A molecular approach to genetic improvement of South African Angora goats

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ABSTRACT

South Africa is considered the primary producer and most reliable source of high quality clip mohair in the world. The application of molecular technologies to improve mohair quality is relatively new to this industry. The aim of the study was to use a molecular approach to genetically improve South African Angora goats, with emphasis on mohair production. A reference population of Angora goats was firstly established consisting of twelve sire families with half-sib offspring (1067 individuals in total). The genetic variation of this population was evaluated using microsatellite markers and the average gene diversity was found to be above 60%. Ninety four microsatellite markers were then genotyped on the reference population, spanning 23 chromosomes (total length 1352cM) with an average marker interval of 23.0cM. This information was used to improve previously published goat linkage maps. Unmapped microsatellite markers were incorporated and previously published inter-chromosomal rearrangements between the goat and sheep genetic maps were confirmed or rejected. Nine new markers were mapped to the goat genome, and six chromosomes showed rearrangement when compared to the previous goat map. Four previously reported intra-chromosomal rearrangements were shown to be either population specific or mapping errors. Variance components and genetic parameters of mohair traits (FW, FD, CVFD, SDFD, CF, SF and SDA) were estimated; including the fibre diameter profile measured using OFDA technology that has not yet been included in genetic evaluations. Heritability estimates ranged between 0.14 (SDA) and 0.63 (CF). OFDA-measured traits should be considered for inclusion into the national breeding strategy. The reference population was lastly analysed to identify QTL associated with fleece traits. Eighteen putative QTL were identified for seven mohair traits on 13 chromosomes. Three putative QTL were detected for FW on CHI 2, 5 and 24 corresponding with KRT and KAP gene locations. Two QTL associated with mohair FD (on CHI 4 and 24) were detected. QTL contributions to variance ranged between 7.44% (CF) and 19.69% (SDA). The results of this study should form part of an integrated approach where both quantitative and molecular tools are applied for genetic improvement of South African Angora goats.

A fact is a simple statement that everyone believes. It is innocent, unless found guilty. A hypothesis is a novel suggestion that no one wants to believe. It is guilty, until found effective.

Edward Teller

DECLARATION

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Science investigates; religion interprets. Science gives man knowledge, which is power; religion gives man wisdom, which is control. Science deals mainly with facts; religion deals mainly with values. The two are not rivals.

Martin Luther King, Jr.

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Chapter 1: Introduction

The *Capra aegagrus* was one of the first wild herbivores to be domesticated (Gentry *et al.,* 2004; Pereira *et al.*, 2009) around 9 000 to 11 000 years ago, giving rise to the domestic goat (*Capra hircus*). Recent genetic analyses have suggested that there were likely separate domestication events in several continents (Fernandez *et al.,* 2002; Gentry *et al.,* 2004), resulting in at least three matrilineal lines. Lineage A (from the Fertile Crescent region) is estimated to have started expanding 10 000 years ago, predominates across the globe and is thought to be the origin of the modern Angora goat. Lineage B seems limited to breeds from southern Asia and Lineage C is very rare and only present in a few European breeds (MacHugh & Bradley, 2001). This was supported by the six mitochondrial DNA haplogroups for domesticated goats that were identified by Naderi *et al.* (2008), of which the A haplogroup represented more than 90% of goats worldwide. Due to their small and portable size, goats have the advantage of being a very mobile species and spread rapidly worldwide owing to trade, warfare, thievery and human migration (Pereira *et al.,* 2009).

Goats play an important agricultural role in present-day society as they are a reliable source of meat, milk, skins and fibre. The goat population has increased dramatically worldwide by 66% from approximately 485.1 x 10^6 animals in 1985 to 808.9 x 10^6 animals in 2005. Dubeuf & Boyazoglu (2009) postulated that the increase is likely due to the growing poor population's need for subsistence farming, and not necessarily because of development of the species. The majority of goats are kept in extensive / low-input systems (Peacock & Sherman, 2010) and many are kept because of their role in traditional and cultural ceremonies (e.g. burials and weddings) (Drum, 1991), for everyday use (Boyazoglu *et al.,* 2005) and because they are still regarded as almost sacred in many cultures. In the African context, cattle represent wealth and symbolizes power (Sikhondze, 2008) while goats do not give the same status to farmers. The goat sector has historically received significantly less support compared to other livestock industries from both the scientific and commercial sides (Dubeuf *et al.,* 2004) and the industry remains underestimated and under-utilized (Dubeuf & Boyazoglu, 2009).

Goat farming is however slowly leaving behind its world-wide negative image of being livestock for poor people, and developed countries are starting to view goats more favourably. Dairy goat farming is now an established sector in many European countries, with the majority of the goat milk sold as cheese (Dubeuf *et al.,* 2004). Both the United States of America and Canada have also established dairy goat industries. Although the world production of goat meat has more than tripled from 1961 (1.1million tons) to 2001 (3.7 million tons), its production remains much lower than that of other meat industries, and centres around China, India, Nigeria and Iran (Dubeuf *et al.,* 2004). Globally, the goat meat market is still marked by very little innovation, processing and a strong bias

against the product, as it is still known as the "meat of the poor". In northern Europe goat meat products are still poorly valued and large scale goat meat production systems are currently limited to the Mediterranean, while a few USA states also contribute to the industry. Australia is the main goat meat exporting country, with a market share in excess of 60% (Dubeuf *et al.,* 2004). Emerging countries with high income (e.g. Malaysia, Oman, Japan and Korea) are the main importers of goat meat, with an increasing demand for the product. In Southern Africa meat goat farming is mainly part of mixed cropping-livestock systems in rural areas where goats have to provide milk, dairy products and meat (Casey & Webb, 2010). A very small proportion of the goats (<1%) are slaughtered at central abattoirs and form part of the commercial red meat supply chain (Simela, 2005), while most meat goats are sold and slaughtered in the informal market (both peri-urban and rural) as well as for house-hold consumption. Abattoirs consider the supply of goats too irregular and insufficient to justify regular slaughtering. The goats that do pass through the formal channels are usually sourced from specialised meat goat breeds (SA Boer goat, Savanna and Kalahari Red goats) which are farmed in commercial systems.

Angora goats do not play a significant role in the above-mentioned markets, but instead serve a specialized industry focusing on the lustrous mohair produced by them. Mohair is a luxury fibre, admired for its superior lustre, handle and high quality. In contrast with the global goat meat sector, the mohair industry is well organised. South Africa is the major role player, producing approximately 55% of the world product, while 25% - 30% of the global product is purchased by one factory in the UK (Dubeuf *et al.,* 2004). World production of mohair decreased sharply between 1988 and 2003, with a drop of 70% to 6.6 million kg (Van der Westhuysen, 2005) owing mostly to a shift towards synthetic fibres in the fashion industry. Global production has decreased further over the last five years, owing mostly to the global financial crises. Although it is a sought-after commodity in its specific niche market, its demand is also heavily dependent on the unpredictable fashion industry. Goat-produced fibres make up about 0.04% of the world textile fiber production (Lupton, 2004), to which the total mohair production contribute less than 0.02%.

1.1 The South African Angora goat industry

The Angora goat was domesticated in Turkey, in the region of Angora (now the city of Ankara), from where they were exported to Europe during the sixteenth century in an attempt to establish a wider mohair industry. The European climate was however not suited to these goats and South Africa (a British colony at the time) was identified as a favourable region for Angora goat production. During 1838 the first Angora goats were imported to South Africa, with more that 3000 animals following these between 1856 and 1896 (Friedrich, 2009). The Karoo (meaning "thirsty land") and harsh, semi-arid Eastern Cape region proved to be well-suited to the Angora goats, and a

flourishing industry evolved from this humble beginning. Today the mohair industry in South Africa consist of approximately 870 000 Angora goats, most of which are still farmed in the Eastern Cape. The Angora ram Breeders' Association consist of 59 stud breeders who supply high quality genetic material to the almost 1000 commercial producers (The Angora Goat & Mohair Journal, 2009).

South Africa is widely considered as the most reliable producer of a good quality clip of mohair worldwide, and produces in excess of 50% of the world product annually (NDAFF, 2009). Early processing (e.g. scouring and combing) of the mohair is occasionally performed in South Africa and then sold as "tops", while raw (unprocessed) mohair is also sold. Mohair in either of these forms are mainly sold at orderly "open-cry" auctions, although branding also account for limited trading (Van der Westhuysen, 2005). Mohair production in South Africa has decreased by 21.6% from 3.7 million kg in 2004 to 2.9 million kg in 2008. This decrease has been attributed to a continuing drought in large parts of the production area, while the global financial crises also contributed to a large extent.

Despite the troubling economic times, the South African National Department of Agriculture, Forestry and Fisheries' (NDAFF) mohair outlook for the 2009 / 2010 season remained positive. While the demand for kid mohair was expected to remain low, young goats' hair was recovering. The first two auctions of the 2010 season fared better than expected, with an increase in market indicators (http://www.cmw.co.za/market_reports/files/CMWMOHREP_2010S-01.pdf). There is a continuing fashion trend in Europe which favours animal fibres and the demand for mohair (especially young goat and adult hair) is expected to remain stable (http://www.mohair.co.za/index.php/news/39-mohair-market-indicator-up-more-than-10).

A number of companies play major roles in the South African industry. Mohair South Africa is responsible for the advancement of a sustainable local industry through the creation of international partnerships. Cape Mohair and Wool is the largest mohair brokerage firm in South Africa. Approximately 50% of all mohair produced globally passes through this firm's handling and warehouse facilities. Producers in the Camdeboo region decided to establish an exclusive high-quality brand ("Camdeboo") which is now recognised globally as an authoritative mohair trademark. Mohair producers are supported by the South African Mohair Growers' Association, which is responsible for promoting and guiding its members through their 33 branches in the Eastern Cape (The Angora Goat & Mohair Journal, 2009).

In the past selection of South African Angora goats have focussed on three main traits, namely body weight, fleece weight and fibre diameter. Fibre diameter is the most economically important of

these, determining both the price and the processing of the fibre (Qi *et al.,* 1994). Staple length and the general appearance of the fleece (including style, character and evenness of the fleece) also influence the price (refer to Addendum A for the detailed Mohair Classing Standards of Mohair South Africa). During the mid 1980s, the South African clip became stronger, possibly due to selection for increased fleece weight, while disregarding fibre diameter (Snyman, 2002). Since then, selection in the national herd has been performed based on a selection index including body weight, fleece weight and fibre diameter. Body weight is included, as selection for decreased fibre diameter results in a corresponding body weight decrease, resulting in lowered robustness and increased mortalities. The unfavourable positive genetic correlation between fleece weight and fibre diameter also necessitates emphasis to be put on both these traits.

1.2 The current status of Angora goat research

Quantitative research

Conventional selection was practiced for the past two decades with the aim of improving phenotypic mohair characteristics. Genetic parameters assisted breeders to make informed decisions on the inclusion of selection criteria into breeding programs (Nicoll, 1985). Genetic parameters have previously been estimated for French (Allain & Roguet, 2003), Argentinean (Taddeo *et al.,* 1998), Australian (Gifford *et al.,* 1991) and South African (Snyman & Olivier, 1996; 1999) Angora goats. Traits included varied over studies, with only fleece weight and fibre diameter being included in all these studies, although measured at different ages. The ranges of heritability estimates for and correlations between these two traits vary extensively (from high to low for both traits) as reported by Pattie *et al.* (1990) and Sumner & Bigham (1993), and are discussed in detail in Chapter 4.

An important shortcoming of previously published results is the lack of newly-measured quality traits; especially those measured using Optical Fibre Diameter Analyser (OFDA) technology. OFDA2000 technology is used for the accurate, objective measurement of the full fibre diameter profile (Qi *et al.,* 1994), and is currently routinely used for fleece measurement in most animal fibre producing countries, including South Africa (details on these parameters are provided in Chapter 4). The measurement of fibre diameter, its variation and distribution as well as other important quality traits have introduced the possibility of including new criteria in a selection objective aiming to improve the quality of mohair. These quality traits have however largely been ignored by the mohair industry, and there is a dearth of information on genetic parameters for them, with estimates only for French Angora goats (Allain & Roguet, 2006).

Research on South African Angora goats were primarily performed by the Grootfontein Agricultural Development Institute (GADI) in Middelburg, South Africa. The industry realized during

the 1990s that although considerable progress has been made at increasing fine mohair production, the inability of Angora goats to survive sub-optimum conditions has become a great concern. This was mainly due to the unfavourable positive genetic correlation between body weight and fibre diameter resulting in small, unthrifty animals (Snyman *et al.,* 1996). As the general practice of energy supplementation to increase survivability and production has become economically unacceptable, more emphasis was placed on increasing body weight of the goats. This led to the review of breeding objectives and selection criteria used by breeders. Heritability and repeatability of and correlations between body weight, fleece weight and fibre diameter was estimated on data obtained from the Angora goat performance testing pilot scheme in 1996 (Snyman & Olivier, 1996; Snyman *et al*., 1996). These results were used to design a selection index (SI) aimed at increasing body weight, decreasing fibre diameter and maintaining fleece weight (SI = (13 x body weight) + (4x fleece weight) - (23 x fibre diameter)). This index was accepted by the breeders, and used to select replacement bucks and does throughout the industry. Breeders still tended however to place emphasis on subjective fleece traits (softness of face and ears, evenness of fleece, style, character etc.) of which no reliable genetic parameters were available (Snyman & Olivier, 1999). Snyman and Olivier (1999) found that these traits all had low repeatabilities and recommended that emphasis should rather be placed on economically important traits, such as body weight and fibre diameter. In 2002 the selection strategy followed by the industry was evaluated, and Snyman (2002) concluded that selection for decreased fibre diameter, while maintaining or increasing body weight and fleece weight leads to the genetic improvement of South African Angora goats.

Body weight is not only important as a factor influencing mohair production. It is also directly related to the survivability of young goats, as well as the reproductive ability of young does. Snyman (2007) reported that post-weaning growth rate of kids without supplementary feeding was unacceptably low. Most farmers do not supply ewe kids with supplementary feeding after weaning, because of the direct financial implication. The most important result of this is that many young does don't conceive when they are mated for the first time at 18 months, as they have not yet reached the target weight of 25kg. It is evident that this area should receive attention from researchers as it affects the economic profitability of the industry directly. Another factor leading to low profitability is the well-recognised poor reproductive efficiency of adult Angora goats (Snyman, 2010). Mortality rates of South African Angora goats have however been very poorly documented, making it very difficult to improve the trait genetically. Snyman (2010) recommended that does with udder problems or giving too little milk should be culled, while care should be taken to avoid using bucks that produce small, unthrifty and deformed kids.

The South African Angora industry has been hampered by the severe loss of young, newly shorn goats, especially during cold spells (Storbeck *et al.,* 2009). The physiological cause of these deaths seem to be linked to the goats' glucose metabolism, with lower cortisol production in this breed compared to other breeds such as the Boer goats and Merino sheep (Goosen *et al.,* 2010). As high quality mohair traits have been found to be negatively related to fitness traits (Snyman & Olivier, 1999; Webb & Casey, 2010), it was postulated by Goossen *et al.* (2010) that the selection for fleece traits have resulted in a reduced adrenal function, characterized by a decrease in cortisol production. Based on this information, breeding trials have been introduced to determine whether more robust Angoras can be bred while maintaining their favourable mohair characteristics.

Although significant genetic progress has been made in the past decades through the use of conventional selection methods, consumer demands for improved quality lead to the incorporation of biotechnology. It is believed that DNA marker information will assist conventional selection by increasing selection accuracy and improving the rate of genetic improvement, as well as leading to a better understanding of the physiological background of quantitative traits (Pollak, 2005; Jeon *et al.,* 2006; Dodds *et al.,* 2007). Mohair SA has taken the initiative to evaluate how molecular technology can advance the Angora goat industry, through the integration of molecular and quantitative information.

Molecular research

Since the advent of molecular genetics including the discovery of the PCR procedure and abundant, polymorphic markers, livestock breeding has entered a new era. The almost unlimited possibilities and application of biotechnology in farm animals have become the focus of many research groups and institutes. Proof of this can be seen by the whole-genome sequencing efforts for many farm animal species, e.g. chicken, cattle, sheep, swine and horses (http://www.ncbi.nlm.nih.gov), the improvement of and value-adding to animal products and the impact on reproduction technology.

Although certain farm animal species (mainly those with a huge economic impact e.g. beef and dairy cattle, poultry and pigs) have received constant attention from researchers, molecular studies on goats have remained relatively limited. Biotechnology and genomics can benefit the goat industry in a number of ways, including pedigree verification, traceability of products, diagnostic tests and marker assisted or gene assisted selection (MAS or GAS), as presented by Dodds *et al.* (2007) and Barrera-Saldaña *et al.* (2010). More recently genomic selection (GWAS) in which an estimation of the total genetic value of breeding animals are estimated, have been proposed (Goddard, 2009). Some of these applications of DNA technology are briefly discussed in this section. Since almost none of

these have been applied on Angora goats, a more general overview on molecular research on goat breeds is given.

A genetic map is the cornerstone of molecular research for any species, detailing the locations of DNA markers on the specific genome. The goat linkage map is relatively underdeveloped (Maddox & Cockett, 2007) and in effect still quite primitive when compared to most other livestock species. The most recent goat map was published in 1998 by Schibler *et al.,* using 307 microsatellite markers. Currently the goat map contains 731 loci with 271 genes, and 423 microsatellite markers (no SNPs have yet been published for goats). Only two regions have so far been mapped in any detail; the flanking areas of the Polled Intersex Syndrome and the αs1-casein genes have been saturated with markers and their molecular basis is well-understood (Van der Werf, 2007). The remaining part of the goat genome is poorly covered with markers (average distance between markers is 14.5 cM) and a number of discrepancies between the goat and the ovine and bovine maps have been identified.

The estimation and maintenance of genetic variability in conservation of breeds is one of the advantages of using DNA-based technology. Genetic diversity, phylogenetic relationships between breeds and co-ancestry can be investigated using genotypic information. Various Chinese (Qi *et al.,* 2009), Indian (Kumar *et al.*, 2005; Gour *et al.,* 2006) and European (Martinez *et al.,* 2004; Iamartino *et al.,* 2005) goat breeds have been included in genetic characterization studies. Globally, goats have however not yet been described in sufficient detail, lagging behind other livestock species. Genetic diversity studies on South African goats are limited to a study on commercial meat goat breeds (Visser *et al.,* 2004) and one broader phylogenetic study that included Angora goats, three meat goat breeds as well as indigenous populations (Pieters, 2007).

The improvement of pedigree integrity is another economically important application of DNA technology. Angora goats are farmed mainly under extensive systems in South Africa, with herd sizes ranging between 1000 and 2500 animals. Breeders primarily make use of group mating, with only stud breeders considering individual mating. Both these practices are however followed by over-mating (the practice of pooling all females and males after individual or group mating, in order to increase pregnancy rates), rendering accurate paternal pedigree recording impossible (Friedrich, 2009). Apart from this, females are generally prone to poor mothering ability – leaving offspring unattended and stealing of offspring. Bolormaa *et al.,* (2008) showed that a high rate of incorrect maternal pedigree is observed in Angora goats, even when kids are assigned to their dams in the first hours or days after birth. Approximately 23% of Angora goat kids born in both commercial and stud herds between 2000 and 2004 had incomplete or inaccurate pedigree records (Friedrich, 2009). Correct pedigrees are essential for accurate genetic parameter estimation, which has an impact on

selection program development and rate of genetic progress (Visscher *et al.,* 2002; Bolormaa *et al.,* 2008). Due to these factors limiting parentage recording, a study was conducted to compile a microsatellite marker set for parentage verification. It is envisaged that this will benefit the industry and will result in greater selection accuracy in future.

The identification of specific regions of interest and the selection thereof (either through the use of closely linked markers (MAS) or of the causative mutation itself (GAS)) has been the aim of many molecular studies. The selection for chromosomal areas that directly contribute to the genetic variation of traits of economic importance will lead to increased genetic progress and offers the opportunity to better understand and exploit phenotypic variation (Dekkers, 2004). This has however mainly remained a theoretical concept in many species. Implementation of marker assisted selection in goats have been limited to the use of selection against diseases (scrapie, CAEV and Johne's disease), and for casein genes in dairy goats (Van der Werf, 2007), but no MAS / GAS are currently practiced for fibre traits (Dodds *et al.,* 2007). Genomic selection has been proposed as a more efficient way of using molecular information. This method requires that the genome should be densely populated with markers in order for them to explain the total genetic variance of a quantitative trait (Moser *et al.,* 2009). Genomic Estimated Breeding Values (GEBV) can then be calculated and should lead to rapid genetic improvement due to higher selection accuracy and reduced generation intervals. One of the main challenges to the feasibility of this method in goats is the absence of SNP for the species. As soon as a SNP discovery project for goats has been performed and the results made publicly available, GWAS could play an important role in genetic improvement of goats. The rate of genetic response to GWAS will however also decline over subsequent generations (Goddard, 2009) and this methodology is not without its own challenges and limitations.

1.3 Aim of the study

Although animal biotechnology has developed rapidly during the past three decades, it is still a relatively unexploited field in the South African livestock industry. Dairy and beef cattle producers are starting to make use of available technologies (parentage verification, limited use of MAS and genomic EBV calculation), but this is limited to a very small number of farmers. Most species, including the South African Angora goats, still largely select animals based on either mass selection or BLUP. The application of Estimated Breeding Values is limited by poor breeder participation in official animal recording in South Africa. Although progress has been made with the use of quantitative selection, it has several limitations which has been well documented (Andersson, 2001; Dodds *et al.,* 2007).

