

final report

Project code: B.CCH.1021

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Date published: April 2012

PUBLISHED BY Meat & Livestock Australia Limited Locked Bag 991 NORTH SYDNEY NSW 2059

Novel individual enteric methane measuring system for multiple ruminants

Meat & Livestock Australia acknowledges the matching funds provided by the Australian Government to support the research and development detailed in this publication.

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Abstract

The concept of measuring rumen gas concentrations and temperature with miniaturized sensors using wireless telemetry has been developed and achieved as a proof of concept. However, there are limitations with data precision and repeatability with the infrared gas sensors currently available on the market and used in these particular studies. Solutions to the problems with corrosive gases and high humidity leading to corrosion of the infrared gas sensors have been slowly resolved, but not entirely eliminated. The incorporation of activated carbon in the controlled release device (CRD)has reduced the problem significantly in the short term over a couple of days but has not eliminated sensor drift entirely when studies have been undertaken for a period of greater than one week.

Refinement to the original electronics and supporting software that underpins this technology has been continuing concurrently despite having to create a system whereby the sensors can withstand additional non-target gases in the rumen.

The newly acquired e2v sensors for CH_4 and CO_2 have been demonstrated to be more robust as well as requiring less power. Reduction in power consumption is a vital aspect of the latest device prototype which will assist in expanding the number of measurements obtainable from a designated battery source and extend the working life of the unit. Technology development of energy harvesting from kinetic motion energy in the rumen is a potential source of recharging batteries but such emerging technologies have still some way to go before reaching maturity.

Field testing of the membrane mounted CRD that houses the device's electronics has demonstrated that it can maintain its integrity in terms of providing an effective seal to rumen liquor and digesta for more than 3 months under grazing conditions. Devices used in fistulated sheep for up to a week have been cleaned and reused for a second and third run without loss of integrity.

Validation studies have commenced in fistulated sheep and CH₄ and CO₂ concentration data from the rumen is being collected with the device over several days. The compilation of a comprehensive data set for validation of the device's gas concentration data has been limited due to problems with calibration drift and viability of the e2v gas sensors under rumen gas conditions. Possible reasons for the less than optimal sensor data are temperature and pressure fluctuations in the rumen and physical deterioration of the gas sensors despite substantial effort to rectify the problem. Although the technology can detect diurnal variation in rumen gas concentrations, the unresolved issues of calibration drift/sensor deterioration is such that the data lacks the level of precision to validate the measurements. Development of the technology is ongoing with the expectation that reliable data and validation will be achieved.

INTRODUCTION

In Australia, ruminant livestock are the single largest source of methane emissions, accounting for nearly 71% of agricultural methane emissions to about 10% of Australia's net emissions of carbon dioxide equivalents (DCC, 2009). Methane is formed in the rumen when hydrogen, released by other microbes during fermentation of forage, is used by methane-producing archaea called methanogens to reduce carbon dioxide. Approximately 94% of the enteric methane emitted by ruminants is belched out and expired via the lungs (Murray *et al.* 1976). A number of mitigation strategies for decreasing ruminant methane emissions from grazing animals have been suggested (Van Nevel and Demeyer 1995; Mbanzamihigo *et al.* 1996; McCrabb *et al.* 1997; Mathison *et al.* 1998; Hegarty 1999; Joblin 1999; Klieve and Hegarty 1999; Machmüller and Kreuzer 1999; Anderson *et al.* 2003; Machmüller *et al.* 2003a, 2003b; Wright *et al.* 2004), but assessing the effectiveness of many of these options depends on the accuracy of methane measurements.

Techniques for enteric methane emission measurements on individual animals can be classified into direct (total or partial enclosure of animals) and indirect measurements (isotopic and non-isotopic tracers). Both techniques have significant limitations which is problematic for achieving precise and repeatable estimates of GHG emissions from individual grazing animals.

The limitations arising from both the direct and indirect methods for measuring methane emissions has prompted researchers to set up and develop a novel intra-ruminal gas measuring device utilizing the latest electronics to estimate GHG concentrations at the source of production in the rumen. Miniaturization of electronic components in conjunction with CSIRO's Nano technology, improvements in battery power utilization and the development of sensor networks have made the option of deploying an intra-ruminal gas measuring system a real possibility. Measurements of CO_2 , CH_4 , rumen temperature and pressure can now be achieved remotely and integrated with the CSIRO FLECKTM platform with telemetric capability.

