### NOVEL PHENOTYPING TECHNIQUES FOR ENHANCING GENETIC AND GENOMIC PREDICTIONS OF TRAITS THAT ARE DIFFICULT TO MEASURE IN GRAZING LIVESTOCK

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#### SUMMARY

The development of miniaturised wireless sensors and data capture systems now offers the capability to study livestock in their commercial production environment, and to do this in a way that does not constrain the animal from expressing its full range of genetic drivers for the traits under study. In this way, variation in the traits of economic importance which form the breeding objective, can be directly assessed. This will allow appropriate genetic parameters to be estimated for novel and hard to measure traits.

This paper discusses the issues underlying the need for new and novel phenotyping methods and presents early results from studies utilising sensors and sensor networks for predicting feed intake and feed efficiency of individual cattle on pasture.

#### **INTRODUCTION**

With recent advances in high-throughput genotyping technologies for livestock, the ratelimiting step in the conduct of large-scale genetic investigations has become the collection of complex phenotype information from relevant populations (Pollak *et al.* 2012). This is particularly so for grazing ruminants. To date, variation in some of the difficult-to-measure attributes of grazing livestock that have significant economic impacts, such as feed intake on pasture, aspects of reproductive performance and quantification of disease status, have only been quantifiable by constraining animals in artificial (non-grazing) environments. For example, the measurement of feed intake in beef cattle is now usually conducted in a feedlot environment where animals are maintained in group pens and fed diets of very different composition and availability than the pasture swards that often constitute the normal production system in Australia. Similarly, in assessing disease traits, the phenotype is not always measured in the environment where the proximal causes of the disease state are found (Houle *et al.* 2010).

The large international research effort that has delivered a high quality map of the bovine genome has been accompanied by a similar effort in phenotyping. However, some of the traits of major economic importance to beef cattle breeders have not been able to be measured, in part, due to the lack of practical measurement technologies and a focus on early age selection criteria. Whilst indirect or proxy traits have been utilised to acquire some knowledge of the associations between genotype and breeding objective traits, there remains a significant phenotype gap that needs to be filled to improve the return on the investment in genotyping. The capacity to directly measure traits of importance in breeding objectives relevant to pasture-based beef enterprises is critical. This will give breeders the capacity to identify the selection strategies that lead to the most cost effective means of achieving optimal and sustainable progress in the aggregate outcome.

## Efficiency

#### NOVEL METHODS OF PHENOTYPING GRAZING LIVESTOCK FOR ECONOMICALLY IMPORTANT TRAITS

The development of electronic sensing capability has the potential to allow the measurement of traits of economic importance that previously had not been measurable in the commercial environment. Historically, on-animal logging devices for sensors used for phenotyping were bulky, and often heavy enough to raise concerns that the animal may not have exhibited its normal behaviours. However, over recent decades there have been considerable advances in miniaturisation and reduction of power use in electronic devices, such as microcontrollers, Global Positioning System (GPS) chips and in radio technologies. This has allowed ecologists and environmental scientists to collect high quality traces of the movements of free-ranging animal over, often lengthy, time-frames.

Recent technical advances in digital radio communications and microcontrollers has led to the evolution of Wireless Sensor Networks (WSN) which offers the potential for lightweight, small sensing devices for measuring a wider variety of traits relevant to grazing livestock (Hancock *et al.* 2009). However, the constraints imposed by a device that can be practically deployed on livestock, introduce limitations on local storage and communications throughput, which in turn, makes transmission of high-temporal, low-level sensory information difficult, particularly as the system is scaled up to a larger number of devices. This limitation has motivated the development of classifiers on the WSN nodes, which change the high temporal-resolution, but low-level sensed-data, into temporally-sparse high-level behavioural activities. Such an approach can produce a significant reduction of information whilst retaining enough information to still accurately classify phenotype behaviours. This reduction of information saves bandwidth and energy, which positions this approach to enable measurement of large numbers of livestock over long-term periods.

The intersection of the capability to have accurate knowledge of phenotype behaviour in their natural environments, over long periods on large numbers of animals, provides the novel methodology for phenotyping livestock in a practical and economically viable way.

# AN EXAMPLE: DIRECT PHENOTYPING OF FEED INTAKE AND EFFICIENCY OF GRAZING RUMINANTS USING WIRELESS SENSORS AND SENSOR NETWORKS

In Australia the major cost of beef cattle production is associated with the cow-calf unit. It has been estimated that the feed costs of the breeding female and her calf can be 60-70% of the total herd feed costs, and as much as 90%, when account is taken of rearing replacement females. As such, this is a critical component of the input costs of the beef enterprise, and genetic variation in this trait and its association with production efficiency should be key elements of the breeding objectives of breeders of beef bulls.