The mohair industry of South Africa acknowledged both the limitations of quantitative selection and the opportunity that DNA technology poses for livestock improvement. Discussions between Mohair South Africa, the NDAFF (mainly the Grootfontein Agricultural Development Institute) and the University of Pretoria resulted in the initiation of a DNA Bio-bank for small-stock in 2005, with the further intent of molecular research. All the goats at the Jansenville experimental farm of the NDAFF were sampled as a base population and to increase the size of this population, breeders from the stud industry were selected to participate. These animals formed the Angora goat reference population and any findings from this study will therefore be applicable to the whole Angora goat population in South Africa. This project was initiated to evaluate how molecular information could contribute to the genetic improvement of the South African Angora goat.

The study aimed to use molecular technology for the benefit of the Angora goat industry through improved selection accuracy and increased rate of genetic improvement. To achieve these aims, the following objectives were set:

i. Establishing an Angora goat reference population with complete pedigree, phenotypic and genomic data.
ii. Evaluation of the genetic variation within the reference population to estimate the level of genetic diversity.
iii. Improving the accuracy of the caprine linkage map by adding new microsatellite markers and verifying previously reported inter-chromosomal re-arrangements.
iv. The estimation of genetic parameters for newly-measured OFDA fleece traits, in order to include these traits in further research.
v. To identify QTL affecting fleece traits in South African Angora goats, which could possibly identify chromosomal regions which in further research could be used for fine mapping and identification of candidate genes.

Conclusions

These aims have been attained in phases and are reported in this thesis by way of published scientific articles. Chapter 2 contains the manuscript "Genetic variation of the reference population for QTL research in SA Angora goats", which describes the establishment of the reference population and evaluates its genetic variation. In "A genetic linkage map for the South African Angora Goat" (Chapter 3) an improved goat linkage map is generated which will form the basis of a more advanced map and advance the identification of loci contributing to phenotypic variation. The heritability for and correlations between product and quality traits were estimated in Chapter 4, and new traits were evaluated for potential inclusion in the breeding goal. Finally, Chapter 5 focuses on the identification of QTL for economically important mohair traits. In Chapter 6 a critical review on the study is given and future projects are discussed.

References

Allain, D. & Roguet, J.M., 2003. Genetic and non-genetic factors influencing mohair production traits within the national selection scheme of Angora goats in France. *Livest. Prod. Sci.* 82, 129-137.

Allain, D. & Roguet, J.M., 2006. Genetic and non-genetic variability of OFDA-medullated fibre contents and other fleece traits in the French Angora goats. *Small Rumin. Res*. 65, 217-222.

Andersson, L. 2001. Genetic dissection of phenotypic diversity in farm animals. *Nat. Rev. Genet.* 2 (2001), pp. 130–138.

Bolormaa, S., Ruvinsky, A., Walkden-Brown, S. & Van der Werf, J., 2008. DNA-based parentage verification in two Australian goat herds. Small Rumin. Res. 80, 95-100.

Barrera-Saldaña, H.A., Ascacio-Martínez, J.A., Sifuentes-Rincón, A.M., Arellano-Vera, W. & Arbiza, S.I., 2010. Applications of biotechnology and genomics in goats. *Small Rumin. Res*. 89, 81-90.

Boyazoglu, J., Hatziminaoglou, I. & Moarnd-Fehr, P., 2005. The role of the goat in society: Past, present and perspectives for the future. *Small Rumin. Res*. 60, 13-23.

Casey, N.H. & Webb, E.C., 2010. Managing goat production for meat quality. *Small Rumin. Res*. 89, 218-224.

Dekkers, J.C.M., 2004. Commercial application of marker- and gene-assisted selection in livestock: Strategies and lessons. *J. Anim. Sci.* 82 (E Suppl): E313-328.

Dodds, K.G., McEwan, J.C. & Davis, G.H., 2007. Integration of molecular and quantitative information in sheep and goat industry breeding programmes. *Small Rumin. Res*. 70, 32-41.

Drum Publications, 1991. Strange rituals and township burials: township funeral undertakers talk. Oct. 1991, 152: 48-49.

Dubeuf, J.-P. & Boyazoglu, J., 2009. An international panorama of goat selection and breeds. *Livest. Sci.* 120, 225-231.

Dubeuf, J.-P., Morand-Fehr, P. & Rubino, R., 2004. Situation, changes and future of goat industry around the world. *Small Rumin. Res*. 51, 165-173.

Fernandez, H., Taberlet, P., Mashkour, M., Vigne, J & Luikart, G., 2002. Assessing the origin and diffusion of domestic goats using ancient DNA In: First steps of animal domestication. Oxbow Books, Oxford, UK. P51-54.

Friedrich, H., 2009. Evaluation of microsatellite markers for parentage verification in South African Angora goats. MSc thesis, University of Pretoria, Pretoria, South Africa.

Gentry, A., Clutton-Brock, J. & Groves, C.P., 2004. The naming of wild animal species and their domestic derivatives. *Journal of Archaeological Science* 31, 645-651.

Gifford, D.R., Ponzoni, R.W., Lampe, R.J., Burr, J., 1991. Phenotypic and genetic parameters of fleece traits and live weight in South Australian Angora goats. *Small Rumin. Res.* 4, 293-302.

Goddard, M., 2009. Genomic selection: prediction of accuaracy and maximisation of long term response. *Genetica* 136, 245-257.

Goosen, P., Swart, A.C., Storbeck, K-H. & Swart, P., 2010. Hypocortisolism in the South African Angora goat: The role of 3βHSD. *Mol Cell Endocrinol.* 315, 182-187.

Gour, D.S., Malik, G., Ahlawat, S.P.S., Pandey, A.K., Sharma, R., Gupta, N., Gupta, S.C., Bisen, P.S & Kumar, D. 2006. Analysis of genetic structure of Jamunapari goats by microsatellite markers. *Small Rum Res.* 66, 140-149.

Iamartino, D., Bruzzone, A., Lanza, A., Blasi, M. & Pilla, F. 2005. Genetic diversity of Southern Italian goat populations assessed by microsatellite markers. Small Rum Res. 57, 249-255.

Jeon, J.T., Lee, J.H., Kim, K.S., Park, C.K. & Oh, S.J., 2006. Application of DNA markers in animal industries. *Aust J Exp Agric.* 46, 173-182.

Kumar, D., Dixit, S.P., Sharma, R., Pandey, A.K., Sirohi, G., Patel, A.K., Aggarwal, R.A.K., Verma, N.K., Gour, D.S. & Ahlawat, S.P.S. 2005. Population structure, genetic variation and management of Marwari goats. *Small Rum Res.* 59, 41-48.

Lupton, C.J., 2004. Export potential, market outlook and value-added processing of goat fibers. *J Anim Sci. 82 (Suppl 1),* 446 (Abstr.).

MacHugh, D.E. & Bradley, D.G., 2001. Livestock genetic origins: Goats buck the trend. PNAS 98 (10), 5382-5384.

Maddox, J.F. & Cockett, N.E., 2007. An update on sheep and goat linkage maps and other genomic resources. *Small Rumin Res.* 70(1), 4-20.

Martinez, A.M., Carrere, M.P., Acosta, J.M, Rodriguez-Gallardo, P.P., Cabello, A., Camacho, E. & Delgado, J.V. 2004. Genetic characterization of the Blanca Andaluza goat based on microsatellite markers. *SA J Anim Sci.* 34 (Suppl 1), 17-19.

Moser, G., Tier, B., Crump, R.E., Khatkar, M.S. & Raadsma, H.W., 2009. A comparison of five methods to predict genomic breeding values of dairy bulls from genome-wide SNP markers. Genet Select Evol. 41:56.

Naderi, S., Rezaei, H-R., Pompanon, F., Blum, M.G.B., Negrini, R., Naghash, H-R., Balkiz, O, Mashkour, M., Gaggiotti, O.E., Ajmone-Marsan, P., Kence, A., Vigne, J-D. & Taberlet, P., 2008. The goat domestication process inferred from large-scale mitochondrial DNA analysis of wild and domestic individuals. *PNAS*, 105 (46), 17659-17664.

National Department of Agriculture, Forestry and Fisheries, 2009. Trends in the Agricultural Sector. P69-70.

Nicoll, G.B., 1985. Estimates of environmental effects and some genetic parameters for weaning weight and fleece weights of young Angora goats. *Proc. New Zealand Soc. Anim. Prod.* 45, 217-219.

Pattie, W.A., Baker, R.L., Nicoll, G.B., Restall, B.J., 1990. Breeding for cashmere and mohair. *Proc. 4th Wrld Congr Gen Appl Livest Prod.,* 23-27 July, 1990. Edinburgh.

Peacock, C. & Sherman, D.M., 2010. Sustainable goat production – Some global perspectives. *Small Rumin. Res*. 89, 70-80.

Pereira, F., Queiros, S., Gusmao, L., Nijman, I.J., Cuppen, E., Lenstra, Econogene Consortium, J.A., Davis, S.J.M., Nejmeddine, F. & Amorim, A., 2009. Tracing the history of goat pastoralism: New clues from mitochondrial and Y chromosome DNA in North Africa. *Mol. Biol. Evol*. 26 (12), 2765-2773.

Pieters, A., 2007. Genetic characterization of commercial goat populations in South Africa. MSc thesis, University of Pretoria, South Africa.

Pollak, E.J., 2005. Application and impact of new genetic technologies on beef cattle breeding: a "real world" perspective. *Aust J Exp Agric.* 45, 739-748.

Qi, K., Lupton, C.J., Pfeiffer, F.A., Minikhiem, D.L., 1994. Evaluation of the Optical Fibre Diameter Analyser (OFDA) for measuring fiber diameter parameters of sheep and goats. *J. Anim. Sci.* 72, 1675-1679.

Qi, Y., Luo, J., Han, X., Zhu, Y., Chen, C., Liu, J. & Sheng, H. 2009. Genetic diversity and relationships of 10 Chinese goat breeds in the Middle and Western China. *Small Rumin. Res*. 82, 88-93.

Schibler, L., Vaiman, D. Oustry, A., Giraud-Delville, C & Cribou, E.P., 1998. Comparative gene mapping: A fine-scale survey of chromosome rearrangements between ruminants and humans. Genome Res 8, 901-915.

Sikhondze, B.A.B., 2008. Diversity in rural African economic activities: A case study of cattle and modernization in Swaziland, 1930s to 1980s. *Journal of Humanities and Social Sciences*, 2(1), 1-11.

Simela, L., 2005. Meat characteristics and acceptability of chevon from South African indigenous goats. PhD Thesis, University of Pretoria, Pretoria, South Africa.

Snyman, M.A., 2002. Evaluation of a genetically fine mohair producing herd. *Small Rumin. Res*. 43, 105-113.

Snyman, M.A., 2007. Body weight and growth rate of South African Angora goat kids under different pre- and post-weaning management systems. *SA J Anim Sci.* 37 (2), 132-141.

Snyman, M.A., 2010. Factors affecting pre-weaning kid mortality in South African Angora goats. *SA J Anim Sci.* 40 (1), 54-64.

Snyman, M.A. & Olivier, J.J., 1996. Genetic parameters for body weight, fleece weight and fibre diameter in South African Angora goats. *Livest. Prod. Sci.* 47, 1-6.

Snyman, M.A. & Olivier, J.J., 1999. Repeatability and heritability of objective and subjective fleece traits and body weight in South African Angora goats. *Small Rumin. Res*. 34 103-109.

Snyman, M.A., Olivier, J.J. & Wentzel, D., 1996. Breeding plans for South African Angora goats. *Angora Goat Mohair* 38, 23-31.

Storbeck, K-H, Swart, A.C. & Swart, P., 2009. CYP17 causes hypocortisolism in the South African Angora goat. *Mol Cell Endocrinol.* 300, 121-125.

Sumner, R.M.W. & Bigham, M.L., 1993. Biology of fibre growth and possible genetic and non-genetic means of influencing fibre growth in sheep and goats – a review. *Livest. Prod. Sci.* 33, 1-29.

Taddeo, H.R., Allain, D., Mueller, J., Rochambeau, Manfredi, E., 1998. Genetic parameter estimates of production traits of Angora goats in Argentina. *Small Rumin. Res*. 28, 217-223.

Van der werf, J.H.J., 2007. Marker assisted selection in sheep and goats. In: Marker-Assisted Selection: Current status and future perspectives in crops, livestock, forestry and fish. Eds. Guimarães, E., Ruane, J., Scherf, B., Sonnino, A. & Dargie, J. FAO, Rome, Italy. pp. 230-247.

Van der Westhuysen, J.M., 2005. Marketing goat fibres. *Small Rumin. Res*. 60, 215-218.

Visscher, P.M., Woolliams, J.A., Smith, D. & Williams, J.L., 2002. Estimation of pedigree errors in the UK dairy population using microsatellite markers and the impact on selection. *J Dairy Sci* 85, 2368-2375.

Visser, C., Hefer, C.A., Van Marle-Köster, E. & Kotze, A., 2004. Genetic variation of three commercial and three indigenous goat populations in South Africa. *SA J Anim Sci.* 34 (Suppl 1), 24-27.

Webb, E.C. & Casey, N.H., 2010. Physiological limits to growth and the related effects on meat quality. *Livest. Sci.* 130, 33-40.

CHAPTER 2

GENETIC VARIATION OF THE REFERENCE POPULATION FOR QTL RESEARCH IN SA ANGORA GOATS

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Genetic variation of the reference population for QTL research in SA Angora goats

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Summary

The South African Angora goat industry makes the largest contribution to global mohair production. Mohair is a luxury fibre and production of a high quality clip is essential. For many years genetic improvement of Angoras in South Africa was based on quantitative selection. Genome mapping efforts provided new avenues for improvement and a QTL study was initiated to identify QTL associated with mohair traits. The aim of this study was to describe the genetic diversity of the reference population using the available stud and commercial herds with full phenotypic records. The most appropriate QTL design was identified based on the population structure with regard to the families and number of bucks available for breeding. Four herds, consisting of 1067 pure bred goats in 12 half-sib families were included. Blood samples were obtained from the herds and 94 markers were tested and diversity parameters estimated. The average number of alleles per marker varied between 5.4 and 7.2 between the herds, while the observed heterozygosity varied between 0.59 and 0.67. The genetic structure of these herds was found appropriate for use as a reference population as they showed sufficient genetic variability.

Keywords: Angora goats, mohair, genetic variation, QTL design

Introduction

South Africa is the major producer of mohair in the world, with a contribution of between 55 and 60 % of the product to the world market (Loots, 2007). It is therefore imperative to maintain a good quality clip through selection for the desired mohair traits, which to a large extent depend on accurate genetic improvement programs. For many years quantitative studies and research were undertaken with regards to mohair traits and production of Angora goats and results have contributed to increased and improved production (Snyman & Olivier, 1996; Snyman, 2002). Despite this progress made with quantitative selection, it has certain limitations, including the fact that selecting on breeding values doesn't account for population effects or genetic diversity and that selection is optimized for a general response in the next generation, rather than the highest long term response (Andersson, 2001). Advances in genomics have provided new opportunities for animal geneticists and breeders where knowledge of the underlying molecular mechanisms of fiber and fleece characteristics should lead to more efficient selection programs on the long term (Purvis & Jeffery, 2007).

Microsatellite markers have been widely applied as a suitable DNA marker for diversity and genome-wide studies in goats (Iamartino *et al.,* 2005), as no SNPs are yet available for this species (Maddox & Cockett, 2007).

QTL studies have been performed in poultry, beef and dairy cattle for some time (Sonstegard *et al.,* 2001; Tuiskuola-Haavisto *et al.,* 2002; Casas, *et al*., 2004; Boichard *et al*., 2006) and the prerequisites include a suitable reference population. It is also required to test for sufficient with-in breed variation of the reference population, as this knowledge is the first step towards responsible exploitation of domestic animal biodiversity (Beuzen *et al*. 2000; Iamartino *et al.,* 2005; Li *et al*., 2008). The necessity of global diversity surveys for further integration into QTL detection studies was also highlighted by Gibson (2003) (http://www.fao.org/biotech/docs/Gibson.pdf).

A QTL identification study was identified in South African Angoras for potential QTL affecting mohair traits. The most appropriate designs for outbred populations with relatively large families are full- or halfsib designs (Weller, 2001). The challenge of these designs lies therein that on the one hand there is a force towards a small number of sire families with large progeny groups, while on the other hand there is the probability that the sires used in the project is not heterozygous (Bovenhuis, 2005). A halfsib design was identified as being the most appropriate for the South African Angora industry.

The aim of this paper was to describe the establishment of the reference population for QTL research in South Africa through the appropriate selection of the stud families and evaluation of the genetic variation within the herds using microsatellite markers.

Material and Methods

Selection of suitable herds

The majority of Angora goats are farmed in the Eastern Cape province of South Africa. This region referred to as the Karoo has a dry climate and a bush type vegetation, suitable for Angora goat production. Angora goat breeders taking part in the National Small Stock Performance Scheme were approached in the selection of the herds for this study. Only families with complete pedigree and phenotypic records were considered for inclusion. The breeders agreed to use at least two of the same bucks over a three year period to generate sufficient offspring for the reference population. Phenotypic recordings were made on growth (birth and weaning weights, ADG) and mohair (fleece weight, fibre diameter, staple length, standard variation of fibre diameter etc.) traits for all goats.

Animal sampling and Genotyping

Blood samples were collected over a three year period from all the animals of the selected herds and the blood is stored in a DNA bank for small stock research (GADI, National Department of Agriculture). A total of 1124 individual blood samples were used in the study from four different Angora stud herds with suitable families, sufficient progeny and required records.

DNA was extracted from whole blood using respectively the Qiagen DNEasy Tissue kit at the University of Pretoria and the Invisorb blood mini HTS kit (Invitek) for the XtractorGene (Corbett Robotics) at Wageningen University and Research Center according to the protocols of the respective manufacturers with a starting volume of 100µl blood for both protocols.

DNA samples were amplified with 94 microsatellite markers as selected for the QTL study. Incorrect parentage due to recording errors and overmating was identified with Cervus 3.0, and all aberrant individuals were removed from the study. Markers were selected on level of polymorphism, heterozygosity, allele size range and amplification success. The markers were divided into eight genotyping sets, averaging twelve markers per set. PCR was performed in a i-cycler (Bio-rad) and Ti Thermocycler (Biometra) using 100ng DNA, 2.94 µl of the ABgene® PCR Master Mix (ABGene, UK) and 0.03 µl each of 40pmol/µl reverse and forward primer. The PCR amplification was conducted in a 6 µl final volume in 384 well PCR plates at the following conditions: 95°C for 5 minutes, followed by 35 cycles of 96°C for 30s, 45s at Annealing Temperature and 90s at 72°C with a final extension step of 10min at 72°C.

Statistical analysis

The statistical power of a half-sib design depends on the number of sires used, offspring per sire, and statistical parameters i.e. the heritability of traits, heterozygosity and magnitude of the QTL, acceptable Type I Error and the marker–QTL recombination fraction. The statistical power was calculated using the 'Power of Daughter design' software by Bovenhuis (2005).

Genetic variability of the families selected for the reference population was analysed using MS toolkit (Park, 2001). Genetic parameters estimated included allelic frequencies, mean number of alleles and heterozygosity values per locus and for each population. The Polymorhpic Information Content (PIC) for each locus, and across loci were estimated using Cervus 3.0 software (Marshall *et al.,* 1998). The FSTAT2.9.3 program (Goudet, 1995) was used to compute Wright's *F*-statistics for each locus, including F, θ and f (analogous to Wright's (1978) F_{IT}, F_{ST} and F_{IS}, respectively). The statistical significance of the obtained values was estimated by bootstrapping using 1000 replications.

Population structure and F_{ST} values was inferred by using the STRUCTURE program (Pritchard *et al.*, 2000), a Bayesian approach based on the genotypes of the individuals collected. Individuals were assigned to *K* (unknown) populations, where *K* is varied across runs of the program, and individuals have membership assigned to them over all the different clusters (number of clusters = *K*). The sum of the probabilities to belong to a population equals one. STRUCTURE was run with 10^6 iterations, and a burn-in period of 10 000 iterations in order to assure a random starting point for the algorithm. The runs were repeated 20 times for $2 > K < 10$, in order to check the consistency of the results. An admixture ancestry model was assumed, which makes provision for the individuals to have a mixed ancestry. This is modeled by assuming that a certain individual (*i*) has inherited some fraction of its genome from ancestors in population *k*.

Results & Discussion

The results obtained from testing for statistical power ("Power of Daughter", Bovenhuis, 2005) of the half-sib design in this study were based on a heritability of 0.32, Type 1 error of 0.05 and Recombination fraction of 0.1. A 12 sire design with approximately 100 offspring per sire was predicted to yield sufficient (0.910) power to detect QTL, and this was identified as the most appropriate experimental design for QTL detection in the South African Angora goat population. The family structure of the four stud herds with full phenotypic and pedigree information selected for the reference population are shown in Table 2.1. These animals were part of 12 half-sib families, ranging between 44 and 140 offspring with an average of 88 half-sib offspring per sire. All possible sires were screened for heterozygosity over loci and sires with the highest heterozygosity values were selected.