Executive Summary

Australia's ruminant livestock are the single largest source of methane emissions, accounting for nearly 71% of agricultural methane emissions. Mitigation strategies for decreasing ruminant methane emissions from grazing animals have been suggested but assessing the effectiveness of many of these options depends on the accuracy of methane measurements. Hence, the livestock industries require technology to measure enteric methane emissions from large numbers of individual animals simply, quickly, accurately and reliably to enable researchers and producers to develop, monitor and validate methane mitigation strategies to reduce emissions from grazing ruminants. The limitations arising from both the direct and indirect methods for measurement in a wide range of applications in the industrial sector which may have potential for application with large numbers of grazing ruminants. The review concluded that the idea of measuring methane in the rumen in grazing ruminants was feasible by using miniaturised infrared sensors, coupled with the technological advances made with wireless platforms.

Collaboration between CSIRO's Division of Livestock Industries and ICT Centre has developed an experimental wireless gas sensor node that uses miniaturized infra-red sensors for methane, carbon dioxide and hydrogen, and is integrated with CSIRO's wireless sensor network platform to provide telemetric capability. The intra-ruminal wireless sensor unit includes the ICT Centre's "Nano" circuit board with flash memory storage and its own battery supply, all of which is enclosed in a high density polyethylene (HDPE) plastic container with a siloxane diffusion membrane sleeve that sits over the slotted barrel of the container. This unit can be dosed orally in cattle and resides in the rumen of the animal. Inside the diffusion cell, the sensors are protected from the corrosive environment of the rumen where they collect and transmit data from the equilibrated rumen gases to the outside world via a 915 MHz radio transceiver. The system logs both methane and carbon dioxide concentrations in the rumen as well as temperature and battery voltage with a date and time stamp. The inbuilt flash drive logs data even when the animal is not in range of a base station for data acquisition and the stored data can later be downloaded at a time when the animal returns within range.

The technology has undergone significant improvements over the course of the project to reduce power requirements and thereby extend battery life. A joint provisional patent has been lodged covering the invention and intellectual property associated with the development of the device titled "System, method and device for measuring a gas in the stomach of a mammal".

Ongoing animal research trials are being performed with sheep in respiration chambers to generate a comprehensive data set to correlate daily emissions of expired methane with methane concentration measured intra-ruminally. Currently, the device is capable of detecting diurnal variations in methane and carbon dioxide concentrations in the rumen of experimental sheep in relation to feeding patterns. In addition, rises in temperature within the rumen due to fermentation and changes in pressure can be monitored by the latest prototype of the intra-ruminal device. A shift in the ratio of carbon dioxide to methane gas levels has been detected with the device in the same animal fed above and below maintenance rations. Measurement of hydrogen gas with the intra-ruminal device and the development of algorithms to predict methane emissions from gas concentrations in the rumen are still to be achieved. Unavoidable delays in the development and commissioning of the hydrogen sensor has meant that no data was obtainable at this time. Data will be generated in the coming weeks under field conditions. The problems experienced with the sensors have taken longer to resolve than first thought and hence the relationships between intra-ruminal gas concentrations with actual emissions is still in progress.

Experimentation and validation of the units is currently progressing which is expected to allow for large scale screening of animals for methane emissions to discriminate phenotypic differences (low *vs* high methane emitters) and effects of variation in feed base, climate and management. This technology is aimed at providing scientists and ultimately livestock producers with a tool to develop, monitor and validate methane mitigation strategies to reduce emissions from grazing ruminants.

