GENETIC FRONTIERS IN THE DEVELOPMENT OF 'CLEAN, GREEN AND ETHICAL' MANAGEMENT SYSTEMS FOR THE EXTENSIVE SHEEP INDUSTRY

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SUMMARY

In 2004, the concept of "clean, green and ethical (CGE)" management was presented with a view to helping producers to respond to developments in societal demands. The initial focus was on efficient reproduction in small ruminants in grazing systems, but subsequent versions have expanded to other animal production systems, all the while aiming to minimise drug use, minimise the environmental footprint, and maximise animal welfare. To date, much of our research has targeted the physiological, behavioural and managerial limitations to implementation of CGE management at flock or herd level. Here, we consider the role of genetics, particularly within the context of Merino sheep under extensive grazing. Our aim is to stimulate discussion and promote research in quantitative and molecular genetics as a means of finding solutions to major limitations in the CGE framework: 1) drug-free control of reproduction; 2) fecundity; 3) fertility; 4) colostrum production; 5) mother-young bonding; and 6) weaner mortality. These new directions in research expand the scope of the CGE concept in animal production and might help producers respond to the increasing intensity of demands for 'clean and green' food and fibre as well as high standards in animal welfare. Importantly, CGE management is low-cost and low-tech, so it is perfectly suited to extensively grazed sheep.

BACKGROUND AND CONTEXT

Changing attitudes in society, and therefore consumers, led to the development of the concept of 'clean, green and ethical' (CGE) animal production, in which we aim to limit the use of drugs, chemicals and hormones (clean), minimise environmental impact (green), and pay attention to ethics and animal welfare (ethical) in all links in the supply chain (Martin *et al.* 2004, 2009; Martin 2009; Bickell *et al.* 2010). The most obvious evidence of market demand for CGE production has been the growing popularity of 'organic' products. However, the CGE concept differs from the organic industry in that it offers a science-based framework that can help transfer innovations derived from research and development to mainstream animal production (Martin *et al.* 2004).

The CGE concept began with sheep reproduction and the implementation of practices such as 'focus feeding' (short, precisely timed nutritional management) and natural and non-invasive methods for controlling the timing of the different stages of the reproductive cycle. Briefly, focus feeding is used to boost sperm production before mating, maximise potential litter size by increasing ovulation rate, maximise postnatal survival and development, and minimise non-productive periods caused by delays in puberty or post-partum fertility. The full implementation of focus feeding is only possible when we have precise control over the timing of reproductive events – for example, by using the 'ram effect' ('teasing'). These concepts were then combined into a "CGE Management Package", such as the one illustrated in Figure 1.

The CGE principles can be applied to any type of animal production, including high-input intensive systems as practiced with dairy cattle (Kadokawa and Martin 2006; Martin *et al.* 2009) but, for the present paper, we will focus on low-input, extensively grazed sheep in Australia. To date, much of our research has targeted the physiological, behavioural and managerial limitations

to implementation of CGE management at flock or herd level. Here, we turn our attention to the role of genetics.



Figure 1. A 'CGE Management Package' for sheep in which periods of focus feeding are used to maximise reproductive success. For accurate timing of the periods of feeding, mating must be controlled (e.g. with the ram effect) and brief, or ultrasound in early pregnancy must be used to estimate fetus age. Finally, the survival and development of the new-born must be maximised. The numbered circles indicate points in the process where we speculate on potential genetic input. Redrawn after Martin 2009.

Our aim is to stimulate discussion and promote research in quantitative and molecular genetics as a means of targeting six major limitations in the CGE framework: 1) drug-free control of the timing of reproduction; 2) fecundity; 3) fertility; 4) colostrum production; 5) mother-young bonding; and 6) weaner mortality. Our choice of topics was guided by evidence of genetic variation (known breed differences or within-breed variation) and of heritability of the trait under consideration. We have high expectations because we are on the verge of a technology-led revolution in the generation of genetic data: electronic identification, DNA pedigrees, and the automatic recording of body weights and number of lambs born, will all combine make it possible to assess large numbers of sheep for a wide variety of production traits under extensive production systems.