A total of 800 alleles from 94 loci were detected in the 1067 individuals genotyped. All markers were found to be polymorphic in each of the four herds evaluated. The number of alleles identified per locus averaged 7.99, with variation from two (BM4630) to 23 (INRA011). The mean PIC value across loci was 0.57, indicating a medium level of information (Table 2.2), corresponding closely to values reported by Kumar *et al.* (2005), Martinez *et al.* (2006) and Traore *et al.* (2009).

The observed and expected heterozygosity values over all loci for all herds averaged 0.63 and 0.62 respectively. Individual markers varied significantly, ranging from as low as 0.14 (CSSM32) to as high as 0.83 (BM1329) for unbiased Heterozygosity. These mean values correspond closely to those reported by both Kumar *et al.* (2005) (H_O = 0.45, H_E = 0.63) and Martinez *et al.* (2006) (H_O = 0.62, H_E = 0.66), although both higher (Qi *et al.,* 2009) and lower (Gour *et al*., 2006) values have previously been reported for various microsatellite panels tested in different goat populations.

Table 2.1 Family structure of herds for the reference population in the study

	Offspring Year 1	Offspring Year 2	Offspring Year 3	Total
Herd 1				
Sire 1	41	33	36	110
Sire 2	18	38	59	115
Sire 3	34	42	8	84
Sire 4	31	46	27	104
Herd 2				
Sire 1	9	99	32	140
Sire 2			84	84
Herd 3				
Sire 1		41	23	64
Sire 2		41	76	117
Sire 3	38	54		92
Sire 4	54	37		91
Herd 4				
Sire 1	27	40		67
Sire 2		37	7	44

The *f*, *F* and θ values estimated for the 96 loci across all populations are indicated in Table 2.2. The mean θ value (0.069, ranging between 0.002 and 0.161) was similar to that found by Gour *et al*. (2006), but was marginally higher compared to other previously reported estimates in goat breeds (Martinez *et al*., 2006; Kumar *et al*., 2005; Dalvit *et al*., 2008). The highest within-population fixation index (*f*) was estimated for BM4630 (0.175), which indicate a heterozygote deficit. Of the 94 markers, 74 showed negative *f* values, indicating no inbreeding but rather outbreeding. Overall, the microsatellite loci included were useful to obtain a reliable assessment of the genetic variability within the population.

Table 2.2 Number of alleles per marker (k), observed (Hobs) and expected (HExp) heterozygosity, polymorphic information content (PIC) and F-statistics per marker

Locus	N samples	k	HObs	HExp	PIC	$F(F_{IT})$	θ (F_{ST})	f (F_{IS})
BM0121	848	9	0.64625	0.6685	0.61475	0.05	0.07	-0.023
BM0321	1101	9	0.54575	0.5175	0.484	-0.03	0.032	-0.063
BM0719	933	6	0.715	0.733	0.69025	0.075	0.06	0.016
BM1225	874	5	0.6175	0.57925	0.519	0.002	0.073	-0.077
BM1258	1098	10	0.71275	0.672	0.61975	0.046	0.1	-0.06
BM1312	524	10	0.5905	0.676	0.625	0.197	0.06	0.145
BM1329	635	7	0.8755	0.82525	0.64175	-0.05	0.036	-0.089
BM143	1018	6	0.70375	0.67075	0.62075	0.033	0.091	-0.064
BM1818	1051	9	0.7565	0.7215	0.67925	0.019	0.073	-0.058
BM2830	931	9	0.6425	0.606	0.52725	-0.038	0.02	-0.059

BM3205	467	9	0.50675	0.576	0.526	0.129	0.034	0.098
BM3517	681	12	0.722	0.723	0.6805	0.057	0.068	-0.011
BM415	919	9	0.842	0.785	0.7515	-0.011	0.058	-0.074
BM4208	874	11	0.8455	0.78825	0.7575	0.019	0.058	-0.041
BM4621	1002	6	0.54	0.51875	0.45975	0.046	0.089	-0.047
BM4630	959	2	0.37025	0.4105	0.32375	0.205	0.037	0.175
BM6526	627	12	0.6555	0.7115	0.6625	0.122	0.062	0.063
BM7160	848	6	0.6975	0.673	0.61425	0.074	0.051	0.024
BM8125	890	8	0.657	0.63	0.58625	-0.006	0.05	0.058
BMC1009	897	8	0.64625	0.64275	0.589	0.058	0.063	-0.005
BMC1222	852	6	0.54875	0.6445	0.5885	0.182	0.112	0.079
BMC8012	872	3	0.526	0.4765	0.36825	-0.076	0.002	0.078
BMS0712	904	9	0.745	0.737	0.6935	0.029	0.042	-0.013
BMS0745	860	10	0.8375	0.73575	0.69775	-0.021	0.078	0.107
BMS1248	869	9	0.2375	0.25125	0.23625	0.082	0.051	0.032
BMS1332	1044	7	0.68575	0.6	0.53075	-0.009	0.012	-0.021
BMS1714	863	5	0.762	0.71975	0.66675	-0.03	0.029	-0.061
BMS1788	919	11	0.73925	0.69	0.644	0.021	0.063	-0.045
BMS2252	920	6	0.62825	0.61425	0.55675	0.001	0.034	-0.035
BMS2526	1085	7	0.756	0.737	0.6895	0.037	0.072	-0.037
BMS2782	781	11	0.7745	0.73	0.68775	-0.011	0.07	-0.087
BP28	855	9	0.62475	0.6995	0.6575	0.221	0.103	0.132
CSRD247	1083	8	0.691	0.64375	0.59275	0.049	0.1	0.057
CSSM19	957	5	0.3155	0.3065	0.27275	-0.008	0.048	-0.059
CSSM32	892	5	0.1415	0.1385	0.132	0.034	0.059	0.026
CSSM43	881	6	0.6175	0.59275	0.5215	-0.035	0.038	-0.075
CSSM47	945	6	0.317	0.29425	0.2745	-0.068	0.014	-0.083
CSSM54	893	12	0.42825	0.54425	0.48975	0.257	0.161	0.115
DRBP1	222	8	0.67575	0.673	0.619	-0.207	0.148	-0.417
HEL11	668	14	0.68775	0.7225	0.682	0.14	0.074	0.071
HUJ614	1108	7	0.55125	0.51275	0.423	-0.048	0.015	-0.063
IL2RA	936	8	0.599	0.5835	0.55	0.052	0.08	-0.031
ILSTS011	1076	7	0.73525	0.6765	0.6315	0.028	0.051	-0.025
ILSTS033	1104	9	0.5895	0.585	0.543	0.083	0.093	-0.012
ILSTS034	898	6	0.60625	0.5785	0.513	0.043	0.045	-0.002
ILSTS045	1094	6	0.633	0.6225	0.5555	0.082	0.116	-0.039
ILSTS058	814	11	0.7325	0.7455	0.7045	0.032	0.107	-0.084
ILSTS059	1111	4	0.50975	0.4965	0.4215	0.083	0.069	0.016
ILSTS087	1070	9	0.524	0.49075	0.46025	0.022	0.079	-0.062
INRA003	919	3	0.574	0.5	0.39475	0.028	0.068	-0.043
INRA005	957	4	0.52125	0.47075	0.37075	-0.027	0.098	-0.139
INRA006	1052	11	0.7745	0.74025	0.698	0.003	0.06	-0.06
INRA011	1097	23	0.74225	0.73125	0.70475	0.038	0.093	-0.061
INRA040	644	8	0.56575	0.5905	0.5525	0.011	0.034	-0.024
INRA063	1082	5	0.6705	0.66775	0.60525	0.032	0.033	-0.002
INRA177	858	9	0.45375	0.4435	0.396	0	0.054	-0.057
INRA206	729	8	0.76	0.7595	0.71875	0.033	0.059	-0.027
INRA210	820	7	0.44875	0.44275	0.38925	0.099	0.103	-0.004
INRABERN192	912	8	0.72525	0.66	0.616	0.025	0.102	-0.086
INRABERN172	1072	6	0.7265	0.6965	0.64925	0.003	0.036	-0.034
LSCV25	877	10	0.738	0.76375	0.728	0.112	0.055	0.06
LSCV36	1098	7	0.62625	0.60725	0.5515	0.001	0.025	-0.024
LSCV46	1114	3	0.284	0.2455	0.22	-0.126	0.009	0.137

LSCV52	1114	7	0.71375	0.68025	0.62225	-0.02	0.025	-0.046
MAF050	894	9	0.74125	0.74325	0.69725	0.02	0.036	-0.016
MAF214	646	12	0.649	0.6745	0.61975	0.19	0.143	0.055
MAF64	1084	7	0.77525	0.75125	0.71125	0.043	0.072	-0.032
MAF70	1083	8	0.70925	0.6875	0.637	0.044	0.083	-0.043
MCM104	1115	6	0.7095	0.659	0.60425	0.003	0.075	
MCM136	1118	3	0.36425	0.358	0.3005	0.157	0.154	
MCM210	788	6	0.57775	0.535	0.4655	0.03	0.078	-051
MCM527	923	5	0.64275	0.63225	0.5685	0.07	0.112	-0047
MCM58	909	17	0.7455	0.73925	0.701	0.036	0.04	-0.004
MCM64	734	9	0.599	0.56025	0.52	0.08	0.116	-0.041
OARAE129	935	4	0.558	0.5635	0.4755	0.093	0.072	0.023
OARCP26	955	7	0.4475	0.525	0.47525	0.16	0.034	0.131
OARCP34	1004	8	0.74925	0.72025	0.67925	0.027	0.073	-0.049
OARCP73	719	15	0.837	0.801	0.7715	0.015	0.057	-0.044
OARFCB005	961	9	0.3515	0.412	0.3795	0.082	0.045	0.039
OARFCB11	891	4	0.34975	0.4195	0.354	0.106	0.035	0.074
OARFCB193	1032	6	0.703	0.66625	0.6205	0.017	0.1	-0.092
OARFCB48	898	8	0.76775	0.70425	0.65525	0.028	0.066	-0.04
OARHH35	728	10	0.77575	0.758	0.7195	0.005	0.031	-0.027
OARHH64	819	6	0.6805	0.68675	0.6285	0.049	0.053	-0.004
OARVH098	949	6	0.69325	0.69575	0.64425	0.098	0.101	-0.003
OLADRB	767	13	0.73825	0.72375	0.67775	0.022	0.051	-0.03
SRCRSP05	1111	8	0.75775	0.74475	0.7075	0.071	0.147	-0.09
SRCRSP08	1089	8	0.64	0.6255	0.57475	-0.005	0.063	0.072
SRCRSP09	1073	9	0.73725	0.65975	0.604	0.02	0.139	-0.139
SRCRSP10	1098	11	0.775	0.727	0.68275	0.009	0.039	-0.031
SRCRSP24	1083	8	0.73475	0.69225	0.65825	0.02	0.064	-0.047
TGLA040	903	6	0.5135	0.55025	0.496	0.12	0.095	0.027
TGLA179	1088	9	0.82975	0.773	0.74075	-0.022	0.046	0.072
TGLA304	977	8	0.6695	0.618	0.55525	0.007	0.067	-0.065
Over all loci		7.989	0.6346356	0.6210346	0.56934574	0.04	0.069	-0.031

Genetic variability in the SA reference population was found to be relatively high with the average number of alleles varying between 5.41 and 7.21 in the four herds. The estimated unbiased H or gene diversity was well above 60%, except for one herd with a value of 56.5%. These levels of heterozygosity for the different herds (Table 2.3) were in the same order as that reported by Martinez *et al.* (2006) for Canary goat populations, but lower compared to values reported by Iamartino *et al.* (2005) for Italian goat populations, Li *et al.* (2008) for Chinese goat breeds and Dalvit *et al.* (2008) for Alpine sheep breeds.

With regards to population subdivision the F_{ST} value (0.182) for herd 2 indicated a reduction of heterozygosity supporting the unbiased H estimation (Hartl, 1988). These levels of H exceeded expectations as the Angora goat population in South Africa is relatively small, and high selection pressure has been applied to the animals over several generations.

Table 2.3 Measures of genetic variation in the population studied

Herd	Sample size	Loci typed	Unbiased Hz ± SD	Obs Hz ± SD	N Alleles	F_{ST}
1	400	94	0.627 ± 0.015	0.637 ± 0.003	6.98	0.0658
2	218	93	0.565 ± 0.018	0.592 ± 0.004	5.41	0.1818
3	338	94	0.633 ± 0.014	0.652 ± 0.003	7.21	0.0659
4	111	93	0.634 ± 0.016	0.671 ± 0.005	6.87	0.0486

The population structure and level of admixture were estimated using Structure. The most likely number of clusters (*K*) was four, as shown in Figure 2.1, and inferred by the LnPr(X/K) value. The variability of this value across runs for a given *K* gives a good indication of the most likely number of clusters for the population. The smallest *K* with the least variability is often the one that bests explains the data (Pritchard, 2000; Sollero *et al.,* 2009), as was the case when *K*=4.

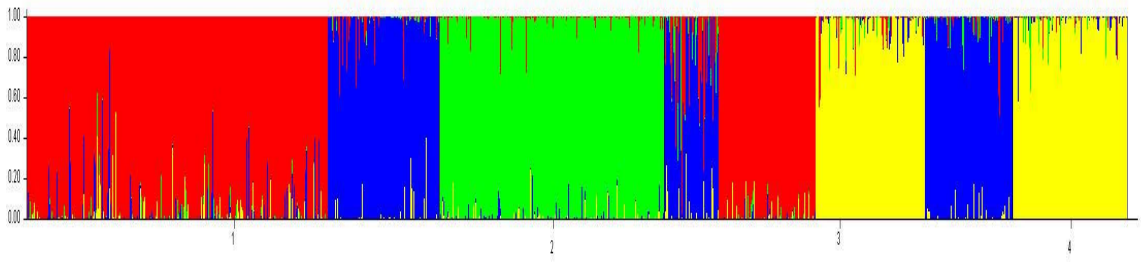

Figure 2.1 Summary plot of estimates of Q. Each individual is represented by a single vertical linebroken into K coloured segments, with lengths proportional to each of the 4 inferred clusters. The numbers (1-4) correspond to the herds

Table 2.4 shows the proportion of individuals of each of the herds in the four most likely clusters inferred by Structure and this corresponded to the four different herds included in the study. Herd 1 were mostly divided between cluster 1 (69%) and 3 (28%). A total of 97% of herd 2 were assigned together in cluster 2, while 96% of the population of herd 4 belonged to cluster 4. Animals in Herd 3 were almost equally divided between cluster 1 (31%), 3 (36%) and 4 (31%). The considerable gene flow between herds 1 and 3 (as well as their almost identical F_{ST} values) are most likely due to interchanging bucks during successive mating seasons, resulting in a lack of divergence due to the recent common ancestors of offspring. In contrast to this, herd 2 forms an individual cluster with a high F_{ST} value, which is probably due to the breeder buying in new bucks on an annual or bi-annual basis. The source(s) of this new genetic material is likely not included in this study.

The genetic structure of these herds were found appropriate for use as reference populations as the genetic diversity is sufficient and the herds showed a level of differentiation. The levels of genetic diversity compared favourably with genetic diversity studies performed previously on various goat populations, indicating that there is a possibility to exploit natural variation on molecular level within the population for improved production.

Table 2.4 Proportion of membership of the analysed goat herds in each of the four clusters inferred in Structure

	Inferred clusters				
Herd	1	2	3	4	N
1	**0.691**	0.008	0.281	0.020	400
2	0.011	**0.969**	0.010	0.010	218
3	0.310	0.020	**0.360**	0.311	338
4	0.007	0.018	0.013	**0.962**	111

Current selection on the Angora goat breed in South Africa aims to establish a balance between production and survival traits as there is a limit to the harshness of environment in which animals can produce viable amounts of mohair, and a limit to the quality of mohair that such an adapted animal will be able to produce. Although the focus of the current project is on mohair production, all recorded traits (including body weights and efficiency parameters) will be included in future research programs. South Africa needs to develop a competitive, sustainable fast-growing economy and therefore need to apply modern technology available. This study was the first attempt to explore the genetic variation available within the Angora goat reference population.

Conclusion

This study confirmed that there is sufficient genetic diversity within the South African Angora goat reference population to utilise molecular research in the genetic improvement of the breed. The establishment of this reference population forms part of a comprehensive, integrated approach where both quantitative and molecular tools are applied for genetic improvement of South African Angora goats. An in-depth knowledge of the genetic diversity of the analyzed populations will help to structure future molecular studies on this newly established reference population.

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References

Andersson, L. 2001. Genetic dissection of phenotypic diversity in farm animals. *Nat. Rev. Genet.* 2 (2001), pp. 130–138.

Beuzen, N.D., Stear, M.J. & Chang, K.C. 2000. Molecular markers and their use in animal breeding. *Veterinary Journal* 160, 45-52.

Boichard, D. Fritz, S., Rossignol, M.N., Guillaume, F., Colleau, J.J. & Druet, T. 2006. Implementation of Marker-Assisted Selection: Practical lessons from dairy cattle. 8th World Congress on Genetics Applied to Livestock Production, August 13-18, 2006. Belo Horizonte, MG, Brazil

Bovenhuis,H. 2005. "Power of a Daughter Design" statistical program. Animal Breeding and Genetics Group, Wageningen UR, Netherlands.

Casas, E., Lunstra, D.D & Stone, R.T. 2004. Quantitative trait loci for male reproductive traits in beef cattle. *Anim. Gen.* 35, 451-453

Dalvit, C., Sacca, E., Cassandro, M., Gervaso, M., Pastore, E. & Piasentier, E. 2008. Genetic diversity and variability in Alpine sheep breeds. *Small Rumin Res.* 80, 45-51.

Goudet, J. 1995. FSTAT (version2.9.3): a computer programme to calculate F-statistics. *J.Hered.* 8, 485-486.

Gour, D.S., Malik, G., Ahlawat, S.P.S., Pandey, A.K., Sharma, R., Gupta, N., Gupta, S.C., Bisen, P.S & Kumar, D. 2006. Analysis of genetic structure of Jamunapari goats by microsatellite markers. *Small Rumin Res.* 66, 140-149.

Hartl, D.L. 1988. A primer of population genetics, 2nd Ed. Sinauer Associates, Inc. Publishers, Sunderland, Massachusetts, USA. P 78.

Iamartino, D., Bruzzone, A., Lanza, A., Blasi, M. & Pilla, F. 2005. Genetic diversity of Southern Italian goat populations assessed by microsatellite markers. *Small Rumin Res.* 57, 249-255.

Kumar, D., Dixit, S.P., Sharma, R., Pandey, A.K., Sirohi, G., Patel, A.K., Aggarwal, R.A.K., Verma, N.K., Gour, D.S. & Ahlawat, S.P.S. 2005. Population structure, genetic variation and management of Marwari goats. *Small Rumin Res.* 59, 41-48.

Li, J.Y., Chen, H., Lan, X.Y., Kong, X.J. & Min, L.J. 2008. Genetic diversity of five Chinese goat breeds assessed by microsatellite markers. *Czech J. Anim. Sci.* 53, 315-319.

Loots, F. 2007. Seisoenoorsig. The Angora goat and Mohair Journal P O Box 50, Jansenville, 6265 l South Africa.

Maddox, J.F. & Cockett, N.E. 2007. An update on sheep and goat linkage maps and other genomic resources. *Small Rumin Res.* 70(1), 4-20.

Marshall, T.C., Slate, J., Kruuk, L.E.B. & Pemberton, J.M. 1998. Statistical confidence for likelihood-based paternity inference in natural populations. *Molecular Ecology* 7: 639-655.

Martinez, A.M., Acosta, J., Vega-Pla, J.L. & Delgado, J.V. 2006. Analysis of the genetic structure of the canary goat populations using microsatellites. *Livest Sci.* 102, 140-145.

Park, S.D.E. 2001. Trypanotolerance in West African cattle and the population genetic effects of Selection. Phd thesis. University of Dublin.

Pritchard, J.K., Stephens, M. & Donnely, P. 2000. Inference of population structure using multilocus genotype data. *Genetics* 155, 945-959.

Purvis, I.W. & Jeffery, N. 2007. Genetics of fibre production in sheep and goats. *Small Rumin. Res.* (70) 42-47.

Qi, Y., Luo, J., Han, X., Zhu, Y., Chen, C., Liu, J. & Sheng, H. 2009. Genetic diversity and relationships of 10 Chinese goat breeds in the Middle and Western China. *Small Rumin Res.* 82, 88-93.

Snyman, M.A. 2002. Evaluation of a genetically fine mohair producing herd. *Small Rumin. Res.* (43) 105-113.

Snyman, M.A. & Olivier, J.J. 1996. Genetic parameters for body weight, fleece weight and fibre diameter in South African Angora goats. *Livest. Prod. Sci.* (47) 1-6.

Sollero, B.P., Paiva, S.R., Faria, D.A., Guimaraes, S.E.F., Castro, S.T.R., Egito, A.A., Albuquerque, M.S.M., Piovezan, U., Bertani, G.R. & Mariante, A. da S. 2009. Genetic diversity of Brazilian pig breeds evidenced by microsatellite markers. *Livest Sci.* 123, 8-15.