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1 Background

1.1 Purpose and description

In Australia, the livestock industries require technology to measure enteric methane emissions from large numbers of individual animals simply, quickly, accurately and reliably to enable researchers and producers to develop, monitor and validate methane mitigation strategies to reduce emissions from grazing ruminants. An intra-ruminal battery powered device incorporating miniaturised infra-red gas sensors, coupled with CSIRO's wireless sensor network platform has been developed to measure rumen methane concentrations under the auspices of RELRP (BCCH 1003). The device is currently being tested under controlled conditions in sheep confined to respiration chambers. Further development is needed to get a better understanding of the utility of the device for determining methane yield by the animal under a range of feeding systems. Measurement of yield and concentration allows emissions intensity, total emissions and efficiency of rumen fermentation processes to be predicted. These data are important for NGGI, NCAS and emerging policies under the carbon farming initiative.

Issues relating to deterioration of the performance of the infra-red gas sensors under rumen gas conditions are being investigated to enhance data quality and extend the lifetime deployment of the device for field applications.

Currently there are no methods available to accurately and reliably measure methane production from large numbers of grazing animals, technologies such as these devices have the potential to fill the knowledge gap that currently exists for the grazing livestock industries. Methane emissions data from respiration chambers do not take into account diet selection of grazing ruminants and day to day variation on total feed intake amongst animals.

2 **Project Objectives**

2.1 Project Objectives

1. Evaluate the utility of the intra-ruminal device to sense changes in methane production under varying feeding conditions in sheep

2. Improve the energy efficiency, transmission capability and size of the device for use in both small and large ruminants

3 Methodology

3.1 Progress on Project Objectives

Despite the robust stainless steel casement of the e2v sensor, the double gold plating of reflective surfaces and even in the presence of 18g of activated carbon as a sink for H_2S , there is some evidence of diminished performance of the sensor over time (approximately 5-7 days). The zero drift that was earlier observed with the Dynament sensors under similar conditions in the rumen has not been an issue over weeks of testing. However, there appears to be a modest drift over time for the span gases during later calibrations. This issue has been under investigation for several months to resolve the calibration drift and despite considerable input being assigned to the task, some progress has been made but the drift has not been entirely eliminated.

The dual e2v gas sensor has demonstrated much lower power requirements then the earlier sensors used in this project and their deployment has extended battery life of the unit quite significantly. There is opportunity to extend this further by reducing the 'powered up' time to achieve stable readings for the infrared lamp in the gas sensor. Currently the sensor is powered on for 30 seconds at each reading but there is the possibility of reducing this time by 20-30% and still obtaining stable gas readings. This would have marked effects on the life of the battery and extend the number of readings significantly. A great deal of effort has been directed to extending battery life at the latter stage of the project. Battery life under the current regime is in the order of weeks (approximately 2-3 weeks of measurements) depending on sampling rates or 1800 data points. This can possibly be extended by a modest level with further refinements until such time as technology development provides a system to harvest energy internally from the animal (eg. Nano-sized piezoelectric films and materials that can convert small mechanical movements into electricity that are jointly being developed by researchers at RMIT and the Australian National University). Energy harvesting may extend the life of the unit for several months or possibly beyond a year.

3.2 Development of the Diffusion Cell

The thin membranes constructed from polydimethyl siloxane (0.2mm) have demonstrated robustness and suitability for withstanding the rigours of the rumen environment and have maintained their integrity whilst allowing gas to diffuse in both directions. There is scope for reducing the thickness of the siloxane membrane to make the system more dynamic with faster gas diffusion times but the manufacture of a single moulded membrane sleeve is outside the capabilities or the expertise of the project team and would require specialist input from CSIRO's Division of

Manufacturing and Materials Science or another agency such as the Royal Melbourne Institute of Technology.

The siloxane membrane has been mounted onto a controlled release device (CRD) capsule, with approximately sixteen, 10mm diameter holes pre-drilled into the barrel of the unit. To adequately seal the unit and prevent liquids moving inside the capsule, the membrane ends have been overlapped and joined with a silicone adhesive. The barrel of the CRD has been rebated at each end to allow a compression ring to be seated in conjunction with the adhesive. This effectively blocked rumen fluids from gaining access to the electronics inside the device.

The open end of the CRD has a specifically designed nylon cap to seal the unit after the electronics have been placed inside. The nylon end-cap is rebated with a groove to allow an "o" ring to be seated in the groove and provide a good moisture barrier to the electronics inside.

