruminal hydrogen sink, was also observed, and there were changes in the methanogen and bacterial communities. However, based on our initial results, homoacetogenesis will not become a major sink for ruminal hydrogen if methanogenesis is inhibited.

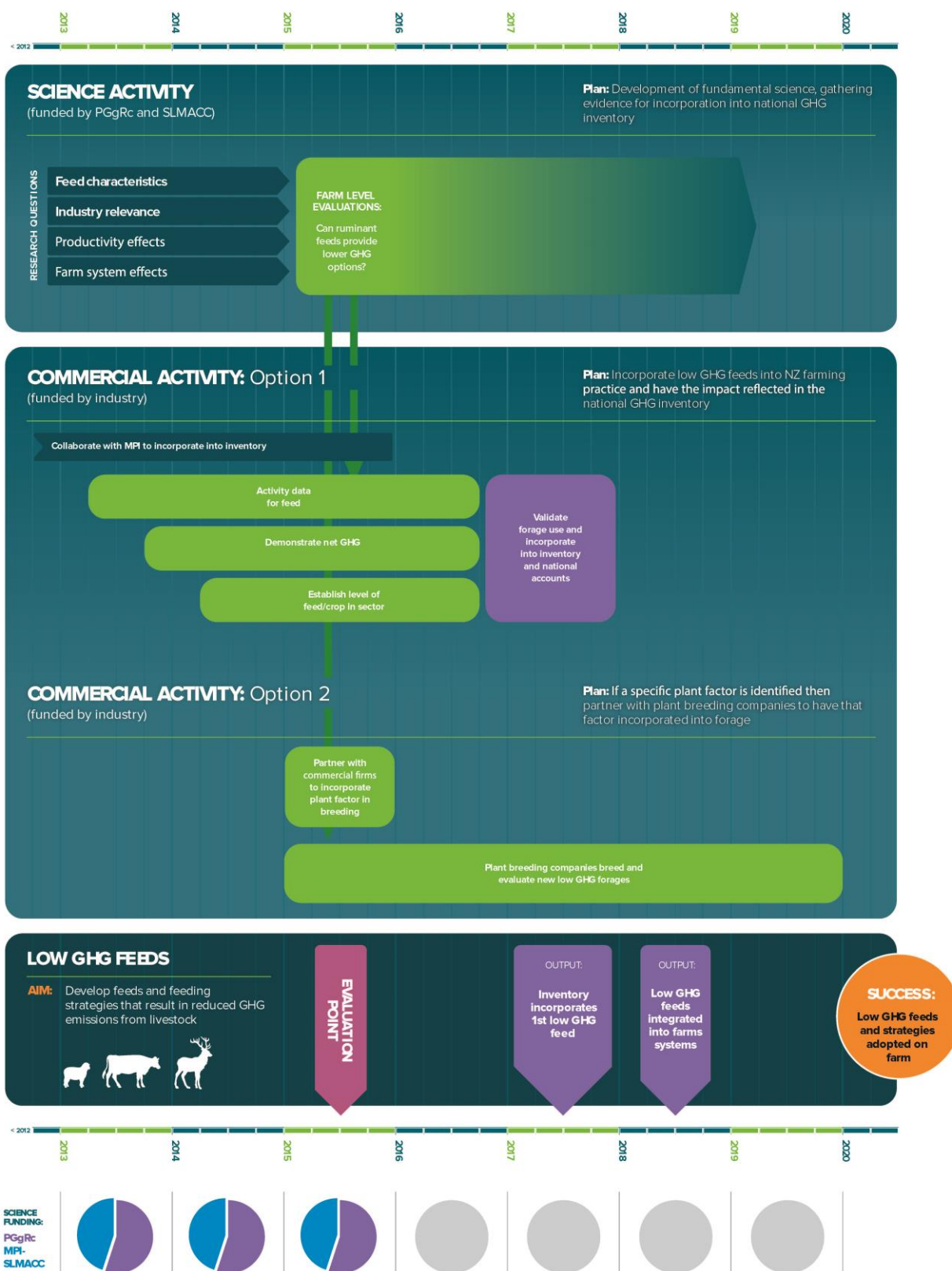
In the modelling area a manuscript describing a model to predict methane emissions from sheep (MollyRum14) has been published in the Journal of Animal Science. We have evaluated this sheep model's predictions for a wider range of fresh forages and found it to have a good predictive performance across the range of diets. The team have also developed a model of microbial interactions, based on metabolic pathways and with a thermodynamic control that predicts co-existence of bacterial groups using a common substrate and allows the production of volatile fatty acid to be an emergent property of the model.

One new methane project was contracted in 2014/15. The aim of this study is to test the practicality of capturing methane emitted by housed cattle and their waste and breaking it down by flowing it into the soil for oxidation by methanotrophs. This project started in late 2014/15 with establishment of the experimental site and experimental protocols.

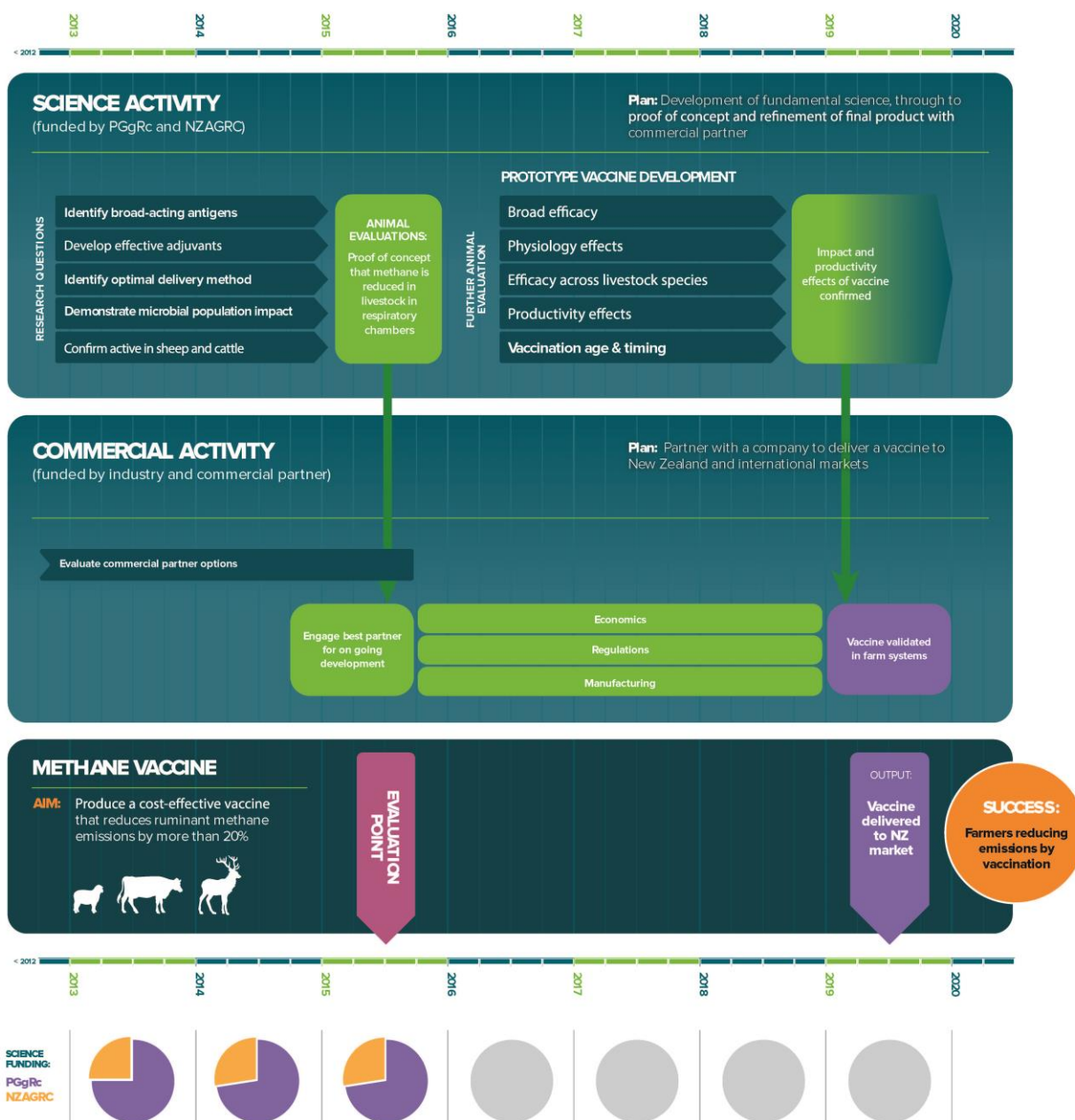
RESEARCH PIPELINE: LOW METHANE ANIMALS



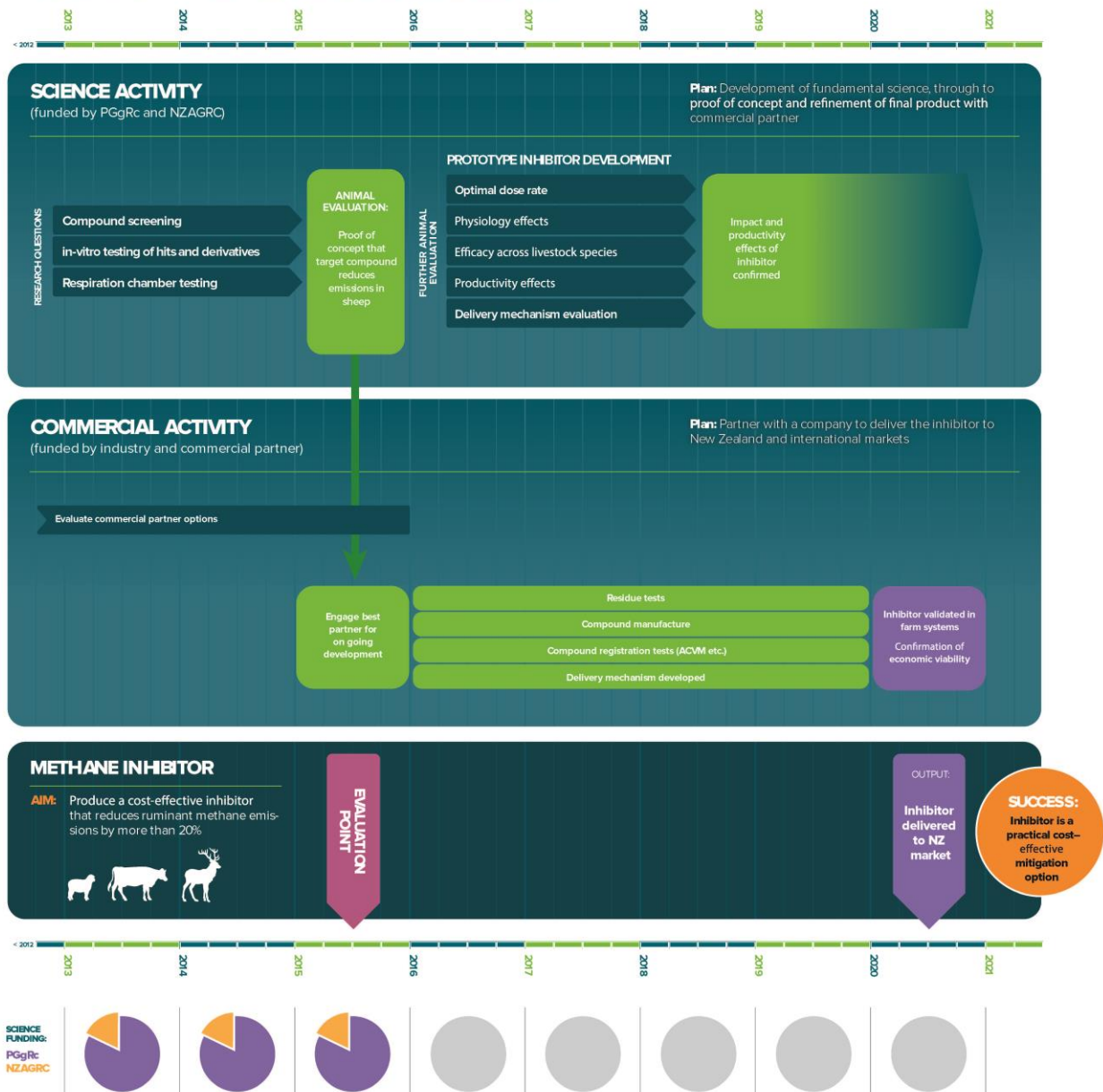
RESEARCH PIPELINE: LOW GHG FEEDS



RESEARCH PIPELINE: METHANE VACCINE



RESEARCH PIPELINE: METHANE INHIBITOR



Nitrous Oxide Research Programme Report - 2014/15

**Principal Investigators: Dr Cecile de Klein and
Prof Hong Di**



The nitrification inhibitor DCD is a proven nitrous oxide mitigation technology, but its withdrawal from the market means that the focus of the NZAGRC's new nitrous oxide research programme is currently on measuring the effects pasture plants and pasture plant communities have on nitrous oxide emissions. This work is closely aligned to the MBIE P21 and Forages for Nitrate Leaching programmes (FRNL). In addition we have commissioned work to better quantify the GHG impacts of the use of stand-off pads/housing by looking at differences in emissions between pasture applied urine and pasture applied farm dairy effluent (FDE). A high-level product development pipeline, involving the NZAGRC and potential activities by industry and government, has been developed to guide and summarise the overall research effort (see following page).

Plants can influence nitrification in soils by a variety of mechanisms: (a) they may secrete inhibitory compounds known as biological nitrification inhibitors (BNi compounds) that directly influence nitrifying organisms, (b) they may compete strongly for nitrogen and thus reduce the substrate for soil nitrifiers, and (c) they may alter the identity of the microbial community and/or microbial activity by altering the soil environment e.g. soil pH and moisture content. This year a field experiment involving 18 different plant species cultivars confirmed the findings of our laboratory studies that plant genotype can influence nitrous oxide emissions with emissions varying between -58 and 120% of the median value. The lowest value came from the Italian ryegrass cultivar Moata and the highest from the brome cultivar Gala. The finding that a simple soil nitrogen potential (SNP) test was highly correlated with N₂O emissions holds out the prospect that potential N₂O emissions can be predicted from a simple and inexpensive laboratory test. Current work is testing whether the effects of different plant genotypes is consistent over time.

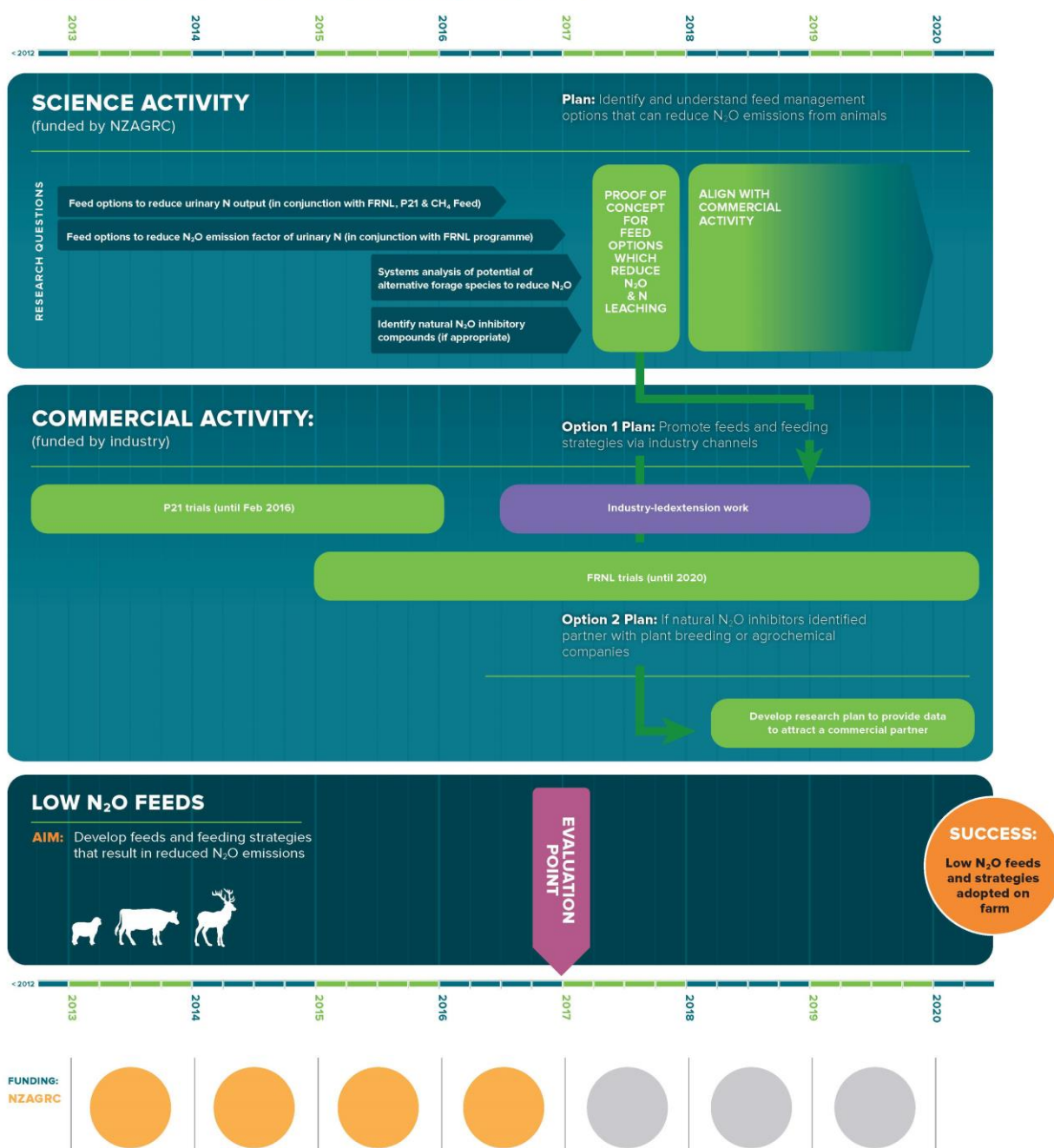
In addition to affecting soil microbial communities plants can also affect the quantity of N consumed by grazing ruminants, how N losses are partitioned between dung and urine and, perhaps, by the excretion of specific compounds that influence emissions per unit of soil deposited urine. In collaboration with the MBIE funded 'Forages for Reduced Nitrate Leaching' (FRNL) research programme we are determining N₂O emission factors from animal urine (EF₃) applied to a range of different plant species. For example in Canterbury we are measuring EF₃ from 'Italian Ryegrass plus Plantain' vs 'Perennial ryegrass' pastures using urine collected from cows grazing on the Italian/Plantain plots as well as cows on the P Ryegrass plots. A control treatment (no urine) and standard fresh urine at 700 kg N/ha treatment are also included on both pasture mixes. Highlights include the following:

- The successful development of a rapid screening methodology for rapid assessment of N₂O mitigation potential of chemical compounds.
- Preliminary results from the rapid screening technique suggesting that glucosinolate hydrolysis products can reduce N₂O emissions.
- The finding in Canterbury that gibberellic acid addition had no direct effect on the N₂O emission factor (EF₃) from urine applied to pasture species, but may allow reduction of fertiliser use without yield penalty.
- Urine collected from cows grazing different pastures has been analysed and applied to the lysimeters.
- EF₃ on lucerne was lower than EF₃ on Italian ryegrass/white clover and perennial ryegrass/WC.

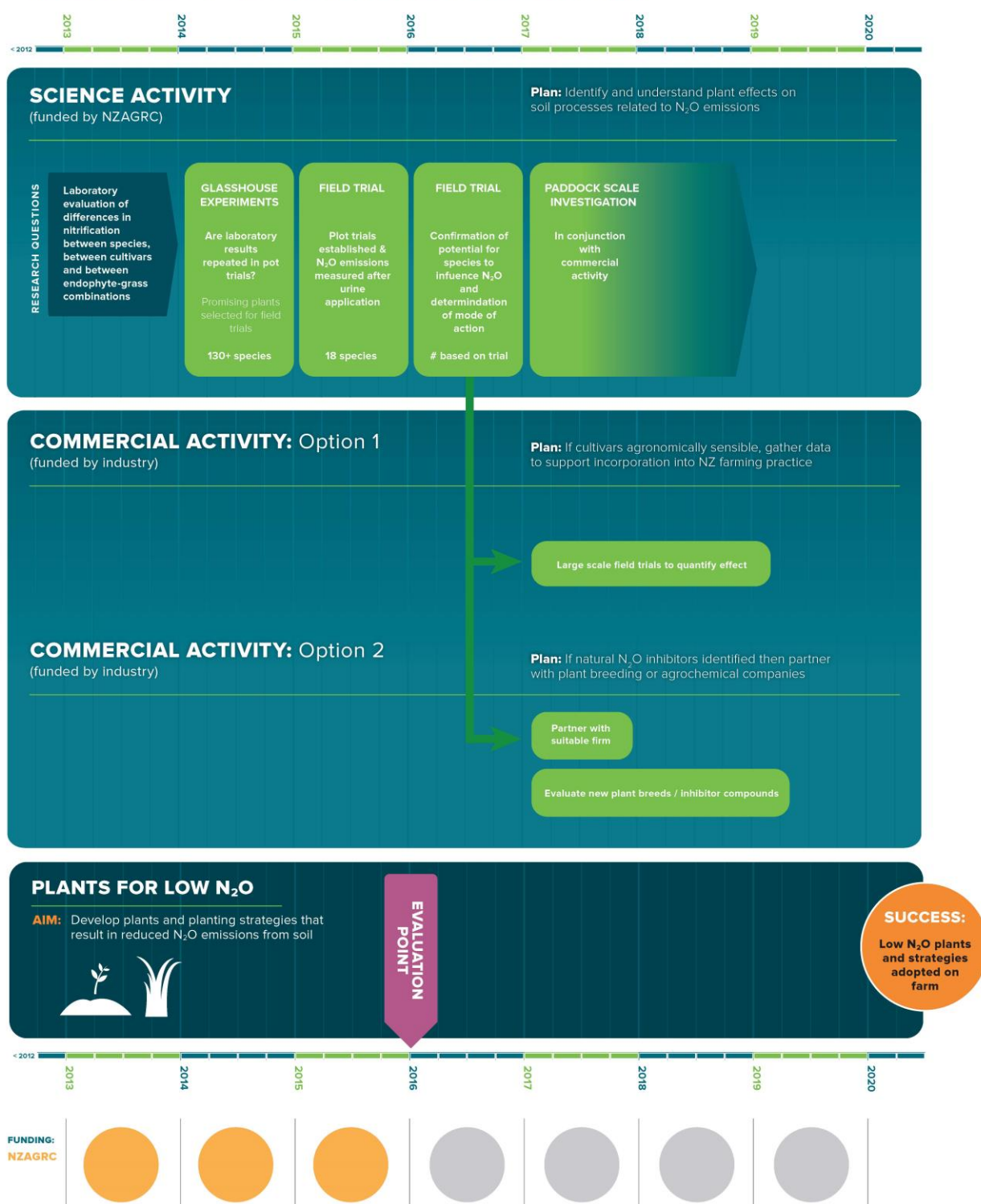
In collaboration with the NZAGRC soil C programme at Troughton farm in the Waikato, the N_2O emission factor (EF_3) for animal urine applied to diverse pasture (lucerne, plantain, chicory plus ryegrass/white clover) is being compared with that for animal urine applied to conventional ryegrass/white clover pasture. This will help us build a full picture of the total GHG effect of a change targeted at increasing soil carbon storage.

Any positive effect of housing/temporary removal of animals from pasture on N_2O emissions, relies on emissions from directly deposited animal urine being more than those from subsequently applied farm dairy effluent (FDE). In April 2015 a field trial was established on a free-draining soil supporting a ryegrass/white clover mixed sward in Otago. A range of soil moistures were established by covering some plots and applying varying amounts of irrigation to others. At the start of the trial, soil moisture content ranged from 25 to 43% volumetric water content, equivalent to 44–78% water filled pore space (WFPS). FDE, with an N content of 0.07%, was applied at an equivalent rate of 70 kg N/ha to the 14 plots, while cattle urine, with a nitrogen (N) content of 0.57%, was applied at an equivalent rate of 570 kg N/ha to 7 of the plots. Control treatments (no applied N) were also established on all 14 plots to allow a calculation of EF_1 across the range of soil moisture contents. The urine treatment was restricted to 7 plots as this was considered sufficient to determine if EF_1 and EF_3 differ. Gas and soil sampling was initiated on the day of treatment application (16 April), and continued for 2.5 months (up to the end of June).

RESEARCH PIPELINE: LOW N₂O FEEDS



RESEARCH PIPELINE: PLANTS FOR LOW N₂O



Soil Carbon Research Programme Report - 2014/15

**Principal Investigators: Prof Frank Kelliher
and Dr David Whitehead**



Increasing the quantity of carbon stored in agricultural soils has the potential to offset emissions of greenhouse gases to the atmosphere. However, realising this potential is technically challenging when soil carbon stocks are already high as they are in New Zealand, potential changes in soil carbon are small and spatial variability is high. The current NZAGRC programme has three distinct components (1) testing specific management practices that may increase the long term soil carbon store in field situations (2) developing and using models to predict how a range of management practices may influence long and short term soil carbon storage and (3) identifying those factors that influence the stability of current or newly added soil carbon. A high-level product development pipeline, involving the NZAGRC and potential activities by industry and government, has been developed to guide and summarise the overall research effort (see following page).