Sonstegard, T.S., van Tassel, C.P. & Ashwell. 2001. Dairy cattle genomics: Tools to accelerate genetic improvement? *J. Anim. Sci.* 79(E.Suppl) E307-E315

Traore, A., Alvarez, I., Tamboura, H.H., Fernandez, I., Kabore, A., Royo, L.J., Gutierrez, J.P., Sangare, M., Ouedraogo-Sanou, G., Toguyeni, A., Sawadogo, L. & Goyache, F. 2009. Genetic characterization of Burkino Faso goats using microsatellite polymorphism. *Livest Sci.* 123, 322-328.

Tuiskula-Haavisto, M., Honkatukia, M., Vilkki, J., De Koning, D.J., Schulman, N.F. & Mäki-Tanila, A. 2002. Mapping of Quantitative Trait Loci Affecting Quality and production traits in Egg Layers. *Poultry Sci.*81: 919-927

Weller, J.I. 2001. Quantitative Trait Loci Analysis in Animals. CABI Publishing, UK.

Wright, S. 1978. Evolution and the genetics of populations. Variability within and among natural populations, Vol 4. University of Chicago Press, Chicago.

CHAPTER 3

A GENETIC LINKAGE MAP FOR THE SOUTH AFRICAN ANGORA GOAT

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A genetic linkage map for the South African Angora goat

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Abstract

Despite their economical importance, relatively few molecular studies have been made on goats compared to other livestock species. The most recent goat map was published in 1998, and lacks complete genome coverage. A large number of discrepancies and especially inter-chromosomal re-assignments were reported between the 1998 goat linkage map and the sheep map. In this study 94 microsatellite markers were amplified in 12 half-sib South African Angora goat families for compilation of a genetic map, aiming to confirm or reject previously reported rearrangements and to improve the alignment between the ovine and caprine maps. The number of informative meiosis per marker ranged from 69 to 836, with an average of 518. The microsatellites were mapped to 23 chromosomes, spanning 1352cM and resulting in an average marker interval of 23.0cM. Marker orders were compared to the previously published goat maps, as well as to the ovine map. Six chromosomes (CHI 2, 4, 5, 11, 13 and 19) showed rearrangements in marker order compared to the 1998 Schibler *et al.* goat map, while nine previously unmapped markers were conclusively assigned to eight chromosomes. Four of the previously reported intra-chromosomal rearrangements between the goat and sheep maps were confirmed to be either population specific or mapping errors. The verification of rearrangements in loci order will lead to improved alignment between the two maps, as well as improved efficiency of genome and fine mapping efforts in goats.

Keywords: Linkage map, Angora goats, microsatellite markers

Introduction

Sheep and goats were the first domestic animals to be used for food production and contribute significantly to the agricultural sectors of small stock producing countries, including South Africa (Notter *et al.*, 2007). The world goat population has grown by 66% from 1985 to 2005, while the sheep population decreased and the cattle population showed only a 9% growth (Dubeuf & Boyazoglu, 2009). Both goat milk and goat meat production has increased significantly during this period. The growth in the goat population was mostly due to an increase in the goat numbers in the developing countries in Asia and Africa.

The Angora goat is the major contributor to goat production in South Africa. Since the early nineteen nineties South Africa is the major producer of quality mohair in the world, with a market share of 55 % of the world market (Loots, 2007). Therefore the Angora goat farmers in South Africa play a crucial role in enhancing the constant availability of quality natural fibers worldwide. The natural animal fiber market, including mohair, is also facing competition from synthetic fibers. As mohair is a specialist fiber with a niche market, producers will always face challenges to remain productive and competitive. Therefore, the South African producers have to adapt to demands for finer fiber, decreasing profit margins, a challenging production environment and changed land-use patterns.

Despite the important contribution of goats, the molecular tools developed for goats during the past decade are relatively limited compared to cattle and sheep (Maddox, 2005). Physical and genetic linkage maps have been developed for most species, and together with other physical resources like libraries (BAC and cDNA), GenBank sequences, microarrays and proteomics have found direct application in direct selection on genetic differences underlying phenotypic variation (Sonstegard *et al.,* 2001; Vignal *et al*., 2002; Baumung *et al.*, 2004). Moreover, the bovine genome has been sequenced and hundreds of thousands of single nucleotide polymorphisms (SNPs) have been detected (Allan & Smith, 2008). Although the ovine genome sequence is lagging behind thousands of SNPs have also been identified (Maddox & Cockett, 2007). The bovine whole-genome radiation hybrid and linkage maps are created and updated at regular intervals (Prasad *et al*., 2007; Faraut *et al*., 2009), however the most recent version of the goat linkage map was published in 1998 and lacks complete genome coverage (Maddox & Cockett, 2007).

Genetic linkage maps are essential for identifying specific loci for quantitative traits. The current caprine maps available are however limited to the first low-resolution genetic map for the male goat genome constructed by Vaiman *et al.* (1996), followed by an improved male goat map by Schibler *et al.* (1998). The mapping of traits of economic importance in goats has mainly focussed on milk proteins (Moioli *et al.,* 2007; Cosenza *et al*., 2008), the polled intersex syndrome mapped on chromosome 1 (Vaiman *et al.,* 1997) and growth hormones (Gupta *et al.,* 2009). This resulted in specific regions (i.e. the areas flanking the PISRT1 locus) with microsatellite marker saturation, while other regions were neglected. Although several QTL studies have been performed on small-stock species and partial linkage maps were created as by-products of these studies, these maps are mostly not publicly available (Maddox & Cockett, 2007).

At this stage the most common type of marker used in goat linkage map studies is microsatellite markers. The present goat map contains 307 loci, mainly microsatellite markers, with the majority of bovine origin. Although microsatellite markers are still useful, SNP markers, if

available, are the best choice in molecular studies, mainly due to their high abundance and increased automation coupled with low cost (Toro *et al.*, 2009). No mapped SNP markers have been reported for goats (Maddox & Cockett, 2007) and only 1.2% of bovine SNPs were proved to be polymorphic in goats (Maddox & Cockett, 2007), and is thus at present of little use in goat studies. SNP discovery projects for caprine SNPs are required and are currently underway.

There are a relatively large number of discrepancies between the goat map and both the sheep and cattle maps (Maddox, 2005), including many rearrangements of locus orders between the different maps. It is not certain which of these discrepancies are mapping mistakes, and which might be genuine inversions between species. The use of small populations and small numbers of microsatellite markers in conjunction with less error checking for the goat map, has lead to a less robust map with a need for further genetic map development.

This study reports a sex averaged genetic linkage map of the South African Angora goat containing 94 microsatellite markers. The markers were mapped on a large three generation half sib population and will be used to perform a QTL study.

Materials and Methods

Family structure:

The genetic linkage map was constructed by genotyping three generations of Angora goat half-sib offspring, belonging to 12 different families. The families ranged from 44 to 140 offspring, with an average of 88 half-sib offspring per sire, as described in Table 3.1. All the herds were kept outdoors and farmed under extensive systems. Farmers made use of group mating during the breeding season, while some practice over-mating at the end of the mating season.

Whole blood samples (5ml) were collected at the stud farms, refrigerated and transported to the University of Pretoria. DNA was extracted from 100µl whole blood using respectively the Qiagen DNEasy Tissue kit at the University of Pretoria and the Invisorb blood mini HTS kit (Invitek) for the XtractorGene (Corbett Robotics) at Wageningen University according to the protocols of the respective manufacturers. Both blood and DNA samples are stored at the Grootfontein Agricultural Development Institute's BioBank, South Africa.

Table 3.1 Family structure of herds included in the study

	Offspring Year 1	Offspring Year 2	Offspring Year 3	Total
Herd 1				
Sire 1	27	40		67
Sire 2		37	7	44
Herd 2				
Sire 1	41	33	36	110
Sire 2	18	38	59	115
Sire 3	34	42	8	84
Sire 4	31	46	27	104
Herd 3				
Sire 1	9	99	32	140
Sire 2			84	84
Herd 4				
Sire 1		41	23	64
Sire 2		41	76	117
Sire 3	38	54		92
Sire 4	54	37		91

Microsatellite markers and genotyping:

One hundred thirty four microsatellite markers were selected from the existing goat map database (http://locus.jouy.inra.fr/cgi-bin/lgbc/mapping/common/intro2.pl?BASE=goat) and previously published literature to obtain sufficient genome coverage. Of these markers, 9 were unassigned by Schibler *et al.* (1998). For some chromosomes certain markers were included, even if they were the only marker on the chromosome, as this linkage map study was the first phase of a QTL project. Coverage of at least 3 markers per chromosome was aimed for, but this was not always feasible due poor amplification, informativeness of some markers, as well as financial constraints. Markers were first screened for polymorphicity and amplification success in the 19 potential Angora sires. Finally, the 12 sires (with offspring) that expressed the highest levels of heterozygosity across all loci tested were selected for inclusion in the study. PCR was performed in a i-cycler (Bio-rad) and T1 Thermocycler (Biometra) using 30ng DNA, 2.94 µl of the ABgene® PCR Master Mix (ABGene, UK) and 0.03 µl each of 40 pmol/µl reverse and forward primer. The PCR amplification was conducted in a 6 µl final volume in 384 well PCR plates at the following conditions: 95°C for 5 minutes, followed by 35 cycles of 96°C for 30s, 45s at Annealing Temperature and 90s at 72°C with a final extension step of 10min at 72°C.

Linkage analysis:

Due to misidentification and recording errors that occur during mating under extensive production systems, the data was analysed using Cervus 3.0 (Marshall *et al.*, 1998) and all aberrant individuals were removed from the study. Linkage analysis was performed with CRI-MAP 2.4 (Green *et al*., 1989) compiled for Windows XP. A LOD threshold of 6 was chosen to detect significant linkage, and lowered in increments (up to 1) until all markers could be assigned. Linkage groups were further analysed using the "build" option. Markers that could not be placed accurately with this option, were then included in alternative "fixed" orders. The final order (which resulted in the shortest map length) was verified with the "flipsn" option.

Results

From the 134 microsatellites initially selected, 94 markers were used in genotyping the whole population. The panel of 94 microsatellites included have a bovine (65), ovine (24) and caprine (5) origin. Of these, 90 markers were mapped to 23 chromosomes covering 1352cM over and resulting in an average marker interval of 23.0cM. The remaining four markers were singletons and mapped to chromosomes CHI9, CHI21, CHI22 and CHI29. No markers could be selected or were associated with CHI10 and CHI15. Four markers (LSCV46, HEL11, TGLA040 and INRABERN172) were mapped to CHI26 and two markers to CHI28 (BMS1714 and ILSTS087) but due to poor informativeness, no orders could be estimated.

The number of alleles, PIC and number of informative meiosis for each marker is indicated in Table 3.2. The average number of alleles per marker was 8.6, whereas only 8 markers had less than 5 alleles. The average PIC value across all loci was 0.57. Twelve markers had a PIC value below 0.4. The average number of informative meiosis per marker was 518, ranging between 69 (DRBP1) and 836 (SRCRSP10).

Table 3.2 Description of microsatellite markers used in linkage groups

Microsatellite	Chromosome assignment	No of alleles	PIC	No of informative meiosis	TA
BM1312	1	11	0.648	374	56
BM3205	1	9	0.513	266	55
CSSM19	1	5	0.267	270	54
CSSM32	1	6	0.142	88	54
MAF64	1	7	0.716	676	58
INRA011	1	27	0.711	824	55
BMS2782	2	12	0.674	543	54
OarFCB11	2	5	0.322	184	56
INRA040	2	8	0.525	260	55
SRCRSP24	2	9	0.673	690	58
CSSM 54	3	13	0.513	460	54

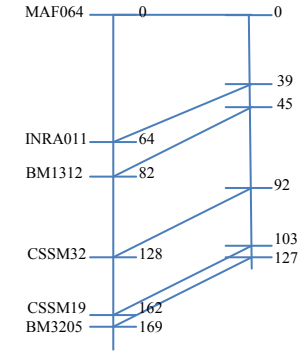
MCM 58	3	20	0.698	696	54
INRA003	3	3	0.392	207	56
INRA006	3	11	0.703	820	55
OarHH64	4	6	0.633	385	56
OarCP26	4	8	0.485	519	56
MAF 050	4	9	0.687	739	60
OarHH 35	4	10	0.723	493	50
BMS1788	4	12	0.655	673	55
MAF70	4	8	0.645	721	58
OARFCB005	5	9	0.41	263	56
LSCV25	5	11	0.726	637	56
BMS1248	5	9	0.241	180	54
BM2830	5	9	0.517	539	56
BMC 1009	5	9	0.593	361	54
ILSTS034	5	7	0.542	548	55
BM0321	5	12	0.485	491	58
BM143	6	6	0.64	685	56
BM415	6	10	0.769	735	50
BM1329	6	7	0.713	450	54
BM4621	6	6	0.449	464	56
ILSTS087	6	9	0.484	518	58
SRCRSP08	6	8	0.578	547	58
INRABERN192	7	7	0.606	540	56
OarAE129	7	4	0.509	370	54
MCM527	7	5	0.571	565	56
CSSM47	8	7	0.306	221	56
MCM64	8	9	0.526	411	58
SRCRSP10	8	11	0.678	836	55
BM4208	9	12	0.776	*	54
INRA177	11	9	0.398	308	56
OarCP34	11	10	0.668	682	56
ILSTS045	11	5	0.549	524	55
BMS0712	12	9	0.689	675	56
INRA005	12	4	0.358	302	54
BMS2252	12	7	0.561	573	55
SRCRSP09	12	11	0.613	443	58
ILSTS033	12	10	0.562	575	55
IL2RA	13	9	0.543	504	50
BMC1222	13	6	0.585	529	56
ILSTS059	13	4	0.445	468	55
BM4630	14	2	0.312	326	54
ILSTS011	14	7	0.625	684	58
BM0719	16	7	0.682	705	56
BM0121	16	9	0.609	522	56
HUJ614	16	7	0.413	451	55
OarVH098	17	6	0.647	610	56
OarFCB48	17	8	0.69	658	60
ILSTS058	17	12	0.722	682	56
BM8125	17	8	0.555	515	55
INRA210	18	9	0.403	264	54
INRA063	18	5	0.61	776	55
MCM104	18	7	0.603	774	55
McM210	19	6	0.478	305	54

BMS0745	19	12	0.692	608	55
LSCV36	19	8	0.564	478	55
OARFCB193	19	6	0.641	610	55
MAF214	20	17	0.618	406	54
TGLA304	20	8	0.533	506	56
BM1225	20	6	0.513	398	55
BM3517	20	15	0.671	441	55
SRCRSP05	21	9	0.714	*	58
BM7160	22	7	0.597	*	55
OarCP73	23	17	0.775	583	56
OLA-DRB	23	15	0.721	575	60
BM1818	23	9	0.683	674	55
DRBP1	23	7	0.648	69	55
BM1258	23	10	0.643	640	55
MCM136	24	3	0.348	450	55
BMS1332	24	6	0.524	399	55
BMS2526	24	7	0.688	775	55
BP28	25	10	0.625	581	50
INRA206	25	9	0.747	572	56
TGLA040	25	7	0.472	464	56
HEL11	26	14	0.687	518	56
INRABERN172	26	8	0.657	823	55
LSCV52	26	7	0.648	821	55
LSCV46	26	3	0.205	199	55
BM6526	27	16	0.686	455	56
CSSM43	27	8	0.5	442	56
TGLA179	27	9	0.757	828	55
BMS1714	28	5	0.679	576	55
ILSTS087	28	9	0.484	518	58
BMC8012	29	3	0.368	*	55

* Singletons

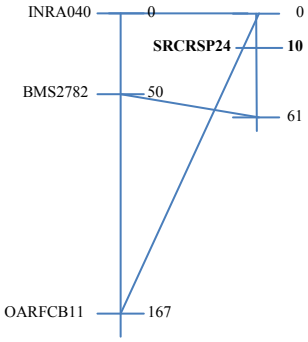
Linkage groups were created using the two-point option with a LOD threshold of 6. A total of 75 markers showed significant linkage (LOD > 6) to at least one other marker. Seven more markers could be linked when the LOD threshold was decreased to 3. The remaining markers formed part of the linkage groups with a LOD >1, and were verified to belong to the same linkage groups as previously reported by Schibler *et al.* (1998). The linkage groups covered 23 autosomes spanning 1326cM, and ranged in size from 26cM to 129cM. Linkage groups generally exceeded 40cM, except on chromosomes CHI7 (27cM), CHI8 (26cM), CHI13 (31cM), CHI18 (26cM), CHI25 (26cM) and CHI27 (30cM). The intervals between consecutive markers within linkage groups ranged between 2 and 51cM. The final linkage map generated in this study is presented in Figure 3.1.

CHI1



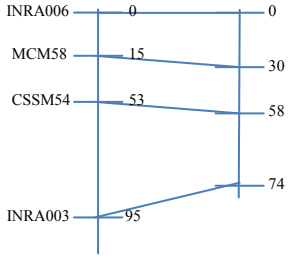
Schibler map 186.0 cM
Current study 127cM

CHI2



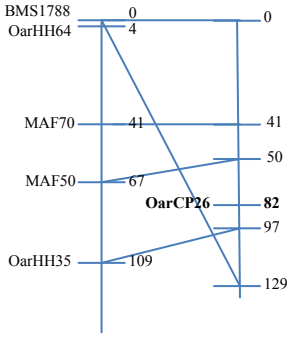
Schibler map 180 cM
Current study 61.2 cM

CHI3



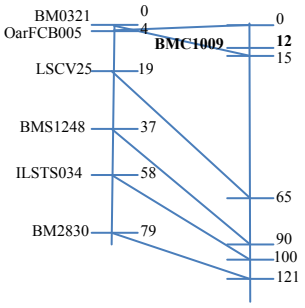
Schibler map 102 cM
Current study 74 cM

CHI4



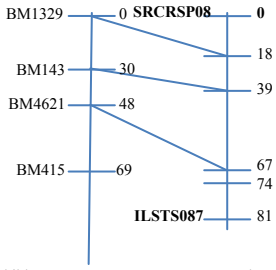
Schibler map 140 cM
Current study 129cM

CHI5



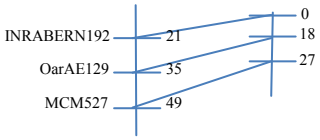
Schibler map 79 cM
Current study 121 cM

CHI6



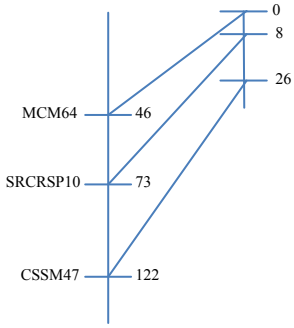
Schibler map 94 cM
Current study 81 cM

CHI7



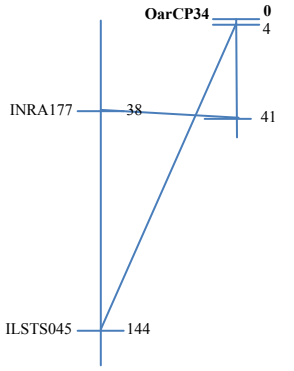
Schibler map 52 cM
Current study 27 cM

CHI8



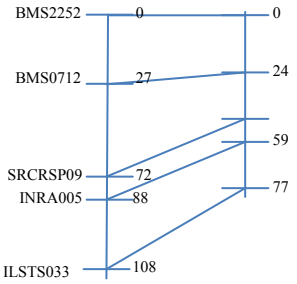
Schibler map 137 cM
Current study 26 cM

CHI11



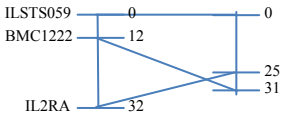
Schibler map 156 cM
Current study 41 cM

CH112



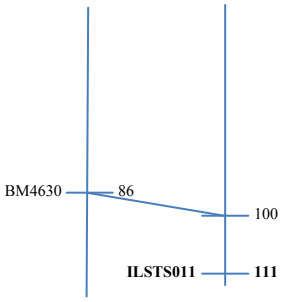
Schibler map 108 cM
Current study 77 cM

CHI13



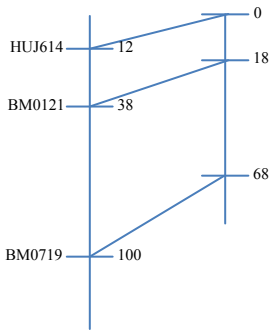
Schibler map 32 cM
Current study 31 cM

CHI14



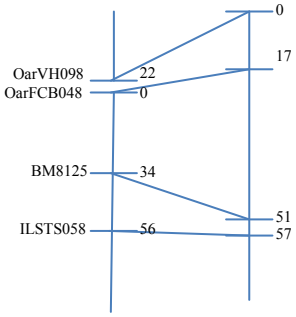
Schibler map 119 cM
Current study 111 cM

CHI16



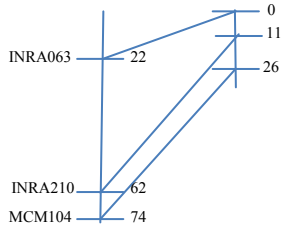
Schibler map 128cM
Current study 68 cM

CHI17



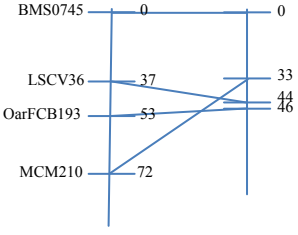
Schibler map 22 + 76 cM
Current study 57 cM