3.3 Overcoming Infra-Red Gas Sensor Contamination

The inert properties of the construction materials of the e2v® sensor make it more suitable for the harsh environment within the animal's rumen. A partial solution to the gas sensor contamination problem was the identification and incorporation of specialised grade pelletised activated carbon (Acticarb EA1000K, Activated Carbon Technologies Pty Ltd). This product is primarily used to remove hydrogen sulphide from sewage plants in Australia. A small amount of activated carbon, 20g, is incorporated in two satchels within the CRD to minimize the damage to the pyroelectric detectors inside the gas sensors. The material however can saturate and has a finite life.

3.4 Experimental Procedure

3.4.1 Experiment 1

Initially, the device was tested in four fistulated animals fed a maintenance ration of 700g DM/day for 5 days prior to being placed in respiration chambers for 2 consecutive 23 hour evaluation periods to measure CO_2 and CH_4 emissions over the experimental period. The ration consisted of 86% lucerne pellets (Ridley) and 14% oaten chaff. The sheep were weighed during this period and the four animals with similar weights were randomly assigned to one of four respiration chambers. Thirty six hours before experimentation, the infra-red gas sensors were calibrated with certified gas standards (CH_4 [100%, Air Liquide]; CO_2 [80.0±0.2%, BOC, NATA]) and then sealed inside the CRD with 18g activated carbon present split between 2 satchels. The devices were then placed inside the rumen of the animals via their fistula and allowed to equilibrate prior to respiration chamber measurements.

The Oxymax gas analyser (Columbus Instruments, USA) attached to the respiration chambers was calibrated 24 hours prior to experimentation with certified gas standards ($0.11 \pm 0.01\%$ CH₄, $1.08 \pm 0.04\%$ CO₂ [Air Liquide]).

Following the 36h equilibration period, the sheep were moved into the chambers at 08:30 and given access to their rations as well as water *ad lib*. Gas samples from all four chambers were sampled and analysed sequentially, taking 15 minutes to complete one cycle and hence 92 measurements per chamber were performed over the 23h experimental period. After the final measurement, the sheep were removed from the chambers and placed in an adjacent pen. This allowed for the emptying of waste trays, placing new rations inside the chambers and the reallocation of sheep to different chambers for the second 23h period to minimize the risk of potential bias associated with chamber allocation.

All sheep ate the full ration provided, although one animal dislodged it's cannula in transit from the animal house with subsequent spillage of rumen fluid (approximately 1-1.5 litres). This animal was atypical for its CH_4 profile for the first 23h period, being substantially lower in methane output over that period and a suggestion of a carryover for the next period, although not as pronounced. After the second day of experimentation, all sheep were returned to the animal house.

3.4.2 Experiment 2

The second experiment was conducted 10 days later using the same 4 sheep after the animals had acclimatized to a change in rations with half receiving slightly less than maintenance (600g DM/day) and the other 2 animals fed above maintenance rations (850g DM/day) which equated to a 42% increase in rations between the lowest and highest treatment rations.

3.5 Results and Discussion

3.5.1 Maintenance rations

Testing of the dual e2v IR gas sensor has been progressing to evaluate performance in the intraruminal device under *in vivo* conditions. Validation studies have commenced in fistulated sheep and CH₄ and CO₂ concentration data from the rumen is being collected with the device over several days. The same animals have been simultaneously run through the respiration chambers at CSIRO Chiswick and CH₄ and CO₂ emissions from the same animals recorded. So far to date, animals have been fed a maintenance diet of pellets and chaff (700g DM) during the study period. Gas concentration in the rumen has consistently demonstrated diurnal variation for a once daily feeding regime (Figures 1-2).

Rumen gas concentrations and temperature



Figure 1. The device was placed in sheep approximately 30 minutes after being switched on. Gas concentrations and temperature were collected every 15 minutes till sample 360. The sheep was in the chamber from time point 140 to 355.

Due to differences in the siloxane membrane permeability to different gases (CO_2 permeability 3 x greater than for CH_4) and gas concentration gradients in the rumen, CO_2 readings on the intraruminal device are more dynamic and responsive than that of CH_4 . Hence, CH_4 concentrations are lowest as measured by the device when CO_2 concentrations peak. The counter-cyclical results in the gas concentrations of interest are a very consistent observation with this technology. This observation is in contrast to respiration chamber data demonstrating maximum methane emissions are strongly associated and aligned with feed intake (Figure 2).