At our field site in the Waikato soil carbon losses following cultivation were not greater than losses following no-till and direct drilling treatments to re-establish the pasture sward. Rather, the soil water content conditions at the time of pasture renewal and duration between spraying and seedling emergence are more important drivers of changes in carbon storage. The main implications for the farming community are that (a) the site preparation for pasture renewal is considerably less important than rapid re-establishment of the new sward and (b) selecting a critical time for the conversion when soil water content allows good pasture growth but when weather conditions minimise carbon losses.

Early results from the two-year comparison of changes in soil carbon storage under conventional ryegrass/clover and a mixed sward incorporating deep-rooting species show that the potential for net carbon uptake into the soil for the mixed sward is greater because of increased carbon inputs from roots. Preliminary findings on the water use efficiency of high and low diversity swards also show that the high diversity swards can produce equivalent amounts of biomass while using less water than a conventional ryegrass-clover sward.

Overall, from our work at the dairy farm site in the Waikato, we conclude so far that (i) the dual goal of milk production and increasing carbon storage in dairy grasslands may be possible, (ii) carbon losses due to pasture renewal are less when time between spraying and sowing is minimised, (iii) carbon losses are minimised if regrassing takes place late summer/autumn rather than spring and (iv) more diverse sward offers potential for increasing soil inputs from roots.

A second experimental site is now being established on a recently converted dairy farm on shallow, stony soils at Ashley Dene, Canterbury. The question we are asking is does conversion from dryland farming to intensive dairy farming using irrigation and addition of nitrogen fertiliser result in increases in soil carbon storage? This site and the experimental treatments are in the process of being set up so our work has not yet started. Our work will contribute to a larger project to address carbon and nitrogen cycling in relation to conversion to intensive dairy management practices in collaboration with Lincoln University, Landcare Research and Plant & Food Research. This site will also be used in a multi-site comparison of processes regulating changes in soil carbon turnover in a GPLER project led by Landcare Research.

The CenW model is being assessed for its suitability to simulate and forecast the impacts of changing climate and management practices on long-term soil carbon storage. We have demonstrated the success of using the model in comparison with 28 models used internationally and in simulating the short-term changes in net carbon exchange that we have observed from the

field measurements at the Waikato site. These comparisons have provided confidence that we are able to apply the model across a much wider range of climate and management scenarios, including changes in fertiliser application, grazing regime, pasture root:shoot ratios, water availability (i.e. climate change or irrigation) and the environmental changes of increasing temperature and atmospheric carbon dioxide concentration. While we have identified practical management modifications that could lead to enhanced productivity, most of them led to only small changes in increased soil carbon storage.

Developing and deploying management practices that maximise the long-term storage of soil carbon depend on understanding the capacity of soils to stabilise carbon and its vulnerability to loss. We aim to identify the soil properties and management practices that most affect the stability of soil carbon and its vulnerability to loss. A second generation statistical model is being developed to better predict the soil carbon stabilisation capacity for New Zealand's grassland soils and soil chemical and physical fractionation methods and a soil carbon mineralisation assay is being used to quantify the stability and vulnerability to loss of soil carbon. We will synthesise findings to identify soil properties and grassland management practices that most affect soil carbon stabilisation and vulnerability to loss.

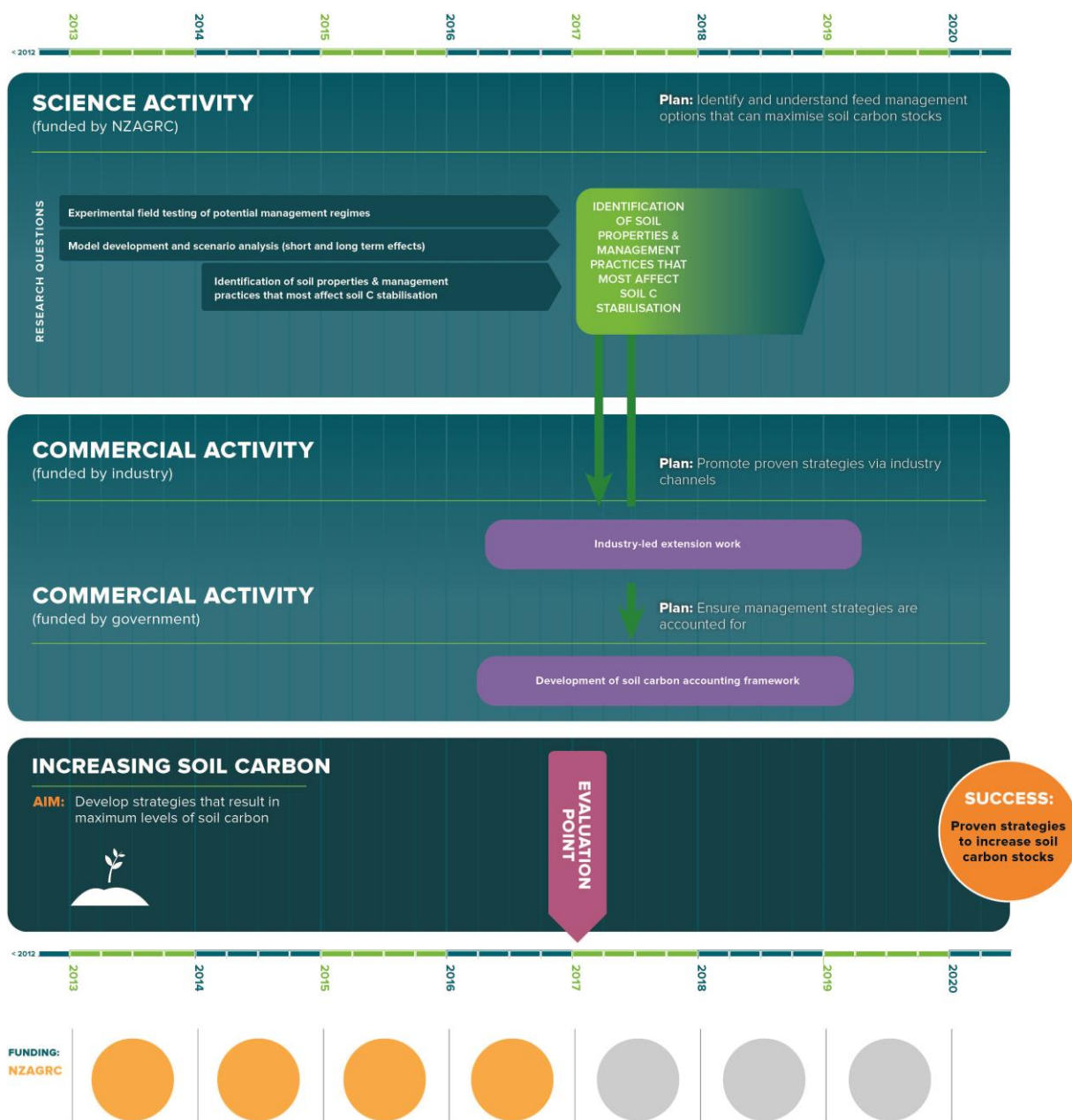
In 2014/15 we sampled soils representative of the major orders including Allophanic, Brown, Gley, Pallic and Recent at 130 sites across NZ which have been under long-term pastoral agriculture management. This was done in collaboration with the Plant & Food Research CORE-funded LUCI (Land Use Change Intensification) programme. These samples were prepared and measured using mid-infrared (MIR) spectroscopy measurements. On the basis of selected, preliminary results, we sampled soils at a further 40 long-term pasture sites. These samples will be used to help develop the statistical model.

Alternative methods to determine soil properties which can affect carbon stabilisation are being tested. For this study, allophanic and non-allophanic soils were sampled for mineral surface area measurement and two methods of drying the samples tested, one recommended for delicate structure because we postulate allophane to be a gel-like substance. Two fractionation methods to examine the possible, confounding effects of organic matter are also being tested. Mineral surface area is being measured using four absorbates and examining the samples using an electronic microscope.

Understanding and predicting changes in soil carbon storage has to be based on an understanding of the effects of changed practices on both C and N cycles. Intensification, for example, increased fertiliser input, increased livestock intake demand, changes in the offtake of nitrogen and carbon as dairy cows replace dry stock (e.g. sheep), increased milk yield per cow, the increased use of purchased feed, cutting (vs grazing) of forages and the increased use of irrigation lead to major changes in the C and N cycles, because animals 'uncouple' the C and N cycles, so the more animals that are involved, the more that cycles are uncoupled. Such manipulations give rise to transient changes that are not necessarily indicative of the longer term changes that can occur when a particular management(s) is sustained. Understanding and clearly separating long term sustainable (c.f. steady-state) changes from transient changes is a priority if we are to understand how changed management practices and system manipulations will impact soil carbon storage in the future. We also need improved understanding of how climate change itself will interact with anticipated management changes. To do this we started a new programme of work in the winter of 2015 using the Hurley Pasture Model (HPM), a mechanistic C and N cycling model that incorporates soil, plant and animal processes. Results are already giving insight that indicators such as emission factors and C and N balances based on measurements collected during the period of transition (some 2 to 10 years minimum) between managements can be misleading. Changed management rarely involves a single action and we will use the model to analyse systematically just which of the components of a management change have the largest impact. The model tracks GHG gaseous emissions, all fluxes of C and N to the environment, through plants, soils and animals, as well as C and N sequestration. Its capacity to help foresee the transient, as well as (often counter-intuitive) long term outcomes for emissions (gaseous and all

other), fluxes and C/N stocks, is therefore considerable. The model predictions include increasing CO₂ and/or temperature.

RESEARCH PIPELINE: INCREASING SOIL CARBON



Integrated Farm Systems Research Programme Report - 2014/15

Principal Investigator: Dr Robyn Dynes



The overall aim of this programme of work is to identify and demonstrate that management strategies to reduce **GHG emissions intensity** already exist and that they are practical and cost effective. The programme covers dairy, beef and sheep farms and is closely aligned to the dairy industry's P21 programme and the Beef+Lamb NZ environment focussed farm programme. A high-level product development pipeline, involving the NZAGRC and potential activities by industry and government, has been developed to guide and summarise the overall research effort (see following page).

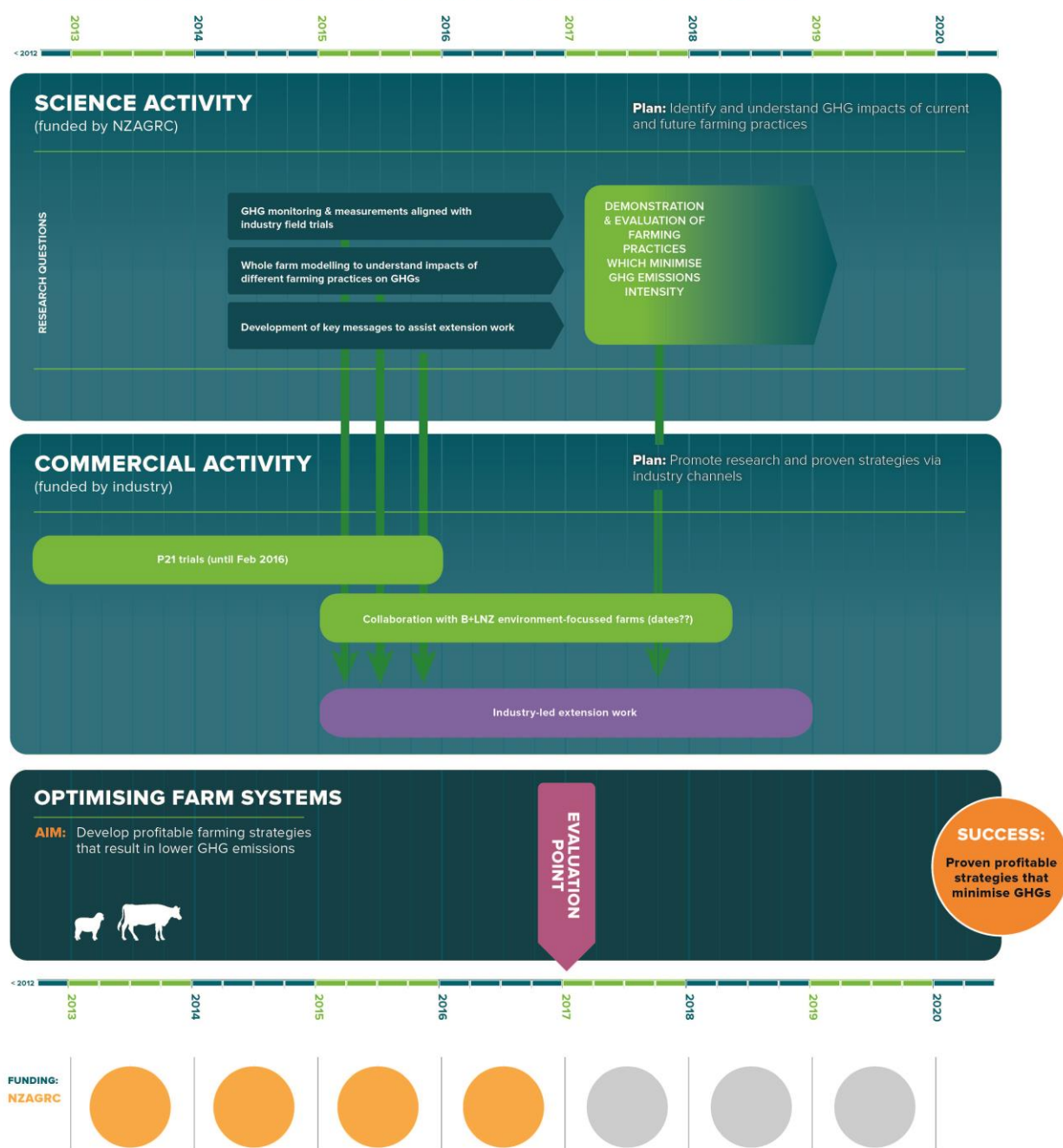
The sheep and beef programme will identify the drivers of GHG emissions intensity on two S+B farms and determine whether these drivers result in decreased GHG emissions intensity when integrated into commercial farm systems. The first step has been a farm systems analysis, including GHG and nutrient losses to the environment, using historical and current farm operational data and we are now working on future scenarios including farming within limits, with change to stocking policy and forage supply. This programme commenced late autumn/winter 2015.

The dairy sector programme will assess the GHG emissions for dairy systems demonstrating a range of practical mitigation options including high genetic merit cows (Waikato), diverse pastures and low stocking rate efficient systems (Canterbury) and off pasture systems (South Otago). The programme partners with established P21 farmlet systems in the Waikato, Canterbury and South Otago, and NZAGRC funding will fund additional data collection and analysis of GHG emissions to:

- Assess whether new mitigations within farming systems will also reduce GHG footprint.
- Validate previous farm systems modelling by demonstrating that these new mitigations deliver real GHG benefits within functional and practical farm systems
- Identify risk areas for pollution swapping within the farm system.

Measurements commenced in autumn 2015.

RESEARCH PIPELINE: OPTIMISING FARM SYSTEMS



Māori-focussed Research Programme Report - 2014/15

Project Manager: Phil Journeaux



This programme aims to assist the Māori pastoral sector to improve its collective capacity to increase resource efficiency and farm productivity while lowering greenhouse gas emissions.

The approach is to develop a set of Māori farm typologies which represent the predominant pastoral farming systems, identify key factors that underpin farm productivity, resource and emission efficiency and sustainable profitability, and then identify and test a range of mitigation strategies. Farm typologies are important to avoid the problems of homogenizing a heterogeneous group that range from very small farms to large multi-enterprise corporates. These typologies will be compared against existing databases and help in the selection of in-depth representative case study farms where emissions from alternative farm system configurations will be evaluated. Two metrics will be used in parallel to identify and assess mitigation scenarios, their costs, and options for implementation: absolute reduction in GHG emissions, and reduction in emission intensity (the ratio of emissions per unit of output, e.g. Kg CO₂-eq/KgMS).

Farm system mitigation scenarios will be based on the interaction and knowledge sharing that will occur between the farmers (including land entities), scientists and industry advisors that will take place in case study workshops around the country.

A key contribution to the literature will be an enhanced understanding of the Māori farm typologies with economic, environmental, social and cultural implications of low emission farming systems within the Māori sector, with wider implications across NZ.

Progress to date includes:

- Development of a typology of Māori farming
- The collection of farm and GHG emission profiles on 29 Māori farms from around the country. This includes 18 sheep & beef farms and 11 dairy farms
- The selection of 4 focus farms; 2 dairy (Bay of Plenty, Taranaki) and 2 S&B (Northland, East Coast)
- The development of Farmax files for each focus farm to allow for farm system modelling, and Overseer files to (a) establish the base GHG emission profile and (b) model the impact of change scenarios
- Data collated to allow for national benchmarking of the emission profiles
- Agreement gained from Trustees of the 4 focus farms for participation in the project
- Development of priority scenarios for modelling of change in farm systems and subsequent impacts on GHG emissions.

FINANCIAL SUMMARY

\$

EXPENDITURE	
<u>Core research spending</u>	
Methane	1,627,395
Nitrous Oxide	1,202,500
Soil Carbon	964,547
Integrated Farm Systems	742,075
Māori	300,793
<u>Research Total</u>	4,837,310
<u>Other research costs</u>	
Additional Fellowships and Studentships	66,000
Planning, engagement & knowledge transfer	95,425
Policy support	144,800
Special IT and communications	35,902
<u>Other Total</u>	342,127
<u>Administration</u>	584,838
<i>Total Expenditure (actual)</i>	5,764,275
<i>REVENUE*</i>	6,071,424
<i>Balance unspent carried over**</i>	307,149

*Includes \$1,221,424 carried over from 2013/14.

**\$105,000 of unspent funding committed to a soil carbon PhD scholarship (candidate starts Oct 2015) and \$137,000 reserved for Phase 2 of Overseer GHG module update (Phase 1 finishes 31st August 2015).

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NZAGRC Director

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NZAGRC Operations Manager

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Project Analyst

Dr Andy Reisinger
Deputy Director (International)

Dr Victoria Hatton
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Vacant at 30 June 2015
Tania Brown from Sept 2015
NZAGRC Administrator

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Chair
Research Director
AgResearch

Dr Rick Pridmore
Strategy and Investment Leader for
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DairyNZ

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Landcare Research

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Professor Mike Hedley
Professor Soil and Earth Sciences
Massey University

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Chief Scientist, Atmosphere, Natural Hazards &
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APPENDIX 1 – COMPOSITION OF NZAGRC SG, ISAG and MAG

Compositions of the SG, ISAG and MAG

The tables below set out the compositions of the SG, ISAG and MAG and the dates of governance meetings held during the course of the financial year.

Steering Group

Four Quarterly meetings were held in 2014/15 (19th August 2014, 19th November 2014, 19th February 2015 and 20th May 2015).

Name	Organisation
Prof. Warren McNabb	AgResearch (Chair)
Dr Rick Pridmore	DairyNZ
Dr Peter Millard	Landcare Research
Dr Stefanie Rixecker	Lincoln University
Prof. Mike Hedley	Massey University
Dr Murray Poulter	NIWA (to 31 Dec 2014)
Dr Rob Murdoch	NIWA (from 1 Jan 2015)
Mr Warrick Nelson	Plant & Food Research
Mr Mark Aspin	PGgRc
Dr Brian Richardson	Scion
Dr Gerald Rys	MPI (Observer*)
Dr Marc Lubbers	MBIE (Observer*)
Dr Andrea Pickering	MPI (Observer**)

*MPI and MBIE hold Observer (non-voting) positions on the Steering Group.

**Dr Andrea Pickering was invited to attend SG meetings in 2011/12 following recommendation from MPI that an Alliance representative attend SG meetings to ensure coordination.

International Science Advisory Group

The ISAG conducted a formal review of the NZAGRC-PGgRc methane programme in April 2015. The group were also invited to attend the conference and workshops following the review to keep abreast of developments in the other NZAGRC-funded programmes.

Name	Organisation
Prof Dorian Garrick	Iowa State University, USA
Prof Keith Goulding	Rothamsted Research, UK
Prof Peter Grace	Queensland University of Technology, Australia
Dr Tim McAllister	AgCanada, Canada
Dr Frank O'Mara	Teagasc, Ireland
Prof Pete Smith	Aberdeen University, UK
Prof Dick Strugnell	University of Melbourne, Australia

Membership runs from 1 March to 28 February in each year.

Māori Advisory Group

Two meetings held in Palmerston North with some members joining by TC/VC (10th September 2014 and 13th February 2015).

Name	Organisation
Lorraine Stephenson	Independent
Jamie Tuuta	Māori Trustee
Dr Tanira Kingi	AgResearch
Tony Finch	DairyNZ
Marino Tahī	Landcare Research
Prof. Hirini Matunga	Lincoln University
Dr Nick Roskrige	Massey University
Alby Marsh	Plant & Food Research
Richard Burton	Scion
Erica Gregory	MPI

APPENDIX 2 – ANNUAL OBJECTIVE SUMMARY SCIENCE REPORTS (SUBMITTED)

Objective Level Summary – 2014/15

Key:

Objective completed
Objective on track
Objective on track with agreed revisions
Objective on track apart from publications
Current issues with Objective (e.g. behind on experimental work)

Those programmes marked with a dagger (†) are co-funded with the PGgRc and/or PGgRc/MPI and those marked with a diamond (◊) are solely funded by the PGgRc. Those with no markings are solely NZAGRC funded.

Area	#	Objective Title	Objective Leader	Objective Leader Organisation	2014/15 \$NZ NZAGRC (GST excl)	Status End 2014/15
Methane	5.1 [†]	Breed low methane ruminants	S Rowe & A Jonker	AgResearch	338,000	Delayed publications
	5.2 [†]	Identifying low GHG feeds	D Pacheco	AgResearch	40,000	Extra work added runs until Dec 15
	5.3 [†]	Vaccine	N Wedlock	AgResearch	280,000	
	5.4 [†]	Identify inhibitors that reduce ruminant methane emissions	R Ronimus	AgResearch	357,500	Extra work added runs until Sept 15
	5.5 [†]	Microbial genomics to underpin methane mitigation	E Altermann & S Leahy	AgResearch	230,000	
	5.6 [◊]	Microbiology to underpin methane mitigation	S Kittelmann & P Janssen	AgResearch	0	
	5.7 [†]	Understanding the low methane rumen	M Tavendale & G Henderson	AgResearch	89,000	PhD due date extended
	5.8	Modelling rumen methane production	D Pacheco	AgResearch	200,000	Paper due date extended
	5.9	Dairy housing methane capture and mitigation by soil	S Saggar	Landcare Research	55,000	
Nitrous Oxide	6.1	Plant Effects on N ₂ O Emissions	S Bowatte	AgResearch	276,500	
	6.2	Denitrification Processes	S Saggar	Landcare Research	90,000	Contract extended to Dec 15
	6.3	Feed management options for mitigating N ₂ O emissions from grazed systems	C de Klein	AgResearch	447,000	
	6.4	Additional N ₂ O work	H Di, J Luo & T van der Weerden	Lincoln University, AgResearch	154,000	

Soil Carbon	7.1	Manipulation of carbon inputs to stabilise and enhance soil carbon stocks	D Whitehead	Landcare Research	409,000	Delay in recruiting PhD student. Student to start Oct 2015
	7.2	Tools to quantify the stabilisation capacity and vulnerability of carbon in grassland soils	F Kelliher	AgResearch	276,000	
	7.3	Modelling management manipulations using the HPM	J Rowarth	Waikato University	75,000	
Integrated Farm Systems	8.1	GHG Emissions on Sheep and Beef Farms	R Dynes & K Hutchinson	AgResearch	124,025	Minor delays in protocol development
	8.2	GHG Emissions from Dairy Systems	R Dynes & K Hutchinson	AgResearch	618,050	Farm related issues to negotiate with possible new site for some CH ₄ measurements
Māori	20.1	Low emissions for the Maori sector	P Journeaux	AgFirst	300,793	Delayed publication

*Minor revisions agreed to Methane 14/15 work plans following Annual Review meetings in May 2014 and 2015. Methane 15/16 work plans will be finalised and new contracts issued in early 15/16.

5.1 - Breed low methane ruminants



Jointly supported programme

Objective Leader – Drs Suzanne Rowe & Arjan Jonker (AgResearch)



The aim of this research is to understand the genetics of host control of ruminant methane emissions. If this is successful, the aim is to develop and make genetic and genomic selection technologies available to reduce methane yield (gCH₄/kgDMI) and methane intensity (gCH₄/kg product) in sheep by June 2016 and in cattle by June 2018. This would be via a beta test format with subsequent full scale industry implementation.

Current research priorities for the next 2 years are:

- Determine the genetic correlation of methane emissions with maternal production traits and anatomical changes in the rumen;
- Test the stability of the results across a variety of feedstuffs and ages;
- Improve genomic predictions and attempt to localise loci with major effects.