CHI18



Schibler map 74cM
Current study 26 cM

CHI19



Schibler map 97cM
Current study 55 cM

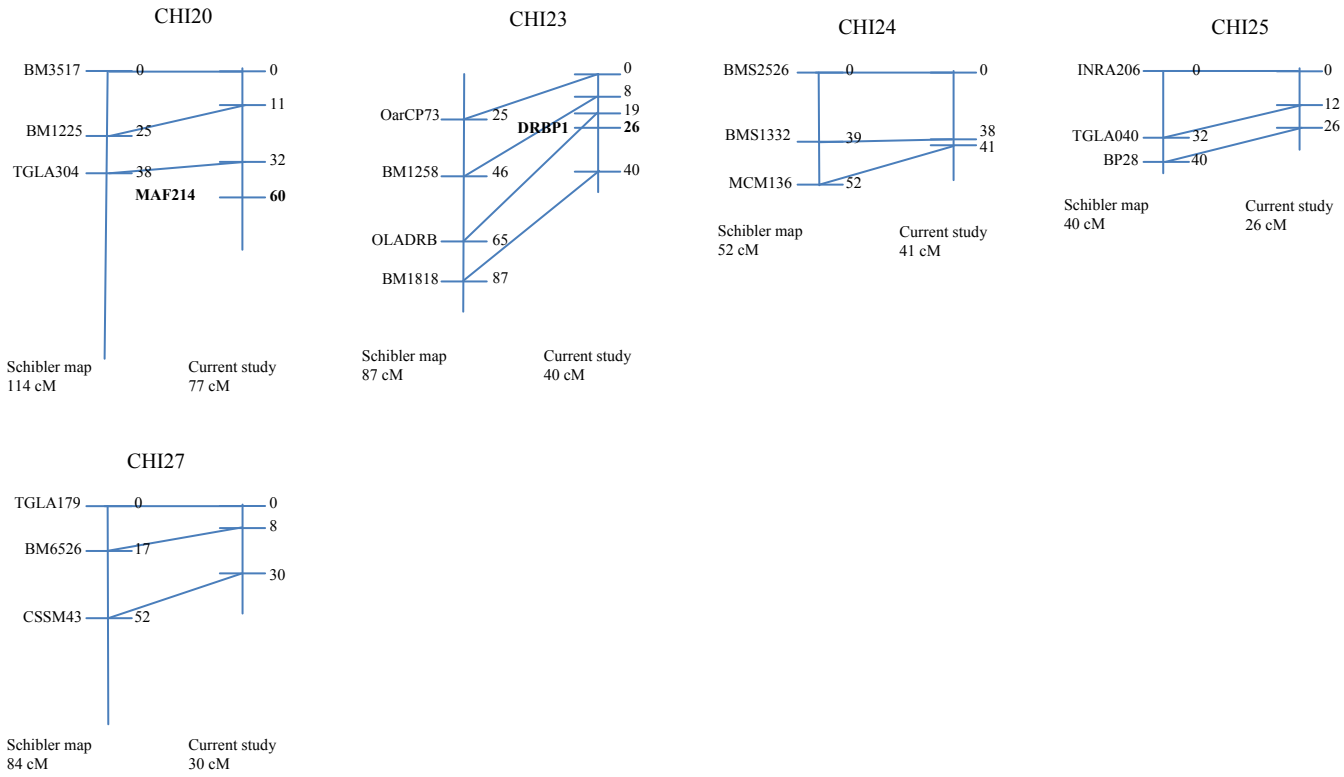


Figure 3.1 Alignment of the marker order of the current study with the most recent goat linkage map (Schibler *et al.,* 1998). Marker names and positions are only given for microsatellites that have been mapped in this study.

Several chromosomes showed marker order differences when compared to the previously published map by Schibler *et al.* (1998). Some of these markers have either not previously been conclusively assigned to specific chromosomes (e.g. SRCRSP08, OarCP26, OarCP34 and DRBP1), or to specific orders (e.g. SRCRSP24, BMC1009, ILSTS087, ILSTS011 and MAF214). Inversions to the Schibler *et al*. (1998) map were found on CHI2, CHI4, CHI5, CHI11, CHI13 and CHI19, and matrices indicating the recombination fractions and LOD scores between the markers on these chromosomes are presented in Table 3.3.

These re-assignments were then compared with the Vaiman *et al*. (1996) goat map and the ovine map, both (Maddox *et al.,* 2001) and the updated SheepMap 4.7 (http://www.ncbi.nlm.nih.gov/mapview/static/sheepsearch.html).

Table 3.3 Matrices indicating linkage groups per chromosome for chromosomes with discrepancies compared to Schibler *et al.* (1998). Recombination fraction above and LOD score below the diagonal

CHI2	SRCRSP24	OARFCB11	BMS2782	INRA040
SRCRSP24		0.49	0.10	0.25
OARFCB11	NS		0.45	0.00
BMS2782	32.39	NS		0.42
INRA040	1.01	2.11	0.07	

CHI4	BMS1788	MAF70	MAF50	OARCP26	OARHH35	OARHH64
BMS1788		0.36	0.36	0.45	0.47	0.37
MAF70	3.24		0.08	0.34	0.37	0.43
MAF50	4.08	58.88		0.28	0.36	0.43
OARCP26	0.07	3.66	9.52		0.14	0.37
OARHH35	NS	1.58	2.16	21.45		0.27
OARHH64	0.53	0.15	0.16	0.42	2.83	

CHI5	BM0321	OARFCB005	LSCV25	BMS1248	BMC1009	BM2830	ILSTS034
BM0321		0.14	0.37	NS	0.03	0.37	NS
OARFCB005	16.56			0.27	0.22	0.39	0.46
LSCV25	1.26	NS		0.16	0.40	0.39	0.31
BMS1248	NS	1.34	11.88		NS	0.17	0.08
BMC1009	36.36	2.96	0.73	NS		NS	0.43
BM2830	0.75	0.08	1.07	8.49	NS		0.19
ILSTS034	NS	0.03	5.98	14.75	0.06	19.31	

CHI11	INRA177	OARCP34	ILSTS045
INRA177		0.37	0.31
OARCP34	0.64		0.04
ILSTS045	1.70	67.12	

CHI13	ILSTS059	IL2RA	BMC1222
ILSTS059		0.27	0.23
IL2RA	4.37		0.06
BMC1222	8.97	50.66	

CHI19	BMS0745	MCM210	LSCV36	OARFCB193
BMS0745		0.26	0.33	0.32
MCM210	4.42		0.11	0.09
LSCV36	5.07	13.08		0.02
OARFCB193	4.65	18.27	70.52	

Discussion

The use of half-sib paternal families for mapping projects is popular because of the relative ease with which sufficient data can be generated. The disadvantage of not having maternal linkage information is a reduced efficiency of the mapping project (Crawford *et al.*, 1994). Despite this limitation, it is still the most popular experimental design in livestock and half-sib population structures of a reasonable size can be generated. The previously published goat maps (Vaiman *et al.,* 1996; Schibler *et al*., 1998) made use of this design, as did the current study. Although marker phase

is difficult to infer in natural populations, it was shown through simulation studies by Slate (2008) that linkage maps constructed from natural populations are reasonable robust and accurate.

The marker order found in this study corresponded with that of the most recent goat map (Schibler *et al.,* 1998) for fifteen of the chromosomes, although shorter map lengths are generally reported in this study. The marker order on chromosomes CHI2, CHI4, CHI5, CHI11, CHI13 and CHI19 however varied from those reported by Schibler *et al*. (1998). The relative marker distances between the studies also differed, which was expected as chromosome lengths is a property of the population under study (Slate, 2008) and incorrect mapping usually result in inflated maps. The average number of informative meiosis (518±179) in our study was much higher than the 114±70 reported by Schibler *et al.* (1998), resulting in a more accurate linkage map. Furthermore, both Vaiman *et al.* (1996) and Schibler *et al.* (1998) reported results without taking Kosambi's correction into account, while the current study made use of Kosambi's mapping function. This function assumes a moderate amount of positive interference between adjacent regions (Weller, 2001) and generally results in shorter map lengths. Many published linkage maps use the function as the recombination data recorded over the past decades mostly corresponds to Kosambi's function (Huehn, 2010).

Despite the overall good agreement of the caprine and ovine maps, a large number of discrepancies have been reported between them (Maddox, 2005), including inter-chromosomal re-assignments. Many of these discrepancies are thought to be artefacts, due to the relatively low robustness, small number of markers and little error checking of the much older goat maps (Maddox, 2005). In the case of any inversions between the current study and the Schibler *et al.* (1998) map, further comparisons were made to the ovine map (SheepMap 4.7: http://www.ncbi.nlm.nih.gov/mapview/static/sheepsearch.html) and / or to the earlier goat map (Vaiman *et al*., 1996). The relative marker orders for the chromosomes with potential rearrangements are shown in Figure 3.2. Vaiman *et al.* (1996) didn't report map positions, but only intervals between markers, making it difficult to compare locations. Many of the chromosomes are also represented in more than one fragment. For this reason, it was not included in the graphical representation of Figure 3.2. Rearrangements are reported in Table 3.3, with their respective pair-wise recombination fractions and LOD scores. Not all markers within the specific linkage groups for CHI2, CHI4 and CHI5 showed linkage to each other. In these cases both markers could however be linked to a common third marker.

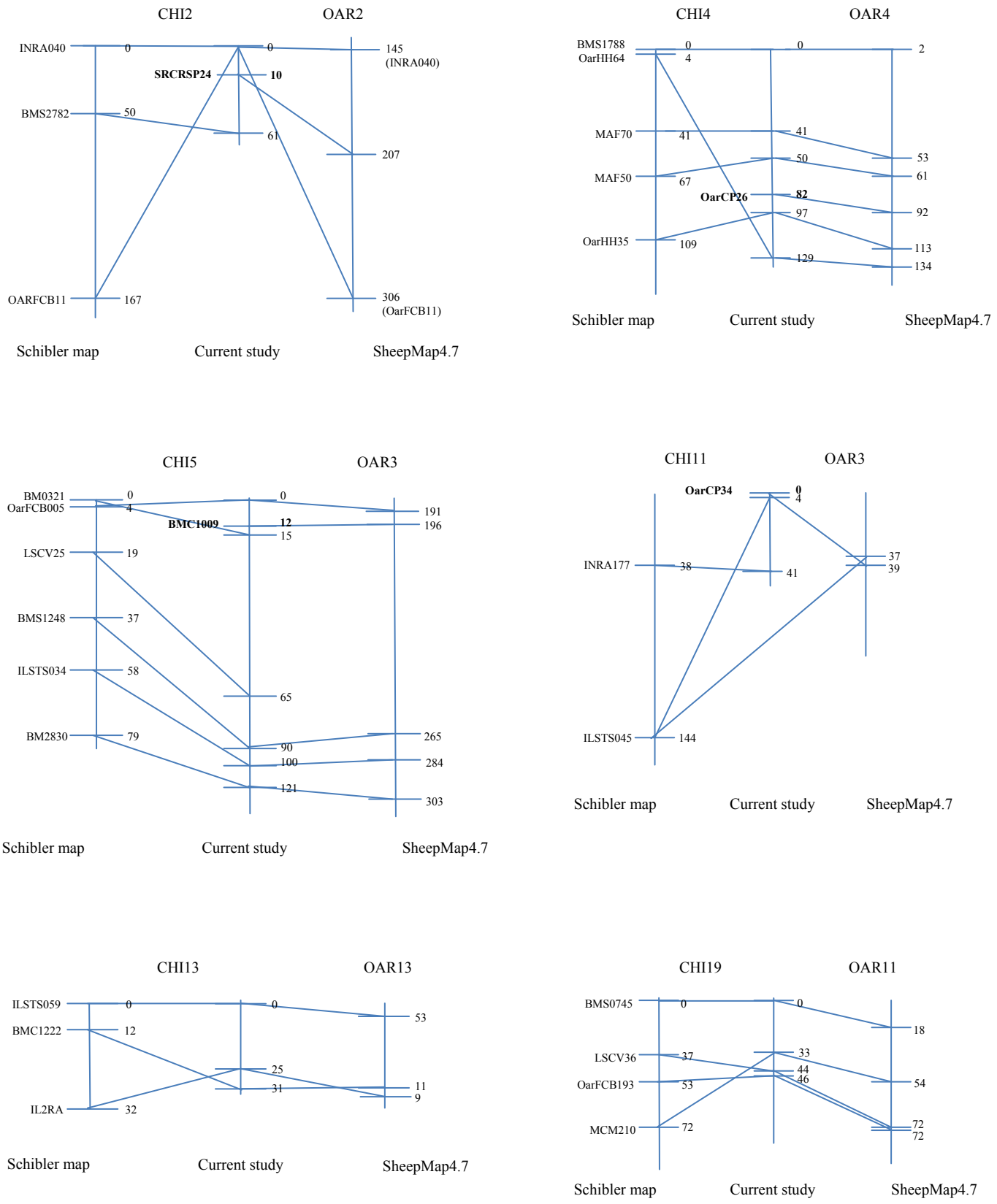


Figure 3.2: Alignment of marker orders on CHI 2, 4, 5, 11, 13 and 19 with orders compared between the current study, the goat map of Schibler *et al.* (1998) and SheepMap4.7 (http://www.ncbi.nlm.nih.gov/mapview/static/sheepsearch.html)

The new position of OarHH64 on CHI4 corresponds to its position on OAR4. Both Vaiman *et al.* (1996) and Schibler *et al.* (1998) divided CHI4 into two linkage groups, making it difficult to compare positions. The inversion of markers BM0321 and OarFCB005 on CHI5 is however consistent with the order reported by Vaiman *et al.* (1996). BM0321 has not yet been mapped to any ovine chromosome, thus map positions could not be compared. The relative positions of OarFCB005 and BMC1009 (adjacent to BM0321) do however correspond to those reported on SheepMap4.7. OarCP34 has not previously been ordered on CHI11, while the inversion of ILSTS045 and its close proximity to OarCP34 corresponds to its relative position on OAR3. Vaiman *et al.* (1996) only reports the position of INRA 177 on bovine BTA11, while OarCP34 was not mapped at all. Markers LSCV36 and MCM210 on CHI19 were inverted in the current study. This order is in agreement with OAR11 on SheepMap4.7. These rearrangements in chromosomal order seem to be the correct order of the markers, and the previously reported orders which showed inversions compared to the ovine map were most likely either population specific or due to problems with map assembly. The rearrangements are also supported by the reduced genetic length of the new map.

Linkage map CHI2 was mapped in two linkage groups by Vaiman *et al.* (1996), making it difficult to compare map positions. The two markers INRA040 and OarFCB11 on CHI2 are spaced far from each other on both the goat map of Schibler *et al.* (1998) and the SheepMap4.7. Their inversion and close proximity reported in this study seems to be population-specific to the South African Angora goat. The inversion of markers BMC1222 and IL2RA on CHI13 corresponds with position reported by Vaiman *et al.* (1996), but is in disagreement with the order on OAR13.

The new positions of anonymous markers on the goat map, as well as the correction of previously reported inversions (compared to the ovine map) will contribute to the improvement and accuracy of the goat map. The improved caprine linkage maps will assist with comparative mapping of economically significant loci, which could result in the application of marker assisted selection.

Conclusion

The accuracy and coverage of the goat map will be increased significantly once SNPs are available for genome mapping in goats. Until then, microsatellite markers have made an essential contribution to the development of caprine genome maps. No effort has been made to date to verify previously reported inter-chromosomal rearrangements. The correction of these inversions (compared to the ovine map) will contribute to the development of a robust caprine linkage map. The use of a large population which resulted in a significant improvement in the number of informative meiosis and shorter mapping distances has lead to the advancement of the caprine linkage map, and should in future be the basis of an advanced linkage map. A more complete and accurate map should lead to the

opportunity of mapping genetic variation that is responsible for the phenotypic differences in economically important traits.

References

Allan, M.F. & Smith, T.P.L., 2008. Present and future applications of DNA technologies to improve beef production. Meat Science 80, 79-85.

Baumung, R., Simianer, H. & Hoffmann, I., 2004. Genetic diversity studies in farm animals – a survey. J. Anim. Breed. Genet. 121, 361-373.

Cosenza, G., Pauciullo, A., Gallo, D., Colimolro, L., D'Avino, A., Mancusi, A. & Rammuno, L. 2008. genotyping at the CSN1S1 locus by PCR-RFLP and AS-PCR in a Neapolitan goat population. Small Rumin. Res. 74, 84-90.

Crawford, A.M., Montgomery, G.W., Pierson, C.A., Brown, T., Dodds, K.G., Sunden, S.L.F., Henry, H.M., Ede, A.J., Swarbick, P.A., Berryman, T., Penty, J.M. & Hill, D.F., 1994. Sheep linkage mapping: Nineteen linkage groups derived from the analysis of paternal half-sib families. Genetics 137, 573-579.

Dubeuf, J.P. & Boyazoglu, J., 2009. An international panorama of goat selection and breeds. Livest. Sci. 120, 225-231.

Faraut, T., De Givry, S., Hitte, C., Lahbib-Mansais, Y., Morisso, M., Milan, D., Schiex, T., Servin, B., Vignal, A., Galibert, F. & Yerle, M., 2009. Contribution of radiation hybrids to genome mapping in domestic animals. Cytogenet Genome Res 126, 21-33.

Green, P., Falls, K. & Crooks, S., 1989. Documentation for CRI-MAP, version 2.4. Washington University, St Louis.

Gupta, N., Pandey, A., Malik, G. & Gupta, S.C., 2009. Single nucleotide polymorphism (SNP) in growth hormone gene of Jakhrana, a prominent milk goat breed in India. Small Rumin Res. 81, 35-41.

Huehn, M., 2010. Random variability of map distances based on Kosambi's and Haldane's mapping functions. *J Appl Genet*. 51(1), 27-31.

Loots, F., 2007. Seisoenoorsig. The Angora goat and Mohair Journal P O Box 50, Jansenville, 6265, South Africa.

Maddox, J.F., 2005. A presentation of the differences between the sheep and goat genetic maps. Genet. Sel. Evol. 37 (Suppl 1), S1-S10.

Maddox, J.F. & Cockett, N.E., 2007. An update on sheep and goat linkage maps and other genomic resources. Small Rumin Res. 70(1), 4-20.

Maddox, J.F., Davies, K.P., Crawford, A.M., Hulme, D.J., Vaiman, D., Cribiu, E.P., Freking, B.A., Beh, K.J., Cockett, N.E., Kang, N., Riffkin, C.D., Drinkwater, R., Moore, S.S., Dodds, K.G., Lumsden, J.M., Van Stijn, T.C., Phua, S.H., Adelson, D.L., Burkin, H.R., Broom, J.E.,

Buitkamp, J., Cambridge, L., Cushwa, W.T., Gerard, E., Galloway, S.M., Harrison, B., Hawken, R.J., Hiendleder, S., Henry, H.M., Medrano, J.F., Paterson, K.A., Schibler, L., Stone, R.T. & Van Hest, B., 2001. An enhanced linkage map of the sheep genome comprising more than 1000 loci. Genome Res 11, 1275-1289.

Marshall, T.C., Slate, J., Kruuk, L.E.B. & Pemberton, J.M., 1998. Statistical confidence for likelihood-based paternity inference in natural populations. Molecular Ecology 7 (5), 639-655.

Moioli, B., D'Andrea, M.. & Pilla, F., 2007. Candidate genes affecting sheep and goat milk quality. Small Rumin Res. 68, 179-192.

Notter, D.R., Baker, R.L. & Cockett, N.E., 2007. The outlook for quantitative and molecular genetic applications in improving sheep and goats. Small Rumin Res. 70, 1-3.

Prasad, A., Schiex, T., McKay, S., Murdoch, B., Wang, Z., Womack, J.E., Stothard, P. & Moore, S.S., 2007. High resolution radiation hybrid maps of bovine chromosomes 19 and 29: comparison with the bovine genome sequence assembly. BMC Genomics 8, 310-319.

Schibler, L., Vaiman, D. Oustry, A., Giraud-Delville, C & Cribou, E.P., 1998. Comparative gene mapping: A fine-scale survey of chromosome rearrangements between ruminants and humans. Genome Res 8, 901-915.

Slate, J., 2008. Robustness of linkage maps in natural populations: a simulation study. Proc. R. Soc. B 275, 695-702.

Sonstegard, T.S., Van Tassell, C.P. & Ashwell, M.S., 2001. Dairy cattle genomics: Tools to accelerate genetic improvement. J. Anim. Sci. (E suppl.): E307-E315.

Toro, M.A., Fernandez, J. & Caballero, A., 2009. Molecular characterization of breeds and its use in conservation. Livestock Science 120, 174-195.

Vaiman, D., Schibler, L., Bourgeois, F., Oustry, A., Amigues, Y. & Cribiu, E.P., 1996. A genetic linkage map of the male goat genome. Genetics 144, 279-305.

Vaiman, D., Pailhoux, E., Schibler, L., Oustry, A., Chaffaux, S., Cotinot, C., Fellous, M & Cribou, E.P., 1997. Genetic mapping of the polled/intersex locus (PIS) in goats. Theriogenology 47, 103-109.

Vignal, A., Milan, D., SanCristobal, M. & Eggen, A., 2002. A review on SNP and other types of molecular markers and their use in animal genetics. Genet. Sel. Evol. 34, 275-305.

Weller, J.I., 2001. Quantative Trait Loci analysis in animals. CABI publishing, Oxon, United Kingdom. P 10-11.