Figure 2. Methane emission data estimates for 4 respiration chambers measured by the Oxymax system over a 23 hour period demonstrating the elevation in methane shortly after animals eat their rations upon entering the chambers. The 'x' axis represents 15 minute intervals.



Rumen gas concentrations and temperature

Figure 3. Fistulated sheep (No. 2) fed a once daily maintenance ration as in Figure 1, showing a similar response both in gas concentration and temperature profiles in the rumen. **Note**, the slight lag in the rise in CO_2 concentration post feeding compared to the rise in temperature due to gut fermentation. The final calibration for CO_2 and CH_4 were approximately 10-15% lower than the initial calibration.



Figure 4. CH_4 emissions from the same fistulated sheep for 23h in the respiration chambers replicated over four days. They were fed a maintenance diet of 700g DM/day.



Figure 5. CO_2 emissions from the same fistulated sheep for 23h in the respiration chambers replicated over four days. They were fed a maintenance diet of 700g DM/day.



Time points (15 min)

Figure 6. The ratio of $CO_{2}CH_4$ over two consecutive days for 2 sheep estimated by the intra-ruminal device whilst the animals underwent testing in the respiration chambers. The above ratios are markedly lower than those calculated from chamber emissions which includes both respired and eructated CO_2 .

It is worth noting that the higher CO₂/methane ratio in sheep 2 compared with sheep 1 correlated with lower daily methane emissions as measured by the respiration chambers but these observations are still preliminary in nature.

3.5.2 Response to varying rations

Data from only 3 animals was obtained due to a failure of one of the intra-ruminal devices and there were significant problems with the data for CO_2 final calibration for the remaining three units in this particular study. Methane values were reasonably consistent from first to final calibration (100%), whereas CO_2 values at the final calibration were only 50-55% of the initial measurement.

There was a 1.0° C increase in temperature during fermentation in the rumen for the sheep fed a below maintenance ration (500g pellets, 100g chaff) as compared to a 1. 5° C rise for the sheep on above maintenance ration (750g pellets, 100g chaff). The pressure in the rumen was similar for both diets peaking at 105kPa after 24h in the rumen and then dropping to around 101 and 98kPa on subsequent days (figures 7a and 8a).

Further data replicated over time is needed to get an accurate estimate of these parameters as well as the gas concentrations.

Sheep3 (750g pellets & 100g chaff) sensor SN9542



Figure 7. The data demonstrates a rise in CO_2 and temperature soon after the once daily morning feed. During fermentation, temperatures rise by more than $1.5^{\circ}C$ for the maintenance ration.



Figure 8. Data from sheep 2 demonstrates a problem occurred with the CO₂ sensor during the testing period, with low readings during testing and at the final calibration. The rise in temperature during fermentation is approximately 1.1°C for the lower than maintenance ration.



Figure 8a. Sheep 2 showing pressure readings taken inside the intra-ruminal device exhibiting peaks in pressure at similar time points to temperature peaks which are aligned with gas concentration peaks. Subsequent pressure responses are less pronounced and step down over time.



Figure 9. Gas concentration data showing sub optimal (50%) final calibration for CO_2 and a reduction in CH_4 values for the final calibration. The temperature rise is of similar magnitude (1.5°C) to sheep 5 which was on the highest ration.



Pressure and temperature readings for sensor SN9517 in the rumen



The project team is focussing on ways to improve the reliability of the data emanating from the device by modifying calculations performed on raw data readings allowing for changes in pressure as well as temperature.

Despite the robust stainless steel casement of the e2v sensor and in the presence of 18g of activated carbon, there is some evidence of diminished performance of the sensor over time (1 week). The cause of this variability in performance remains to be defined but may be mulitfactorial (eg, pressure, temperature and biological spoiling of the sensors by corrosive gas).