In order to undertake these tasks a structured approach has been taken:

- Determine the heritability and repeatability of methane emissions using a wide range of New Zealand maternal sheep;
- Estimate genetic and phenotypic relationships with economically important traits;
- Create divergent selection lines and examine the detailed changes in: anatomy, emissions at different ages, feeding levels and feed stuffs, production traits in young and adult sheep, including carcass and meat quality traits;
- Make the selection lines progeny and samples available for other research e.g., rumen microbial changes, vaccine and forage trials as required.

The second component is to extend these results into industry via:

- Development and validation of low-cost rapid measurement technologies.
- Implementation via genomic selection which is being separately developed in sheep and cattle industries, because it can be applied at effectively zero additional cost to the breeder.
- Development of appropriate selection index weighting based on expected long term methane emission costs.

An important aspect of using genetic change is that it is slow, but permanent and cumulative. As a consequence, it is important that on-going monitoring of genetic changes in other traits is undertaken to detect any unfavourable changes at an early stage. Sheep are being used for initial development as they are markedly cheaper and we expect broad consistency of results across other ruminant species.

After an initial pilot project in 2008, a formal research programme commenced in with 2009 born progeny. To date heritability and repeatability of methane emissions has been estimated and published, as have correlations with production traits to one year of age. The measured animals and subsequent selection lines are being monitored for responses at later ages and detailed studies of anatomical changes are being undertaken. Similarly, studies have commenced using rapid measurement techniques and examining responses on pasture under a variety of

physiological ages and seasons. Sufficient genotyping has been done to identify certain host genome regions associated with methane emission and validate that genomic selection can be undertaken.

5.1 – Progress in 2014/15

Methane is a waste product generated by methanogenic archaea in the rumen of the host. We have shown that methane varies both with the genetics of the individual ruminant host and with the microbial populations present in the rumen. Genetic and genomic selection offers a potential method to reduce gross ruminant methane emissions (g CH₄/d), methane yield (g CH₄/kg dry matter intake) and to improve methane efficiency ((g CH₄/kg end product). Our previous work has shown that gross methane emissions and methane yield are heritable and repeatable traits when measured in respiration chambers. These results led to the development of selection lines of low and high methane yield animals. The work undertaken in the current year has focused on continuing this selection for divergent methane phenotypes. The current cohort differ in methane yield by ~7% in respiration chambers and by ~14% using a proxy for methane yield when measured in portable accumulation chambers (PAC) off pasture.

The lines are used as a research tool to monitor the effects of breeding for methane yield on a variety of physiological and productive traits. This year we have included maternal traits such as body condition score and number of lambs born. For methane yield, no genetic correlations were significant but they reflected trends seen in previous published work where higher fat content and lower dressing out percentage were associated with higher methane yield. Research in the current year also validates previous findings that, under the current SIL index, lower methane emitting animals having higher EBVs and the expectation of greater profit. This gain in profit comes from a tendency for low emitters to have a higher dressing out percentage and leaner growth. The respiration chamber measures are also used as a standard to evaluate alternative methods for monitoring of methane in the commercial setting. The many thousands of methane measures required for a commercial breeding program would be prohibitively difficult and expensive to collect using respiration chambers. Furthermore, the monitoring of methane emissions in the respiration chamber environment, although extremely accurate, may not be the ideal scenario for ranking animals at pasture. For an effective breeding program to reduce methane on a national/international scale, alternative predictors are needed.

Research has been carried out to determine the effectiveness of genomic selection, rumen microbial profiling, volatile fatty acids (VFA), and PAC chamber measures as alternative predictors for animals at pasture under commercial production systems. We have shown that distinctive VFA profiles exist with high emitting animals having a high acetate phenotype seen across multiple scenarios. Although it is known that methane production varies with rumen microbial community structure, it was not known whether the host genetics controlled the structure of RMC nor whether RMC could be used to predict methane directly. Further evaluation of rumen microbial populations present in low and high emitting animals has now been carried out and in particular research into how to statistically handle this complex source of information. Reducing this information to as few as two dimensions and looking at genetic correlations with methane yield suggests that RMC could be highly predictive of the methane status of the animal. Although preliminary and based on low animal numbers, these results may present a further low cost method that can potentially be used across species.

To validate that differences in the low and high methane selection lines were still present under typical New Zealand grazing conditions, trials using the SF₆ tracer gas technique and PAC were carried out. These measures enabled a greater number of animals in different physiological states and on different feeding levels to be monitored. The portable chambers were used to measure methane across different seasons, in young and adult animals and for production animals on commercial properties.

The greatest challenge of monitoring methane yield or methane emitted per kg of feed ingested (g CH₄/kg DMI) at pasture is obtaining an accurate measure of feed intake. CO₂ information obtained

from animals in respiration chambers consuming a pelleted diet were evaluated as a proxy for intake. Results showed that the total gas emitted by an animal ($\text{CO}_2 + \text{CH}_4$ expressed in moles) could be used as a proxy for voluntary feed intake. Further analysis of the data using this proxy and CO_2 information obtained from PAC chambers and a grazing trial where gas was collected in a yoke over a 24 hour period attached to the animals neck inferred that the low methane emitting animals that had been selected on a fixed feeding regime in respiration chambers, could in fact be eating 8% more than the high emitters on pasture. This finding is being followed up via a comprehensive 6-month study of the high and low selection lines in which pasture intake is ad-lib

Key achievements for 2014/15:

- Invited talk on genomic selection for methane at the World Congress in Genetics Applied to Livestock Production. Co-organised the ASGGN satellite meeting attended by 98 registrants.
- SF_6 trial showing that methane yield differences between lines persist at pasture
- Results showing that rumen microbial profiles can be reduced to 2 dimensions and were highly correlated with methane yield and contain a heritable and repeatable component.
- Initial results indicate that moles of total gas may be used as a proxy for feed intake and this is a heritable trait even after adjustment for liveweight when used in PAC chambers on animals grazing pasture.
- Invitation to present at NZSAP contract session exemplifying the use of genomics in the NZ sheep industry.

5.2 - Identifying low GHG feeds



Jointly supported programme



Objective Leader – Dr David Pacheco (AgResearch)

We aim to develop feeds and feeding strategies that result in reduced GHG emissions from ruminants. Nutritional strategies will take advantage of desirable characteristics of different forages, feed crops and feed ingredients to design appropriate feeding systems. Such feeds and feeding systems will then direct rumen fermentation towards pathways that are conducive to reduced methane and increased nitrogen utilisation efficiency. The devised nutritional strategies will provide benefits on GHG that are above and beyond any benefits in emission intensity achievable via increased productivity when current feed supplementation practices are used. We will not only identify nutritional strategies that work, but also understand the mechanisms behind the GHG reductions so that low GHG feeding systems can be designed for current and future markets of livestock feed.

Initially, we have used the leads from forage brassica experiments to develop hypothesis of mechanisms underpinning the reduction in GHG emissions. From a mechanistic perspective, the initial lines of enquiry have been the quantification of the contribution of pH and carbohydrate structure as factors affecting fermentation patterns leading to lower methane production in the rumen. The purpose of understanding these rumen processes is to design manipulations of rumen fermentation to reduce GHG emissions, while maintaining or increasing animal productivity. In all animal experiments, nitrogenous aspects of rumen fermentation are being studied to ensure that feeds and feeding strategies will deliver reductions for both methane and nitrous oxide.

The knowledge generated will be used by scientists in the NZAGRC-PGgRc programme. For example, it is being used to help define the role of digestive processes as a contributor to the

variation in animal to animal methane emissions and also to understand their potential role as modulator of responses to methanogen inhibitors and vaccines.

In the medium term, knowledge generated from this objective will be used by nutrition scientists, applied nutritionists, forage and crop breeders to develop feeds, feeding practices and forages that will result in reduced methane emissions from the process of feed digestion.

5.2 – Progress in 2014/15

The work in the last year continued to build on the finding that feeding some brassicas results in lower methane emissions when compared with perennial ryegrass diets. The work in this objective has sought to i) understand the mechanisms behind the low methane emissions measured from brassicas, using forage rape as the 'model' crop in indoors trials, ii) use this understanding to identify or develop feeds with methane mitigation properties.

- We evaluated methane emissions from sheep offered different mixes of forage rape (cultivar Titan: 0, 25, 50, 75 and 100%) with the remainder (on dry matter basis) offered as ryegrass-based pasture. Methane emissions from sheep linearly decreased with the levels of forage rape included in the diet. It suggests that reductions in methane emissions observed in brassicas occur through a different mechanism to those elicited by feeding grains. Understanding the interactions between feeds require further research to progress our understanding on ways to reduce methane emissions from ruminants in practical feeding situations.
- A Feeds Workshop gathered scientists involved in research related to feeds and GHG, industry stakeholders (nutrition consultants, seed companies) and funder representatives to discuss trends in feed usage across the NZ livestock systems. A key conclusion reached at this workshop was that the feed base for future ruminant systems in NZ will continue to be predominantly feeds grown on-farm or locally. The home-grown feed will be used as a tool to mitigate risks related to supply, quality and cost of imported feed. A key knowledge gap identified is "understanding pasture quality", because the quality of ryegrass-based pasture has a large range, making it difficult to have general recommendations for improvement of nutrient supply to ruminants. The use of alternatives to ryegrass, such as herbs and crops, will grow in importance. Thus, it will be important to understand their nutritional value both as monocultures and in mixes with other feedstuffs. Because of the extremely large number of possible combinations of feeds and ruminant species, an implication is that the mechanistic understanding of the contribution of different physical and chemical characteristics of feeds will be critical to predict methane emissions and animal production in New Zealand livestock systems.
- Fodder beet as a 'low GHG' feed: Informed by the findings from the brassica work and the discussion with nutrition stakeholders, we have evaluated methane emissions from fodder beet. Out of the 'emergent' crops identified in the industry workshop, fodder beet was selected based on its high ratio of readily fermentable to structural carbohydrates. Also, due to its low nitrogen content, fodder beet has been the focus of allied research programmes aimed at minimising nitrate leaching. In one trial, feeding 90% of the diet as fodder beet reduced methane emissions by 50%, compared to ryegrass. We are currently conducting experiments to validate this observation and to assess responses in methane emissions across a wider range of fodder beet inclusions.
- Analysis of our combined data indicate that, within and across feeds, there is a wider variation when methane emissions are expressed per unit of digestible organic matter than when expressed per unit of dry matter intake. Because organic matter digestibility is a better descriptor of feed quality than any of the feed chemical constituents alone or combined, our assumption that feed quality (expressed in terms of chemical composition variables) does not explain variation in methane emissions requires revision. We have assembled data that allow exploration of the relationship between organic matter digestibility and metabolisable energy (ME). This relationship is expected to be strong.

- We conducted an initial assessment of the potential of near-infrared reflectance spectroscopy (NIRS) to predict methane yields from forages fed to ruminants. Although modest, the predictive ability of the NIRS calibration could be considered useful for screening purposes, particularly in terms of the predicting the potential methane yield of a feed, rather than predicting it from a particular feeding experiment. It is recommended that the databases for NIRS calibrations are expanded by collecting feed information from future experiments in which methane emissions are measured.

Key achievements for 2014/15:

- Our summary of work on dietary mitigation practices was presented at the combined International Symposium of Herbivore Nutrition/International Symposium in Ruminant Physiology, held in Canberra Australia in September 2014. This was the Stobbs Memorial Lecture, which was the opening of the conference.
- Our study investigating the mechanisms behind the reduction in methane emissions observed when forage rape was fed has been published in PLoS ONE (Impact factor 3.53). This paper describes a variety of factors including feed composition, rumen fermentation, animal digestion physiology and rumen microbiology to determine their potential involvement in the consistent reduction of methane emissions from ruminants fed forage rape.
- We have generated evidence of the potential utility of near-infrared reflectance spectroscopy (NIRS) to do a rapid assessment of the methane yield potential of a feed. Despite the low number of samples included in our study with NIRS, we were able to obtain a predictive model that explains twice as much of the variance in methane yield relative to multiple regression models.
- We have been able to translate knowledge generated as part of our brassica studies into identifying fodder beet as a potential low GHG feed. In the first study, feeding 90% of fodder beet in the diet, mixed with ryegrass was able to reduce methane emissions by 50%. We are currently following up and confirming results from our first study.
- Our research on methane emissions from ruminants fed brassicas has been covered in public media and farmer magazines. For example:

<http://www.stuff.co.nz/business/farming/discovery/63941914/Methane-cut-link-explored>.

5.3 – Vaccine



Jointly supported programme

Objective Leader – Drs Neil Wedlock & Art Subharat (AgResearch)



The immediate goal of the vaccine programme is to produce a prototype vaccine that has efficacy in either sheep or cattle such as a change in methanogen communities in the rumen. Further development of the vaccine (by optimising antigens, adjuvants and delivery) will lead to a vaccine which is targeted at reducing methane emissions in sheep and cattle by at least 20%.

The programme in Y13-14 aimed to provide antigens and adjuvants for a prototype vaccine and 'proof-of-concept' that a vaccine will work, i.e., sufficiently high levels of antibodies can be produced following vaccination of sheep or cattle and there is evidence of vaccine-mediated changes in methanogen populations in the rumen. To achieve this, experimental vaccines formulations, consisting of new antigens selected by bioinformatics analysis of genomes from the most rumen-abundant methanogens, and formulated with current 'best' adjuvants will be

administered to sheep and cattle. In each trial, animals were monitored for their antibody responses to the methanogen antigens, anti-methanogen activity measured in *in vitro* assays and rumen microbial profiling undertaken to determine antibody induced changes in microbial populations in the rumen.

From these vaccine trials we aim to determine:

1. Which is the best model – sheep or cattle? After two trials, we expect to understand if cattle are more promising, or if sheep are the most suitable species for further research.
2. Do the adjuvants increase salivary IgA, and ruminal IgA (and other classes of antibody) resulting in very high levels of antibody in the rumen?
3. Do the serum antibodies inhibit the target methanogens in pure culture?
4. Do any combinations of adjuvant and antigen change the ruminal methanogen community?

The scheduled work makes efficient use of resources and effectively makes every set of experiments an animal trial, and also a search for further ‘proof-of-concept’.

Because of the structure of the process, if both the right antigen and the correct adjuvant are administered, positive results will be gained for points 2 and 3, and possibly 4. If the right adjuvant is combined with an ineffective antigen, increased IgA (or IgG) will be measured in the saliva and rumen (point 2), but there will be no impact on pure cultures (point 3) or on methanogens in the rumen (point 4). If an effective antigen is tested with an ineffective adjuvant, results from points 2 and 4 will be negative, but from point 3 will be positive. In each round of trials, we will formulate the best antigens with the best adjuvants and test those combinations, and also introduce new antigens or adjuvants.

Once we have obtained positive results in point 4, we will have the next ‘proof- of-concept’ step needed. Depending on the nature/magnitude of the change in the rumen methanogen community, we can then proceed to conduct a larger vaccination trial in either sheep or cattle with quantification of the reduction in methane emissions using respiratory chambers. This will be negotiated with the funders, since it may require reallocation of resources, and changes in milestones.

The prototype vaccine arising from research will be available for scientists within the NZAGRC-PGgRc programme to develop further and along with critical knowledge of vaccination (antigens/adjuvants/route of vaccination and relevant IP will be available for entering negotiation with an Animal Health Company for commercialisation.

5.3 – Progress in 2014/15

A technical problem with the *in vitro* methanogen culture assay used to evaluate antisera produced against potential vaccine targets has been solved by a modification to the protocol. This will now enable us to continue using the *in vitro* pure culture system for testing antisera produced against new candidate vaccine antigens and also retest some of our existing antisera as appropriate. Using the modified assay, antisera against two new vaccine targets have shown inhibitory effects against the model rumen methanogen *Methanobrevibacter ruminantium* M1.

The presence or absence, degree of conservation as well as the level of expression in the rumen of a chosen vaccine candidate gene across different methanogens are all critical for the successful selection of vaccine antigens. The programme is using all genomes from the previous genome sequencing work performed in this and earlier years to identify conserved targets and antigen motifs. The two best candidates identified to date were further analysed and this has provided important information on the suitability of these potential targets for a vaccine. A comparative analysis of one of the lead antigens in known rumen methanogen genomes has revealed the presence of novel and highly conserved sequence motifs and these were tested further in the four animals trials (described below). A strategy for testing the other lead target, to determine its

suitability for a prototype vaccine was developed and a vaccine trial in sheep is being conducted (described below).

Four vaccine trials (3 in sheep and 1 in cattle) were conducted to follow up on promising results observed with the lead vaccine antigen that showed promising results from trials conducted in 2013-2014 (PGgRc-NZAGRC and GPLER). These four animal trials tested different variants of the antigen with adjuvants. Sera, saliva and rumen contents were collected from all vaccinated and control animals pre- and post-vaccination for analysis. Antibody levels in sera and saliva in the animals from these trials were of a similar level and followed similar patterns of antibody response to animals in the previous trials that provided the promising leads. Strong serum and salivary antibody responses were observed in all vaccinated animals with evidence of cross-reaction of antibody between different methanogens. A 2-fold higher dose of the antigen was used to vaccinate the cattle compared to the 'standard' dose used for sheep. This dose produced strong antibody responses in cattle but may still require further optimisation to achieve maximal antibody responses in these ruminants. Rumen microbial analysis indicated that there were no differences in rumen methanogen or bacterial populations between the vaccinated and control groups in three of the four trials. However, in one trial there was an apparent effect in some of the vaccinated animals at 6 weeks post-vaccination with a relative decrease in abundance of the target methanogens, consistent with a vaccine effect. To follow up this promising result, the sheep from this group were revaccinated with the same vaccines and route of vaccination and additional rumen content samples were collected 2 weeks after the boost, for analysis. This is expected to be completed within the next 2 months.

A vaccine trial was conducted in sheep to identify additional potential vaccine antigens, produced as either peptides or recombinant proteins. A comparison was made between immune responses produced in animals vaccinated with different formulations, two different doses of vaccine (standard and 10-fold lower) and between two vaccination routes. The results have produced important information on dose and route of vaccination in sheep. While strong antibody responses were observed in all animals vaccinated with the various targets, there was animal-to-animal variation in antibody responses. Results from testing antisera in the *in vitro* methanogen cultures indicated that antibodies against one of these targets inhibited the target methanogen, suggesting that this antigen is a promising lead antigen. This antigen will now progress to larger animal trials and further analysis has commenced to identify the best variants of this target. An analysis of rumen microbial profiles in the vaccinated animals from this trial is underway and results are expected within 1-2 months. A second vaccination trial in sheep is being conducted to further investigate another antigen as a candidate for a prototype vaccine. This trial is testing variants from multiple methanogens. Sera, saliva and rumen contents have been collected for analysis of antibody responses and rumen microbial populations.

Key achievements for 2014/15:

- Two vaccine candidates have progressed to larger scale vaccine trials.
- Two potential new targets have been tested in sheep with promising results. Antisera against some of these targets have shown inhibitory effects in *in vitro* methanogen culture assays and these targets will now be tested further using larger groups of animals.
- Improvements have been made to the *in vitro* methanogen pure culture assay system for methanogen which has meant that routine testing of antisera from vaccinated animals can occur. An *in vitro* culture assay has been established for *Methanobrevibacter thaueri* D5.
- Data on targets and vaccine formulation have been provided for patenting purposes.

5.4 - Identify inhibitors that reduce ruminant methane emissions



Jointly supported programme

The aim of this objective is to develop cost-effective inhibitors that reduce methane emissions by at least 20% in sheep and cattle without reducing productivity. The research will use an already established “pipeline” that uses rumen methanogen enzyme assays and enzyme structures to screen for inhibitors in a cost-effective and rapid way. The pipeline has produced a number of “hits” (compounds that inhibit in simple tests). Taking these compounds through to testing in animals has the highest priority. The discovery of novel inhibitors will be also accelerated using the developed enzyme assays to screen large compound libraries for potential inhibitors (5,000 compounds) and by enhancing the efficacy of hit compounds using drug design techniques. The compounds will be tested in animal trials for efficacy and productivity effects. The data from the animal trials will be used to engage a commercial partner (or partners) for the development of appropriate technologies for delivering the inhibitors.

5.4 – Progress in 2014/15

The inhibitor development programme seeks to discover specific and novel small molecule inhibitors of rumen methanogens. Methane emissions from ruminants are produced by a unique group of archaeal microorganisms termed methanogens and are widely recognised as contributing to climate change. Inhibitors of rumen methanogens are likely to play a significant role as one of a suite of strategies used in methane mitigation. Inhibitors offer several advantages in that they can be easily produced, enabling rapid uptake on a global basis, incorporated into slow release boluses, and are able to cause large reductions in methane.

Rumen methanogens are the only Archaea in the rumen, and are quite different in a number of features compared to the fibre-degrading bacteria, fungi and protozoa. Such methanogen features include cell wall structures, lipids, metabolic cofactors, and unusual metabolic pathways including amino acid synthesis and their signature methane production pathway. Methanogens represent only about 1-3% of the rumen microbial community. Due to the unique metabolic pathways of methanogens, and their relatively low abundances, it is thought that methanogen-specific inhibitors can be developed that do not adversely affect the fermentation of feed. Identification of suitable inhibitors within the programme involves several screening strategies including: (1), high-throughput direct enzyme assays; (2), *in silico* screening of large compound libraries using methanogen enzyme structures; (3), pure culture-based 96 well plate assays; and (4), rumen fluid-based assays (both a miniaturised format, developed in a MPI Global Partnerships in Livestock Emissions Research 2 (GPLER 2) programme and in 60 ml volume methods). The latter two methods simultaneously screen all ~500 essential genes and enzymes of the rumen methanogens in the assays. The above methods are complementary to each other and all have yielded active inhibitors for further testing.

Derivatives of any promising hit compounds identified using the various methods are ordered off the shelf or custom synthesised in an attempt to improve the potency and other properties such as stability and toxicology. Derivatives can potentially lead to dramatic increases in potency of 1,000-10,000-fold. The ultimate goal is to move compounds that have confirmed activity in rumen fluid-based assays to short-term, and then long-term animal trials.

In the last year, good progress has been made in several milestones relating to the programme: animal trial testing; rumen *in vitro* assay testing; enzyme preparation and assay development; enzyme structure development; *in silico* screening; enzyme assay screening; screening using pure cultures of methanogen; and longer term trials with promising candidate inhibitors. Over a million compounds were screened *in silico* against newly determined methanogen enzyme structures, tens of thousands against enzyme or pure cultures, and thousands using rumen fluid-based methods. This has identified >100 compounds of interest, currently at various stages of development. The most promising of the compounds have been assessed in short term animal trials, which confirmed the ability of five of the compounds to significantly reduce methane emissions (in one case showing >90% inhibition of methane emissions) and to be safe to animals.

These were highlighted in the media through multiple broadcast, printed, and online outlets, including comment by MPs after the results were presented at the New Zealand Agricultural Greenhouse Gas Mitigation Conference 2015 in Palmerston North on April 28, 2015. The data are preliminary in that the next stage in the research would be to test the compounds in longer-term animal trials. The demonstration that high levels of methane emission mitigation can be obtained using small molecule inhibitors in an animal-safe manner is very encouraging, and highlights the potential for this technology in aiding the long-term control of ruminant methane emissions for farmers in both New Zealand, and globally.

Key achievements for 2014/15:

- A short-term sheep trial with an animal-safe inhibitor that produced >90% inhibition (despite it only being dosed once-daily in the morning via drenching).
- Four other short-term sheep trials, one 16 day, all showed significant (~20% or greater) methane inhibition, with brief levels of inhibition >90% just after feeding in three cases.
- *In silico* screening of >1,000,000 compounds, >20,000 against enzymes and >15,000 against *Methanobrevibacter boviskoreani* AbM4.
- Identification of numerous other potential inhibitors using *in silico*, direct enzyme screening and AbM4 screening techniques.
- Multiple media announcements associated with the New Zealand Agricultural Greenhouse Gas Mitigation Conference announcement on 'five animal-safe compounds' identified, showing significant methane inhibition.

5.5 - Microbial genomics to underpin methane mitigation



Jointly supported programme

Objective Leader – Drs Eric Altermann and Sinead Leahy (AgResearch)



A comprehensive set of reference genomes is essential to understanding the role of specific groups of rumen methanogens to methane emissions from ruminants and to underpin current and future vaccine and inhibitor development pipelines. Closed genomes are important in this regard to ensure the entire gene complements of methanogens are included in the comparative gene analyses and to enable genome-wide functional studies via protein identification and RNA sequencing-based technologies.