GENETIC FRONTIERS IN THE DEVELOPMENT OF CGE MANAGEMENT SYSTEMS

Target 1: Drug-free control of reproduction. From Figure 1, it is clear that we need to be able to predict accurately the timing of the events in the reproductive process. Until now, we might have considered using exogenous hormones, but progestagen devices are too expensive and impractical in extensive systems, raise market concerns about food safety, and, upon disposal, are seen as an 'environmental endocrine disruptor'. However, in many genotypes, there is a 'natural' alternative if the ewes are mated before February – the *ram effect ('teasing')* can be used to assist in controlling the time of ovulation and thus conception and lambing. The scope for genetics-driven research on the ram effect is clear: i) it is highly likely that all breeds have the anatomy and

physiology, and thus the genes, that underpin the ram effect; ii) there are profound differences among genotypes in responsiveness to the ram effect; iii) there is considerable variation among genotypes, and among individuals within a genotype, in the way they express their breeding season (e.g. Pearce and Oldham 1988). Differences in seasonality will be reflected in differences in the strength of the photoperiod-drive 'filter' and therefore their responsiveness to the ram effect (Fig. 2). The power of genetics is clear in the work of Notter *et al.* (2005) who showed that selection for reduced seasonality could be achieved by using spring fertility records – in other words, the strength of the 'filter' can be modified through genetics.



Figure 2. A schema of the relationships among the major environmental signals that affect the reproductive system of the sheep. Our observations suggest that photoperiod acts as a genotype-dependent 'filter' that modifies the responses to nutritional and socio-sexual signals (Blache *et al.* 2003). Redrafted after Martin *et al.* (2010).

Genotype differences. The ram effect works well in Merinos because the ewes are sufficiently responsive to photoperiod to have clear breeding seasons, yet not so responsive that photoperiod blocks the induction of ovulation by socio-sexual signals. However, with genotypes that originate from higher latitudes, amongst which are the meat breeds, the photoperiodic filter dominates the reproductive system of both sexes. In the male, the production of the socio-sexual signals seems to be reduced; in the female, there seems to be a break in the physiological and anatomical chain from perception of the socio-sexual signals to the stimulation of GnRH secretion. We need to consider the interaction between these processes and those that implement the photoperiodic strategy for reproduction – we still do not know why, for example, Suffolks are more responsive to photoperiod than Merinos.

Genetic research on teasing -a) Male factors. Teasing is not like mating where an oestrous ewe only has to encounter a ram once or twice in 24 hours to conceive. Rather, each anoestrous ewe needs a sustained and intense olfactory, behavioural, visual and auditory experience, probably for 48-72 hours. Thus, teasing will fail to induce ovulation or will lead to poor synchrony among the flock if the males produce stimuli of low quality and intensity. There are a few published comparisons on this topic: it appears that Dorset rams are more effective than Suffolk, Romney,

Romney x Finn, or Coopworth rams, with Merino rams being intermediate between Dorsets and Romneys (Meyer 1979; Tervit *et al.* 1977; Knight and Lynch 1980; Nugent *et al.* 1988; Scott and Johnstone 1994). There is some evidence also that the ram effect results in more twin ovulations than expected (Cognié *et al.* 1980) and that this outcome might be affected by the genotype of the stimulus ram. For example, King (1990) showed that, when Merino ewes were mated to Ronderib Afrikaner rams, they had a 22% higher fecundity (and therefore ovulation rate) than when mated to Merino rams.

Genetic research on teasing – b) Female factors. Considering the overwhelming commonality of genes among sheep genotypes, and the fact that the reproductive processes in all genotypes are virtually identical, the genotypic 'filter' can only be acting on a specific link in the physiological chain of events between perception of the socio-sexual signals and the secretion of GnRH (for detail, see review by Delgadillo *et al.* (2009). We need to look for genotypic variation in this chain and, as with the males, we need to consider the way the females respond to photoperiod. In the ewe, however, there are a number of extra levels of complexity. First, memory comes into play because a ewe will only respond to a ram that is 'new' to her – 'familiar' rams cannot switch on the reproductive centres in the ewe brain (Hawken *et al.* 2009). This involves 'olfactory memory' because the whole process is driven primarily by the odour of the ram and ewes can recognise individual males by their smell in the same way as they remember their offspring. Olfactory memory involves the production of new cells in the memory centres of the brain (Hawken *et al.* 2009). Second, ewes are not born with the complete ability to respond to the ram stimulus – they need to learn the process through sexual experience (review: Delgadillo *et al.* 2009).