CHAPTER 3.1

IMPROVED ALIGNMENT BETWEEN THE SHEEP AND GOAT LINKAGE MAPS

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Improved Alignment Between The Sheep And Goat Linkage Maps

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Introduction

Linkage maps are an essential tool in identifying specific loci associated with phenotypic variation in economically important traits. The current caprine maps available are however limited to the first low-resolution genetic map for the male goat genome constructed by Vaiman et al. (1996), followed by a more densely populated map on the male chromosome by Schibler et al. (1998). The focus on mapping specific traits of economic importance, such as milk proteins (Barillet (2007); Moioli et al. (2007)), polled intersex syndrome (Vaiman et al. (1997)) and growth hormones (Gupta et al. (2009) has resulted in the creation of unbalanced maps, with an uneven distribution of markers. Specific regions (i.e. the areas flanking the PISRT1 locus) have been very well described and populated densely with microsatellite markers, while other regions were neglected. Although several QTL studies have been performed on small-stock species and partial linkage maps were created as by-products of these studies, these maps are mostly not publicly available (Maddox & Cockett (2007)).

Despite the generally good alignment between the caprine and ovine linkage maps, a relatively large number of discrepancies between both species have been reported (Maddox (2005)), including many inversions in locus orders between the two maps. It is not certain which of these discrepancies are artefacts, and which might be genuine. The use of a small number of microsatellite markers in conjunction with less error checking for the goat map, has lead to a less robust map with a need for further genetic map development. This study aimed to improve the alignment between the ovine and caprine maps by confirming or rejecting previously reported rearrangements of loci order.

Material and methods

The genetic linkage map was constructed by genotyping three generations of Angora goat half-sib offspring, belonging to 12 different families. The families ranged from 44 to 140 offspring, with an average of 88 half-sib offspring per sire. DNA was extracted from whole blood samples using respectively the Qiagen DNEasy Tissue kit at the University of Pretoria and the Invisorb blood mini HTS kit (Invitek) for the XtractorGene (Corbett Robotics) at Wageningen University according to the protocols of the respective manufacturers. Incorrect parentage was identified with Cervus 3.0 (Marshall (1998)), and all aberrant individuals were removed from the study.

96 Microsatellite markers were selected from the existing goat map database (http://locus.jouy.inra.fr/cgi-bin/lgbc/mapping/common/intro2.pl?BASE=goat). PCR was performed

in a i-cycler (Bio-rad) and T1 Thermocycler (Biometra) using 30ng DNA, 2.94 µl of the ABgene® PCR Master Mix (ABGene, UK) and 0.03 µl each of 40pmol/µl reverse and forward primer. The PCR amplification was conducted in a 6 µl final volume in 384 well PCR plates at the following conditions: 95°C for 5 minutes, followed by 35 cycles of 96°C for 30s, 45s at Annealing Temperature and 90s at 72°C with a final extension step of 10min at 72°C.

Linkage analysis was performed with CRI-MAP 2.4 (Green et al. (1989)) compiled for Windows XP. Linkage groups were assigned on the basis that the markers have been mapped to specific goat chromosomes. The "two point' option was used to verify these linkage groups. A LOD threshold of 6 was chosen to detect significant linkage, and lowered in increments (to 1) until all markers could be assigned to a specific chromosome. Linkage groups were further analysed using the "build" option. Markers that could not be placed accurately with this option, were then included in alternative "fixed" orders. The final order (which resulted in the shortest map length) was verified with the "flipsn" option.

Results and discussion

Despite the overall good agreement of the caprine and ovine maps, a large number of discrepancies have been reported between them (Maddox (2005)), including inter-chromosomal re-assignments. Due to the limited development of the goat genetic map since the first loci were mapped, the discrepancies reported were not verified. The marker order generated in this study is in agreement of Schibler et al. (1998) except for the marker orders on CHI4, CHI11 and CHI19. The relative marker orders for the chromosomes with potential rearrangements are shown in Figure 3.1.1. For these three chromosomal linkage maps our marker order is in agreement with the marker order reported in sheep (SheepMap 4.7 ; http://www.ncbi.nlm.nih.gov/mapview/static/sheepsearch.html). This indicates that the three inversions observed by Schibler et al. (1998) were either population specific or incorrect marker assignments and is supported by the reduced genetic length of the linkage map. However, the rearrangement on CHI3 detected by Schibler et.al. (1998) is validated in this study, and therefore a true inter-chromosomal rearrangement. The relative map lengths between the studies also differed. This could be explained by the higher average number of informative meiosis (518±179) in our population, compared to 114±70 by Schibler et al. (1998) resulting in a more accurate linkage map. Furthermore, Schibler et al. (1998) reported results without taking Kosambi's correction into account, while the current study made use of Kosambi's mapping function.

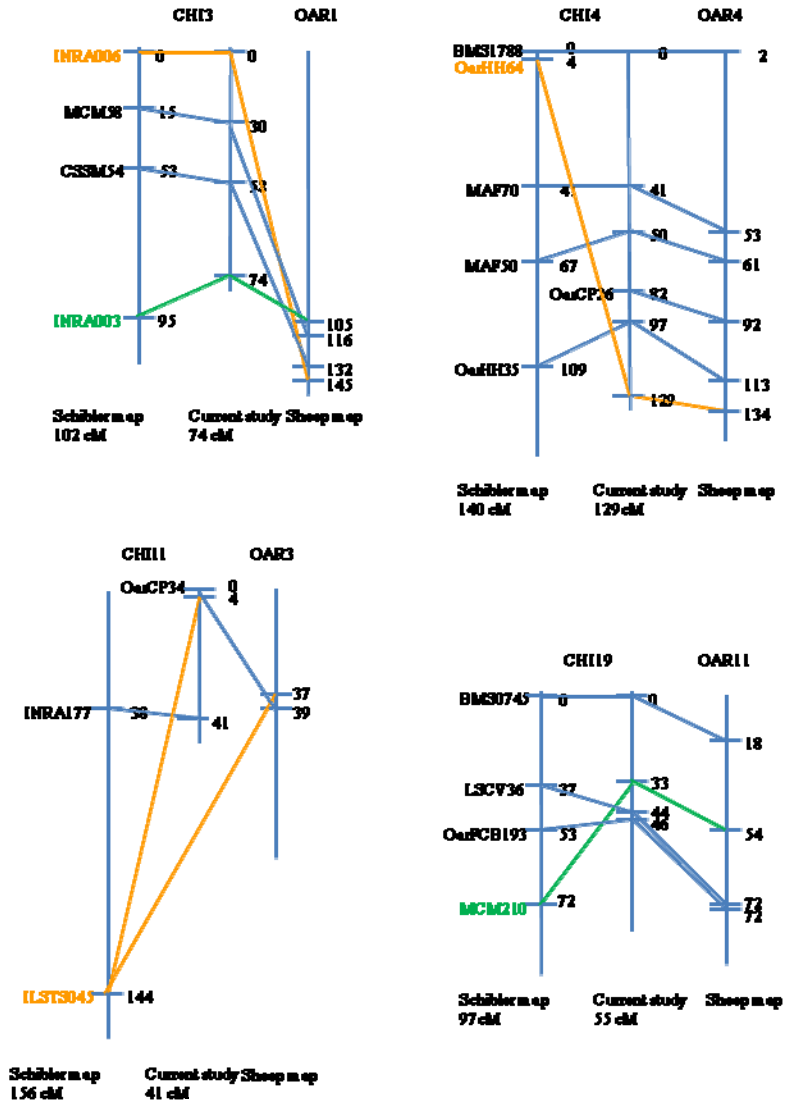

Figure 3.1.1: Alignment of marker orders on CHI3, 4, 11 and 19 with orders compared between the current study, the goat map of Schibler et al. (1998) and SheepMap4.7 (http://www.ncbi.nlm.nih.gov/mapview/static/sheepsearch.html)

The most common marker used in goat linkage map studies is still microsatellite markers. Single Nucleotide Polymorphisms (SNPs) are becoming the marker of choice in many molecular studies, mainly due to increased automation coupled with low cost (Toro et al. (2009)). However, while millions of SNPs have been identified for cattle, and about 5000 potential SNPs have been identified for use in ovine studies, no mapped SNP markers have been reported for goats (Maddox & Cockett (2007)). Only 1.2% of bovine SNPs were proved to be polymorphic in goats (Maddox & Cockett (2007)), and is thus of little use in goat studies. SNP discovery projects for caprine SNPs are required before these markers can be utilized for molecular research on goats.

Conclusion

The accuracy and coverage of the goat linkage map will be increased significantly once SNPs are available for genome mapping in goats. Until then, microsatellite markers have made an essential contribution to the development of caprine genome maps. The correction of previously reported inversions (compared to the ovine map) will contribute to the improvement and accuracy of the goat map. The improved caprine maps will help with comparative mapping of economically significant loci, which could result in the application of marker assisted selection.

References

Barillet, G. (2007). Small Rumin Res. 70: 60-75.

Green, P., Falls, K. and Crooks, S. (1989). Documentation for CRI-MAP, version 2.4. Washington University, St Louis.

Gupta, N., Pandey, A., Malik, G. et al. (2009). Small Rumin Res. 81: 35-41.

Maddox, J.F. (2005). Genet. Sel. Evol. 37 (Suppl 1): S1-S10.

Maddox, J.F. and Cockett, N.E. (2007). Small Rumin Res. 70: 4-20.

Marshall, T.C., Slate, J., Kruuk, L.E.B. et al. (1998). Mol Ecol 7 (5): 639-655.

Moioli, B., Andrea, M.D. and Pilla, F. (2007). Small Rumin Res. 68: 179-192.

Schibler, L., Vaiman, D. Oustry, A. et al. (1998). Genome Res 8: 901-915.

Toro, M.A., Fernandez, J. and Caballero, A. (2009). Livest Sci 120: 174-195.

Vaiman, D., Schibler, L., Bourgeois, F. et al. (1996). Genetics 144: 279-305.

Vaiman, D., Pailhoux, E., Schibler, L. et al. (1997). Theriogenology 47: 103-109.

CHAPTER 4

GENETIC PARAMETERS FOR PHYSICAL AND QUALITY TRAITS OF MOHAIR IN SOUTH AFRICAN ANGORA GOATS

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Genetic parameters for physical and quality traits of mohair in South African Angora goats

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Abstract

The continuous evaluation and genetic improvement of fleece traits in Angora goats are of major importance to the Angora goat industry. The objective of this study was to estimate variance components and genetic parameters of for both physical and quality traits of mohair in South African Angora goats. The data used for this study were collected on between 898 and 6211 kids (depending on the trait measured) born between 2000 and 2006 in 11 different Angora goat studs. Variance components and genetic parameters were estimated with the ASREML programme for fleece weight, fibre diameter, coefficient of variation of fibre diameter, standard deviation of fibre diameter, comfort factor, spinning/effective fineness and standard deviation of fibre diameter along the length of the staple. Most of these traits (excluding fleece weight) are measured using OFDA technology exclusively, and this study was the first attempt to calculate genetic parameters for these traits in South Africa. A model including the direct additive genetic effects only, were the most appropriate for estimation of all parameters. Heritability estimates were 0.24±0.03 for fleece weight, 0.45±0.03 for fibre diameter, 0.37±0.10 for coefficient of variation of fibre diameter, 0.32±0.11 for standard deviation of fibre diameter, 0.63±0.10 for comfort factor, 0.61±0.10 for spinning/effective fineness and 0.14±0.08 for standard deviation of fibre diameter along the length of the staple. Results from this study can now be applied for the estimation of breeding values for fleece production of South African Angora goats and in incorporation into selection programs.

Keywords: Angora goats, fleece weight, fibre diameter, genetic parameters, quality traits, OFDA

Introduction

The improvement of fleece traits has for many years been of major concern to the Angora goat stud breeders in South Africa. In excess of 55% of the world's mohair is produced by the South African Angora goat industry and continuous improvement of selection methods and genetic analyses are therefore of major importance to maintain a quality clip (Loots, 2007). Fleece traits determine the quality of the mohair product and the potential profits to be made, with fibre diameter being the most

important profit-influencing trait (Qi *et al*., 1994; Snyman 2002). Fibre diameter has been a focus point of research over the last few decades. Quality traits that contribute to both the processing performance and consumer satisfaction of natural fibers (i.e standard deviation of fibre diameter and spinning fineness) have however been ignored to a large extent.

Selective breeding has been the major tool for improvement of fleece traits in South African Angora goats. In 1996 a breeding plan was designed for the industry by the National Department of Agriculture, with the aim of implementing a performance testing scheme with data collection of all relevant traits (Snyman & Olivier, 1996, 1999). A fine hair flock was established at the Jansenville Experimental Station in the Eastern Cape province of South Africa where production of both the fine hair flock and a control flock were evaluated. Both young replacement bucks and does were selected with emphases on increasing bodyweight, while decreasing fibre diameter using a selection index (Snyman & Olivier, 1996). This breeding strategy was evaluated by Snyman (2002) who concluded that selection for increased body weight and a high quality fine fleece with style and character was possible.

Most studies on Angora fleece traits have in the past included fleece weight, fibre diameter, standard deviation of fibre diameter and sometimes kemp score. Heritability and correlations have been reported for Australian (Gifford *et al,* 1991), Argentinean (Taddeo *et al.*, 1998) and New Zealand (Nicoll *et al.*, 1989) Angora goats, but in these studies not all fleece traits were included and the estimates obtained tend to vary based on different model structures, sample sizes and measuring techniques. The objective evaluation of fleece traits in the Angora goats worldwide was complicated for many years by the lack of efficient methods to measure traits such as fiber diameter. This also resulted in less objective measurements available for estimation of accurate genetic parameters for genetic analyses.

Due to fibre diameter being the most important trait in determining the price of mohair (McGregor and Butler, 2008), there was considerable interest in the development of cost-effective and rapid methods for objective measuring of the trait. Various methods such as projection microscopy (PM) and Airflow methods are available and recognised as standard methods (IWTO, 1989) but these are either rather time consuming, laborious and expensive or don't measure all required traits (Qi *et al*., 1994; Brims *et al.*, 1999). The Optical Fibre Diameter Analyser (OFDA) technology developed in the early 1990s provided an objective method for accurate and rapid determination of fibre diameter. The OFDA system was shown to be effective for measuring staples, tops and cores in Angora goats (Qi *et al*., 1994). OFDA100 technology was developed in 1989, introduced to and tested in South Africa during 1992 and has since been applied commercially (Baxter *et al.,* 1992). OFDA2000

technology has since been developed to be a portable instrument capable of real-time measurements of the fibre properties of greasy fibre with minimal sample preparation as an aid to selection or clip classing (Brims *et al.*, 1999) and is currently routinely used for mohair fleece measurements in South Africa. Before these objectively and accurately measured traits can be considered as selection criteria, genetic parameters need to be estimated. If the traits associated with the full diameter profile are shown to have medium heritabilities and favourable correlation with other traits they could be considered for inclusion in a selection index, depending on their economic value.

Although several studies have been performed to investigate the usefulness of the diameter profile in wool sheep (Greeff, 2006; Smith *et al.*, 2006), very limited research has been conducted in the mohair industry. It was proposed by Butler & Dolling (1992) that spinning fineness might be preferable to mean fibre diameter as a selection criterion for wool sheep. Additional information on fibre properties that are obtained from the OFDA analysis, including coefficient of variation, spinning fineness and comfort factor, could have direct impact on processing performance and skin comfort of mohair fibres (Lamb, 1992). Inclusion of these new quality traits as selection criteria is however dependent on their heritabilities and genetic correlations with other traits. To date only Alain & Roguet (2006) has yet reported genetic parameters for OFDA-measured traits in mohair, including coefficient of variation of fibre diameter, OFDA kemp and OFDA medullated fibres.

The objectives of this study were to estimate heritabilities and genetic correlations for physical and quality traits of mohair in South African Angora goats.

Material and methods

Data for this study were obtained from 11 different Angora goat herds, consisting of phenotypic records for kids born during the 2000 to 2006 kidding seasons. Data were recorded on the second (8 to 12 months) and third (16 to 18 months) shearings. Bucks were mostly sheared at eight to 12 months, while the does were mostly sheared at 16 to 18 months. Only one shearing record was included per animal in the analysis. A description of the dataset with the number of records analyzed for each trait, as well as the average and coefficient of variation per trait is given in Table 4.1.

For the analyses of fleece weight (FW) there were 302 sires and 3602 dams with progeny, while there were only 27 sires and 510 dams with progeny for the OFDA2000 fibre traits in the data set. A total of 1073 animals in the pedigree file had unknown sires, but all dams were known.

Table 4.1 Description of the dataset for estimation of genetic parameters for fleece traits of Angora goats

Trait	Abbreviation	N records	Measurement	Mean	CV (%)	Rams second shearing	Does second shearing	Rams third shearing	Does third shearing
Greasy fleece weight (kg)	FW	6211	Scale	1.44	20.23	1627	1328	535	2721
Fibre diameter (µm)	FD	6041	OFDA100 + OFDA2000	27.97	8.40	1617	1326	532	2566
Coefficient of variation of fibre diameter (%)	CVFD	898	OFDA2000	25.43	11.98	393	-	374	131
Standard deviation of fibre diameter (µm)	SDFD	898	OFDA2000	7.29	13.53	393	-	374	131
Comfort factor (%)	CF	898	OFDA2000	64.30	20.66	393	-	374	131
Spinning/effective fineness (µm)	SF	898	OFDA2000	29.13	8.85	393	-	374	131
Standard deviation of fibre diameter along the length of the staple (µm)	SDA	898	OFDA2000	1.33	37.54	393	-	374	131

Greasy fleece weight (measured to the nearest 0.1 kg) was determined just after shearing. Individual midrib samples were taken from each animal for determination of fibre diameter. From 2000 to 2003, fibre diameter of mohair samples was determined with the OFDA100 at the Wool Testing Bureau. Owing to the availability of OFDA2000 technology, micron testing of mohair samples collected since 2004 was done using an OFDA2000 instrument. OFDA 2000 provides the same accuracy, but faster measurement than the previously used OFDA 100 and correlations between the measurements of the two instruments are high (AWI Project EC 397 Final Report, 2004). A single sub-sample (prepared from three different locks) was analysed for each individual to obtain fibre properties with the latter instrument. OFDA measured traits include the Standard Deviation of fibre diameter, that measures in microns the distance either side of the average fibre diameter in which two thirds of the fibre diameter are found. The Coefficient of Variation is the SD expressed as a percentage of the average micron (Baxter et al., 1992) and Comfort factor is the percentage of fibres finer than 30 microns. These traits were all included as no parameters for them exist.

The final data file comprised of 6211 records which included the following traits: greasy fleece weight (FW; kg), fibre diameter (FD; µm), coefficient of variation of fiber diameter (CVFD; %), standard deviation of fiber diameter (SDFD; µm), comfort factor (CF; %), spinning/effective fineness (SF; µm) and standard deviation of fiber diameter along the length of the staple (SDA; µm). The original raw data were edited. Animals with missing information on birth date, sex, age of the dam or dam identification, were omitted. All the available pedigree information (7119 records) was included in the analyses.

In the model building phase an ANOVA was used to identify effects which contributed significantly to variation, using a General Linear Model (GLM) procedure of the SAS computer package (SAS, 2004). The fixed effects tested in the models for all traits were herd-year of birth (HY), sex (male or female), birth status of the kid (single offspring or twin), age of dam at kidding (2 to 12 years of age) in classes and a covariate for age of the animal at shearing (in days).

Variance components were estimated using the ASREML programme of Gilmour *et al.* (2002). A single trait animal model was fitted for all traits. Direct additive and maternal additive genetic effects, with or without a covariance between them, and common environmental effects were tested in different combinations to obtain the final model. The final model was as follows:

$\mathbf{y} = X\mathbf{b} + Z_1\mathbf{a} + \mathbf{e}$

Where:

y is a vector of observed traits of animals;

b, **a** are vectors of fixed effects and direct additive genetic effects, respectively;

X, Z_1 are incidence matrices respectively relating fixed effects and direct additive genetic effects;

e is the vector of residuals.

It was assumed that:

$V(a) = A\sigma^2_a$; $V(e) = I\sigma^2_e$

Where:

I is an identity matrix,

$\boldsymbol{\sigma^2_a}$ and $\boldsymbol{\sigma^2_e}$ are the direct additive genetic variance and environmental variance respectively.

All traits were analysed using a model that included only direct additive genetic effects. Maternal additive genetic effects and common environmental effects were evaluated for inclusion in the final model, but didn't cause a significant increase in the Log likelihood. For the purpose of this study a significance level of $P<0.01$ was applied throughout. Snyman & Olivier (1996) fitted several models for fleece traits (FW and FD) and concluded that the model which included only the direct additive genetic effects to be the most appropriate for parameter estimation. The fixed effects finally included in the models for all traits were herd-year of birth, sex , birth status, age of dam and a covariate for age of the animal at shearing, applied over sex groups. All effects were significant at $P<0.001$.

The final model was fitted for all the traits. All components, with the phenotypic variance (σ^2_p), being the sum of σ^2_a and σ^2_e, were derived at convergence. Direct heritabilities ($h^2 = \sigma_a{}^2/\sigma_p{}^2$) were computed for each trait. Multi-trait analyses were done to estimate covariance components and correlations among the traits, using the most suitable model for each trait, as determined under single trait analyses.