In two years we will have reference genomes for each of the main groups of hydrogenotrophic (*Methanobrevibacter ruminantium* M1 and *Methanobrevibacter olleaye* YLM1 (both members of the “ruminantium” clade, *Methanobrevibacter millerae* SM9 and *Methanobrevibacter thaueri* D5 (both members of the “gottschalkii clade”, *Methanobrevibacter boviskoreani* AbM4 and *Methanobacterium formicicum* BRM9) and methylotrophic rumen methanogens (*Methanosarcina barkeri* CM1, *Methanosphaera* sp. ISO3-F5, and *Methanomassiliicoccales* strains ISO4-H5, ISO4-G1 and ISO4-G11). Comparative genome analysis is being used to inform target selection in the established vaccine and chemogenomic pipelines and to confirm the presence and variability of the target genes in the methanogen population. Additionally, the information is available to underpin future mitigation approaches, in particular, the interpretation of metagenomic datasets from high and low methane emitting sheep. The genomes of the three strains from the poorly-studied *Methanomassiliicoccales* are a particular focus, as their metabolism and physiology is not well understood, and they are not yet well targeted by either the vaccine or inhibitor pipelines. The

current PhD thesis research by Mr Yang Li on the ISO4-H5 genome will improve knowledge in this area.

To maximize the value of the genome sequences and to improve the international reputation of the PGGRC and NZAGRC methane mitigation programmes, we plan to publish the genomes in peer reviewed journals after IP assessment and where appropriate, patent protection.

5.5 – Progress in 2014/15

The rumen is home to a complex community of bacteria and archaea (the microbes that produce methane). To develop effective products that inhibit these archaea – known more specifically as methanogens – knowledge of their genetic background is paramount. Specifically, which genes are critical for the survival and fitness of methanogens, and what is their genetic diversity?

To address these questions, rumen methanogens have been isolated and grown in pure (or enriched) cultures. DNA isolated from these archaea has been used to determine entire genome sequences of a number of key microbes. Presently, the genomes of *Methanobrevibacter ruminantium* M1, *Methanobrevibacter olleyae* YLM1, *Methanobrevibacter thaueri* D5, *Methanobrevibacter millerae* SM9, *Methanobacterium formicicum* BRM9, *Methanobrevibacter boviskoreani* AbM4, *Methanosarcina barkeri* CM1 and *Methanosphaera* sp. ISO3-F5 (not yet annotated) have been assembled, annotated and analysed for their metabolic capabilities, their cell surface proteins and for the presence of other key genetic elements suitable for methane mitigation. Further, enrichment cultures of the novel methanogen order *Methanomassiliicoccales* (ISO4-G1, ISO4-G11 and ISO4-G5) are being similarly sequenced, assembled and analysed. In addition, we have closed, annotated and characterised the genome of *Eubacterium limosum* SA11, a rumen acetogen.

After evaluation and protection of commercially valuable targets, manuscripts reporting the genome sequences of *Methanobacterium formicicum* BRM9 (2014), *Methanobrevibacter boviskoreani* AbM4 (2013), *Methanosarcina barkeri* CM1 (2015), *Methanobrevibacter millerae* SM9 (2015) and *Eubacterium limosum* SA11 (2015) have been submitted and/or accepted for publication.

With the emerging complexity of the rumen methanogen community, it is essential that vaccine and chemogenomics solutions target the widest possible range of methanogens. Furthermore, although genes and proteins may be conserved, genetic variation is common among microbes and such variation may reduce the effectiveness of any vaccine if not recognised. We therefore have initiated and are maintaining one of the world's largest comparative datasets on archaea and (rumen) methanogen genomes with the aim to investigate and assess the frequency and variability of priority targets across cultured microbes. To date, 345 archaeal genomes are part of the growing dataset which is regularly updated. However, only a fraction of the archaeal diversity found in the rumen is presently in culture and we have developed tools to investigate the large metagenomic (what is there) and metatranscriptomic (which genes are turned on or off) datasets created by a collaboration between the Joint Genome Institute (USA) and the GPLER "Deep sequencing the rumen microbiome" programme. These datasets contain information on the uncultured methanogens and therefore provide a true reflection of the genetic diversity within the rumen microbiome and methanogens in particular.

Bioinformatic analyses of priority vaccine targets have led to the discovery of two novel extracellular motifs that are highly conserved throughout a range of rumen methanogens. These motifs are currently being tested in *in vitro* assays and animal trials for their effectiveness against pure cultures and in the rumen. Additionally, they are the basis for a new multi-species patent currently being prepared by the PGgRc.

The analysis of available genomic datasets has highlighted that methane mitigation targets identified for a single methanogen may indeed be conserved across a wide range of different methanogen groups – or in contrast, display great diversity even between closely related species.

To optimise our approaches towards effective methane inhibition, we will interrogate selected genetic targets found through the metabolic analysis of available genome information not only for the presence of suitable target domains, but also their abundance and diversity in the entire methanogen community. Domain profiles will then allow us to assess and rank the most likely candidates that will aid the development of technologies to inhibit a broad range of rumen methanogens.

Key achievements for 2014/15:

- Publication of the genome of *Methanobrevibacter boviskoreani* AbM4 (Stand Genomic Sci)
- Publication of the genome of *Methanosarcina barkeri* CM1 (Stand Genomic Sci)
- Publication of the genome of *Methanobacterium formicium* BRM9 (Stand Genomic Sci)
- Manuscript submissions “The complete genome sequence of *Eubacterium limosum* SA11, a metabolically versatile rumen acetogens” and “The complete genome sequence of the rumen methanogen *Methanobrevibacter millerae* SM9” (Stand Genomic Sci)
- Creation of the largest known comparative dataset on archaea (and methanogens).

5.6 - Microbiology to underpin methane mitigation



PGgRc sole funder

Objective Leader – Drs Sandra Kittelmann & Peter Janssen (AgResearch)



The microbiology objective analyses microbial community structure in rumen samples from animal trials to evaluate the effects of methane mitigation strategies on rumen microbial communities.

Ruminant methane is derived largely from the action of the rumen microbial community. Low methane emitting animals have different microbial community structures from high emitting animals, whether through natural variation or feeding different diets. Understanding the differences in microbial community structure within and between mitigations points to common or different mechanisms that drive the differences in methane. In addition, partial effects on the microbial community, in the absence of methane emissions reduction, will inform vaccine and inhibitor objectives of near successes that deserve to be refined further.

To enable testing on larger numbers of trials envisaged in the overall programme, we will also start using higher-throughput technologies (sampling methods, sequencing methods) to collect these data in a timely, cost-effective and non-invasive way. The outcomes of animal trial analyses will be made available to other scientists within the PGgRc-NZAGRC programme. The outcomes of methodology development will be available to the PGgRc-NZAGRC directly.

5.6 – Progress in 2014/15

Sequencing and analyses of 16S rRNA gene amplicons were performed to estimate the composition of the rumen methanogen community in 252 samples from eight cohorts of sheep and cattle, separated into 16 different sample groups by diet, and to determine which methanogens are most prominent in the rumens of farmed New Zealand ruminants. Methanobacteriales (relative abundance \pm standard deviation, 89.6% \pm 9.8%) and Methanomassiliicoccales (10.4% \pm 9.8%) were the two major orders and contributed 99.98% (\pm 0.1%) to the rumen methanogen communities in the samples. Sequences from Methanobacteriales were almost entirely from only four different species (or clades of very closely related species). Each was detectable in at least 89% of the samples. These four species or clades were the *Methanobrevibacter gottschalkii* clade and *Methanobrevibacter ruminantium* clade with a mean abundance of 42.4% (\pm 19.5% standard deviation) and 32.9% (\pm 18.8%), respectively, and *Methanosphaera* sp. ISO3-F5 (8.2% \pm 6.7%) and *Methanosphaera* sp. group5 (5.6% \pm 5.7%). These four species or clades appeared to be primarily represented by only one or, in one case, two dominant sequence types per species or clade when the sequences were grouped into operational taxonomic units (OTUs) at 99% sequence identity. The mean relative abundance of *Methanomassiliicoccales* in the samples was relatively low but exceeded 40% in some of the treatment groups. Animal feed affected the apparent methanogen community structure of both orders, as evident from differences in relative abundances of the major OTUs in animals under different feeding regimens.

Analysis of rumen microbial community structure based on small-subunit rRNA marker genes in metagenomic DNA samples provides important insights into the dominant taxa present in the rumen and allows assessment of community differences between individuals or in response to treatments applied to ruminants. This information is important to guide development of enteric methane mitigation technologies. However, natural animal-to-animal variation in rumen microbial community composition can limit the power of a study considerably, especially when only subtle differences are expected between treatment groups. Thus, trials with large numbers of animals may be necessary to overcome this variation. Because ruminants pass large amounts of rumen material to their oral cavities when they chew their cud, oral samples may contain good representations of the rumen microbiota and be useful in lieu of rumen samples to study rumen microbial communities. Here, we showed that apparent microbial communities in rumen samples from sheep sequenced using 454 Titanium pyrosequencing and Illumina MiSeq PE300 sequencing were highly similar, and then used the more exhaustive Illumina MiSeq data set to compare rumen microbial communities in the same animals (feeding on four different diets) sampled with a stomach tube and with buccal swabs. After bioinformatic depletion of potential oral taxa from libraries of samples collected via buccal swabs, bacterial communities showed significant clustering by diet ($R = 0.37$, ANOSIM) rather than by sampling method ($R = 0.07$). Similarly, archaeal, ciliate protozoal and anaerobic fungal communities showed a significant clustering by diet and not by sampling method even without adjustment for potential orally-associated microorganisms. These findings indicate that buccal swabs may in future allow quick and non-invasive sampling for analysis of rumen microbial communities in large numbers of ruminants.

We further applied our rumen microbial ecology methods (high-throughput sequencing of microbial marker genes, enumeration of total methanogens by quantitative PCR) to the four research aims and evaluated the effects of different methane mitigation strategies. Results will be discussed in the context of the respective research aims.

Key achievements for 2014/15:

- Identified dominant methanogens in NZ ruminants at species level => limited diversity => impacts RA3
- Established barcoded Illumina MiSeq amplicon sequencing for microbial community analysis => impacts RA1-4
- Validated buccal swabs method for microbial community analysis => impacts RA1

- Applied microbial ecology methods to rumen samples to show effect of mitigation strategies
=> impacts RA1-4

5.7 - Understanding the low methane rumen



Jointly supported programme

Objective Leader – Drs Mike Tavendale and Gemma Henderson (AgResearch)



Methane mitigation technologies must be part of profitable farming systems. Their viability will depend on carbon costs, implementation and material costs, and production impacts. In theory, the reduction of methane emissions per unit of feed intake could result in an increase in production through the capture of metabolisable energy that was previously lost as methane formed from hydrogen produced in the fermentation. This could be through increased production of propionate or other reduced fermentation products or an increase in homoacetogenesis to produce acetate from hydrogen and carbon dioxide. Increased production would improve the uptake and economics of mitigation technologies. In addition, users of technologies will want to be informed of production effects before they use them.

This objective will first predict the impacts of methane inhibition using model inhibitors. As soon as viable vaccines (Objective 5.3) or inhibitors (Objective 5.4) are developed, it will switch to using those to measure production effects.

Work in this objective forms part of a PhD programme being conducted by Ms Preeti Raju.

5.7 – Progress in 2014/15

Ruminant livestock contribute significantly to global greenhouse gas emissions. This is due to rumen microorganisms, known as methanogens, that generate methane from hydrogen and carbon dioxide during the microbial fermentation of feed. Various methane mitigation strategies (vaccines, inhibitors, etc.) are being developed to reduce methane emissions from ruminants. However, inhibiting methane production may cause accumulation of unused hydrogen in the rumen, which may slow down rumen fermentation and thereby affect animal productivity. Homoacetogens, a group of microbes known to reside in the rumen, can use hydrogen and carbon dioxide to form acetate. This process is known as “homoacetogenesis”, and homoacetogens could therefore take over the role of ruminal hydrogen disposal following the inhibition of methanogens.

The aims of this objective are to understand what happens in the rumen when methanogenesis is inhibited, identify alternative hydrogen utilisers, such as homoacetogens, and to quantify their involvement in hydrogen utilisation. To achieve our aim, model inhibitor compounds were used to inhibit methanogenesis in sheep, and the effects on rumen microbial community structure, fermentation and homoacetogenic acetate formation were studied and compared with those of control sheep, in which methanogenesis was not inhibited. Additionally, labelled substrates were added to *in vitro* fermentations of sheep rumen contents and stable isotope probing used to trace their conversion into fermentation products such as volatile fatty acids (e.g., homoacetogenesis was measured *via* incorporation of $^{13}\text{CO}_2$ into ^{13}C -acetate).

We found that inhibition of methanogenesis resulted in

1. a decrease in methane emissions by sheep,
2. a reduction in the number of methanogens and a change in their community composition,
3. a shift in fermentation towards more of the volatile fatty acid propionate and less acetate being formed,

4. bacterial numbers and community compositions remaining stable and broadly comparable with untreated animals,
5. almost a two-fold increase in homoacetogenesis, as evidenced by $^{13}\text{CO}_2$ incorporation into ^{13}C -acetate,
6. greater than 90% of the labelled acetate was formed from CO_2 and H_2 , and not via interconversion of volatile fatty acids such as propionate,
7. homoacetogenesis did not become a significant hydrogen sink after methane inhibition in sheep.

Summary and implications

Homoacetogenesis occurs in the ovine rumen, even when methanogenesis is not inhibited. Ruminal homoacetogenic activity increased when methanogenesis was inhibited with a model inhibitor compound. An increase in propionate, a further ruminal hydrogen sink, was also observed, and there were changes in the archaeal and bacterial communities. However, based on our results, homoacetogenesis did not become a major sink for ruminal hydrogen.

Key achievements for 2014/15:

- Validated, using stable isotope labelled volatile fatty acids, that interconversion of volatile fatty acids does not give false positive results, when measuring homoacetogenic activity by $^{13}\text{CO}_2$ incorporation into acetate
- Showed that homoacetogenesis does not contribute significantly as a hydrogen sink during short-term (5 days) inhibition of methanogenic activity in sheep
- Showed that homoacetogen numbers do not appear to increase in numbers during short-term (5 days) inhibition of methanogenic activity in sheep.

5.8 - Modelling rumen methane production



Jointly supported programme

Objective Leader – Dr David Pacheco (AgResearch)

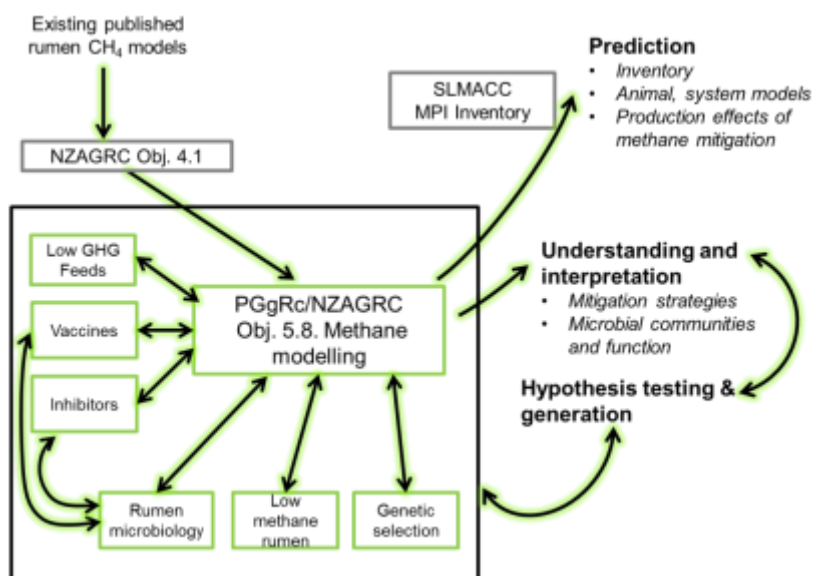


The development and evaluation of methane mitigation strategies requires a mechanistic understanding of the processes influencing methane formation in the rumen. Work in this objective seeks to improve our ability to predict responses in methane formation from NZ ruminants.

The outcomes from this project will be used by scientists in the NZAGRC/PGgRc programme as a tool to develop hypotheses regarding methanogens growth and activity in the rumen in response to current and future intervention such as feeding practices, inhibitors and vaccines. In the medium term, a predictive model of methanogenesis will be available for the wider scientific community for incorporation into whole animal models, which in turn will be able to generate knowledge on methane production and animal productivity. Ultimately, such models will be used to improve inventories and tools for monitoring the effects of mitigation options on farm practices.

The methane modelling objective will serve as an integration point for knowledge related to the development of methane mitigation strategies. This objective builds on previous work conducted as part of the “Integrated Systems” programme of the NZAGRC (Objective 4.1). The relationships between this objective and the rest of the NZ programme on methane accounting and mitigation are presented in the diagram below. The integrative role of the methane modelling project will be formalised in the form of six-monthly meetings with other objective leaders within the programme, to discuss advances in the modelling capabilities, with the purpose of defining simulations to validate the model with empirical findings. Also, as the model progresses, it is expected that

simulations will be conducted to test likely outcomes of empirical research, leading to improved design and power of experiments.



5.8 – Progress in 2014/15

Mechanistic models of rumen function provide the opportunity to explore the impact of feed type and feeding management on methane emissions. Furthermore, they also provide a way to predict the nutrient supply (e.g., volatile fatty acids) for animal production.

We have progressed towards developing a mathematical model that predicts enteric methane production from sheep. Our adaptation of Molly (a model of a cow rumen) to predict methane emissions from sheep has now been published in the *Journal of Animal Science*, one of the top international journals in animal science. In that publication, we described not only a re-sized version of Molly, but also described the addition of a dynamic pool of rumen hydrogen. This addition forms part of a development towards introducing a representation of the hydrogen-methanogen interaction that is a key component of a feedback system that controls not only methane production, but also other key aspects of rumen function such as volatile fatty acid formation. Additionally, in our paper we correct an error in the original Molly model, which could lead to unrealistic methane predictions at low dry matter intakes. Thus, we are contributing to global efforts to improve the ability to predict methane emissions, beyond what is in our model.

The published sheep model was calibrated using data from experiments in which sheep were fed ryegrass diets at several feeding levels. As part of the work this year, we have evaluated the predictive performance of the model for diets other than ryegrass pastures. We conducted an evaluation of the predictions of the model for a variety of forages such as brassica crops (both leafy and bulbs), chicory and white clover. Although the model was developed from ryegrass data, the predictive performance was good for other forages too. Across all the feed types, the model was able to predict methane yields (methane per unit of dry matter intake) with reasonably good precision. However, the prediction of volatile fatty acid ratios was not as accurate, and we have identified possible ways to improve the ability to predict both methane and volatile fatty acid production.

The work of Mr James Wang, a PhD student funded by the programme, has continued towards developing a model of hydrogen-methanogen interactions. The first part of this development is described in a manuscript that is currently under revision for publication in an international peer-reviewed journal. In other parts of this work, the model of hydrogen-methanogen dynamics has been expanded to represent the thermodynamic control of the conversion of reactants such as hydrogen and carbon dioxide into methane. Furthermore, the model provides a scalable

representation of microbial metabolism, which can be parameterised for any number of microbial metabolic pathways. Indeed, Mr Wang has developed a model in which the activity of fermenting bacteria is influenced by the concentration of hydrogen, which is in turn affected by methanogens. Importantly, in this model, there is no need to provide fixed, arbitrary stoichiometry coefficients to estimate volatile fatty acid production (as in the Molly model). Rather, the model that Mr Wang is developing generates volatile fatty acid profiles as an emergent property of the bacterial metabolism, which is in turn influenced by the rumen hydrogen concentration.

Our work has been presented at international conferences such as the “Modelling Nutrient Digestion and Utilization in Farm Animals” workshop in September 2014. Our presentations highlighted our growing collaboration with Prof Mark Hanigan (Virginia Tech University, USA). At the conference, we also strengthened our linkages to the Australian methane modelling project, led by Malcolm McPhee from DPI New South Wales. The Australian team has expressed strong interest in including the sheep model as the tool to predict methane emissions from Australian farms.

Key achievements for 2014/15:

- Our manuscript describing a model to predict methane emissions from sheep (MollyRum14) has been published in the Journal of Animal Science, one of the top journals in the area of animal science and agriculture. Due to the high visibility of this journal, we expect to increase our profile as contributors to the area of methane modelling.
- We have evaluated the sheep model (MollyRum14) predictions for a wider range of fresh forages and found it to have a good predictive performance across the range of diets.
- Mr James Wang has developed a model of microbial interactions, based on metabolic pathways and with a thermodynamic control, that predicts co-existence of bacterial groups using a common substrate and allows production of volatile fatty acid production to be an emergent property of the model.
- We have been approached by the Australian team in the DAFF-funded project “Impacts of CFI methodologies on whole-farms” to access the sheep model developed, rather than re-scaling the beef rumen model developed by Nagorcka et al. This collaboration has been signalled as an important linkage for our group with other international modelling efforts on rumen function and methane emissions.
- Our profile in the area of methane modelling has grown, with new approaches by researchers in INRA, France, interested in collaboration with our group.

5.9 - Dairy Housing Methane Capture and Mitigation by soil - a feasibility study

Objective Leader – Dr Surinder Saggar (Landcare Research)



The aim of this study is to test the practicality of capturing methane (CH₄) emitted by housed cattle and their waste and mitigating by injecting it in the soil for oxidation by methanotrophs.

Research conducted by Landcare Research since 1995 has demonstrated that soils containing active methanotrophs (CH₄-eating bacteria) and exposed to CH₄ can remove these emissions. Our research over the past decade showed that a biofilter made from a suitable soil containing a very active population of methanotrophs could potentially remove almost all of the high CH₄ emissions produced from an average dairy farm waste pond. Thus, soil containing active methanotrophs could also potentially capture and mitigate enteric and waste CH₄ from dairy housing. As methanotrophs are strict aerobes, efficient oxidation of CH₄ requires a well aerated soil environment. This would be hard to achieve in some poorly drained heavier soils, especially in

winter when cows are usually housed. Thus the proposed research aims to assess the capacity of soil, or artificial material mixed with soil, to mitigate the low concentrations of CH₄ produced in dairy housing. This will be achieved by injecting the CH₄-rich air into the soil for oxidation by methanotrophs, and then measuring the potential mitigation by these bacteria, and the influence of changes in soil moisture and aeration conditions. To ensure that the “dairy shed air” is representative of the air in a dairy house, a suitable level of ammonia will be included in the enriched air.

5.9 – Progress in 2014/15

To initiate the research by testing the practicality of capturing methane and ammonia emitted by housed cattle and their waste and mitigating by injecting it in the soil for oxidation by methanotrophs, the experimental design has been finalised and site is established for the delivery of methane and ammonia into the soil.

Key achievements for 2014/15:

- The experimental design meets the requirements for the flow rates and the site is set-up for delivery of methane and ammonia into soil.

6.1 – Plant Effects on N₂O Emissions

Objective Leader – Dr Saman Bowatte (AgResearch)



It is well established that nitrification rates in soil are strongly influenced by the presence of plants and can differ markedly depending on the plant species. Plants can influence nitrification in soils by a variety of mechanisms: (a) they may secrete inhibitory compounds known as biological nitrification inhibitors (BNI compounds) that directly influence nitrifying organisms, (b) they may compete strongly for nitrogen and thus reduce the substrate for soil nitrifiers, and (c) they may alter the identity of the microbial community and/or microbial activity by altering the soil environment e.g. soil pH and moisture content.

Our previous work has found differences in nitrification between species, between cultivars and between endophyte-grass combinations. This programme will test whether differences apparent in these initial experiments are evident in a field situation, and if so, whether the effect is quantitatively important and whether there are trade-offs (e.g. in forage production) that might reduce the desirability of a low emitting species as a mitigation option. Our broad screening approach will also complement industry testing of alternative pasture species by providing valuable information on the most suitable material for testing in grazing trials.

6.1 – Progress in 2014/15

The main focus of the project is to a) determine whether there were differences in N₂O emissions among pasture species and cultivars of sufficient magnitude to warrant further investigation and investment, b) identify potential mechanisms responsible for any differences and c) test whether an assay for potential nitrification could be used as a predictor of species N₂O emissions.

Eighteen plant species/cultivars, with a wide range of plant influence on nitrification potential when evaluated in a pot experiment with the same soil type, were selected for this experiment. A six week N₂O measurement experiment was conducted in late spring-early summer 2014 (30 October - 12 December), six months after sowing. The experiment included a cattle urine treatment (530 kg N/ha; 10 l/m²) and a water (control) treatment, applied to all plots. The highest emissions were observed from bare soil plots but the percentage of bare ground inside the chamber area of plots which did contain plants did not correlate with the total amount of emissions. Emissions from the urine treated plots over the six week measurement period were markedly different among plant genotypes with the lowest emitter being the Italian ryegrass cv. Moata and the highest being grazing brome cv. Gala. This represented a range of about -58 to +120% of the median value for the dataset. We measured soil pH and moisture inside the chambers at the end of the experiment. There were no correlations between either pH or soil moisture 6 weeks post urine application, and total N₂O emissions. Considering all the plant types together, there was no evidence of a strong relationship between the amount of emissions and the amount of N taken up by the plants.

There was a general positive relationship between total emissions of N₂O from the urine treatment and both soil nitrification potential (SNP) in sampled soil and *in situ* soil nitrification rate measured indirectly as the average of the weekly ratio of plant available soil NO₃⁻/NH₄⁺. As the SNP assay did not include plants and was done with unlimited NH₄⁺, we postulate that the difference in SNP exhibited during the 16 h assay was due to difference in the abundance or activity of the soil borne nitrifying community induced by the different plant genotypes.