GENETICS OF RATE OF REPRODUCTION

All components of reproduction rate are heritable traits. Safari *et al.* (2005) summarized the literature for ovulation rate ($h^2 = 0.15$), embryo survival ($h^2 = 0.01$), litter size $h^2 = (0.13)$ and lamb survival rate ($h^2 = 0.03$). These h^2 estimates are generally low, but the highly variable nature of these traits makes it possible to increase reproduction rate by selection. This was clearly demonstrated by Cloete *et al.* (2009) who showed that selection for the ability to rear multiple lambs results in an increase in the number of lambs weaned per ewe mated. Breeding values for the number of lambs weaned are now routinely provided by Sheep Genetics Services in Australia. However, focussing on specific components, such as ovulation rate, might lead to better outcomes.

Target 2: Fecundity (ovulation rate). The genetics of ovulation rate needs to be divided into two subsets: i) single genes, such as the Booroola mutation, that have a profound impact on a critical step in the process controlling follicle development in the ovary (review: Davis *et al.* 2005); ii) polygenic effects for which we have a large body of data and for which there are now standard breeding values available to industry. We will ignore the single-gene mutations because they are not a realistic option for industry and focus on the polygenic effects that bestow upon an animal its maximum potential litter size, with the final outcome depending on a variety of environmental factors, such as nutrition.

Basically, our aim should be for all ewes to bear twins because we know that, in our extensive production systems, it is disastrous for Merino ewes to bear triplets. Therefore, our major goal will be the identification of animals that have the genetic potential to produce a maximum of two ovulations, perhaps with the final outcome of single or twin births being decided by the breeder using focus feeding.

Genetics may also offer opportunities to reduce the variability in litter size as Hanrahan (2003) reported that 80% of adult ewes of the Icelandic breed had twin ovulations. He also found differences in the variability of ovulation rate between the Romanov and Finn sheep breeds. This suggests that it may be possible to select for reduced variation in litter size whilst maintaining a

potential ovulation rate of two. The genes that control ovulation rate and how they work are being revealed (review: Scaramuzzi *et al.* 2011) and it is essential to combine this understanding with our genetic goals.

Target 3: Fertility. The major advantage of a brief, concentrated period of lambing is that management strategies for maximising lamb survival become affordable. The ram effect can be use to synchronise lambing for flocks that are bred before February. For mating after February, when the ewes are ovulating spontaneously, we do not have a simple, effective and reliable CGE tool for synchronising cycles. In this situation, the ideal is to mate the ewes for only 17 days. The reality is that the fertility of Merino ewes is low so a significant proportion of the flock requires a second mating to become pregnant. This is surely an area where genetic selection could be used to improve efficiency.

LAMB SURVIVAL

We are probably losing about 10 million lambs per year, mostly in the first few days after birth. The economic impact of this problem can be seen simply by comparing that numerical loss with estimates of the number of lambs needed to satisfy our market. In addition, we have a potential disaster awaiting us if our domestic and export markets decide that a high rate of perinatal mortality is an ethical issue.

Genetic methods to select directly for perinatal survival have not been successful so alternative methods are being researched. Brien *et al.* (2010) have shown that lamb survival is lowly heritable and that selection for a multi-trait objective including reproduction rate, but not lamb survival, could result in an actual decline in lamb survival. Very little information is available on the importance of maternal genetic effects on lamb survival. An alternative approach is to increase survival rate by selection for reduced variation in birth weight in multiple births (Bodin 2010).

A focus on the causes of perinatal mortality might offer new opportunities for selection. The problem has been studied intensively for at least 50 years so we know that perinatal mortality is a multifactorial problem involving managerial as well as sheep-based factors. Here we will focus on two of the sheep problems: i) the timing of colostrum and the quantity of colostrum produced; ii) the behaviour of the ewe and lamb as they attempt to form their mother-young bond.

Target 4: Colostrum production. The importance of colostrum in perinatal survival and postnatal development has long been recognised. Recently, it has become clear that the quantity of colostrum that is available to the newborn depends greatly on the nutrition of the mother in the final week of pregnancy (review: Banchero *et al.* 2006) and we have incorporated this into the CGE program (Fig. 1). In addition, two sources of variation could also be exploited.

Genetic research on colostrum production – quantity produced. There are clear differences between genotypes (milk breeds vs meat and wool breeds) in milk production, with Merinos near the bottom of the table, and wide variation between ewes within genotypes (Bencini *et al.* 1992). Udder size, the quantity of milk produced, and the components of milk, are all heritable traits and respond to selection (Barillett 1997). There is therefore no reason that we cannot improve the ability of Merino mothers to feed their lambs. Obviously, greater capacity to produce milk will need to be balanced by feed supply, but lactation often falls in the peak period for quality and quantity of pasture production.