The dual CO_2 and CH_4 sensor has demonstrated much lower power requirements then the earlier Dynament sensors and has extended battery life of the unit quite significantly. There is an opportunity to extend this further by reducing the 'powered up' time to achieve stable readings for the infrared lamp in the gas sensor. Currently the sensor is powered on for 30 seconds at each reading but there is the possibility of reducing this time by 20-30% and still obtaining a stable gas

reading. This would have marked effects on the life of the battery and extend the number of readings significantly.

3.5.3 Siloxane membrane

The siloxane membrane used in the manufacture of the device has been very resilient during testing both in the rumen of sheep and cattle with very few failures. Many of the units have undergone repeated testing in sheep with no deleterious effects. A single moulded piece of siloxane membrane manufactured specifically for the CRD barrel would be even more robust over time in the rumen. Future developments in membrane design may result in a finer membrane than the current one in use and thereby provide faster response times to reach equilibrium. Testing of the longevity of siloxane mounted CRD over time in the rumen of a fistulated steer has been achieved for an extended period of greater than 3 months. The integrity of the liquid barrier (siloxane membrane) over 3 months with the most recent prototype design allows for a reasonable period of evaluation of rumen gas concentrations for research purposes. Also, testing of the membrane mounted capsule for periods of 1-2 weeks in fistulated sheep has been achieved. Additionally, these units have been washed and the outer surface cleaned and reused for follow up studies.

3.5.4 Activated carbon in the capsule

The inclusion of activated carbon (Acticarb EA1000K) in the device to act as a capture reagent and sink for H_2S has shown promise. Not only does it bind H_2S entering the diffusion cell across the siloxane membrane but takes up water vapour as well. The inclusion of activated carbon with the sensors may not have eliminated deterioration of the units over time.

3.5.5 Cut-off valve to minimize exposure to hydrogen sulphide

The inclusion of a cut-off valve within the CRD to reduce the duration of exposure IR gas sensors to hydrogen sulphide and moisture has progressed slowly due to size constraints within the capsule and the need to develop specific software to run the valve. The small valve mechanism has the ability to open and expose the e2v IR sensor to the intra-ruminal gases for measurement. After the gas measurement is complete the valve closes and then activated carbon is used to reduce the reactive gases (eg. H_2S) and limit the amount of moisture coming into contact with the sensor for extended periods. Due to space limitations, the valve has not been deployed in a CRD.

3.5.6 Hydrogen sensor

The new intra-rumen wireless sensor unit, shown in Figure 10, is based on the PACP circuit board and was designed and developed by the CSIRO ICT Centre Engineering Team. The miniaturized electrochemical e2v® gas sensor for hydrogen is mounted on one end of the gas sensor node.



Figure 10. PACP hydra Intra-rumen hydrogen sensor node. The silver sensor is 20 mm x 20mm in dimension

The new intra-rumen wireless sensor has a number of new key features and specifications including;

- Socket for one miniaturized electrochemical e2v® gas sensor to measure 0 to 1000 parts per million (PPM) concentrations of hydrogen gas.
- Texas Instruments CC430F5137 microcontroller and 916 MHz radio transceiver
- 16-Bit, 8-Channel Serial Output Sampling Analog-To-Digital Converter with precision voltage reference
- Pressure and air temperature sensing
- Very low powered gas sensor filter circuits for the reference and hydrogen gas signals
- Battery voltage, charging voltage and current monitoring
- 1 MByte serial-interface data Flash memory
- Switchable regulators for the gas sensor and gas circuit power
- Efficient on-board Li-ion battery charger which can be used for energy harvesting
- 916 MHz quarter wave wire antenna
- Improved radio range from inside the rumen and up to 3 Km line of sight
- Size, including two gas sensors and battery, is 100 mm long x 20 mm round
- Board mounted rechargeable 3.6 volt @ 3A hour Li-ion battery

The schematic and printed circuit board (PCB) is designed using the very small components including 0201 sized surfaced mount capacitors and resistors. A conformal coating is applied to protect the electronics against moisture, chemicals, and temperature extremes inside the harsh environment of the rumen.

The initial hydrogen sensor node prototype has been manufactured and tested under laboratory conditions. The sensor device has a normal gas measurement range from 0 to 1,000 parts per million (PPM).