Plantain and forage rape stand as outliers in the relationship between SNP and total emissions. They had low emissions relative to their high rates of potential nitrification. One explanation for this combination could be that there is an active nitrifying population associated with these species (as

shown by the high SNP) but when the plant is present this activity is suppressed potentially by a biological nitrification inhibition (BNI) compound.

N₂O emissions were strongly correlated with SNP measured after, but not before, the application of urine. If this result can be repeated and shown to be robust then the inexpensive and simple SNP assay could provide a useful method for assessing N₂O emissions potential where large sample numbers are involved.

Next, we will conduct experiments confirming that plant genotypes have a consistent effect on N₂O emissions and improve our understanding of the mechanisms responsible for plant effects. In particular we will resolve whether the variation in emissions is related to plant effects on soil moisture and pH. It is often stated that plant uptake of N removes the substrate for microbial production of N₂O, that plants differ in their uptake rates and affinity for different N forms and therefore plant uptake is the most likely explanation for differences in N₂O emissions associated with different plant species. Results of this experiment provided only weak support for this argument. However, given the prevalence of the argument and the interest in plant N uptake and N use efficiency, this is a critical area to understand. In addition, if plant N uptake has only a small effect on emissions we would have to postulate an effect of plants on soil microbes that is not mediated simply by competition for N and would therefore provide new avenues for potential mitigations based on plant/microbe interactions. Therefore we propose a further piece of work (that would require additional funding) designed to explicitly test the importance of plant/microbe competition for N as a mechanism responsible for plant effects on N₂O emissions.

Key achievements for 2014/15:

- We found evidence for markedly different N₂O emissions after urine applications to soils under different pasture genotypes.
- Our first assessment of the mechanisms driving differences among plants suggests plant uptake of N or effects on soil moisture or pH cannot fully explain the species differences.
- N₂O emissions were predicted by the potential nitrification rate associated with each species after urine was applied. This implies a role for nitrification in determining emissions and offers the possibility of using the simple nitrification assay as a predictor of N₂O emissions.
- We published results of the 6.1.2 experiment in an international journal: Bowatte et al (2015) Wide variation in nitrification activity in soil associated with different forage plant cultivars and genotypes. Grass and Forage Science doi: 10.1111/gfs.12175. The results of plant genotype effects on N₂O emissions after urine application were presented to the NZAGRC-PGgRc linkages between soil C and N₂O Workshop 29 April 2015 and to the Methanet/NZOnet meeting in Wellington 7-8 May 2015

6.2 – Denitrification Processes (Wrap-up of previous Objective 2.3)

Objective Leader – Dr Surinder Saggar (Landcare Research)



Denitrification is the primary process of N₂O production in New Zealand pasture soils. However, we lack a comprehensive, quantitative understanding of denitrification rates and controlling factors across agrosystems. Denitrification is a facultative anaerobic microbial process producing nitric oxide, nitrous oxide and N₂ from nitrate and nitrite. Abiotic denitrification can occur under some conditions. Understanding those mechanisms (microorganisms; biotic processes and mineral oxide; abiotic processes) and soil & environmental factors that have the potential to reduce the production of N₂O during denitrification is vital to the development of new and effective N₂O mitigation technologies.

This objective will wrap up the previous NZAGRC Objective 2.3 which focussed on testing and improving the latest microbiological techniques to identify pathways to reducing N₂O production during denitrification and develop mitigation technologies that reduce N₂O emissions by lowering N₂O/N₂ ratio during denitrification, including in areas where denitrification is maximised to reduce nitrate leaching losses (e.g. riparian buffer zones). Mitigation option of liming for modifying denitrifier community structure, accelerating complete denitrification and mitigating N₂O emissions from urine is evaluated.

6.2 – Progress in 2014/15

Laboratory experiments to determine the efficacy of lime for modifying denitrifier community structure and accelerating complete denitrification with soil samples from the non-allophanic Manawatu and allophanic Horotiu soils treated with different rates of lime incubated to stabilise at increased pH levels were completed together with the analyses of soil and gas samples and qPCR of the extracted DNA. The data collation and processing to write up this work will be due to the researcher Neha Jha being on maternity leave from 19 Jan 2015. The Contract was varied to extend the End Date for this Milestone and Payment Schedule to 31 December 2015 to allow for a period of Maternity leave and LWOP for Neha Jha (from mid-January 2015 to 30 June 2015). The Quarter 3 and 4 payments for Financial Year 2014-15 have been reallocated to Quarter 1 and 2 in 2015-16.

The metagenomic analysis from soils obtained from an incubation using three New Zealand dairy pasture soils (Manawatu sandy loam, MW; Tokomaru silt loam, TM; and Otorohonga Silt Loam, OH), saturated with cattle urine (700 mg N kg⁻¹ dry soil) and water to was added to soils with saturated water content showed dairy pasture soils examined harbour different bacterial communities though the most abundant phyla in all three field moist soils belong to Proteobacteria, Actinobacteria, Verrucomicrobia, Bacteroidetes and Acidobacteria. This study showed that the three dairy pasture soils examined harbour different bacterial communities but their response pattern to wetting and addition of cattle urine was generally similar and showed immediate shifts in bacterial community structure. Also, the results suggest that for comparative metagenomics work and assessing the impacts of N input or irrigation events the soils should be sampled at field moist conditions to provide representative bacterial community structure.

Publications: 1 submitted MS (ROI 000378) has been published in SBB (ROI 000511), 2nd (ROI 000480) is in press in SBB after revision, and 3rd on metagenomics is submitted (000602). The MS on liming effect will be drafted and submitted by December 2015. Additional 2 posters (000404, 000417) were presented at International conference and one oral conference presentation (000488) was made at the Soil Science Conference in Hamilton.

Key achievements for 2014/15:

- Soil denitrification increases as the sites move further North towards the equator – direct response to temperature or indirect response to moisture. These changes are associated to change in microbial populations at geographical scales.
- Microbial community composition (based on 16S rRNA gene sequencing) can be used to predict both the gas species and quantity emitted.
- Typical response of nitrification and the response of functional groups linked to denitrification after DCD application are soil specific. At a total community level, urine deposition resulted in a decrease in richness both in the presence and absence of DCD, confirming that the inhibitor does not contribute to reducing richness further.
- For comparative metagenomics work and assessing the impacts of N input or irrigation events the soils should be sampled at field moist conditions to provide representative bacterial community structure.

6.3 – Feed management options for mitigating N₂O emissions from grazed systems

Objective Leader – Dr Cecile de Klein (AgResearch)



Animal urine patches are the key source of N₂O emissions from grazed systems and the amount of urinary N excreted is largely determined by plant and dietary N content. A key issue is that in New Zealand pasture-based systems, animals consume more N than they need for growth and production, and hence surplus N is excreted and contributes to N₂O emissions.

Key research questions are whether feed management options can (a) reduce N urinary output, and (b) reduce the N₂O produced per unit N excreted?

Forage species (pasture species and forage crops) differ in N content and some of them (e.g. tannin containing plants) can also influence how N is partitioned between dung and urine. Furthermore, there is evidence that plant species can affect N₂O emissions through their impact on N cycling and on soil microbial processes e.g. increased N use efficiency and/or biological nitrification inhibition. Finally, plant chemical composition may influence the chemical composition of the urine which in turn may influence the amount of N₂O released from a urine patch.

6.3 – Progress in 2014/15

The aim of this objective is to develop feed management options for mitigating N₂O emissions from NZ systems. The research is conducted in three key regions in NZ (Waikato, Canterbury and Southland) and is run in close association with the DairyNZ-led Forage for Reduced Nitrate Leaching (FRNL) programme. The work focuses on two key aspects of feed management options to reduce N₂O emissions: 1) plant species that reduce total N excretion in urine; and 2) plant species that reduce the N₂O emission factor of urine (e.g through Root exudates effects on N cycling in the rhizosphere; or through the effect of urine composition on N₂O emissions).

The evaluation of the effects of plant species on N₂O emissions is conducted in different stages: 1) rapid screening of potential reductions; 2) small plots or lysimeter studies; and 3) grazed field plots. For stage 1) a novel 'rapid screening' methodology was developed that can evaluate the effect of urine from animals on different types of feed and/or the effect of specific urinary compounds on N₂O emissions. This methodology is now being used to evaluate the effects of different urine compounds on N₂O emissions.

We also screened a large range of plant species, firstly in pot trials and then in small field plot trials for their effect on the N₂O emission factor for urine. The results show that there is a large variability between plants and the mechanisms behind these differences are being further investigated.

Linking in with field studies associated with the FRNL programme, the N₂O emission factor was determined for fresh cow urine deposited on Lucerne, Italian ryegrass/white clover and perennial ryegrass/WC. Other FRNL-linked field studies are now being conducted to assess the N₂O emission factor for fresh cow urine on mixtures of Italian ryegrass/plantain and ryegrass/white clover (Canterbury), monocultures of ryegrass, white clover, lucerne and plantain (Hamilton), and grazed plots of Italian ryegrass and ryegrass/white clover (Southland).

Key achievements for 2014/15:

- Two PhD students have been recruited (Sheree Balvert and Camilla Gardiner). Sheree is enrolled at Waikato University, and Camilla is enrolled at Lincoln University. Sheree started her PhD project in Nov 2014, while Camilla will arrive from UC Berkeley late June/early July.
- A screening methodology for rapid assessment of N₂O mitigation potential of compounds has been successfully developed.
- Preliminary results from a study using the rapid screening methodology are promising and suggest that glucosinolate hydrolysis products can reduce N₂O emissions.
- Field trials in association with the Forage for Reduced Nitrate Leaching (FRNL) programme have been set up in Hamilton, Canterbury and Southland to test the effect of different forages on N₂O emissions from urine.
- The results of the measurements in Canterbury found that gibberellic acid addition had no direct effect on the N₂O emission factor (EF₃) from urine applied to pasture species, but may allow reduction of fertiliser use without yield penalty – thus reducing N₂O from fertiliser. EF₃ on lucerne was lower than EF₃ on Italian ryegrass/white clover and perennial ryegrass/WC.

6.4.1 – Effects of ‘Italian Ryegrass plus Plantain’ versus ‘Perennial ryegrass’ on N₂O emission factor for urine



Objective Leader – Prof Hong Di (Lincoln University)

In collaboration with the MBIE funded ‘Forages for Reduced Nitrate Leaching (FRNL)’ research programme, determine N₂O emission factors from animal urine (EF₃) applied to ‘Italian Ryegrass plus Plantain’ vs ‘Perennial ryegrass’ pastures using urine collected from cows grazing on the Italian/Plantain plots as well as cows on the P Ryegrass plots (i.e. urine inputs of 0, rate as collected from different pasture mixes, and standard urine at 700 kg N/ha). Thus 2 pasture mixes x 3 urine rates x 5 reps gives a total of 30 lysimeter gas chambers.

6.4.1 – Progress in 2014/15

Excellent progress has been made as scheduled in the contract. In collaboration with MBIE-funded research programme (FRNL), lysimeters have been collected and installed at a field lysimeter facility at Lincoln University. Urine collected from Italian Ryegrass plus Plantain and from Perennial ryegrass pastures has been applied to the lysimeters to determine the N₂O emission factors from the urine. N₂O gas collection and analysis are progressing according to plan. Results will show the effect of different pasture mixtures on N₂O emissions.

Key achievements for 2014/15:

- Consent has been obtained from managers of the MBIE funded programme ‘Forages for Reduced Nitrate Leaching’ (FRNL) to use of the lysimeters for this N₂O programme.
- Lysimeters have been collected and installed in the field lysimeter facility at Lincoln University.
- Urine collected from cows grazing different pastures has been analysed and applied to the lysimeters.
- N₂O gas has been collected according to schedule.
- Analysis of N₂O samples have been carried out at the National Centre for N₂O Measurement at Lincoln University.

6.4.2 – Effect of diverse pasture on the N₂O emission factor for urine



Objective Leader – Dr Jiafa Luo (AgResearch)

In collaboration with the NZAGRC soil C programme on Troughton farm in the Waikato, compare the N₂O emission factor (EF₃) for animal urine applied to diverse pasture with that for animal urine applied to conventional ryegrass/white clover pasture.

N₂O emissions will be measured following application of synthetic urine at 500 and 700 kg N /ha (to mimic expected urine N concentrations of diverse and conventional pasture, respectively) to both diverse and conventional pasture plots at Troughton farm. A non-urine control treatment will also be included to allow emission factors to be calculated. Measurements will continue for 4 months to allow the full emission envelop to be captured.

Soil mineral N and soil moisture contents will also be measured to provide additional information on explanations of any differences in EF₃ found.

6.4.2 – Progress in 2014/15

In collaboration with the NZAGRC soil C programme on Troughton farm in the Waikato, the N₂O emission factor (EF₃) for animal urine applied to diverse pasture is being compared with that for animal urine applied to conventional ryegrass/white clover pasture.

Measurement areas were identified (with assistance of Prof Louis Schipper, NZAGRC soil C programme) during a farm visit. Each area covered conventional ryegrass/ white clover pasture and adjacent diverse pasture consisting of lucerne, plantain and chicory as well as ryegrass/clover. A trial protocol was prepared and, after discussion with the principal investigator (Dr de Klein), it was decided that real urine would be used for the gas sampling plots in this field study.

On 16/4/15, 3 the identified areas were fenced off to exclude stock for 4 weeks before applying the treatments. On 22/4/15, 36 gas sampling chambers were installed and soil sampling plots marked out in the fenced areas. On 11/5/15 pre-treatment sampling was conducted to establish a baseline for emissions. On 13/5/15, real cow urine was applied to the chambers at rates of 500 and 700 kg/ha. Artificial urine was applied to the adjacent soil sampling plots at a rate of 700 kg/ha. Gas and soil sampling commenced immediately after treatment application and was repeated twice a week until 24/6/15 when it was reduced to once a week. Gas and soil sampling is ongoing and samples are currently in the process of being analysed.

Key achievements for 2014/15:

- Discussion was held with Ben Troughton, the farm owner in association with Louis Schipper from the University of Waikato to negotiate access to the site.
- A protocol describing the site preparation and sampling procedures was developed.
- The site was identified and prepared.
- Treatments were applied to the trial areas.
- Gas and soil samples have been collected and analysis of the samples is under way. Sampling is currently ongoing.

6.4.3 – N₂O emission factor for dairy farm effluent (FDE) applied to pasture

Objective Leader – Dr Tony van der Weerden (AgResearch)



To enable a more robust assessment of the effect of housing systems on N₂O emissions, determine whether the emission factor for FDE applied to pasture (EF₁) differs from the emission factor for urine deposited onto pasture (EF₃).

N₂O emission measurements will be conducted following FDE application at 50 kg N/ha and real cow urine application at 700 kg N/ha to pasture field plots at c 70% water filled pore space (WFPS) in autumn. A non-FDE/non-urine control treatment will also be included to allow emission factors to be calculated. Measurements will continue for 4 months to allow the full emission envelop to be captured.

In addition, the effect of soil moisture content (to mimic a 'timing of application effect') on the N₂O emission factor will be determined following FDE application in autumn to pasture field plots at an additional three soil moisture contents (c. 40, 60 and 80% WFPS). Measurements will continue for 4 months to allow the full emission envelop to be captured.

6.4.2 – Progress in 2014/15

A field trial was established on a free-draining soil supporting a ryegrass/white clover mixed sward in Otago in April 2015. As one of the objectives of this project was to determine the influence of the initial soil moisture content on the emission factor for farm dairy effluent (FDE EF1), 14 plots were marked out and a range of soil moistures were established by covering some plots and applying varying amounts of irrigation to others. At the start of the trial, soil moisture content ranged from 25 to 43% volumetric water content, equivalent to 44 – 78% water filled pore space (WFPS). A second objective of this project was to determine if FDE EF1 and cattle urine EF3 differ.

The FDE, with an N content of 0.07%, was applied at an equivalent rate of 70 kg N/ha to the 14 plots, while cattle urine, with a nitrogen (N) content of 0.57%, was applied at an equivalent rate of 570 kg N/ha to 7 of the plots. Control treatments (no applied N) were also established on all 14 plots to allow a calculation of EF1 across the range of soil moisture contents. The urine treatment was restricted to 7 plots as this was considered sufficient to determine if EF1 and EF3 differ.

Gas and soil sampling was initiated on the day of treatment application (16 April), and has continued for 2.5 months (up to the end of June). Gas samples collected over the first month have been analysed: results to date suggest that N₂O fluxes from urine were greater than from the control treatment, whereas fluxes from the FDE treatment were initially higher than the control, but have returned to levels similar to the control treatment. Gas measurements will continue until mid-August, after which the data will be analysed and reported by December 2015.

Key achievements for 2014/15:

- Field trial established and gas and soil sampling in progress

7.1 - Manipulation of carbon inputs to stabilise and enhance soil carbon stocks

Objective Leader – Dr David Whitehead (Landcare Research)



Measurements and models will be used to quantify and forecast changes in soil carbon attributable to manipulation of carbon input, incorporation and retention. The experimental manipulation procedures (including diverse swards, irrigation, addition of nitrogen fertiliser, grazing intensity and imported feed) are selected to test the following hypotheses:

1. Conversion of ryegrass clover to diverse grassland swards increases root inputs of carbon
2. Irrigation and associated management intensification increases carbon inputs to the soil

Farm carbon balances will be constructed from measurements of carbon exchange and carbon imports and exports and used to estimate conversion of carbon inputs to soil carbon.

Detailed process-based dynamic modelling will be used to forecast the combined impacts of different management options and environmental changes on soil carbon inputs, incorporation and stabilisation.

The research will focus on experimental sites at Troughton Farm (Waikato) and an irrigated dairy farm in Canterbury (location to be confirmed).

7.1 – Progress in 2014/15

We have continued our integrated approach of field measurements and modelling to determine and forecast the effects of management practices on soil carbon storage in grasslands. Our work has centred on two dairy farm platforms with different management practices.

In the Waikato, we have been working on the effects of sward diversity on soil carbon storage in non-irrigated grassland for five years. The questions we are asking are (i) how much soil carbon loss occurs during pasture renewal with or without full cultivation? and (ii) can replanting to more diverse pasture with more and deeper roots increase long term soil carbon storage?

The work this year has shown that soil carbon losses following cultivation were not greater than losses following no-till and direct drilling treatments to re-establish the pasture sward. Rather, the soil water content conditions at the time of pasture renewal and duration between spraying and seedling emergence are more important drivers of changes in carbon storage. The main implications for the farming community are that (a) the site preparation for pasture renewal is considerably less important than rapid re-establishment of the new sward and (b) selecting a critical time for the conversion when soil water content allows good pasture growth but when weather conditions minimise carbon losses.

Early results from the two-year comparison of changes in soil carbon storage under conventional ryegrass/clover and a mixed sward incorporating deep-rooting species show that the potential for net carbon uptake into the soil for the mixed sward is greater because of increased carbon inputs from roots.

We have also undertaken two projects with PhD students at the site. We quantified root turnover rates for both the ryegrass and diverse pasture swards using isotopic labelling. Differences are small but preliminary results for root carbon inputs following pasture renewal show that the root turnover following spray-off accelerated rapidly and carbon input from the sprayed-off sward was approximately twice that for the existing ryegrass-clover during 88 days of spring growth. These findings suggest that pasture renewal including spraying off the existing sward could increase carbon inputs to the soil at least temporarily.

The second PhD project has investigated differences in evaporation and water use efficiency between the sward types. Grazing events were found to have very little to no effect on evaporation, probably due to soil evaporation compensating when transpiration rates were lower. Preliminary findings on the water use efficiency of high and low diversity swards show that the high diversity swards can produce equivalent amounts of biomass while using less water than a conventional ryegrass-clover sward. This suggests that, if mixed swards were incorporated into farm systems, they may be able to improve the resilience at non-irrigated sites especially during drought periods.

Overall, from our work at the dairy farm site in the Waikato, we conclude so far that (i) the dual goal of milk production and increasing carbon storage in dairy grasslands may be possible, (ii) carbon losses due to pasture renewal are less when time between spraying and sowing is minimised, (iii) carbon losses are minimised if regressing takes place late summer/autumn rather than spring and (iv) more diverse sward offers potential for increasing soil inputs from roots.

Our second field platform is at a recently converted dairy farm on shallow, stony soils at Ashley Dene, Canterbury. The question we are asking is does conversion from dryland farming to intensive dairy farming using irrigation and addition of nitrogen fertiliser result in increases in soil carbon storage? This site and the experimental treatments are in the process of being set up so our work has not yet started. Our work will contribute to a larger project to address carbon and nitrogen cycling in relation to conversion to intensive dairy management practices in collaboration with Lincoln University, Landcare Research and Plant & Food Research. This site will also be used in a multi-site comparison of processes regulating changes in soil carbon turnover in a GPLER project led by Landcare Research.

In parallel with the field measurements at our two farm sites, we have been testing the CenW model for its suitability to simulate and forecast the impacts of changing climate and management practices on long-term soil carbon storage. We have demonstrated the success of using the model in comparison with 28 models used internationally and in simulating the short-term changes in net carbon exchange that we have observed from the field measurements at the Waikato site. These comparisons have provided confidence that we are able to apply the model across a much wider range of climate and management scenarios, including changes in fertiliser application, grazing regime, pasture root:shoot ratios, water availability (i.e. climate change or irrigation) and the environmental changes of increasing temperature and atmospheric carbon dioxide concentration. While we have identified practical management modifications that could lead to enhanced productivity, most of them led to only small changes in increased soil carbon storage. We presented our findings at two stakeholder workshops and to scientists and our work continues to forecast the long-term effects of the management practices at our two field platforms.

Key achievements for 2014/15:

- Rutledge S, Mudge PL, Campbell DI, Woodward SL, Goodrich JP, Wall AM, Kirschbaum MUF, Schipper LA 2015. Carbon balance of an intensively grazed temperate dairy pasture over four years. *Agriculture, Ecosystems and Environment*, 206:10-20. This publication provides evidence for changes in the components of carbon exchange for a grazed dairy farm with a conventional ryegrass/clover sward. The paper provides credibility for the techniques we use and confirms that, despite variability associated with weather conditions, notably a marked drought period, the site was a net carbon sink or carbon neutral on an annual basis. Rates of carbon exchange were comparable with those from other managed grassland sites, confirming a general trend for increased carbon storage with increasing productivity, suggesting that it may be possible to meet the dual goal of increased pasture production and increased soil carbon storage in managed temperate grasslands.
- McNally SR, Laughlin DC, Rutledge S, Dodd MB, Six J, Schipper LA. Root carbon inputs under moderately diverse sward and conventional ryegrass-clover pasture: implications for soil carbon sequestration. To be submitted to *Plant and Soil*. This paper confirms that moderately diverse pasture systems offer scope to increase soil carbon storage under grazed pastures through increased root mass inputs and rooting depth.

- Kirschbaum MUF, Kelliher FM, Beare M, Rutledge S, Mudge PL, Puche N, Schipper LA, Campbell D, Camps Arbestain M, Schon N, Whitehead D 2015. Overview presentations of findings from soil carbon research programme at a stakeholder workshop held in Wellington on 12 March. This presentation provided a full update of findings from the first four years of research funded by NZAGRC and inform stakeholders of the implications.
- Kirschbaum MUF, Rutledge S, Mudge PL, Puche N, Schipper LA, Campbell DI 2015. Changes in soil carbon stocks of a New Zealand grazed pasture in response to variations in management and environmental factors. In: Moving farm systems to improved nutrient attenuation, Currie LD, Burkitt LL, eds. Occasional Report No. 28. Fertiliser and Lime Research Centre, Massey University, Palmerston North, New Zealand, 19 pp. This provides an update of the likely impacts of management options for minimising nutrient and carbon losses directed to an end user audience.
- *Reducing New Zealand's agricultural greenhouse gases: Soil carbon*. Factsheet 3 produced by NZAGRC and PGgRc, with substantial input from the University of Waikato. An informative publication written for farmers and the general public summarising what is known about the management and environmental factors regulating soil carbon storage in grasslands.

7.2 - Tools to quantify the stabilisation capacity and vulnerability of carbon in grassland soils

Objective Leader – Prof Frank Kelliher (AgResearch)



Developing and deploying management practices that maximise the long-term storage of soil carbon depend on understanding the capacity of soils to stabilise carbon and its vulnerability to loss. We aim to identify the soil properties and management practices that most affect the stability of soil carbon and its vulnerability to loss. This will contribute to objective 7.1 by collaboration on the modelling of soil carbon incorporation, distribution and stabilisation. We will:

1. Test key assumptions to develop a second generation statistical model (developed from previous milestones) for predicting the soil carbon stabilisation capacity for New Zealand's grassland soils
2. Apply soil chemical and physical fractionation methods and a soil carbon mineralisation assay to quantify the stability and vulnerability to loss of soil carbon
3. Synthesise findings to identify soil properties and grassland management practices that most affect soil carbon stabilisation and vulnerability to loss

7.2 - Progress in 2014/15

At a workshop in Wellington, representatives from science providers and stakeholder organisations reviewed findings from the first four years of the NZAGRC soil carbon programme and discussed future research priorities to ensure translation and uptake of the findings by land managers. The workshop recommended developing greater opportunities to publicise research findings and promote the value of the research. A further recommendation was scenario modelling to explore the effects of land management practices on soil carbon stocks and nitrous oxide and enteric methane emissions at a national scale. While unprecedented for soil carbon, stakeholders consider such analysis to be warranted. We responded to this feedback by a second presentation of our findings and new programme objectives at a workshop in Palmerston North.