Results

The variance components and heritability estimates for the traits using the single trait analyses are shown in Table 4.2. All heritability estimates were based on one shearing record per animal that was included in the analysis. The estimated heritability values vary from as low as 0.14 for SDA to as high as 0.63 for CF.

Table 4.2 Results of a single trait analyses: variance components and heritability estimates for fleece traits

	FW (kg)	FD (µm)	CVFD (%)	SDFD (µm)	CF (%)	SF (µm)	SDA (µm)
σ^2_p	0.080	5.586	8.454	0.921	192.530	7.285	0.242
σ^2_a	0.019	2.495	3.128	0.294	121.219	4.447	0.034
σ^2_e	0.061	3.091	5.326	0.627	71.311	2.838	0.208
h^2	0.24±0.03	0.45±0.03	0.37±0.10	0.32±0.11	0.63±0.10	0.61±0.10	0.14±0.08

Where: σ^2_p phenotypic variance, σ^2_a direct additive variance, σ^2_e residual variance, and h^2 direct heritability.

The phenotypic and genetic correlations estimated for the fleece traits using multi-trait analyses are summarized in Table 4.3. Most of the traits had medium to strong positive (0.35 to 0.78) or medium to strong negative (-0.30 to -0.97) genetic correlations with each other, the exceptions being FW with FD, CVFD and SDA. The SE of the genetic correlation between FW and FD was unexpectedly high, given that in excess of 6000 records were included. The negative genetic correlation between SF and CF approached 1 (-0.97), while the genetic correlations between FD and CF (-0.81) and FD and SF (0.78) were also strong.

Table 4.3 Genetic (above diagonal) and phenotypic (below diagonal) correlations among fleece traits in Angora goats using a multi trait analyses

	FW	FD	CVFD	SDFD	CF	SF	SDA
FW		0.08±0.22	-0.17±0.27	0.35±0.26	-0.46±0.18	0.46±0.18	0.11±0.43
FD	0.39±0.04		-0.36±0.17	0.51±0.15	-0.81±0.04	0.78±0.05	0.44±0.31
CVFD	-0.04±0.04	-0.19±0.04		0.41±0.19	0.50±0.16	-0.31±0.21	0.71±0.45
SDFD	0.30±0.03	0.45±0.03	0.71±0.02		-0.55±0.14	0.74±0.09	-0.30±0.54
CF	-0.46±0.03	-0.86±0.01	0.21±0.04	-0.48±0.03		-0.97±0.01	-0.46±0.27
SF	0.47±0.03	0.82±0.01	0.08±0.04	0.75±0.02	-0.93±0.01		0.41±0.28
SDA	0.15±0.03	0.23±0.03	0.15±0.04	0.34±0.03	-0.26±0.03	0.34±0.03	

For a number of traits, the genetic and phenotypic correlations were not in agreement. The phenotypic correlations between FD and FW (0.39) and SDA and SDFD (0.34) were both medium positive, while the genetic correlations between these traits were 0.08 and -0.30 respectively. Phenotypic correlations between CVFD and SF (0.08) and between CVFD and SDA (0.15) were low positive, in contrast with genetic correlations of -0.31 and 0.71 respectively. The Standard Error for all genetic correlations with SDA were relatively high, as the direct additive variance component were low for this trait.

No genetic trends could be included as breeding values were not estimated for these traits. Too few records are currently available, and this will be included in future research papers as the number of animals measured increases.

Discussion

Before OFDA technology became available, genetic parameters for fleece characteristics of Angora goats have been largely limited to fleece weight, fibre diameter and in some cases kemp scores. Estimates of these genetic parameters are also scarce and highly variable (Taddeo *et al.,* 1998). In this study additional traits could be included from the OFDA data that Angora breeders have been recording since 2004.

Two shearing age periods were included (8 to 12 months and 16 to 18 months) with an average fleece weight of 1.44 kg while in a previous study (Snyman & Olivier, 1999) on data from the Angora Goat Performance Testing Scheme, the average fleece weights reported varied between 1.97 kg at 10 months of age to 2.33 kg at 16 months of age. The fibre diameter of 27.97 µm in this study was lower than Snyman (2002) reported for South African Angoras at 14 months of age (31.13 µm) and than Allain & Roguet (2003) reported for French Angoras measured at 18 months of age (30.4 µm). The CVFD however corresponded very closely to that reported in the French Angora goat study (25.43 % vs. 25.3 %).

The environmental effects (sex, birth status and herd year of birth and age of the dam) that had a significant effect on the different traits were similar to effects studied by Nicoll *et al.,* (1989), Gifford *et al.,* (1990) and Gertsmayr & Horst (1995). The variance component results reported in Table 2 correspond to values reported by Taddeo *et al*. (1998), who reported direct additive genetic variance of 0.04 and residual variances of 0.12 for fleece weight and additive genetic variance of 1.83 and residual variances of 3.72 for fibre diameter.

The estimated heritability values vary from as low as 0.14 for SDA to as high as 0.63 for CF, and are in the general range of values cited in literature (Nicoll, 1985; Gifford *et al.*, 1991; Sumner & Bigham, 1993, Taddeo *et al.,* 1998). Pattie *et al*. (1990) reported values ranging from 0.13 to 0.50 for greasy fleece weight and 0.12 to 0.51 for fibre diameter, for studies performed in Turkey, the USA, New Zealand and Australia.

The heritability estimate calculated for fleece weight was 0.24 (±0.03), which is similar to estimates of 0.22 (±0.04) and 0.19 (±0.04) reported by Snyman & Olivier (1996 and 1999) on South African Angora goats and 0.25 (±0.04) in French Angora goats (Allain & Roguet, 2006), but lower than estimates reported for Australian goats of 0.45 (±0.23) for greasy fleece weight (Gifford *et al.,* 1991).

Estimates for OFDA-measured fibre diameter were higher (0.45) than previous estimates of 0.30 by Snyman & Olivier (1999) on South African goats and 0.33 by Taddeo *et al.,* (1998) on Argentinean Angora goats. These studies were performed before OFDA technology became commonly available. A similar trend was observed where Allain & Roguet (2003) estimated a heritability of 0.32 for fibre diameter of French Angora goats, but found a much higher estimate of 0.51 three years later when using OFDA measures (Allain & Roguet, 2006). This trend could be attributed to the more accurate measurements performed with OFDA, leading to higher estimates of heritability.

Most of the OFDA-measured quality traits analysed in this study were included for first-time estimation of genetic parameters in South Africa. Limited results have previously been reported globally on genetic parameters for these traits. CVFD and SDFD both had a moderate heritability of 0.37 and 0.32 respectively, while CF and SF had high heritability estimates as indicated in Table 2. Spinning Fineness is a combination of FD and CVFD, and has a significantly higher heritability estimate than FD (0.61 vs. 0.45). Derived heritability values for SF were reported by Butler & Dolling (1992), which ranged between 0.43 and 0.70. The Coefficient of variation for fibre diameter reported by Allain & Roguet (2006) from ODFA data in French Angoras was lower (0.29) than that found in the present study (0.37) and Nicoll *et al.* (1989) reported a lower heritability value (0.21) for SDFD. SDA was the only trait with a low heritability of 0.14.

The low positive genetic correlation and high SE (0.08±0.22) between FW and FD does not correspond to those in the literature. Medium to strong positive correlations ranging between 0.35 (±0.04) (Allain & Roguet, 2003) and 0.551 (±0.07) (Snyman & Olivier, 1996) have been reported. In a review by Pattie *et al.* (1993) genetic correlation values for these two traits ranging from -0.29 to 0.98

were reported, reflecting the great variation in these genetic parameters. The strong negative genetic correlation found in this study between FD and CVFD of -0.36 corresponded to the values of -0.33 (Allain & Roguet, 2003) and -0.44 (Allain & Roguet, 2006) reported previously. The genetic correlations between SF and FD (0.78) and CF (-0.81) was very strong, indicating this trait's direct impact on skin comfort.

Heritability estimates indicate that FD, CVFD, CF and SF are moderately to highly heritable and should respond positively to selection. Selection for decreased SF should result in decreased FD, SDFD and increased CF. FW will however also decrease with selection for decreased SF, and SF should thus only be included in a selection index, which will limit the corresponding decrease in FW. CVFD is noted as an important measure of the relative distribution of fibre diameter around the mean within a fleece and therefore a standardized measure of the variation in the FD. The ideal would be a lower FD that will result in increase in CF and lower CVFD.

It has been shown that the fibre diameter distribution has a significant effect on yarn properties and processing performance of wool (Qi *et al.,* 1994; Smith *et al.,* 2006). A decrease in CVFD is associated with more sound wool in Merino sheep and can be used as an indirect selection criterion to improve staple strength in Merinos as well as decreasing susceptibility to fleece rot (Greeff, 2006). Smith *et al.* (2006) concluded that the fibre diameter profile could be a useful tool to improve fleece quality in fine-wool sheep breeds. Much less is known regarding the effect of quality traits such as CVFD of mohair on its processing performance. Fleece homogeneity is however of economic importance to Angora farmers (Allain & Roguet, 2003), and information on the fibre diameter profile gives opportunity to select for less variability in fleece quality. Lupton & Pfeiffer (1998) stated the efficiency of both quality assessment and selection programs should be improved by using OFDA technology in Angora goat programs.

Fibre diameter distribution has a significant effect on processing and fabric properties. Factors contributing to the variation in mohair fleeces are the physical area where the sample is taken from (Taddeo *et al.,* 2000; McGregor & Butler, 2008) and secondly between-staple variation and variation along the length of individual fibres (Venter, 1959; Grobbelaar & Landman, 1984). The full fibre diameter profile, including the width of the distribution in terms of the coefficient of variation, can be measured with OFDA (Kritzinger, 1992; Hunter, 1993).

From the results reported here it is clear that OFDA-measured quality traits are moderately to highly heritable. Future studies should focus on the economical importance of the various traits in

mohair production and processing, to evaluate their possible inclusion in selection strategies for Angora goats.

Conclusion

Since the development of OFDA technology, additional traits for improvement of mohair quality are available to use as possible selection criteria. This was the first attempt to estimate genetic parameters for these traits. Some OFDA-measured fleece traits should receive serious consideration for inclusion in a selection index for increased genetic progress in South African Angora goats, as they have medium to high heritabilities, as well as favourable correlations with other economically important traits. The economic value of these traits needs to be estimated in future studies. The simultaneous measurement of medullated fibres by using OFDA medullation measurements, as performed by Allain & Roguet (2006), should also be taken into account as a way to improve selection efficiency.

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References

Allain, D., Roguet, J.M., 2003. Genetic and non-genetic factors influencing mohair production traits within the national selection scheme of Angora goats in France. *Livest. Prod. Sci.* 82, 129-137.

Allain, D., Roguet, J.M., 2006. Genetic and non-genetic variability of OFDA-medullated fibre contents and other fleece traits in the French Angora goats. *Small Rumin. Res*. 65, 217-222.

AWI Project EC397, 2004. Final Report On Farm Fibre Measurement (OFFM) Instrument Evaluation Trial April 2004. Australian Wool Innovation Limited.

Baxter, B.P., Brims, M.A., Teasdale, D.C., 1992. The optical fibre diameter analyser (OFDA). New technology for the wool industry. *Wool Tech. Sheep Breed.* 40, 131–134.

Brims, M.A., Peterson, A.D., Gherardi, S.G., 1999. IWTO Tech. Rep. RWG 04, Florence

Butler, K.L., Dolling, M., 1992. Calculation of the heritability of spinning fineness from phenotypic and genetic parameters of the mean and CV of fibre diameter. *Aust. J. Agric. Res*. 43, 1441-1446.

Gerstmayr, S., Horst, P., 1995. Estimates of performance traits in Turkish Angora goats. *Small Rumin. Res*. 16, 141-157.

Gifford, D.R., Ponzoni, R.W., Burr, J., Lampe, R.J., 1990. Environmental effects on fleece and body traits of South Australian Angora goats. *Small Rumin. Res*. 3, 249-256.

Gifford, D.R., Ponzoni, R.W., Lampe, R.J., Burr, J., 1991. Phenotypic and genetic parameters of fleece traits and live weight in South Australian Angora goats. *Small Rumin. Res*. 4, 293-302.

Gilmour, A.R., Gogel, B.J., Cullis, B.R., Welham, S.J., Thompson, R., 2002. ASReml User Guide Release 1.0. VSN International Ltd, Hemel Hempstead, HP1 1ES, UK.

Greeff, J., 2006. Coefficient of variation of wool fibre diameter in Merino breeding programs. Farmnote 46/98. Department of Agriculture, Government of Western Australia, Australia.

Grobbelaar, P.D., Landman, C.M.M., 1984. Effek van energiebyvoeding op jong groeiende Angorabokkies "Effect of energy supplementation on young growing Angora kids". *Karoo Agric* 3 (4), 11-15.

Hunter, L., 1993. Mohair: A review of its properties, processing and applications. CSIR Division of Textile Technology, NMB Printers (Pty) Ltd, Port Elizabeth, South Africa, 278 pp.

IWTO, 1989. IWTO. In: International Wool Textile Organisation, Specification IWTO-8-89E, International Wool Secretariat, Ilkley

Kritzinger, N.M., 1992. Selection for more uniform fibre diameter – a future reality? *Angora Goat and Mohair Journal* 34(1), 51-52.

Lamb, P.R., 1992. The effect of fibre diameter distribution on yarn properties. *Wool Tech. Sheep Breed.* 40(2), 65-68.

Loots, F., 2007. Seisoenoorsig. The Angora goat and Mohair Journal P O Box 50, Jansenville, 6265 South Africa.

Lupton, C.J., Pfeiffer, F.A., 1998. Measurement of medullation in wool and mohair using an Optical Fibre Diameter Analyser. *J.Anim. Sci.* 76, 1261-1266.

McGregor, B.A., Butler, K.L., 2008. Variation of mean fibre diameter across mohair fleeces: Implications for within flock animal selection, genetic selection, fleece classing and objective sale lot building. *Small Rumin. Res.* 75, 54-64.

Nicoll, G.B., 1985. Estimates of environmental effects and some genetic parameters for weaning weight and fleece weights of young Angora goats. *Proc. New Zealand Soc. Anim. Prod*. 45, 217-219.

Nicoll, G.B., Bigham, M.L., Alderton, M.J., 1989. Estimates of environmental effects and genetic parameters for live weights and fleece traits of Angora goats. *Proc. New Zealand Soc. Anim. Prod.* 49, 183-189.

Pattie, W.A., Baker, R.L., Nicoll, G.B., Restall, B.J., 1990. Breeding for cashmere and mohair. *Proc. 4th Wrld Congr Gen Appl Livest Prod.* 23-27 July, 1990. Edinburgh.

Qi, K., Lupton, C.J., Pfeiffer, F.A., Minikhiem, D.L., 1994. Evaluation of the Optical Fibre Diameter Analyser (OFDA) for measuring fiber diameter parameters of sheep and goats. *J. Anim. Sci.* 72, 1675-1679.

SAS Institute Inc., 2004. SAS Procedures Guide, Version 9.1. Cary, NC, SAS Institute Inc.

Smith, J.L., Purvis, I.W., Lee, G.J., 2006. Fibre diameter profiles – potential applications for improving fine-wool quality. *International Journal of Sheep and Wool Science* 54, 54-61.

Snyman, M.A. 2002. Evaluation of a genetically fine mohair producing herd. *Small Rumin. Res*. 43, 105-113.

Snyman, M.A., Olivier, J.J., 1996. Genetic parameters for body weight, fleece weight and fibre diameter in South African Angora goats. *Livest. Prod. Sci.* 47, 1-6.

Snyman, M.A., Olivier, J.J., 1999. Repeatability and heritability of objective and subjective fleece traits and body weight in South African Angora goats. *Small Rumin. Res*. 34 103-109.

Sumner, R.M.W., Bigham, M.L., 1993. Biology of fibre growth and possible genetic and non-genetic means of influencing fibre growth in sheep and goats – a review. *Livest. Prod. Sci.* 33, 1-29.

Taddeo, H.R., Allain, D., Mueller, J., Rochambeau, Manfredi, E., 1998. Genetic parameter estimates of production traits of Angora goats in Argentina. *Small Rumin. Res*. 28, 217-223.

Taddeo, H.R., Duga, L., Almeida, D., Willems, P., Somlo, R., 2000. Variation in mohair quality over the body in Angora goats. *Small Rumin. Res.* 36, 285-291.

Venter, J.J., 1959. A study of mohair classing. *S. Afr. J. Agric. Sci.* 2(1), 119-131.

CHAPTER 5

QTL FOR MOHAIR IN SOUTH AFRICAN ANGORA GOATS

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QTL for mohair traits in South African Angora goats

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Abstract

The aim of this study was to identify QTL associated with mohair production and quality traits in South African Angora goats. Limited research has been performed on QTL influencing the economically important mohair traits of Angora goats. A genome scan using 88 microsatellite markers covering 22 autosomes was conducted. Twelve half-sib families with an average of 58 offspring per sire were genotyped. Phenotypic data was collected at second and third shearing for males and females respectively. One or more QTL were identified for each trait investigated. Three putative QTL were detected for fleece weight on CHI 2, 5 and 24 which corresponds with the locations of keratin and keratin-associated proteins. This study detected the first QTL associated with mohair fibre diameter (on CHI 4 and 24, respectively). Four QTL were detected on CHI 8, 13, 18 and 20 which influence both comfort factor and spinning fineness. The variance explained by the QTL ranged between 6.9% for fibre diameter and 33.6% for standard deviation along the length of the staple. These results reveal segregation of QTL influencing mohair production and quality, and contribute to the understanding of the genetic variation of mohair traits.

Keywords: Angora goats, QTL, mohair

Introduction

DNA technology has resulted in the identification of loci and chromosomal regions that contribute to phenotypic variation in economically important traits (Dekkers, 2004). Identifying and confirming Quantitative Trait Loci (QTL) is the first step in the process that could lead to Marker Assisted Selection (MAS) or Gene Assisted Selection (GAS). Since the first QTL mapping studies in livestock (Andersson, 2001) the advantages and complications of implementing genomic information in genetic improvement programs have been well documented (Dekkers, 2004; Allan & Smith, 2008; Goddard, 2009). Using genomic information in selection especially has the potential to increase the rate of genetic progress for traits with low heritability, sex-limited traits and those that are difficult to measure (e.g. Pollak, 2005). Over the past two decades QTL have been confirmed in several farm animal species and some are applied in MAS, i.e. meat tenderness and marbling in beef cattle and meat quality and feed intake in pigs (Jeon *et al.,* 2006). The benefits of MAS is driven by an increased selection accuracy, and the successful implementation thereof is dependent amongst others on the economic relevance of the trait studied (Van der Werf, 2007).

In goats QTL studies have been mainly limited to disease resistance and milk proteins. Implementation of MAS in goats has been limited to the selection against diseases like scrapie, CAEV and Johne's disease (Dodds *et al.,* 2007) and for casein genes (Van der Werf, 2007). Wool and fibre traits seem to remain a challenge, despite moderate to high heritability for fleece weight and fibre diameter – the two traits that have usually been included in studies. The unfavourable genetic correlation that exist between fleece weight and fibre diameter as well as with body weight in wool sheep and mohair goats have prompted several studies on candidate genes.

The genetic improvement of production and quality mohair traits of Angora goats in South Africa has been based on phenotypic information for the past two decades. Emphasis was originally placed only on fibre diameter, but the unfavourable positive correlation with body weight resulted in small, unthrifty animals with low survivability. The national selection strategy was changed to include three primary traits, namely fibre diameter (the most important price-determining trait), fleece weight and body weight which were combined into a selection index (Snyman & Olivier, 1996). This policy was evaluated in 2002 (Snyman, 2002) and confirmed to be successful in improving production and quality of the national clip, while maintaining body weight. If QTL explaining significant fractions of the genetic variance in these traits could be identified, it should however lead to increased accuracy of EBVs with a corresponding faster rate of genetic improvement (Van der Werf, 2007). QTL mapping should also lead to increased knowledge of the underlying molecular mechanisms of fibre and fleece characteristics which could result in more efficient selection programs on the long term (Purvis & Jeffery, 2007).

Although several QTL identification studies have been undertook for wool sheep (Allain *et al.,* 2006; Bidinost *et al.,* 2006; Bidinost *et al.,* 2008; Roldan *et al.,* 2010), relatively few studies have been conducted to identify linkage with goat fibre production. Chromosome segments that affect mohair were identified by Cano *et al.* (2007) in Argentinean Angora goats and this resulted in further investigation into goat chromosome 19 (Cano *et al.,* 2009). These results, together with the QTL affecting conformation traits in Angora goats (Marrube *et al.,* 2007) paved the way for a candidate gene approach by Mohammad Abadi *et al.* (2009) for improved cashmere yield in Rayini goats. Recently QTL affecting fleece traits were identified by Debenedetti *et al.* (2010) on CHI 5 in a backcross Angora x Creole population. No QTL for the most important price-determining traits of mohair (fleece weight and fibre diameter) have however yet been identified. As new technology (i.e. OFDA measurements) was developed, it became clear that newly measured quality traits (e.g. coefficient of variation, comfort factor, spinning fineness) could possibly play a role in breeding objectives (Visser *et al.,* 2009). Traits that could result in a more uniform, fine mohair staple are especially of increasing importance. These are currently being considered for inclusion in the South African Angora goat selection index. Of these quality traits, only CVFD have been included in any of the previous studies.