Figure 11 shows the hydrogen gas measurements against the new intra-rumen hydrogen sensor node. The EK-3 evaluation board uses a cable and custom connector to enable measurements of gas concentration within a glass vessel and the intra-rumen hydrogen sensor unit was also placed within the same vessel and transmitted data wirelessly. During the study, hydrogen gas was added in increasing concentration to a level of 2000 PPM.



Figure 11. Gas concentration measurements using the EK-3 evaluation board and the new intrarumen hydrogen sensor node under *in vitro* conditions.

The hydrogen concentration results show a line through the origin with a slope that varies in amplitude depending on the individual hydrogen sensor sensitivity (nA/PPM). The hydrogen sensor used on the EK-3 evaluation board therefore has a lower sensitivity than the sensor used on the

intra - rumen hydrogen sensor node. The gas concentration errors appear to be partly related to the method of injecting hydrogen as both sensors have the same rise and fall pattern for each reading. Also of note is the saturation of each sensor occurring at 2,000 PPM.

The sensors are ready to be to be trialled in rumen fistulated animals. The project team is awaiting the arrival of a certified hydrogen gas standard in the coming days to calibrate each hydrogen sensor before animal testing can be undertaken.

3.6 Overall progress of the project

Steady progress has been achieved in both the sensor hardware logistics in association with the intra-ruminal diffusion cell and further refinement of the wireless sensor device to capture the specific gas concentrations in the rumen remotely.

Although validation studies have commenced, achieving a large comprehensive data set replicated over time has been delayed. This delay is in part due to unforeseen circumstances relating to the occasional sub-optimal performance of the infrared sensors at varying times. The variances observed in the sensors are not consistent and thereby creates issues in dealing with this particular problem. The research team has been working towards eliminating the problem by incorporating temperature compensation of the algorithms for the calculations of the estimates of gas concentration and recently pressure compensation has become an issue to consider.

Possible causes for inconsistent data generation could be due to temperature and pressure variation in the rumen impacting on the electronic gas sensors as well as physical deterioration of the sensors within a corrosive gas environment. Recent pressure measurements taken from inside the device demonstrate diurnal variation but also a step-down over subsequent days. This observation could be as a consequence of faster rates of CO₂ diffusion into the device compared to the much lower gas permeability of siloxane membranes for oxygen and nitrogen. During the process of residual air (78% nitrogen; 20.9% oxygen) diffusing out of the device following placement inside the rumen, then greater pressures could be experienced during the state of gas flux into and out of the diffusion cell in attaining a state of equilibrium.

Further studies are planned in the coming weeks to expand on the data set already generated for the validation process. To date, animals fed on either a maintenance ration or above and below maintenance have been studied with limited replication. Intermittent failures in the technology due to battery failure, data acquisition problems and total system shutdowns have hindered the collection of complete data sets during experimentation. Data losses have been in the order of 25% during testing and thereby hindering progress in a timely fashion. Plans are in place to vary the diet ration

in more detail and examine the relationship with CO_2 and CH_4 concentrations in the rumen with the GHG emissions from the experimental animals.

4 Success in Achieving Objectives

4.1 Latest Prototype

The device is presently capable of detecting diurnal variations in carbon dioxide and methane concentrations in the rumen of experimental sheep in relation to feeding patterns. In addition, rises in temperature within the rumen due to fermentation can be monitored by the latest device and have varied in relation to feed intake. Although measurement of absolute gas concentrations utilizing algorithms to predict emissions from gas concentrations has not been achieved to date, progress is being made in meeting this objective.

The hydrogen gas device is to be deployed shortly in animals following tests and evaluation already completed in the lab. Deployment in fistulated animals is expected in the next week or two following receipt of a certified H₂ gas standard to the laboratory.

An apparent deterioration in sensitivity of the gas sensors after 4 days in the rumen of experimental animals has limited the project team's ability to collect a comprehensive data set to complete this stage of the project. Additional time is required to generate sufficient data for the validation process to develop relationships between gas concentrations with actual emissions. Reuse of sensors is being examined despite the apparent calibration drift but it appears that the reference peak to peak voltage range is reduced as a consequence of H_2S exposure and as a consequence the sensitivity of the unit is reduced for subsequent measurements.