At 130 sites across NZ which have been under long-term pastoral agriculture management, we sampled soils representative of the major orders including Allophanic, Brown, Gley, Pallic and

Recent. This was done in collaboration with the Plant & Food Research CORE-funded LUCI (Land Use Change Intensification) programme. The samples were prepared and sent to CSIRO for mid-infrared (MIR) spectroscopy measurements. The MIR results will guide us to select soils for physical and chemical analyses for milestone 7.2.2. On the basis of selected, preliminary results, we sampled soils at a further 40 long-term pasture sites.

We are examining alternative methods to determine soil properties which can affect carbon stabilisation. For this study, we sampled allophanic and non-allophanic soils for mineral surface area measurement. Based on a literature review, this included two methods of drying the samples, one recommended for delicate structure because we postulate allophane to be a gel-like substance. We are in the process of using two fractionation methods to examine the possible, confounding effects of organic matter. We are also measuring the mineral surface area using four absorbates and examining the samples using an electronic microscope.

A paper by Curtin, Beare, and Qiu (Texture effects on carbon stabilisation and storage in New Zealand soils) was accepted for publication by *Soil Research*.

Waikato University PhD candidate Sam McNally has been appointed to the soil C stabilisation post-doc position. It was agreed Sam will take up the appointment in Q1 of 2015/16 after completing the submission of his PhD Thesis under the supervision of Prof Louis Schipper.

Key achievements for 2014/15:

- Curtin D, Beare MH, Qiu W. 2015. Texture effects on carbon stabilisation and storage in New Zealand soils containing predominantly 2:1 clays. *Soil Research* (In press)
- Kelliher FM, Whitehead D. 2015. Overview presentation of findings from soil carbon research programme at a stakeholder workshop in Wellington on 12 March. This presentation provided plain language background and summarised the research questions and outcomes from the programme's first four years.
- Kelliher FM, Whitehead D. 2015. Soil carbon research: Digging for solutions. Building on stakeholder feedback from the 12 March presentation, this overview presentation of soil carbon research in NZ was given at the NZAGRC Conference in Palmerston North on 28 April. This presentation began with a clear statement of purpose, compelling research questions, more insightful background and progress and outcomes from NZ's soil carbon research to date.

7.3 - Modelling management manipulations using the HPM



Objective Leader – Prof Jacqueline Rowarth (University of Waikato)

There are substantial changes in management associated with the intensification seen in NZ agriculture. These include increases in fertiliser input, increases in livestock intake demand, changes in the offtake of nitrogen and carbon as dairy cows replace dry stock (e.g. sheep) and/or there are increases in milk yield per cow, the increased use of purchased feed, in cutting (vs grazing) of forages, and the increased use of irrigation. Individually and together these changes lead to major changes in the C and N cycles, because animals ‘uncouple’ the C and N cycles, so the more animals that are involved, the more that cycles are uncoupled. Prospects for future development in the NZ pastoral industry also include making fundamental changes to the fate of C and N in plants (via new species/traits), animals (altering N partition), and soils (e.g. reducing SOM decomposition). Very importantly intensification (and the reverse), and all such manipulations, give rise to transient changes that are not necessarily indicative of the longer term changes that can occur when a particular management(s) is sustained. Understanding and clearly separating long term sustainable (c.f. steady-state) changes from transient changes is a priority if we are to understand how changed management practices and system manipulations will impact soil carbon storage in the future. We also need improved understanding of how climate change itself will interact with anticipated management changes.

The Hurley Pasture Model (HPM) is a mechanistic C and N cycling model that incorporates soil, plant and animal processes, in depth; it is an appropriate model for studying short and long term changes in C and N cycling in response to management and environment. It works from first principles and does not rely on ‘tuning’ using experimental data when making predictions. Results from the HPM are already giving insight that indicators such as emission factors and C and N balances based on measurements collected during the period of transition (some 2 to 10 years minimum) between managements can be misleading. This applies to identification of the fundamental drivers of changes in soil carbon and nitrogen, and to the general direction of changes in, and the scale of, these changes.

Our aim is to use the HPM to provide indications of how, and in what direction, C and N emissions, fluxes, and sequestration (of both) will change, what the changes will look like over different time scales after changes in management are introduced, and whether the outcome is sustainable, and at what state. Changed management rarely involves a single action and we will use the model to analyse systematically just which of the components of a management change have the largest impact. This is of value as in any system several factors are often changed simultaneously, potentially leading to sustained false impressions of what needs to be focussed on to offer mitigation.

7.3 - Progress in 2014/15

During this first quarter of this new contract, all of the components of the model listed above and so necessary to conduct the analysis stated in ‘Deliverables’ have been tested and shown to function correctly. The routines to impose the management changes and alter the function of the ecosystem in the model all operate as intended and the integrity of the C and N and water cycles in the model sustained.

Overcoming previous confusion over its application, the model now describes the uptake cycling and fate of all aspects of the C and N cycles (and water cycle) through diurnal and seasonally varying environments using NZ met. data and soil types (variable if required). Long term steady states are now derived NOT by making INPUTS fixed, (a practice used previously only for clarity of how functions interact) but in terms of repeated annual cycles in the diurnal and seasonally varying OUTPUTS. Hence no component (flux or state) in the model is ever unchanging. ‘Steady states’ refers to the ultimately emerging sustained mean annual balances derived as a consequence of the repeating seasonal patterns. Every day, and every change in all process is accounted for, hence realistically and not “conceptually”. The model tracks GHG gaseous emissions, all fluxes of

C and N to the environment, through plants soils and animals, as well as C and N sequestration. Its capacity to help foresee the transient, as well as (often counter-intuitive) long term outcomes for emissions (gaseous and all other), fluxes and C/N stocks, is therefore considerable. By default (switchable) the model predictions include increasing CO₂ and/or temperature.

The model has undergone considerable re-development (funded internally by Massey) since the last NZAGRC contract (ended 2014) and in preparation for future applications, of which this new contract is a case in point. This model is likely unique in its capacity to consider the likely benefits (or otherwise) of proposed alteration of plant or animal traits, or of fundamental changes in soil function derived from these (eg by changes in soil fungal and microbiological communities). These trait changes (eg fructan storage cf 'High Sugar Grasses', or alternative feeds) and proposed alterations directly or indirectly in N partitioning in animals are uniquely integrated into the fundamental processes (photosynthesis, respiration (cf CO₂ fluxes), soil processes and C and N emissions).

Developments in several of these areas rely on very recent advances in the fundamental knowledge of plant growth strategies, as published by Parsons/Rasmussen/Thornley. Likewise soil community functional responses depend on the insights gained from our previous MBIE contract (C10X0903). The model has been tested for its capacity to emulate phenomena observed in our 'plant growth with less N requirement', ex Rasmussen's nitrous oxide NZAGRC programme.

We have the tool for the job and routines to run it consistently and comprehensively. The number of management (and trait) changes we are asked to consider, is large, so running is a substantial task. But the most foreseeable challenge is in extracting key, critical insights for policy and industry from the predictions. This is the recognised forte of Prof Jacqueline Rowarth.

Key achievements for 2014/15:

- Testing of new routines for plant trait changes completed successfully
- Testing of new routines to evaluate changes in N partition in animals (eg milk v urine) completed successfully
- Testing reveals this model produces diurnal and seasonal patterns in a multitude of components of the system, very close to those measured in NZ practice, wherever these have been measured (indeed model predictions are far more comprehensive than in any known series of trials).

8.1 - GHG Emissions on Sheep and Beef Farms

Objective Leader – Drs Kathryn Hutchinson & Robyn Dynes (AgResearch)



The programme will identify the drivers of GHG emissions intensity on two S+B farms and determine whether these drivers result in decreased GHG emissions intensity when integrated into commercial farm systems. Key outputs will include a peer-reviewed journal article, a popular fact sheet and alignment with B+LNZ's extension programme to communicate the findings of the research.

The programme proposes to align with a new environment extension program within B+LNZ and partner with its extension activities. The North Island and South Island environment-focussed farms (EFF) will be evaluated for suitability as study farms. Drivers of GHG emissions intensity on S+B farms will be identified using data from previous projects. These mitigations will then be evaluated within commercial farming systems via a comprehensive measurement and monitoring programme which will include measurement of components of farm systems efficiency as a proxy for direct measurement of GHG which will not be undertaken. A farm systems analysis, including GHG and nutrient losses to the environment will be completed with historical and current farm operational data and for future scenarios including farming within limits, with change to stocking policy and forage supply.

8.2 - GHG Emissions from Dairy Systems

Objective Leader – Drs Kathryn Hutchinson & Robyn Dynes (AgResearch)



The dairy sector programme will assess the GHG emissions for dairy systems demonstrating a range of practical mitigation options including high genetic merit cows (Waikato), diverse pastures and low stocking rate efficient systems (Canterbury) and off pasture systems (South Otago) for management of environmental impact. The programme will partner with established P21 farmlet systems in the Waikato, Canterbury and South Otago, and NZAGRC funding will fund additional data collection and analysis of GHG emissions to:

- a. Assess whether new mitigations within farming systems will also reduce GHG footprint
- b. Validate previous farm systems modelling by demonstrating that these new mitigations deliver real GHG benefits within functional and practical farm systems
- c. Identify risk areas for pollution swapping within the farm system.

Key outputs will include data and resources contributing to the existing DairyNZ extension program, industry conference presentations, journal publications and new data on emission factors for GHG

8.1 & 8.2 – Progress in 2014/15

The Integrated Farming Systems Project will deliver new knowledge and understanding of the impacts of farm management decisions and mitigation options on the farming system and its GHG footprint in both the dairy and sheep and beef sectors. The early phase of the programme has focused on four areas:

1. On-farm measurement of GHG emissions from existing focus farms within the dairy Pastoral 21 research program. Measurements have commenced in both Waikato and Canterbury. This novel approach will enable researchers to collect GHG data from animals

and soils that are part of farm systems research programs and assess the measured emissions along with the modelled emissions at a farm systems level. These data will provide new knowledge of the GHG emissions from farming systems. The P21 sites have a number of potential component mitigations already being evaluated at the farm systems level. Our collaboration enables comparison of GHG footprint from these mitigation options in addition to the leaching losses being assessed for P21.

2. Delivery and installation of new GreenFeed equipment. To enable on-going in-paddock estimates of methane emissions, new equipment has been purchased and installed.
3. Sheep and Beef: the focus has been on developing a working relationship with Beef + Lamb NZ Farm Environment program and their focus farm Onetai Station. This has been achieved through a series of meetings with Beef + Lamb NZ Environmental Extension Manager to gain an understanding of their vision for the Beef + Lamb NZ project on Onetai Station and how we can integrate our programme to add value for both parties. Farmax and Overseer modelling has commenced for this property and future scenario modelling will be undertaken after our first meeting with the farm manager and steering group in early August. Work on this property provides a valuable opportunity to assess how development of a property and changes to the farm system impacts on GHG emissions intensity using a combination of systems modelling and measurements of performance parameters that can be used as indirect indicators of emissions intensity.
4. Maori Farmer Network: working with the programme team to identify opportunities for complementary research, particularly in farm systems modelling, opportunities for collaboration and how to align communication activities to maximise impact of our research for industry.

A communication plan has focused on ensuring the program has the greatest impact through maximising connectivity with existing sector programmes, plans and extension activities.

Key achievements for 2014/15:

- On-farm measurements have commenced at the Waikato P21 dairy farm to compare the impact of component mitigation options on GHG footprint.
- New Greenfeed equipment has been purchased, assembled on arrival in New Zealand and installed at the Lincoln University Dairy Farm. This equipment enables on-going in-paddock estimates of methane emissions. The first use will be an assessment of methane emissions from wintering forage crops.
- A strong working relationship has been developed with Beef + Lamb NZ Environmental Extension Manager. This is critical to our programme achieving a meaningful and on-going interaction with the Environment Focus Farm project at Onetai station. Work on this property provides a valuable opportunity to assess how development of a property and changes to the farm system impacts on GHG emissions intensity.

20.1 - Low emission farm systems for the Māori sector

Objective Leader – Phil Journeaux (AgFirst)



This programme aims to assist the Māori pastoral sector to improve its collective capacity to increase resource efficiency, farm productivity and while lowering greenhouse gas (GHG) emissions.

The programme will achieve this by developing a set of Māori farm typologies, which represent the predominant pastoral farming systems, identify key factors that underpin farm productivity, resource and emission efficiency and sustainable profitability, and then identify and test a range of mitigation strategies. Farm typologies are important to avoid the problems of homogenizing a heterogeneous group that range from very small farms to large multi-enterprise corporates. These typologies will be compared against existing databases and help in the selection of in-depth representative case study farms for scenarios of alternative farm system configurations that will evaluate mitigation options.

Two metrics will be used in parallel to identify and assess mitigation scenarios, their costs, and options for implementation: absolute reduction in GHG emissions, and reduction in emission intensity (the ratio of emissions per unit of output, e.g. Kg CO₂-eq/KgMS).

Farm system mitigation scenarios will be based on the interaction and knowledge sharing that will occur between the farmers (including land entities), scientists and industry advisors that will take place in case study workshops around the country. The research team will apply a range of suitable tools to model these scenarios including Farmax, Overseer, Mitigator and MyLand along with others where required (e.g. LCA, LP optimisation). The integration of forestry models (MyLand) alongside pastoral sector farm systems (Farmax) and emissions models (Overseer) to derive whole farm mitigation strategies is a key feature of the modelling in this programme.

The programme will improve our understanding of the critical characteristics of GHG profiles (both in terms of absolute emissions and emissions intensity) of existing Māori pastoral farming systems and to produce a range of mitigation options to modify farm systems to lower absolute emissions and/or emissions intensity.

A key contribution to the literature will be an enhanced understanding of the Māori farm typologies with economic, environmental, social and cultural implications of low emission farming systems within the Māori sector, with wider implications across NZ. The programme will build on several research programmes including: "SFF Farmers Climate Change and GHGs (C08/008)" led by Margaret Brown; "Tuhono Whenua Māori Benchmarking Framework (M12/173)" led by Tanira Kingi; "Aohanga Incorporation: Climate change mitigation and adaptation: A social process framework for engagement and the development of a climate change resilience strategy (SLMACC C10x1003)" led by Bruce Small; and "Identifying small and medium sized forest owner typologies", MPI contract 16969, led by Steve Wakelin. The programme will also contribute to the integrated systems programme within the New Zealand Agricultural Greenhouse Gas Research Centre (NZAGRC).

20.1 – Progress in 2014/15

The project is designed to assist Māori farmers in New Zealand to improve their collective capacity to increase resource efficiency and farm productivity while lowering greenhouse gas (GHG) emissions.

Progress to date includes:

- Development of a typology of Māori farming
- The collection of farm and GHG emission profiles on 29 Māori farms from around the country, with 2 more to come. This includes 18 sheep & beef farms, and 11 dairy farms
- The selection of 4 Focus farms; 2 dairy (Bay of Plenty, Taranaki, and 2 S&B (Northland, East Coast)
- The development of Farmax files for each focus farm to allow for farm system modelling, and Overseer files to (a) establish the base GHG emission profile and (b) model the impact of change scenarios
- Data collated to allow for national benchmarking of the emission profiles
- Meetings held with the Trustees of the 4 focus farms and agreement gained re (a) participation in the project and (b) discussion on scenarios for modelling
- Development of priority scenarios for modelling of change in farm systems and subsequent impacts on GHG emissions.
- Development of farm maps to assist with the modelling
- A paper has been produced for submission to an international journal; currently being peer reviewed.

Key achievements for 2014/15:

- Base GHG emission profiles on 29 Māori farms developed
- Scenarios for modelling changes in farm systems/land use and the impact on GHG emissions developed
- Focus farm programme underway
- Paper written on the project to date; undergoing peer review before submission to an international journal

APPENDIX 3 – NZAGRC INTERACTIONS AND OUTPUTS

NZAGRC Meetings and Presentations (New Zealand)

- Workshop: Managing GHG emissions (DairyNZ PGP project): 6 August, 2014 - Hamilton
- Workshop: Ag Emissions White Board session (DairyNZ): 11 August, 2014 - Wellington
- MPI Policy staff visit: 25 August, 2014 - Palmerston North
- Science - New Zealand's Place in the World: 25 August, 2014 - Auckland
- Visit by New Zealand governmental and industry officials: 27 August, 2014 - Palmerston North
- Meeting with MfE re greenhouse gases: 8 October, 2014 - Wellington
- Initial discussion re MPI Grants Management System: 13 October, 2014 - Palmerston North
- Visit to DPMC to discuss agricultural emissions and progress: 15 October, 2014 - Wellington
- FullCAM and Agriculture Inventory Expert Advisory Board meeting: 21 October, 2014 - Teleconference
- Workshop: Importance of Pasture Quality in New Zealand: 3 November, 2014 - Wellington
- Workshop: Agricultural Inventory Advisory Panel: 20 November, 2014 - Wellington
- Meeting: Deep South Challenge strategic meeting (science): 27 November, 2014 - Wellington
- Meeting: NZAGRC Science Leadership Team: 2 December, 2014 - Palmerston North
- Reception: 'Grand Challenges cooperation and the Horizon 2020 Programme': 8 December, 2014 - Wellington
- Workshop: NZ/EU Joint Science and Technology Cooperation Commission: 8 December, 2014 - Wellington
- Meeting: Maori Advisory Group: 13 February, 2015 - Palmerston North/Wellington
- Workshop: 'Decision-centric' approach to help identify research gaps and priorities for adaptation to climate change: 24 February, 2015 - Palmerston North
- Workshop: Vaccine & inhibitor progress updates: 4 March, 2015 - Palmerston North
- Workshop: NZAGRC Soil carbon stakeholder workshop: 12 March, 2015 - Wellington
- Workshop: New Zealand agricultural modelling - future work for agriculture sector: 26 March, 2015 - Wellington
- Meeting: Bill Kaye-Blake (PWC) on economics of climate change: 31 March, 2015 - Wellington
- NZAGRC-PGGRc Methane Review (with ISAG): 23 April, 2015 - Palmerston North
- New Zealand Agricultural Greenhouse Gas Mitigation Conference: 28 April, 2015 - Palmerston North
- Meeting: Sally Garden, Parliamentary Commissioner for the Environment re understanding perspectives on the NZ ETS: 29 April, 2015 - Palmerston North
- NZAGRC-PGGRc Feeds & Nitrous Oxide Workshop: 29 April, 2015 - Palmerston North
- NZAGRC Soil Carbon & Nitrous Oxide Mitigation Workshop: 29 April, 2015 - Palmerston North
- Workshop: Increased N efficiency in pastoral systems: 30 April, 2015 - Palmerston North
- NZAGRC Systems & Maori Project Workshop: 1 May, 2015 - Palmerston North
- Meeting: Ministers Guy & Groser, Chris Carson and Peter Gluckman re GRA: 11 May, 2015 - Wellington
- Meeting: Climate Change Implications Panel: 15 May, 2015 - Wellington
- Meeting: PGGRc Board: 19 May, 2015 - Wellington
- Meeting: NZCCC Annual Meeting: 17 June, 2015 - Wellington

Meetings and Presentations (New Zealand)

- David Pacheco, 'Summary of research in feeds and greenhouse gases' - Presentation at the PGGRc/NZAGRC workshop on "Feeds and GHG" - 24 July, 2014
- James Wang, Peter Janssen, Tammy Lynch, Bruce van-Brunt, David Pacheco, 'A mechanistic model of methanogen-hydrogen dynamics' - Workshop on Modelling of Nutrient Digestion and Utilisation in Farm Animals (ModNut 2014) - 12 September, 2014
- Indrakumar (Kumar) Vetharaniam, Ronaldo Vibart, David Pacheco, 'A modified version of Molly to quantify methane emissions from sheep' - Project team meeting "Impacts of CFI methodologies on whole-farms" - 17 September, 2014
- Yang Li, Presentation at group meeting - 19 November, 2014
- Andy Reisinger, 'Key findings from IPCC 5th assessment report and implications for insurance' - New Zealand Society of Actuaries conference 2014, 19-22 November 2014, Dunedin - 21 November, 2014
- Sam McNally, Louis Schipper, Daniel Laughlin, Susanna Rutledge, Mike Dodd, Johan Six, 'Root turnover and C input between a moderately diverse and a ryegrass-clover pasture' - DairyNZ seminar series - 15 December, 2014
- Andy Reisinger, 'Prospects and challenges for agriculture in efforts to reduce GHG emissions'

- Meeting of the RSNZ Panel for Biological and Life Sciences - 28 January, 2015
- Xuezhao Sun, Edgar Sandoval, David Pacheco, 'Brief Communication: Effects of forage rape inclusion level in the diet on methane emissions from sheep' - The 2015 NZSAP conference - 29 January, 2015
- Suzanne Rowe, Sandra Kittelmann, Michelle Kirk, Graham Wood, Siva Ganesh, Sharon Hickey, Ken Dodds, Peter Janssen, John McEwan, 'Genetic control of the rumen microbiome in sheep' - NZSAP - 02 February, 2015
- Miko Kirschbaum, Susanna Rutledge, Paul Mudge, Nicolas Puche, Louis Schipper, David Campbell, 'Changes in soil carbon stocks of New Zealand's grazed pasture in response to variations in management and environmental factors' - Proceedings of the Fertiliser and Lime Research Centre 28th Annual Workshop - 28 February, 2015
- Andy Reisinger, 'Limiting warming to 2 degrees: Opportunities and challenges for agriculture (and New Zealand)' - Asia-Pacific Carbon Markets Roundtable - 04 March, 2015
- David Pacheco, Garry Waghorn, Peter Janssen, 'Decreasing methane emissions from ruminants grazing forages' - Presentation to AgResearch staff - 05 March, 2015
- Ron Ronimus, 'Methanogen and nitrous oxide inhibitors' - Microbiome workshop at AgResearch - 05 March, 2015
- Peter Janssen, 'Rumen Microbes. Community Profiling, Global Census, and Isolates' - Rumen Microbiome Workshop, AgResearch - 05 March, 2015
- John McEwan, Suzanne Rowe, Arjan Jonker, Peter Janssen, Sandra Kittleman, Sharon Hickey, Cesar Pinares-Patino, 'Linking host genomes with rumen characteristics and its microbiome' - Rumen microbiome workshop - 06 March, 2015
- Nicole Schon, Alec Mackay, Ross Gray, Mike Dodd, 'Do earthworms increase carbon incorporation?' - NZAGRC Workshop - 12 March, 2015
- John McEwan, 'Animal Breeding: mitigation research progress and strategies' - New Zealand Agricultural Greenhouse Gas Mitigation Conference 2015 - 28 April, 2015
- Andy Reisinger, 'Limiting warming to 2 degrees: Opportunities and challenges for agriculture' - New Zealand Agricultural Greenhouse Gas Mitigation Conference 2015 - 28 April, 2015
- Peter Janssen, 'Directing the rumen fermentation towards less methane' - New Zealand Agricultural Greenhouse Gas Mitigation Conference 2015 - 28 April, 2015
- Cecile DeKlein, Keith Cameron, 'New Zealand's nitrous oxide research: plant and animal interventions' - New Zealand Agricultural Greenhouse Gas Mitigation Conference 2015 - 04 May, 2015
- H.J. Di, K.C. Cameron, A. Podolyan, G Edwards, C de Klein, 'Nitrous oxide emissions from different pastures' - Methanet/Nzonet meeting - 06 May, 2015
- Cecile DeKlein, Keith Cameron, Hong Ji Di, Tony van der Weerden, Jiafa Luo, 'Nitrous oxide measurements in farm systems' - Methanet/Nzonet meeting - 08 May, 2015
- Linley Schofield, 'Inhibitors for methane mitigation' - Colbert and Cooper Rural Seminar - 20 May, 2015
- Louis Schipper, 'How can we improve environmental outcomes of farming?' - University of Waikato Agri-event - 09 June, 2015
- Susanna Rutledge, Aaron Wall, Paul Mudge, Jack Pronger, Dave Campbell, Louis Schipper, 'Progress at Troughton Farm: Carbon and water dynamics' - Soil Organic Matter Forum - 17 June, 2015
- Kirsty Hammond, Peter Janssen, David Pacheco, 'Summary of New Zealand research on feeds and methane' - Food Nutrition Network meeting - 25 June, 2015
- Sandeep Kumar, 'Microbiology of sheep rumens emitting different amounts of methane' - Student Symposium, Massey University, Palmerston North - 03 July, 2015
- Yang Li, 'The Genome Sequence of a Rumen Methanomassiliicoccales' - Massey University Wednesday Seminar - 15 July, 2015

NZAGRC Meetings and Presentations (International)