To date, the focus for measuring variation in feed intake has been evaluation of young animals in a feedlot environment where test animals are maintained in group pens and fed *ad lib*. diets of grain-based high energy concentrates. Alternatively, under pasture systems, chemical markers such as N-alkanes (Dove and Mayes 2006), have been used to predict intake, selectivity, and digestibility of the pasture. However, marker methods have limitations, and are difficult to apply for the lengthy periods needed to get robust estimates of an animal's underlying intake of pasture.

The development of a practical measure of feed intake for all classes of animals maintained in a pasture-based environment, would provide a means of estimating the heritability and genetic correlations necessary to evaluate the utility of direct and indirect selection criteria for a range of breeding objectives. **Application of new technologies to measurement of feed intake and efficiency in livestock.** The particular challenge associated with developing a robust and precise method of measuring feed intake in grazing animals is the absence of an existing methodology to use as a high quality benchmark against which to train the predictive algorithms developed from the sensor data.

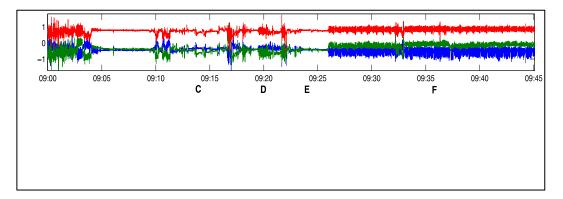
The first step in developing means of economically estimating feed intake *en masse* is to identify a suite of sensors that are likely to exhibit a response that correlates with pasture intake and determining where these sensors are best located in and on the animal. A study jointly initiated by CSIRO and NSW Department of Primary Industries at Armidale in NSW is being employed to test a range of sensors, from the perspective of size, cost, weight, energy usage, sensor longevity and impact of the sensor on the pasture intake of the animal. Two locations have initially been trialled to assess likely survivability (longevity of the device) and non-obtrusiveness. One approach was to mount a suite of sensors on an eartag and the other as a device attached to a halter, adjacent to the mouth of the animal or on the back of the head.

With respect to sensors, two sensing modalities are being initially employed. An Inertial Measurement Unit (IMU), comprised of 3-axis accelerometers, 3-axis magnetometers and a pressure sensor for gross-height change detection were selected due to the need of the animal to move its head (and mouth) in order for feed intake to occur. Similarly, the ripping and chewing of feed matter (and drinking) will necessarily produce sounds which could potentially be used to estimate feed intake (Galli *et al.* 2011).

The second step in developing a practical and economic means of estimating pasture intake is development of algorithms that classify the low-level, high-sample-rate *input* sensor data into *output* behaviours such as foraging, biting, chewing, ripping, ruminating, drinking, sleeping etc. Mapping the inputs directly to pasture intake was deemed impractical as this would have required measuring the pasture intake at a frequency similar to input sample rate (faster than 1Hz). The input features (every accelerometer and magnetometer axis and the pressure value as well as a number of audio statistics over various window sizes) inherently provide different levels of predictive power and so need to be scaled and weighted. However, in order to determine the appropriate predictive power of any feature using a supervised learning approach requires a training dataset to be compiled which is comprised of the potential *input* features and the *outputs* (behaviours / traits). Therefore, a multi-day, multi-animal trial, recording the raw data (IMU and audio) was performed with simultaneous recording of benchmark methods of measuring feed intake and animal behaviour. These benchmark methods of biomass disappearance, chemical markers and highly annotated video by experts, required significant human and technological resourcing per reading and is a key motivation for our use of sensors and predictive algorithms.

Figure 1 shows the uncalibrated ("raw") accelerometer and magnetometer traces over a 45 minute period for an Angus steer in a field-grazing environment. The animal exhibits a variety of behaviours (as evidenced from video footage), ranging from foraging, standing still, visual searching, and continuous episodic grazing. The low accelerometer variation when the animal is still, or visually searching, highlights the advantage of utilising multiple sensors as the magnetometer trace can be used to differentiate these two different behaviours. Similarly magnetometer readings alone do not clearly differentiate the continuous episodic grazing activity correctly.

Based on evidence from other studies the inclusion of acoustic data provides significant additional power to discriminate between biting and chewing actions which allowed accurate estimations to be made of dry matter intake in grazing sheep (Galli *et al.* 2011).



**Figure 1.** Accelerometer (100Hz sampling) and Magntometer (10Hz sampling) trace for 45 minute period for a grazing steer. Annotation of time refers to A) foraging, B) stationary, C) visual searching, D) foraging again, E) visual searching again, and F) continuous episodic grazing.

# CONCLUSIONS

The use of technologies built around electronic sensors and sensor networks offers great promise for the phenotyping of large numbers of animals in their normal commercial environment. Initial experiments on small numbers of animals using video data to benchmark behaviours associated with grazing have provided a platform from which to develop robust predictive algorithms. The particular challenges in the further development of this phenotyping method lie in the management of the very large volumes of data that are an integral part of this methodology and the design and management of the power source.

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