Genetic research on colostrum production – timing of production. There is considerable variation in the synchrony of parturition and colostrum supply, in Merinos in particular (review: Nowak and Poindron 2006). In many cases, colostrum production appears to be delayed, often by many hours, leading to a scenario that is disastrous if the weather is inclement (McNeill *et al.* 1988). It is important to determine the genetic mechanisms that underlie this effect.

Because we are interested in increasing fecundity, we need to take into consideration an important interaction – compared to single-bearing ewes, twin-bearing ewes produce more colostrum but less per lamb, while the onset of lactation is slower (review: Nowak and Poindron 2006). This adds to the disadvantage of low birth weights and reduced energy reserves in twinborn lambs. Thus, a genetic strategy for dealing with colostrum must consider the genetic strategy for fecundity.

Target 5: Mother-young bonding. Variation between genotypes in neonatal survival is well documented, usually with the Merino at the bottom of the table and British breeds at the top. Behavioural studies have shown us why this is the case – compared to Merino cross sheep, Merino ewes take longer to recognise their newborn lambs, and their lambs take longer to recognise their dams. Even among Merino strains (Trangie, Australian Merino Society, Booroola), maternal behaviour differs, with the differences being more apparent in twin-bearing than in single-bearing ewes (review: Nowak 1996).

Rearing performance is repeatable (Piper *et al.* 1982; Haughey 1984) but estimates of repeatability and heritability in the Merino are low. Nevertheless, in Merino lines that have been selected for a decrease or increase in multiple rearing rate, ewes from the high line groomed their lambs quicker and for longer after birth whereas ewes from the low line were more likely to start grazing earlier (Cloete *et al.* 2002). This shows that mothering ability can be improved significantly, even by selection on a trait as complex as multiple rearing rate. It is feasible that focussing attention on specific behaviours, and considering litter size, might increase the rate of improvement.

Target 6: Weaners to survive and thrive. While perinatal mortality often confronts us with mountains of little bodies and worrying numbers for the national industry, there is a risk that we can forget another major source of loss – weaner mortality. Weaner mortality tends to be steady, only a few percent every week, but can accumulate over 9-12 months to become as large as perinatal mortality. The slow but gradual loss of animals makes it very difficult to diagnose the causes, but diseases and parasites, compounded by poor nutritional management, can probably explain much of the problem. Here we will focus on health.

The obvious genetic targets are resistance to flystrike and to internal nematodes, the two most important diseases affecting sheep. Substantial progress has been made in breeding for worm resistance (Woolaston and Piper 1996; Karlsson and Greeff 2006) and for blowfly resistance (Greeff *et al.* 2009; Smith *et al.* 2009). ASBVs are now available for faecal worm egg count and for the indicator traits of breech strike (breech wrinkle, dags, breech cover). All the known factors that could affect breech strike explain only 25% of the variation between animals (Greeff *et al.* 2010), but selecting animals on the three indicator traits for breech strike will improve the health and welfare of the Australian sheep flock. Research is underway to identify other sources of variation. The next health issue that needs to be researched is selection for resistance against lice.

We have made significant gains in these areas and now we need to ensure that the genetic advantages penetrate the national flock. Clearly, this approach fits squarely within our CGE framework because it deals simultaneously with both animal welfare and the reduced use of chemicals and drugs.

CONCLUSION

The CGE concept is a useful framework within which to develop R&D that will ultimately allow us to develop new management strategies that will improve the health, welfare and productivity of ruminants. The new strategies will be based on science so should be reliable and repeatable but, to date, the research has been limited to diseases, and behavioural and physiological studies. We need discussion and research in quantitative and molecular genetics as a means of finding solutions to the major limitations in the CGE framework – we have identified variation in critical components of sheep biology and, if there is some investment in research, we will soon be able to identify gene products that will focus our selection criteria. We will then be in a good position to use the power of genetics to enable management that is low-cost and low-tech and thus perfectly suited to extensively grazed sheep, thus giving us a head start in industry uptake. We will be greatly aided by our developing ability to generate robust genetic data for a wide range of production traits under extensive production systems. Implementation of CGE management will allow us to improve the image of the industry in the marketplace and thus provide a platform for a long and profitable future.

ACKNOWLEDGEMENTS

Much of the research that we have cited was supported by Meat and Livestock Australia and Australian Wool Innovation. Many of our studies could not have been contemplated without the generous assistance of the students and staff of Animal Science (The University of WA) and of the Department of Agriculture and Food Western Australian.

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