- FACCE-JPI SAB Meeting: 9 September, 2014 - London
- IPCC Synthesis Report Approval Meeting: 23 October, 2014 - Copenhagen
- Meeting: FACCE-JPI SAB: 20 January, 2015 - London
- Workshop: European Commission: "Research on Climate Change and Agriculture": 10 February, 2015 - Brussels
- Meeting: President of Embrapa & tours of facilities: 23 February, 2015 - Brasilia, Brazil
- Meeting: FACCE-JPI SAB: 22 April, 2015 - Brussels

Meetings and Presentations (International)

- John McEwan, 'Correlated changes after selection in sheep for methane yield' - ASGGN Vancouver workshop - 18 August, 2014
- Suzanne Rowe, John McEwan, Natalie Pickering, 'Genomic selection to decrease greenhouse gas emission in New Zealand Sheep' - 10th World Congress of Genetics and Livestock Production - 20 August, 2014
- David Pacheco, Garry Waghorn, Peter Janssen, 'Decreasing methane emissions from ruminants grazing forages' - Joint ISNH/ISRP International Conference 2014: Harnessing the Ecology and Physiology of Herbivores - 08 September, 2014
- Andy Reisinger, 'IPCC 5th Assessment Report - Global Overview and key findings for Australia' - Climate Adaptation 2014 Conference, National Climate Change Adaptation Research Facility - 30 September, 2014
- Sinead Leahy, 'The ruminant and its microbes, a genomics perspective' - 4th International Symposium on Gastrointestinal Microbial Ecology and Functionality, China - 29 October, 2014
- Andy Reisinger, 'Limiting warming to 2 degrees: Opportunities and challenges for agriculture and New Zealand' - 2015 Australian Agricultural & Resource Economics Society Conference - 13 February, 2015

International Visitors and Groups

- Visit by Te Tumu Paeroa: 6 October, 2014 - Palmerston North
- Visit: South Korean Delegation: 12 November, 2014 - Wellington
- Visit: Chinese delegation: 22 January, 2015 - Palmerston North
- Meeting: UK High Commissioner: 18 February, 2015 - Wellington
- Meeting: UK High Commissioner: 5 March, 2015 - Wellington
- Meeting: US Embassy Environment desk officials: 14 April, 2015 - Wellington
- Meeting: Tomonori Watanabe regarding feeds: 21 April, 2015 - Palmerston North
- Meeting: US Ambassador to New Zealand: 12 June, 2015 - Palmerston North

NZAGRC Global Research Alliance related interactions

- Conference: RUFORUM (on behalf of MPI): 20 July, 2014 - Mozambique
- World Farmers Organisation Study Tour: 6 November, 2014 - Palmerston North
- Meeting: GPLER Round 3 Assessment Panel: 16 December, 2014 - Wellington
- Meeting: Chris Carson, Peter Gluckman MPI re GRA strategy: 16 December, 2014 - Wellington
- Workshop: Rumen Microbiome: 6 March, 2015 - Palmerston North
- Meeting: CCAC enteric fermentation project meeting: 11 March, 2015 - Teleconference
- Meeting: FAO CCAC/GLEAM planning: 4 May, 2015 - Palmerston North
- Meeting: Vanida Khumnidrpetch, Thai Embassy: 13 May, 2015 - Palmerston North
- Meeting: GRA LRG: 23 June, 2015 - Lodi, Italy

Global Research Alliance related interactions

- Fuyong Li, Xu Sun, Gemma Henderson, Faith Cox, Peter Janssen, Leluo Guan, 'Comparative analyses of the bovine rumen microbiome using RNA and targeted DNA-based sequencing approaches' - American Society of Animal Science (ASAS), and the Canadian Society of Animal Science (CSAS) - 12 February, 2014
- Blaz Stres, Gemma Henderson, Faith Cox, Peter H. Janssen, 'Microbial communities of wild ruminants in an altitude gradient' - ISAM2015 conference (International Symposium on Anaerobic Microbiology), Portoroz, Slovenia, June 2015
- Blaz Stres, Gemma Henderson, Faith Cox, Peter H. Janssen, 'Microbial communities of wild ruminants in an altitude gradient' - 15th ISME Seoul, Korea, 24-29 August 2014
- Jie Li, Jiafa Luo, Y Shi, D Houlbrooke, L Wang, 'Nitrogen gaseous emissions from farm effluent application to grazed pastures and mitigation measures to reduce the emissions' - The Open Agricultural Journal - 25 July, 2014
- Arjan Jonker, German Molano, Christopher Antwi, Garry Waghorn, 'Methane and carbon dioxide emissions by beef heifers fed alfalfa silage at three allowances and four feeding frequencies: measured by respiration chambers, SF6 tracer technique and NZAGRC Annual Report 2015 [89]

- GreenFeed head-chamber system' - Journal of Animal Science - 30 September, 2014
- Bambang Kusumo, Marta Camps Arbustain, Ainul Mahmud, Mike Hedley, Carolyn Hedley, Roberto Calvelo Pereira, Tao Wang, Bhupinderpal Singh, 'Assessing biochar stability indices using near-infrared (NIR) reflectance spectroscopy' - Journal of Near Infrared Spectroscopy - 14 October, 2014
 - Ron Ronimus, Kristy Lunn, Stefan Muetzel, Mike Tavendale, Greg Cook, William Denny, Pat Edwards, Vince Carbone, 'Investigating transformation and degradation of scaffold compounds in the rumen to advance the development of methanogen-specific inhibitors' - ICAR International Conference on Antimicrobial Research (GPLER 2 project-16601) - 12 August, 2014
 - Corentin Leroux, Pierre Roudier, Martina Alvarez, Carolyn Hedley, 'Vis-NIR soil carbon prediction' - Landcare Research - 13 August, 2014
 - Gemma Henderson, Faith Cox, Global Rumen Census Collaborators, Adrian Cookson, Graeme Attwood, Sinead Leahy, Bill Kelly, Peter Janssen, 'Who's there? A global census of rumen microbial diversity' - 15th ISME Seoul, Korea, 24-29 August 2014
 - Eric Altermann, Ron Ronimus, Bernd Rehm, Kerri Reilly, 'Methanogen lytic enzyme PeiR domain interaction with Methanobrevibacter ruminantium M1' - New Zealand Microbiological Society Inc and the New Zealand Society for Bio Conference, 18-21 November 2014, Wellington
 - Siva Ganesh, Gemma Henderson, Faith Cox, Peter Janssen, 'Understanding and characterizing complex microbial community data' - Australasian Applied Statistics Conference 2014, 1-5 December 2014
 - Jie Li, Jiafa Luo, Yuanliang Shi, Stuart Lindsey, David Houlbrooke, Stewart Ledgard, Lingli Wang, 'Nitrous oxide emissions from farm effluent application on a New Zealand pasture' - Journal/ Soil use and management - 14 October, 2014
 - Dragana Gagic, Filomena Ng, Jasna Rakonjac, Graeme Attwood, Mark Patchett, Sandra Kittelmann, Kerri Reilly, Dong Li, Peter Janssen, 'Rumen methanogens molecular bridges' - Annual conference of the New Zealand Microbiological Society Inc and the New Zealand Society for Bio - 31 August, 2014
 - Ron Ronimus, Kristy Lunn, Vince Carbone, Stefan Muetzel, Mike Tavendale, Pat Edwards, Greg Cook, Bill Denny, William Whitman, 'Investigating transformation and degradation of scaffold compounds in the rumen to advance the development of methanogen-specific inhibitors' - New Zealand Microbiological Conference in Wellington, Nov 18-21 2014
 - Eric Altermann, Ron Ronimus, Kerri Reilly, Bernd Rehm, Natalie Burr, 'Reducing methanogen numbers using a nanobead-displayed lytic enzyme' - Invited seminar at Teagasc, Ireland - 01 September, 2014
 - Arjan Jonker, German Molano, Christopher Antwi, Garry Waghorn, 'Effect of feed intake and meal frequency on methane emissions from beef cattle' - ISNH-ISRP joint meeting Canberra - 05 September, 2014
 - Sinead Leahy, 'The rumen and its microbes: genomic insights' - 4th International Symposium on Gastrointestinal Microbial Ecology and Functionality, Nanjing Agricul - 11 September, 2014
 - Neil Wedlock, Supatsak Subharat, Bryce Buddle, Neil Wedlock, 'Saliva, a good source for purification of secretory IgA' - Journal of Immunological methods - 28 November, 2014
 - Gemma Henderson, Faith Cox, Global Rumen Census Collaborators, Adrian Cookson, Graeme Attwood, Sinead Leahy, Bill Kelly, Peter Janssen, 'Who's there? A global census of rumen microbial diversity' - Poster in Beef in BC Magazine (a magazine targeted at our local Canadian producers in British Columbia - 24 September, 2014
 - Ellis Poole, Jasna Rakonjac, Dragana Gagic, 'Multicopy display of archaeal adhesins for methane mitigation' - Massey University Research Methods seminar - 29 September, 2014
 - Ron Ronimus, Vince Carbone, Kristy Lunn, Patrick Edwards, Mike Tavendale, Stefan Muetzel, Greg Cook, William Denny, William Whitman, 'Investigating transformation and degradation of scaffold compounds in the rumen to advance the development of methanogen-specific inhibitors' - ICAR Antimicrobial Conference Madrid Oct 1-3 - 01 October, 2014
 - Jiafa Luo, Stewart Ledgard, Bridget Wise, Brent Welten, Stuart Lindsey, Amanda Judge, Mike Sprosen, 'Evaluation of the effect of dicyandiamide (DCD) on nitrous oxide emissions from cow urine deposited into a pasture soil, as affected by season and application rate' - Biology and Fertility of Soil - 18 November, 2014
 - Lin Bo, Gemma Henderson, Caixia Zou, Faith Cox, Xianwei Liang, Peter Janssen, Graeme Attwood, 'Characterization of rumen microbial community composition of buffalo feeding on diets varying in forage to concentrate ratio' - Animal Feed Science and Technology - 24 November, 2014
 - Supatsak Subharat, Dairu Shu, Tao Zheng, Bryce Buddle, Kan Kaneko, Sarah Hook, Peter Janssen, Neil Wedlock, 'Salivary antibody responses in sheep vaccinated with a methanogen protein formulated with different adjuvants' - Vaccine - 19 December, 2014
 - Ron Ronimus, Kristy Lunn, Greg Cook, Patrick J.B. Edwards, Vince Carbone, Mike Tavendale, Stefan Muetzel, William A. Denny, William Whitman, 'Investigating transformation and degradation of scaffold compounds in the rumen to advance the development of methanogen-specific inhibitors'

- New Zealand Microbiological Society Annual Meeting in Wellington - 16 November, 2014
- Dragana Gagic, Filomena Ng, Jasna Rakonjac, Graeme Attwood, Sandra Kittelmann, Kerri Reilly, Peter Janssen, 'Rumen methanogen "molecular bridges"' - New Zealand Microbiology Society Conference - 19 November, 2014
- Jie Li, Jiafa Luo, Yuanliang Shi, Stuart Lindsey, David Houlbrooke, Stewart Ledgard, 'Nitrous oxide emissions from farm effluent application on a New Zealand pasture' - 2014 NZ Society of Soil Science Conference (NZSSS Conference) - 21 November, 2014
- Gemma Henderson, Faith Cox, Siva Ganesh, Arjan Jonker, Wayne Young, Global Rumen Census Collaborators, Peter Janssen, 'A conserved rumen microbial community supporting ruminant herbivory' - Science (or other high impact journals) - 12 January, 2015
- Jiafa Luo, Stewart Ledgard, Bridget Wise, Brent Welten, Stuart Lindsey, Amanda Judge, Mike Sprosen, 'Effect of dicyandiamide (DCD) delivery method, application rate, and season on pasture urine patch nitrous oxide emissions' - Biology and Fertility of Soil - 18 November, 2014
- Fiona Ehrhardt, Jean-Francois Soussana, Val Snow, Josef Beaudrais, Mark Lieffering, Paul Newton, Many (about 20) Others, 'An international inter-comparison and benchmarking of crop and pasture models simulating GHG emissions and carbon sequestration' - Climate Smart Agriculture, 16-18 March 2015, Montpellier, France - 28 November, 2014
- Siva Ganesh, Gemma Henderson, Faith Cox, Peter Janssen, 'Understanding and characterizing complex microbial community data' - Australasian Applied Statistics Conference 2014 - 04 December, 2014
- Eddy Minet, Karl Richards, Gary Lanigan, John Murphy, Jiafa Luo, Stewart Ledgard, 'Animal delivery of a nitrification inhibitor to urine patches' - RAMIRAN 2015 - 16th International Conference - Rural-Urban Symbiosis - 11 December, 2014
- Graeme Attwood, Janine Kamke, Sandra Kittelmann, Peter Janssen, Siva Ganesh, 'Deep sequencing the rumen microbiome: update for TAP Wellington December 2014' - Presentation to the Technical Advisory Panel for GPLER projects - 16 December, 2014
- Brendon Welten, Stewart Ledgard, Sheree Balvert, Martin Kear, Moira Dexter, 'Effects of oral administration of dicyandiamide to lactating dairy cows on residues on milk and the efficacy of delivery via a supplementary feed source' - Effects of oral administration of dicyandiamide to lactating dairy cows on residues on milk - 24 March, 2015
- William Kelly, 'The HUNGATE1000. A catalogue of reference genomes from the rumen microbiome.' - Rumen Microbiome Workshop - 06 March, 2015
- Graeme Attwood, 'Sequencing the rumen microbiome' - Rumen Microbiome Workshop - 06 March, 2015
- Janine Kamke, Sandra Kittelmann, Weibing Shi, Siva Ganesh, Jeff Froula, Edward M Rubin, Graeme T Attwood, 'Bacterial communities and their activities correlated with methane yields in New Zealand sheep' - DOE Joint Genome Institute Genomics of Energy and Environment Meeting, 23-26 March 2015
- Janine Kamke, Sandra Kittelmann, Weibing Shi, Siva Ganesh, Jeff Froula, Edward M Rubin, Graeme T Attwood, 'Bacterial communities and their activities correlated with methane yields in New Zealand sheep' - 10th annual DOE Joint Genome Institute Genomics of Energy and Environment Meeting - 17 March, 2015
- Gemma Henderson, Faith Cox, Siva Ganesh, Arjan Jonker, Wayne Young, Global Rumen Census Collaborators, Peter Janssen, 'A global census of rumen microbial diversity' - Hungate1000 workshop - CGIF - 19 March, 2015
- Stefan Muetzel, 'JPI Progress March 2015' - Presentation at the JPI workshop in Spain to the Rumen stability consortium - 20 March, 2015
- Fuyong Li, Gemma Henderson, Xu Sun, Cox Faith, Janssen Peter, Guan Leluo, 'Total RNA sequencing represents a suitable alternative to targeted DNA sequencing to characterize the rumen microbiome' - Microbial Ecology or similar journal - 19 May, 2015
- Lei Zhonga, Coby J. Hoogendoorn, Saman Bowatte, Frank Yonghong Li, Yanfen Wang, Dongwen Luo, 'Grazing intensity and microtopography effects on microorganisms and nitrogen transformation processes responsible for nitrous oxide emissions from hill pastures' - Agriculture Ecosystem and Environment Journal - 08 June, 2015
- William Kelly, 'Toward a reference genome set of rumen microbes' - Hungate1000 Genome Workshop Congress on Gastrointestinal Function 2015 - 13 April, 2015
- David Stevens, Seth Laurenson, Tony van der Weerden, 'Developing solutions for restricted grazing managements to mitigate environmental impacts' - Australasia Pacific Extension Network Conference - 15 April, 2015
- David Stevens, Seth Laurenson, Tony van der Weerden, 'Developing solutions for restricted grazing managements to mitigate environmental impacts' - Australasia Pacific Extension Network Conference - 15 April, 2015
- Vanessa Cave, Siva Ganesh, 'Integrating high-dimensional datasets' - AgResearch Science conference - 21 April, 2015
- Sergio Morales, Md Sainur Samad, Lars Bakken, Shahid Nadeem, Timothy Clough, Cecile de Klein, Karl Richards, Gary Lanigan, 'High-resolution denitrification kinetics in pasture soils link N₂O emissions to pH, and

- denitrification to soil respiration and moisture content' - Soil Biology and Biochemistry Journal - 09 July, 2015
- Sinead Leahy, William Kelly, Gemma Henderson, 'Hungate1000 inside story' - AgResearch Internal & Social media - 08 May, 2015
 - Eric Altermann, 'GRA reports' - Reports made available to PolyBatics - 13 May, 2015
 - Janine Kamke, 'Uncultured microbes and their functions in host systems: from complex communities to single cells' - University of Auckland School of Biological Sciences Research Seminar Series - 22 May, 2015
 - William Kelly, 'AgResearch-led project focuses on potential of rumen microbiome' - AgResearch public website - 05 June, 2015
 - Filomena Ng, Dragana Gagic, Graeme Attwood, Sandra Kittlemann, Peter Janssen, Kerri Reilly, Jasna Rakonjac, Mark Patchett, 'Molecular bridges of the rumen microbial alliances' - Poster at 13th Symposium on Bacterial Genetics and Ecology - 15 June, 2015
 - Carolyn Hedley, 'Evaluation of methods to remove moisture effects from Vis-NIR spectra for soil organic carbon prediction models' - 19 June, 2015
 - Tony van der Weerden, Seth Laurenson, Iris Vogeler, Pierre Beukes, Cecile de Klein, 'Evaluation of an on-farm tool to reduce nitrous oxide emissions from dairy cattle grazing' - abstract for GGAA 2016, 14-18 February 2016 - submitted 26 June, 2015
 - Jiafa Luo, Stewart Ledgard, Bridget Wise, Stewart Lindsey, 'Evaluation of the effect of dicyandiamide (DCD) on nitrous oxide emissions from cow urine deposited into a pasture soil, as affected by DCD application rate and delivery method' - abstract for GGAA 2016, 14-18 February 2016 - submitted 29 June, 2015
 - Garry Waghorn, Arjan Jonker, Kevin Macdonald, 'Measuring methane using GreenFeeds from grazing dairy cows' - abstract for GGAA 2016, 14-18 February 2016 - submitted 30 June, 2015
 - David Pacheco, Mark Evered, Aaron B. Ingham, Ermias Kebreab, Holland C. Dougherty, Roger S. Hegarty, Malcolm J. McPhee, 'Evaluation of AusBeef Methane Predictions from Beef Cattle' - abstract for GGAA 2016, 14-18 February 2016 - submitted 30 June, 2015
 - Ron Ronimus, Greg M Cook, James Cheung, Marion Weimar, Chris McSweeney, Mark Morrison, Yasuo Kobayashi, William B. Whitmann, Vince Carbone, Linley Schofield, 'Development and validation of 48-well and 96-well plate methods for screening methanogens to identify novel inhibitors' - abstract for GGAA 2016, 14-18 February 2016 - submitted 01 July, 2015
 - Ron Ronimus, Greg Cook, Gemma Henderson, 'Specific and chemically-defined inhibitors of ruminant methanogens: a review' - abstract for GGAA 2016, 14-18 February 2016 - submitted 01 July, 2015
 - Eddy Minet, SF Ledgard, JB Murphy, G Lanigan, D Hennessy, E Lewis, P Forrestal, KG Richards, 'Mixing dicyandiamide (DCD) with supplementary feeds for cattle' - abstract for GGAA 2016, 14-18 February 2016 - submitted 01 July, 2015
 - Sergio Morales, Timothy Clough, Gary Lanigan, Cecile de Klein, Lars Bakken, Md Sainur Samad, David Rex, Charlotte Johns, Karl Richards, 'Contribution of the co-denitrification process to soil nitrous oxide and dinitrogen emissions under ruminant urine patches.' - abstract for GGAA 2016, 14-18 February 2016 - submitted 07 July, 2015
 - Sergio Morales, Md Sainur Samad, Timothy Clough, Karl Richards, Gary Lanigan, Lars Bakken, Cecile de Klein, 'Response of gene abundance and transcription of N cycling genes linked to denitrification kinetics under ruminant urine patches' - abstract for GGAA 2016, 14-18 February 2016 - submitted 07 July, 2015
 - Sergio Morales, Md Sainur Samad, Lars Bakken, Shahid Nadeem, Timothy Clough, Cecile de Klein, Karl Richards, Gary Lanigan, 'High-resolution denitrification kinetics in pasture soils link N₂O emissions to pH, and denitrification to soil respiration and moisture content' - abstract for GGAA 2016, 14-18 February 2016 - submitted 07 July, 2015
 - Ellis Poole, 'Mitigation of Methane Production by *Methanobrevibacter ruminantium* using Filamentous Phage Display' - MSc Thesis - 24 July, 2015

Media Interactions

Significant media coverage was generated by the NZAGRC conference and associated media release about the inhibitor programme. This is not captured in the interactions below.

- Visit: Peter Burke, Rural News: 5 August, 2014 - Palmerston North
- Peter Janssen, 'Tiny organisms a big challenge to methane researchers' - Rural News - 19 August, 2014
- Journalist visit: Jill Galloway: 22 September, 2014 - Palmerston North
- Andy Reisinger, 'Key findings from latest IPCC report' - radio interview - 07 November, 2014
- Media: TV3 filming methane chambers work (aired 1 Jan 2015): 28 November, 2014 - Palmerston North
- Cecile DeKlein, 'Interview Dominion post on N₂O highlights 2014' - Dominion Post - 20 December, 2014
- Media: The Nation: 21 May, 2015 - Palmerston North

NZAGRC Conference Presentations

- Attended NCCARF conference: 29 September, 2014 - Brisbane
- Presentation: New Zealand Society of Actuaries (IPCC): 21 November, 2014 - Dunedin
- Presentation: Royal Society of NZ Biological and Life Sciences: 28 January, 2015 - Wellington
- Presentation: Australian Agricultural and Resource Economics Society: 11 February, 2015 - Rotorua
- Presentation: Royal Society of New Zealand: Trends in greenhouse gas emissions: 16 February, 2015 - Wellington
- Presentation: Marginal abatement cost curves and mitigation potential for agriculture in France and Europe (sponsorship): 17 February, 2015 - Wellington
- Presentation: Asia-Pacific Carbon Markets Roundtable: 4 March, 2015 - Rotorua
- Presentation: Climate Smart Agriculture Conference: 16 March, 2015 - Montpellier

Conference Presentations

- Sandra Kittelmann, Cesar S Pinares-Patino, Wendy E Bain, Henning Seedorf, Michelle R Kirk, Siva Ganesh, John C McEwan, Peter H Janssen, 'Bacterial community types linked with high- and low-methane emitting sheep' - 15th International Symposium on Microbial Ecology - 12 August, 2014
- Wendy Bain, Louret Bezuidenhout, Neville Jopson, Cesar Pinares-Patino, John McEwan, 'Rumen Differences between sheep identified as being low or high methane emitters' - 10th World Congress of Genetics and Livestock Production - 19 August, 2014
- Henning Seedorf, Sandra Kittelmann, Gemma Henderson, Peter Janssen, Presentation - 15th ISME Seoul, Korea, 24-29 August 2014
- Sergio Morales, Neha Jha, Surinder Sagar, 'Impact of urine and DCD application on microbial communities in dairy-grazed pasture soils' - 15th International Symposium on Microbial Ecology - 26 August, 2014
- Xuezhao Sun, Sarah Maclean, Dongwen Luo, David Pacheco, 'Methane Emissions were less from sheep fed Summer Forage Brassicas than Perennial Ryegrass/White Clover Pasture' - ISNH/ISRP (The joint International Symposium on the Nutrition of Herbivores/International Symposium - 27 August, 2014
- Arjan Jonker, Kate Lowe, Stewart Ledgard, David Pacheco, 'Substitution of lucerne silage by increasing levels of maize silage or maize grain results in a quadratic response in methane emissions from sheep' - ISNH-ISRP joint meeting Canberra, Australia - 03 September, 2014
- Pierre Noziere, Emilie Ollion, David Pacheco, Daniel Sauvage, 'Evaluation of a bovine volatile fatty acids (VFA) prediction model using data on sheep' - 8th International Workshop on Modelling Nutrient Digestion and Utilisation in Farm Animals - 04 September, 2014
- Stewart Ledgard, Andy Reisinger, 'Implications of alternative greenhouse gas metrics for life cycle assessments of livestock food products' - Ninth International Life Cycle Assessment of Foods Conference - 08 October, 2014
- Neil Wedlock, Supatsak Subharat, Dairu Shu, Debjit Dey, Henning Seedorf, Tao Zheng, Sandra Kittelmann, Bryce Buddle, Peter Janssen, 'Development of a sub-unit vaccine to reduce methane emissions in sheep and cattle' - NZMS conference in Wellington - 14 November, 2014
- Preeti Raju, Gemma Henderson, Michael Tavendale, Jasna Rakonjac, Peter Janssen, 'Identifying alternative hydrogen utilisers in the rumen' - NZMS Conference - 18 November, 2014
- Sandeep Kumar, Peter H Janssen, Sandra Kittelmann, Gemma Henderson, 'A closer look at rumen bacterial species associated with low-methane emitting sheep' -