A joint provisional patent has been lodged in the last 6 months covering the invention and intellectual property associated with the development of the device titled "System, method and device for measuring a gas in the stomach of a mammal".

Significant advances have been achieved at this point in time in the development of a modified CRD diffusion cell to house the electronic components, the software underpinning the system including wireless telemetry as well as advanced capability and functionality of this novel technology.

5 Impact on Meat and Livestock Industry – now & in five years time

5.1 Impact on Meat and Livestock Industry

The livestock industry has invested large amounts of time and funds into developing mitigation strategies for reducing ruminant greenhouse gas emissions, particularly methane emissions. However, in order to develop, monitor and validate such mitigation strategies it is necessary to be able to readily measure enteric gas emissions from large numbers of individual animals. It is desirable to measure gas emissions in an autonomous fashion which does not significantly disturb or impede the animals in their natural grazing environment.

The current development of wireless rumen sensor (bolus) technology for use in grazing ruminants provides exciting opportunities for the future. This technology is relatively easy to use and provides an excellent and affordable option for continually measuring rumen gases. The immediate impact of this project on the meat and livestock industries will be the ongoing opportunity to have an impact on reducing methane emissions from livestock with potential for further development through round 2 of the DAFF National Livestock Methane Program that will commence in 2012.

Northern Australian production systems which account for approximately 50% of the national beef herd are typified by low rates of gain (< 1 kg/day), high turn-off ages (~ 3.5 years old), lack of grain for finishing, and an almost complete reliance on low quality forage based diets. It has been estimated that as little as a 5% increase in the efficiency of digestion could yield an economic benefit of at least \$100 million to the cattle industry. Therefore reducing methane production could benefit the ruminant energetically provided the efficiency of ruminal metabolism is not compromised.

Research from the current project is likely to have impact on both the intensive and extensive sectors of the ruminant livestock industries particularly if there is a productivity benefit associated with a reduction in methane emissions. If successful, the technology will allow researchers to discriminate phenotypic differences and the effects of variation in feed base, climate and management on methane emissions. This should lead to feeding management strategies for improved utilisation of the feedbase and a reduction in methane emissions from grazing and intensively fed ruminants.

The intra-ruminal methane sensor will be a powerful tool to advise whether genetic selection for low methane production and superior liveweight gains is feasible using genetic measures of the animal that could be employed commercially. The technology has the potential to provide a major benefit in screening cattle for high and low methane emissions under normal grazing conditions without the

need to handle or modify their behavioural patterns during the testing period. It would be considered by industry and the general public as a very low stress system on animals as well as being far less intrusive than the current options available for estimating methane emissions from grazing ruminants.

6 Conclusions and Recommendations

6.1 Conclusions and Recommendations

Although the technology is still undergoing development, considerable advances have been made in the technology despite major technical problems that have arisen regarding sensor drift over time. Miniaturized sensors and sensor networks are already playing a significant role in manufacturing industries, environmental monitoring of landscapes in conjunction with satellite imaging and monitoring elite athletes during training sessions and in competition. It is now becoming more evident that these sensor technologies have a role to play in agricultural systems, either on or within the animal. As more research groups focus on developing and enhancing sensor systems, then the combined knowledge gained during this process has the potential to accelerate sensor technology outputs.

The provisional patent application on this technology provides MLA and CSIRO protection to further develop the technology in the short to medium term without the threat of other research groups from competing in the same space and utilizing the intellectual property generated from this joint research project. The generation of comprehensive data sets for the validation process has been slower than expected and is continuing in order to determine the relationships between rumen gas concentrations and GHG emissions from ruminants. Building on the substantial achievements and developments to date of this new and novel technology requires further work to be undertaken. Incorporating CO_2 and CH_4 measurements with that of H_2 gas concentrations and possibly temperature variation in the rumen is continuing.

A collaborative project with Royal Melbourne Institute of Technology will commence in 2012 to optimise the performance of the device through improvements to the diffusible gas membranes with the intent of excluding corrosive H_2S gas while making membranes that are selective for specific gases. The new collaborative project will also develop programs and equations to adjust the data for temperature and pressure variations in the rumen which should result in robust and accurate measurements of the target gases.

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