- NZMS/NZSBMB Joint Conference Molecules and Microbes, 2014 - 18 November, 2014
- Eric Altermann, Kerri Reilly, Ron Ronimus, Bernd Rehm, 'Methanogen lytic enzyme PeiR domain interaction with Methanobrevibacter ruminantium M1' - NZMS 2014 - 18 November, 2014
 - Neha Jha, Surinder Saggar, Julie Deslippe, Donna Giltrap, Russ Tillman, 'Effects of urine and DCD application on denitrification and denitrifier communities in soils' - New Zealand Society of Soil Science Conference - 01 December, 2014
 - Saman Bowatte, Paul Newton, Coby Hoogendoorn, David Hume, Alan Stewart, Shona Brock, Phil Theobald, 'Forage plant cultivar and genotype effects on soil nitrification activity' - New Zealand Soil Science Society Annual Conference - 02 December, 2014
 - Susanna Rutledge, Paul Mud, Aaron Wall, Dave Campbell, Louis Schipper, 'CO₂ emissions during pasture renewal via cultivation or direct drilling' - New Zealand Society of Soil Science Conference - 03 December, 2014
 - Nicole Schon, Alec Mackay, Ross Gray, Mike Dodd, 'Do earthworms increase carbon incorporation?' - NZSSS Conference - 03 December, 2014
 - Sam McNally, Louis Schipper, Daniel Laughlin, Susanna Rutledge, Mike Dodd, Johan Six, 'Root turnover and C input between a moderately diverse and a ryegrass-clover pasture' - New Zealand Society of Soil Science - 03 December, 2014
 - Xuezhao Sun, Edgar Sandoval, David Pacheco, 'Effects of forage rape inclusion levels in the diet on methane emissions from sheep' - The 2015 NZSAP conference - 29 January, 2015
 - Ron Ronimus, Linley Schofield, Vince Carbone, Yanli Zhang, Debjit Dey, Carrie Sang, Amy Beattie, 'Expression, purification and characterisation of methanogen enzymes to discover novel inhibitors for reducing ruminant methane emissions' - Lorne Protein Structure and Function conference, Feb 9-13th, Australia
 - Miko Kirschbaum, Susanna Rutledge, Paul Mudge, Nicolas Puche, Louis Schipper, David Campbell, 'Changes in soil carbon stocks of New Zealand's grazed pasture in response to variations in management and environmental factors' - Fertiliser and Lime Research Centre 28th Annual Workshop - 11 February, 2015
 - Ron Ronimus, Yanli Zhang, Carrie Sang, Debjit Dey, Kristy L. Lunn, Andrew J. Sutherland-Smith, Greg Cook, Linley R. Schofield, 'Methanogen Metabolism and How Their Unique Enzymes' - 6th Pan Commonwealth Veterinary Conference of the CVA and the 27th Congress of the Veterinary Association - 09 March, 2015
 - Andy Reisinger, 'Implications of GHG emission metrics for agricultural emission trends and targets' - Climate Smart Agriculture 2015 Open Science Conference - 10 March, 2015
 - Preeti Raju, Gemma Henderson, Michael Tavendale, Jasna Rakonjac, Peter Janssen, 'Homoacetogenic activity in the sheep rumen' - Congress on Gastrointestinal Function 2015 - 10 April, 2015
 - Yang Li, Sinead Leahy, Jeyamalar Jeyanathan, Faith Cox, Eric Altermann, William Kelly, Suzanne Lambie, Peter Janssen, Jasna Rakonjac, Graeme Attwood, 'Genome Sequence of a Rumen *Methanomassiliicoccales*' - 2015 Congress on Gastrointestinal Function - 13 April, 2015
 - Susanna Rutledge, Paul Mudge, Aaron Wall, Dave Campbell, Louis Schipper, 'CO₂ balance of an intensively grazed temperate pasture during pasture renewal via cultivation or direct drilling' - European Geophysical Union General Assembly 2015 - 15 April, 2015
 - Suzanne Rowe, Neville Jopson, Eleanor Linscott, Mark Young, Ken Dodds, Shannon Clarke, Tim Byrne, Graham Alder, 'Contract session: Application of genetic technologies to the New Zealand sheep industry' - New Zealand society of animal production - conference proceedings - 01 July, 2015
 - Saman Bowatte, Coby Hoogendoorn, Paul Newton, Shona Brock, Phil Theobald, 'Pasture species and cultivar effects on nitrous oxide emissions after cattle urine application' - New Zealand Institute of Agricultural & Horticultural Science and the New Zealand Branch of the International Association of Plant Biotechnology, 1-4 July 2015
 - Andy Reisinger, Joeri Rogelj, Adam Daigneault, 'Interactions between agriculture non-CO₂ mitigation choices and fossil CO₂ emissions within integrated low-carbon pathways' - Our Common Future under Climate Change conference, 7-10 July 2015

Submitted

- Bowatte, S., Ganesh, S., Jha, N., & Saggar, S. (Submitted). Metagenomic analysis reveals immediate responses of bacterial communities to water saturation and cattle urine application to pastoral soils. *Soil Biology and Biochemistry*.
- Calvelo Pereira, R., Hedley, M., Camps Arbestain, M., Wisnubroto, E., Green, S., Saggar, S., Mahmud, A. F. (2015). Net changes of soil C stocks in two grassland soils 26 months after simulated pasture renovation including biochar addition. *GCB Bioenergy*. doi: 10.1111/gcbb.12271
- Calvelo Pereira, R., Wisnubroto, E., Hedley, M., Camps Arbestain, M., Green, S., & Saggar, S. (Submitted). Can a deep application of biochar mitigate a pulse of greenhouse gas emissions after a fertilizer application to mature pastures? *Plant & Soil*.
- Gregorini, P., Beukes, P., Waghorn, G., Pacheco, D., & Hanigan, M. (2015). Development of an improved representation of rumen digesta outflow in a mechanistic and dynamic model of a dairy cow, Molly. *Ecological Modelling*, 313, 293-306. doi: 10.1016/j.ecolmodel.2015.06.042
- Jha, N., Deslippe, J., Saggar, S., Giltrap, D., & Tillman, R. W. (2014). The effects of soil properties on denitrifier community structure, abundance, and N₂O and N₂ production in New Zealand dairy grazed pasture soils. *Soil Biology and Biochemistry*.
- Jonker, A., Lowe, K., Ledgard, S. F., & Pacheco, D. (Submitted). Effect of substitution of lucerne silage by increasing levels of maize silage or maize grain on methane emissions and rumen fermentation in sheep and in an automated in vitro gas production system. *Animal*.
- Kelly, W. J., Leahy, S. C., Altermann, E., Attwood, G. T., Henderson, G., Janssen, P. H., Naylor, G. (Submitted). The complete genome sequence of *Eubacterium limosum* SA11, a metabolically versatile rumen acetogen. *Standards in Genomic Sciences*
- Kelly, W. J., Leahy, S. C., Attwood, G. T., Altermann, E., Pacheco, D., & Li, D. (Submitted). The complete genome sequence of the rumen methanogen *Methanobrevibacter millerae* SM9. *Standards in Genomic Sciences*.
- Kittelmann, S., Kirk, M. R., Jonker, A., McCulloch, A., & Janssen, P. H. (2015). Buccal swabbing as a noninvasive method to determine bacterial, archaeal, and eukaryotic microbial community structures in the rumen. *Applied and Environmental Microbiology*, 81(21), 7470-7483. doi: 10.1128/aem.02385-15
- Morales, S. E., Jha, N., & Saggar, S. (2015). Impact of urine and the application of the nitrification inhibitor DCD on microbial communities in dairy-grazed pasture soils. *Soil Biology and Biochemistry*, 88, 344-353. doi: 10.1016/j.soilbio.2015.06.009
- Pickering, N. K., Hickey, S., Pinares-Patino, C., Rowe, S., Dodds, K. G., & McEwan, J. C. (Submitted). Genome wide association study for methane emissions in New Zealand dual purpose sheep, utilising the HD chip. *PLoS ONE*.
- Rasmussen, S., Liu, Q., Jones, C., Xue, H., & Parsons, A. J. (Submitted). Does gibberellin biosynthesis play a critical role in the growth of *Lolium perenne*? Evidence from a transcriptional analysis of gibberellin and carbohydrate metabolic genes after defoliation. *Frontiers in Plant Science*.
- Ronimus, R. S., Carbone, V., Schofield, L. R., & Sutherland-Smith, A. J. (Submitted). Structure files for manuscript titled: Structure and evolution of the archaeal lipid synthesis enzyme sn-glycerol-1-phosphate dehydrogenase. *Proceedings of the National Academy of Sciences*.
- Ronimus, R. S., Carbone, V., Schofield, L. R., Sutherland-Smith, A. J., & Beattie, A. K. (Submitted). The Crystal Structure of GTP and GMP-Bound GTP Cyclohydrolase III from *Methanobrevibacter ruminantium* M1. *Proteins: Structure, Function and Bioinformatics*.
- Ronimus, R. S., Schofield, L. R., Zhang, Y., Al-Attar, S., Beattie, A. K., Tootill, C., Carbone, V. (Submitted). A Class I 3-hydroxy-3-methylglutaryl-CoA reductase (HMGR) from *Methanobrevibacter ruminantium*: cloning, expression, purification, characterisation and statin inhibition. *FEBS Letters, or similar*
- Ronimus, R. S., Zhang, Y., Beattie, A. K., & Schofield, L. R. (Submitted). Expression, purification and characterisation of (R)-sulfolactate dehydrogenase (ComC) from the rumen methanogen *Methanobrevibacter* species SM9. *FEBS Letters, or similar*
- Schon, N., Mackay, A., Gray, R. A., & Van Koten, C. (Submitted). Establishment of *Aporrectodea longa* and quantification of dung carbon incorporation in soils under permanent pasture. *Agriculture, Ecosystems and Environment*.
- Schon, N., Mackay, A., Gray, R. A., Voegler, I., Van Koten, C., & Dodd, M. B. (Submitted). Influence of earthworm abundance and functional diversity on soil structure and implications for plant growth and nitrous oxide emissions. *Applied Soil Ecology*.
- Sparling, G., Chibnall, E. J., Pronger, J., Rutledge, S., Wall, A., Campbell, D. I., & Schipper, L. A. (Submitted). Contribution from leached DOC to C balances in grazed dairy

pasture: Field estimations from Waikato, New Zealand. *NZ Journal of Agricultural Research*.

- Sun, X., Harland, R., & Pacheco, D. (Submitted). Effect of altering ruminal pH by dietary buffer supplementation on methane emissions from sheep fed forage rape. *TBC*.
- Vetharaniam, K., Vibart, R. E., Hanigan, M., Janssen, P. H., Tavendale, M., & Pacheco, D. (Submitted). A modified version of the Molly

rumen model to quantify methane emissions from sheep. *Journal of Animal Science*.

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- Bryce Little, Aaron Ingham, Roger Hegarty, Ermias Kebreab, David Pacheco, Barry Nagorcka, Malcolm McPhee, 'AusBEEF: revision of the model to simulate animal and enterprise impacts of methane mitigation implemented on farm' - 8th International Workshop on Modelling Nutrient Digestion and Utilization in Farm Animals, 15-17th September, 2014, Cairns, Australia
- James Wang, Peter Janssen, Tammy Lynch, Bruce van-Brunt, David Pacheco, 'A mechanistic model of methanogen-hydrogen dynamics' - 8th International Workshop on Modelling Nutrient Digestion and Utilization in Farm Animals, 15-17th September, 2014, Cairns, Australia
- Mark Hanigan, Ronaldo Vibart, Stefan Muetzel, Mike Tavendale, Pablo Gregorini, David Pacheco, 'Predictions of digestion and methane production from fresh grass by the Molly cow model' - 8th International Workshop on Modelling Nutrient Digestion and Utilization in Farm Animals, 15-17th September, 2014, Cairns, Australia
- Peter Janssen, 'Fixed cells of culture ISO4-H5' - Transfer of fixed cells to Lethbridge Research Centre, Canada - 22 August, 2014
- Sergio Morales, Neha Jha, Surinder Saggar, 'Biogeography and biophysicochemical traits link N₂O emissions and microbial communities across New Zealand pasture soils' - 15th International symposium on Microbial Ecology - 24 August, 2014
- Preeti Raju, Peter Janssen, Gemma Henderson, Michael Tavendale, Jasna Rakonjac, 'Identifying alternative hydrogen utilisers in the rumen' - NZMS Conference, 18-21 November 2014
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- Andreas Schielke, 'Crystallisation of G1PDH and GGGPS, Proteins Involved in Phospholipid Biosynthesis in Archaea' - Bachelor thesis, University of Duisburg-Essen, Germany - 30 August, 2014
- Sandeep Kumar, Peter Janssen, Sandra Kittelmann, Gemma Henderson, 'A closer look at rumen bacterial species associated with low-methane emitting sheep' - New Zealand Microbiological Society Conference 2014, 18-21 November 2014
- Saman Bowatte, Paul C.D Newton, Coby Hoogendoorn, David E. Hume, Shona Brock, Phil Theobald, 'Plant effects on soil nitrification potential' - New Zealand Society of Soil Science Conference, December 1-4, 2014
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- Susanna Rutledge, Paul Mudge, Aaron Wall, Dave Campbell, Louis Schipper, 'CO₂ emissions during pasture renewal via cultivation or direct drilling' - New Zealand Society of Soil Science Conference, December 1-4, 2014
- Nicole Schon, Alec Mackay, Ross Gray, Mike Dodd, 'Do earthworms increase carbon incorporation?' - NZSSS Conference 2014, 1-4 December 2014
- Graeme Attwood, 'NZAGRC-PGGRC IP developed in Objective 5.5.6' - Global Partnerships in Livestock Emissions Research (GPLER) project Deep Sequencing the Rumen Microbe - 22 September, 2014

- Peter Janssen, Neil Wedlock, 'GT2 identification through serum testing' - Release of information to Baldwins for patent purposes - 03 October, 2014
- David Pacheco, Sunny Sun, 'Data from experiments with beef cattle (pasture quality and forage rape)' - Data will be used to evaluate methane predictions from Ausbeef model - 23 October, 2014
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- Andy Reisinger, 'Implications of alternative GHG emission metrics for emission trends and targets' - Climate Smart Agriculture 2015 Open Science Conference, 16-18 March 2015, Montpellier, France
- Rogier Schulte, Andy Reisinger, Harry Clark, Trevor Donnellan, Gary Lanigan, 'Opportunities and limitations of emissions intensity as a metric for climate change mitigation from the livestock sector' - Climate Smart Agriculture 2015 Open Science Conference, 16-18 March 2015, Montpellier, France
- Ron Ronimus, Vince Carbone, Linley Schofield, 'Business case study for Australian Synchrotron show casing PGGRC, NZAGRC, NZ government and AgResearch' - Australian Synchrotron in Melbourne (website) - 02 December, 2014
- Xuezhao Sun, Edgar Sandoval, David Pacheco, 'Effects of inclusion level of forage rape in the diet on methane emissions from sheep' - 2015 NZSAP conference, 28 June - 1 July 2015
- Suzanne Rowe, Sandra Kittelmann, Graham Wood, Siva Ganesh, Sharon Hickey, Peter Janssen, John McEwan, 'Genetic control of the rumen microbiome in sheep' - New Zealand Society of Animal Production (NZSAP), 28 June 2014
- "Ron Ronimus, Linley Schofield, Yanli Zhang, Amy Bettie, Carrie Sang, Vince Carbone, 'Expression, purification and characterisation of methanogen enzymes' - Lorne Conference on Protein Structure and function, 8-12 February 2015
- "Susanna Rutledge, Paul Mudge, Aaron Wall, Dave Campbell, Louis Schipper, 'CO₂ balance of an intensively grazed temperate pasture during pasture renewal via cultivation or direct drilling' - European Geophysical Union General Assembly 2015, 12-17 April
- Preeti Raju, 'Identifying alternative hydrogen utilisers in the rumen' - New Zealand Microbiological Society (NZMS) - 15 January, 2015
- Suzanne Rowe, Jenny Jeungel, John McEwan, 'Sharing of data for reproduction research.' - Peer reviewed journal dependent on final results - 23 January, 2015
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- Eric Altermann, Linley Schofield, 'How does it work? Functional characterisation of a unique archaeal lytic enzyme discovered in the *Methanobrevibacter ruminantium* virus phi mru' - Marsden proposal - 05 February, 2015
- Preeti Raju, Gemma Henderson, Michael Tavendale, Jasna Rakonjac, Peter Janssen, 'Identifying alternative hydrogen utilisers in the rumen' - Congress on Gastrointestinal Function 2015, 13-15 April 2015
- Yang Li, 'The Genome Sequencing of a Member of the Rumen Methanomassiliicoccales - Methanogenic Archaeon Isolate ISO4-H5' - 2015 Congress on Gastrointestinal Function, 13-15 April 2015, Chicago
- Ron Ronimus, Yanli Zhang, Carrie Sang, Debjit Dey, Kristy L. Lunn, Andrew J. Sutherland-Smith, 'Methanogen Metabolism and How Their Unique Enzymes Can Be Targeted in the Development of Methanogen-Specific Inhibitors' - 6th Pan Commonwealth Veterinary Conference of the CVA and the 27th Congress of the Veterinary Association, 23-27 March 2015, Kuala Lumpur
- Vince Carbone, Ron Ronimus, 'Fast start Marsden abstract' - Marsden fund council by the royal society of New Zealand - 11 February, 2015
- Miko Kirschbaum, Susanna Rutledge, Paul Mudge, Nicolas Puche, Louis Schipper, David Campbell, 'Changes in soil carbon stocks of New Zealand's grazed pasture in response to variations in management and environmental factors' - Fertiliser and Lime Research Centre 28th Annual Workshop, 12 February 2015
- Ron Ronimus, 'Associate investigator in MBIE Smart Idea Bid' - MBIE Smart Idea Bid Portal - 19 February, 2015
- Andy Reisinger, Adam Daigneault, Joeri Rogelj, 'Interactions between agriculture mitigation choices and fossil CO₂ emissions within integrated low-carbon pathways to limit warming to 2 degrees' - Our Common Future conference, 10-15 July 2015
- Ron Ronimus, Vince Carbone, Linley Schofield, Bishwa Subedi, Andrew Sutherland-Smith, 'Methanogen cell wall evolution Marsden annual report' - Marsden (Royal Society) - 06 March, 2015
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- Conference, Kuala Lumpur, Malaysia 23-27 March 2015
- Susanna Rutledge, Dave Campbell, Aaron Wall, Louis Schipper, 'Scott Farm data' - Contribute to a synthesis paper about managed ecosystems led by Australian researchers. - 26 March, 2015
 - Linley Schofield, Yanli Zhang, Carrie Sang, Renee Atua, Salome Molano, Amy Beattie, Debjit Dey, Greg Cook, James Cheung, Yasuo Kobayashi, William Whitman, Chris McSweeney, Mark Morrison, Bill Denny, Andrew Sutherland-Smith, Vince Carbone, Ron Ronimus, 'High-throughput screening for the discovery of novel inhibitors to mitigate ruminant methane emissions' - Pacificchem 2015 Conference, 15-20 December 2015, USA
 - Eric Altermann, 'Smart Ideas Phase 2, Mitigating methane emission from ruminant livestock using secondary metabolites' - Smart Ideas Phase 2 proposal - 31 March, 2015
 - Ron Ronimus, Charlotte Schafer, 'Investigation into thermophilic methanogen cofactor' - University of Duisburg-Essen Master's thesis - 02 April, 2015
 - Ron Ronimus, Marion Woermann, 'Investigation of methanogenesis marker proteins: implications for environmental processes' - University of Duisburg, Dusseldorf-Essen, MSc thesis - 02 April, 2015
 - Ron Ronimus, Stefan Muetzel, Mile Tavendale, Kristy Lunn, Yanli Zhang, Carrie Sang, Renee Atua, Marion Weimar, James Cheung, Andrew Sutherland-Smith, Patrick Edwards, William Whitman, William Denny, Greg Cook, Vince Carbone, Linley Schofield, 'Targeting rumen methanogens to aid the development of methane mitigation agents' - Annual European Biochemistry Conference, 24-28 August 2015, Brazil
 - John McEwan, 'Abacusbio project' - Confidential communication - 26 May, 2015
 - Sandeep Kumar, 'Walsh Fellowship student progress report' - TEAGASC WALSH FELLOWSHIPS SCHEME - 30 May, 2015
 - Cecile DeKlein, Mike Harvey, 'Nitrous Oxide Chamber Methodologies: Evolving Issues and Collection of Ancillary Data to Broaden Flux Interpretations' - ASA-CSSA-SSSA international annual meeting, 15-18 November 2015, Minneapolis
 - Vince Carbone, Kristy Lunn, Stefan Muetzel, Michael Tavendale, Yan-li Zhang, Carrie Sang, Debjit Dey, Renee Atua, Marion Weimar, James Cheung, Patrick Edwards, William Whitman, William Denny, Siva Ganesh, Mark Morrison, Christopher McSweeney, Yasuo Kobayashi, Linley Schofield, Ron Ronimus, 'High throughput screening of the rumen methanogens in aiding the development of methane mitigation agents' - ComBio 2015, 27 September 2015-1 October 2015, Melbourne
 - Saman Bowatte, Coby Hoogendoorn, Paul C.D Newton, Shona Brock, Phil Theobald, Yang Liu, 'Nitrous oxide emissions after cattle urine application to soils of different pasture species and cultivars' - International Conference The East and Southeast Asia Federation of Soil Science Societies (ESAFS), Nanjing, China, 18-21, September, 2015
 - Hong Xue, Qianhe Liu, Chris S Jones, Anthony J Parsons, Susanne Rasmussen, 'Plant Growth, Animal Nutrition and GHG Mitigation' - NZIAHS/IAPB Conference: Plants for the Future, 1-3 July 2015 - 24 June, 2015
 - Neil Wedlock, Suparsak Subharat, Dairu Shu, Faith Cox, Tania Wilson, Sandra Kittelmann, Tao Zheng, Bryce Buddle, Sarah Hook, Eric Altermann, Sinead Leahy, William Kelly, Graeme Attwood, Peter Janssen, 'Vaccination of ruminants to reduce methane emissions' - GGAA 2016, 14-18 February 2016
 - Xuezhao Sun, David Pacheco, Dongwen Luo, 'Forage brassicas as a mitigation tool for enteric methane emissions' - GGAA 2016, 14-18 February 2016 - 29 June, 2015
 - Saman Bowatte, Coby Hoogendoorn, Paul Newton, Shona Brock, Phil Theobald, 'Nitrous oxide emissions from bare areas in pastures' - GGAA 2016, 14-18 February 2016 - 30 June, 2015
 - Natasha Swainson, Stefan Muetzel, Harry Clark, 'Additional data to the methane inventory for sheep and the effect on the current predictions' - GGAA 2016, 14-18 February 2016 - 01 July, 2015
 - Ron Ronimus, Greg Cook, 'An integrated compound library screening approach for discovery of specific inhibitors for mitigating ruminant methane emissions' - GGAA 2016, 14-18 February 2016 - 01 July, 2015
 - Ron Ronimus, 'Developing methanogen inhibitors to reduce methane emissions: the story behind the story' - AgResearch internal Inside Story - 03 July, 2015
 - Sandeep Kumar, Sandra Kittelmann, Gemma Henderson, Ganesh Siva, Mark Patchett, Sinead Waters, Peter Janssen, 'Diagnostic rumen bacteria associated with low-methane emitting sheep' - GGAA 2016, 14-18 February 2016 - 09 July, 2015
 - Sandra Kittelmann, Michelle R Kirk, Peter H Janssen, 'Deposition of sequence and meta data for PGGRC-NZAGRC objective 5.6.2' - NCBI Short Read Archive (Data repository) - 10 July, 2015
 - Arjan Jonker, Sharon Hickey, Suzanne Rowe, Brooke Bryson, E. Jones, German Molano, Stephen Olinga, Sarah MacLean, Cesar Pinares-Patino, John McEwan, 'Progeny from low methane selection line sheep also have lower emissions when grazing pasture' - GGAA 2016, 14-18 February 2016 - 14 July, 2015
 - Andy Reisinger, 'How much does livestock actually contribute to climate change?' - GGAA 2016, 14-18 February 2016 - 15 July, 2015
 - David Pacheco, Indrakumar Vetharaniam, Ronaldo Vibart, Xuezhao Sun, 'Evaluation of Methane Predictions using a Sheep Rumen Model' - GGAA 2016, 14-18 February 2016 - 15 July, 2015

- Ruidong Xiang, Jody McNally, Suzanne Rowe, Arjan Jonker, Cesar Pinares-Patino, Jude Bond, Hutton Oddy, Phil Vercoe, John McEwan, Brian Dalrymple, 'Transcriptomic analysis identifies rumen volatile fatty acids metabolism as a candidate pathway for host regulation of methane production' - GGAA 2016, 14-18 February 2016 - 15 July, 2015
- Sandra Kittelmann, Peter H Janssen, 'Cheek swabs shortcut rumen sampling process' - NZ Genomics Limited Annual Report - 17 July, 2015
- Sheree Balvert, Jiafa Luo, Louis Schipper, Cecile DeKlein, 'Methodology for rapid and initial assessment of the reduction potential of nitrous oxide mitigation options.' - GGAA 2016, 14-18 February 2016 - 15 February, 2016