Candidate genes for wool production have been studied quite extensively (Itenge-Mweza *et al.,* 2007; Gong *et al.,* 2010; Jin *et al.,* 2010). This is due to the unfavourable genetic correlation between fibre diameter and fleece weight (the two most important price-determining factors in natural animal-produced fibres) and the need for natural fibres with novel properties (Purvis & Franklin, 2005). The same motivation applies to the study of genes influencing mohair traits. The South African mohair clip contributes in excess of 50% to the global market. The constant pressure from synthetic fibres necessitates the need for a high quality clip with little variation within and between fleeces. The production of mohair with new and novel properties might expand the niche market for this quality product. This paper aimed to identify chromosome segments associated with product and quality traits of mohair.

Materials and Methods

Animals and phenotypic data:

Nineteen potential Angora goat bucks were screened for 50 markers and twelve bucks with the highest average heterozygosity were selected for participation in the study. Twelve half-sib stud families from four different farms were included. Family sizes were on average 58 and ranged between 16 and 130 offspring per sire. Families were created over a three-year period. For more detail regarding the family structure, refer to Visser & Van Marle-Köster (2009). Not all individuals that

were genotyped had phenotypic records for fleece traits; therefore fewer animals were included in the QTL analyses.

Phenotypes were recorded on second (8 to 12 months) or third (16 to 18 months) shearings. Bucks were mostly sheared at eight to 12 months, while the does were mostly sheared at 16 to 18 months. Only one shearing record per animal was included in the analysis. Greasy fleece weight (measured to the nearest 0.1 kg) was determined just after shearing. Individual midrib samples were taken and a single sub-sample (prepared from three different locks) was analysed for each individual for determination of average fibre diameter (FD; µm), coefficient of variation of fibre diameter (CVFD; %), standard deviation of fibre diameter (SDFD; µm), comfort factor (CF; %), spinning fineness (SF; µm) and standard deviation of fibre diameter along the length of the staple (SDA; µm). The quality traits were measured using OFDA2000 technology (Visser *et al.,* 2009). One breeder did not send in samples for further testing, and thus the number of records used for the quality traits were less than for FW and FD.

DNA and genotyping:

DNA was extracted from 100µl whole blood samples using respectively the Qiagen DNEasy Tissue kit and the Invisorb blood mini HTS kit (Invitek) for the XtractorGene (Corbett Robotics) according to the protocols of the respective manufacturers.

One hundred thirty four microsatellite markers were selected from the existing goat map database (http://locus.jouy.inra.fr/cgi-bin/lgbc/mapping/common/intro2.pl?BASE=goat) and previously published literature to obtain sufficient genome coverage. Eighty eight markers spaced over 22 autosomes were finally selected based on level of polymorphism, heterozygosity levels, allele size range and amplification success (a complete list of these parameters were reported in Visser *et al.,* 2010). Poor amplification and little informativeness of some markers limited the aim of at least three markers per chromosome. The markers were divided into eight genotyping sets, averaging twelve markers per set. PCR reactions were performed in a 384 well i-cycler (Bio-rad) or Ti Thermocycler (Biometra) in a 6µl final volume using 100ng DNA, 2.94 µl of the ABgene® PCR Master Mix (ABGene, UK) and 0.03 µl reverse and forward primer each of 40pmol/µl. The PCR amplification was conducted at the following conditions: 95°C for 5 minutes, followed by 35 cycles of 96°C for 30s, 45s at annealing temperature and 90s at 72°C with a final extension step of 10min at 72°C. Complete genotyping sets were attached as Addendum B.

Statistical analysis:

Phenotypic records were pre-adjusted for herd (levels 1-12), year of birth (2004, 2005, or 2006), birth status (single or twin) and sex. QTL analysis was performed using half-sib regression (Knott *et al.,* 1996) as implemented in the GridQTL software (Seaton *et al.,* 2006). The following least-squares regression model used was:

$y_{ij} = Sire_i + \beta_i x_{ij} + e_{ij}$

where y_{ij} is the phenotype (corrected for fixed effects) of individual ij, offspring of sire i,

$Sire_i$ is the mean of sire family i,

β_i is the allele substitution effect of the QTL within family i,

x_{ij} is the probability that the animal ij inherited the first allele of sire i and

e_{ij} is the residual.

The 10 000 permutations were used to generate a test-statistic under the null hypothesis and to determine thresholds for both chromosome-wide and experiment-wide Type 1 error rates. The confidence intervals of the QTL locations were estimated using 2 000 bootstraps. The QTL variance was calculated as follows:

$$\text{QTL variance} = 4\left(1 - \frac{\text{Residual MS Full model}}{\text{Residual MS Reduced model}}\right)$$

Results

The phenotypic averages for the mohair product and quality traits of the 12 families belonging to four different breeders are shown in Table 5.1. Families 7-10 were bred by one breeder, and no quality traits were available on these animals. These four families were the smallest sub-sets of the dataset.

Families 5 and 6 had the highest means for FD (coarse fibres) and SF and correspondingly the lowest means for CF. Families 8 and 10 showed the highest means for FW, with little variation between the herds for FD. The lowest means for FD and SF was recorded in families 11 and 12, with correlated low values as expected for FW and high values for CF.

Table 5.1 Summary statistics (means ±SD) for the fleece traits of the 12 Angora goat families

Family	n	FW (kg)	FD (µm)	n	SDFD (µm)	CVFD (%)	CF (%)	SF (µm)	SDA(µm)
1	67	1.27±0.40	28.24±3.16	4	8.15±1.74	31.88±4.17	78.78±14.97	27.63±4.15	1.68±1.23
2	86	1.20±0.29	26.80±2.51	25	7.76±0.99	28.64±3.01	71.52±13.06	28.44±2.82	1.18±0.42
3	79	1.27±0.40	27.09±2.61	11	7.99±0.99	28.11±3.44	68.45±13.78	29.66±2.14	1.51±1.00
4	91	1.35±0.43	25.84±3.24	40	8.35±1.05	31.66±2.99	73.45±12.82	28.55±3.12	1.52±0.68
5	130	1.64±0.44	29.84±2.53	129	7.72±0.89	25.98±2.85	58.57±13.50	30.41±2.44	1.27±0.38
6	80	2.10±0.56	32.60±2.99	80	8.71±1.14	26.80±3.15	46.34±15.20	33.47±2.95	1.60±0.50
7	16	1.73±0.45	29.08±2.10	0	-	-	-	-	-
8	17	2.10±0.37	29.46±2.82	0	-	-	-	-	-
9	29	1.59±0.42	28.28±2.01	0	-	-	-	-	-
10	23	2.47±0.47	29.83±1.94	0	-	-	-	-	-
11	33	1.22±0.18	25.67±2.12	32	7.22±1.16	27.97±3.12	78.08±8.40	26.83±2.59	1.08±0.33
12	44	1.32±0.23	25.14±2.13	44	6.33±0.96	25.17±2.70	81.71±9.32	25.43±2.37	1.02±0.35

Table 5.2 indicates the markers used in the analyses, their locations and genome coverage. Five chromosomes (CHI 9, 21, 22, 28 and 29) were excluded from the study, as only one marker was located on each of these chromosomes and consequently interval mapping was not possible. The intervals between consecutive markers ranged between 1 and 51cM with an average marker interval of 23.0cM, covering a total of 1253cM. The number of informative families used in the GridQTL analysis varied between 11 and 12 per chromosome. The lowest percentage of heterozygous sires on a chromosome was 53% (CHI 8), increasing to 80% on CHI 25. The information content across chromosomes ranged between 35% on CHI 1 and 69% on CHI 26.

Table 5.2 Genome coverage of microsatellite markers

CHI	N markers	% Heterozygous sires[a]	N Informative families	IC[b]	Markers (position in cM)
1	6	57	12	0.35	BM1312 (0), BM3205 (39), CSSM19 (45), CSSM32 (92), MAF64 (113), INRA11 (137)
2	4	54	12	0.40	BMS2782 (0), OARFCB11 (1), INRA40 (11), SRCRSP24 (62)
3	4	71	11	0.47	CSSM54 (0), MCM58 (30), INRA3 (58), INRA6 (74)
4	6	75	12	0.50	OARHH64 (0), OARCP26 (41), MAF50 (50), OARHH35 (82), BMS1788 (97), MAF70 (129)
5	7	57	12	0.42	OARFCB5 (0), LSCV25 (12), BMS1248 (15), BM2830 (65), BMC1009 (90), ILSTS34 (100), BM321 (121)
6	6	71	12	0.58	BM143 (0), BM415 (18), BM1329 (39), BM4621 (67), ILSTS087 (74), SRCRSP08 (81)
7	3	75	11	0.46	INRABERN192 (0), OARAE129 (18), MCM527 (27)
8	3	53	11	0.57	CSSM47 (0), MCM64 (8), SRCRSP10 (26)
11	3	58	11	0.36	INRA177 (0), OARCP34 (4), ILSTS45 (41)
12	5	67	12	0.50	BMS712 (0), INRA5 (24), BMS2252 (43), SRCRSP9 (59), ILSTS33 (77)
13	3	61	11	0.44	IL2RA (0), BMC1222 (25), ILSTS59 (31)
14	2	54	11	0.56	BM4630 (0), ILSTS11 (11)
16	3	75	11	0.39	BM719 (0), BM121 (18), HUJ614 (68)
17	4	75	12	0.52	OARVH98 (0), OARFCB48 (17), ILSTS58 (51), BM8125 (57)
18	3	72	12	0.64	INRA210 (0), INRA63 (11), MCM104 (26)
19	4	64	11	0.40	MCM210 (0), BMS745 (33), LSCV36 (44), OARFCB193 (46)
20	4	75	12	0.44	MAF214 (0), TGLA304 (11), BM1225 (32), BM3517 (60)
23	5	73	12	0.63	OARCP73 (0), OLA-DRB (8), BM1818 (19), DRBP1 (26), BM1258 (40)
24	3	69	12	0.48	MCM136 (0), BMS1332 (38), BMS2526 (41)
25	3	80	12	0.51	BP28 (0), INRA206 (12), TGLA40 (26)
26	4	77	12	0.69	HEL11 (0), INRABERN172 (14), LSCV52 (22), LSCV46 (42)
27	3	78	11	0.55	BM6526 (0), CSSM43 (8), TGLA179 (30)

[a] % of Heterozygous sires averaged over all markers per chromosome
[b] Average Information Content across chromosome

This study identified QTL for mohair traits on thirteen chromosomes at the chromosome-wide significance level (Table 5.3). At least 1 region of interest was detected for each trait included in the analyses. In Table 3 the *F*-statistics, location of the QTL, QTL effect and QTL variance are given. 95% confidence intervals frequently exceeded 50cM and usually included the whole chromosome, and were thus not reported.

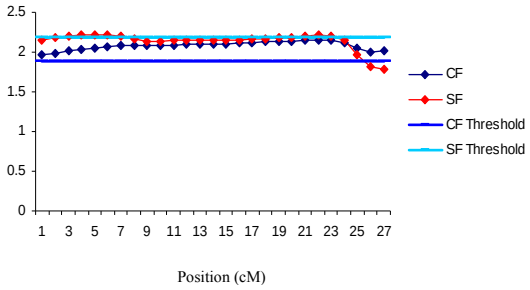
Table 5.3 Putative QTL identified for seven traits by chromosome

CHI	Trait	Position (cM)	*F* Statistic	*F* Threshold	Segregating family	Effect /SD[a]	Variance[b] (%)
1	SDA	121	5.15	5.13*	3	-1.3	33.6
2	FW	1	2.26	2.16*	10	0.4	8.8
3	SDFD	62	2.68	2.67*	4	1.0	14.4
4	FD	71	2.25	2.25*	12	1.7	8.3
5	FW	1	2.52	2.35*	5	0.5	10.3
8	CF	21	2.15	1.88*	5	4.6	9.9
	SF	4	2.22	2.18*	5	-1.3	10.5
12	CF	1	2.22	1.95*	6	0.9	10.5
13	CF	31	2.27	2.02*	6	0. 2	14.6
	SF	31	2.22	2.16*	6	0.03	14.0
16	CF	68	2.15	1.92*	6	0.8	9.9
18	CF	26	2.31	1.84*	6	0.9	11.3
	SF	26	2.07	2.07*	6	-1.1	9.3
20	CF	32	2.15	1.89*	4 & 6	1.3 & 1.1	9.9
	SF	32	2.33	2.24*	4 & 6	-1.3 & -1.4	11.5
24	FW	41	2.93	2.71**	4	0.3	13.4
	FD	31	2.02	1.99*	4	0.7	6.9
25	CVFD	22	2.73	2.47*	4	1.0	14.8

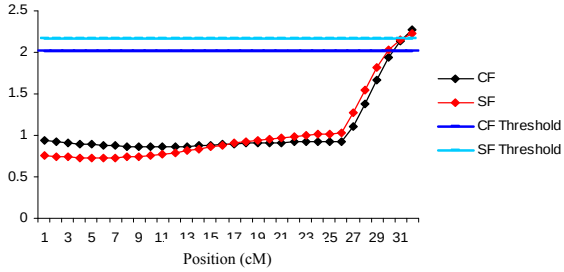
[a] QTL allele substitution effect in significant sire family
[b] % of phenotypic variance explained
* $p < 0.05$ chromosome-wide significance
** $p < 0.01$ chromosome-wide significance

Thirteen putative QTL regions were found in total for the seven traits. Three QTL were identified for FW on chromosomes 2, 5 and 24. Putative QTL for FD were identified on chromosomes 4 and 24. One putative QTL was found for CVFD, SDFD and SDA each on chromosomes 25, 3 and 1 respectively. Chromosomes 8, 13, 18 and 20 each showed segregation for a QTL for both CF and SF. On chromosomes 12 and 16 two more putative QTL were identified for CF. Only one putative QTL (for FW, on chromosome 24) could be identified with $p < 0.01$ chromosome-wide significance, while all other QTL complied to $p < 0.05$, chromosome-wide.

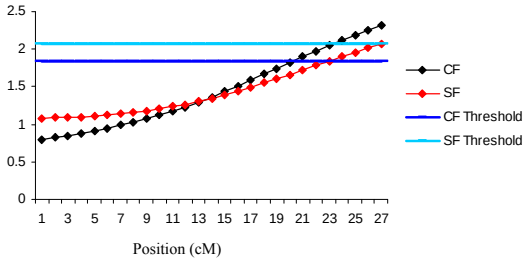
The estimates of QTL contributions to the phenotypic variance ranged between 6.9% for FD on CHI 24 and 33.6% for SDA on CHI 1. Most of the QTL variance estimates were between 9 and 14%. The QTL effects (scaled by the standard deviation of the trait) varied from -1.4 to 4.6 standard deviation units for different traits and families. The plots of the *F*-statistics for the chromosomes with more that one putative QTL are shown in Fig 5.1.



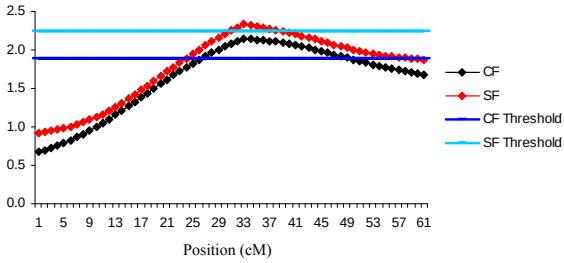
CHI 8



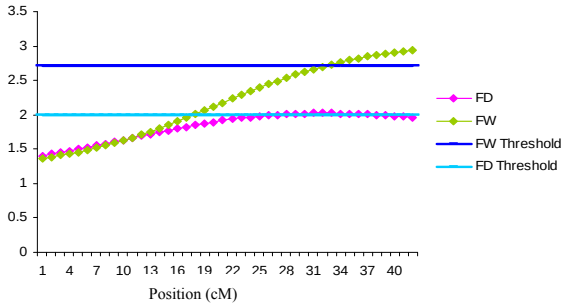
CHI 13



CHI 18



CHI 20



CHI 24

Figure 5.1 F-statistics depicting the locations of the putative QTL

Discussion

In this study out-bred populations were analysed, in contrast to the back-cross populations often used in QTL investigations. Findings reported therefore represent genes segregating in the South African Angora goat population and results can be exploited by means of selection. Heritability estimates reported for these traits in the South African Angora goat population ranged between 0.14 (SDA) and 0.63 (CF) (Visser *et al.,* 2009). The size of the out-bred population directly affects the statistical power of the experimental design (Van der Werf *et al.,* 2007). It was estimated that this study (consisting of 12 sires with an average of 58 offspring, average heritability of 0.35, Type I error of 0.05) will achieve a power of 70% to detect a QTL effect of 0.5 phenotypic SD (Weller *et al.,* 1990). Most of the QTL detected had larger effects than this. This design compared favourably with

those of Cano *et al.* (2007), Bidinost *et al.* (2008), Mohammad Abadi *et al.* (2009) and Roldan *et al.* (2010).

The phenotypic averages over families for FW and FD were in the same range as those previously reported by Snyman & Olivier (1996, 1999) and Visser *et al.* (2009) on South African Angora goats. Differences between families are due to herd effects, breeders following different selection strategies and placing emphasis on varying selection criteria, with limited genetic linkages between farms. Previous reports on QTL associated with mohair traits are limited to one research group (a collaboration between Argentina and France (Cano *et al.,* 2007; Cano *et al.,* 2009; Debenedetti *et al.,* 2010)) and traits investigated varied between studies. In this study several putative QTL were identified for CF, SF, FW and FD and one each for SDA, SDFD and CVFD. It is interesting to note that most QTL detected in the study were segregating in families 4 (fine fibre producers) and / or family 6 (strong fibre producers). These two families were also amongst the largest included, and it was thus more likely that a segregating QTL could be detected in them.

Fleece weight remains one of the major traits that affect profit in the Angora goat industry. Three QTL for FW (on CHI 2, 5 and 24) were identified in this study. The putative QTL on CHI 5 had the largest effect (0.5) on this trait and explained 10.3% of the variance for this trait, while the QTL on CHI 2 and 24 explained between 8.8% and 13.4% of the variance respectively. FW was not included in the study on Argentinean Angora goats by Cano *et al.* (2007). Chromosome 5 was however investigated by Debenedetti *et al.* (2010) and QTL affecting FW were reported in his study using a backcross population. These results are supported by the study of Mohammad Abadi *et al.* (2009) which targeted chromosomes 1, 2, 5 and 13 in a search for QTL affecting cashmere traits in Rayini goats. Two putative QTL for cashmere yield was identified on CHI 2 and CHI 5 respectively. The current study reports the first putative QTL segregating on CHI 24 for FW.

Two QTL associated with FD were identified on CHI 4 and 24 respectively in this study. To date no QTL have been detected for mohair fibre diameter in purebred Angora goats. A putative QTL for FD was reported by Debenedetti *et al.* (2010) on CHI 5 in an Angora x Creole backcross population where only CHI 5 was investigated. QTL for wool fibre diameter have also been reported by Bidinost *et al.* (2008) on OAR 3 for FD at first shearing, and Roldan *et al.* (2010) on OAR 11 for FD at second shearing. When comparing QTL detected for the same traits at different shearing periods, Bidinost *et al.* (2008) reported that for some fleece traits QTL were only revealed at an early age, and not later in life and concluded that different genes could possibly be influencing the same trait at different stages in life. The discrepancy in detecting a QTL for FD between the current study and that of Cano *et al.* (2007) could be due to phenotypic traits measured at either second or third shearing

in the current study, while samples were taken at 4 months of age in the study by Cano *et al.* (2007). It should also be noted that all but one of the QTL detected was at chromosome-wide significance level, as was those reported by Cano *et al.,* (2007, 2009) Further studies should focus on confirming the reported QTL at genome-wide significance levels in experimental designs with higher statistical power.

The unfavourable positive genetic correlation between FD and FW has been a challenge in Angora goat breeding strategies. Medium to strong genetic correlations have been reported, ranging between 0.35 (Allain & Roguet, 2003) and 0.55 (Snyman & Olivier, 1996). These traits are the main price-determining factors in the mohair industry, and optimisation of both has proved difficult. The detection of a QTL for FD on a separate chromosome (CHI 4) as for FW, might pose an opportunity to decrease or maintain fibre diameter in Angora goats using marker assisted selection, while increasing fleece weight. It is however possible that a QTL affecting FW could also reside on CHI 4, but that the experiment didn't have sufficient statistical power to detect it. The second putative QTL for FD reported here is on the same chromosome as a QTL for FW (CHI 24).

Processing and fabric properties are influenced significantly by fibre diameter distribution (Qi *et al.,* 1994; Smith *et al.,* 2006) which are measured using OFDA2000 technology. The use of marker assisted selection to directly improve traits correlated with more uniform production (CVFD, SDFD, SDA) can contribute to a higher quality clip and improved attributes when compared to synthetic fibres. A putative QTL for CVFD was identified on CHI 25 in the current study. Two putative QTL for CVFD on CHI 1 and CHI 13 respectively were reported by Cano *et al.* (2007), while a QTL affecting CVFD was identified by Cano *et al.* (2009) in a further investigation of CHI 19, which was not included in the current study. The quality traits CF, SF, SDA and SDFD have not been included in previous QTL identification studies. The QTL for SDA segregating on CHI 1 explains 33.6% of the variance in the trait and is therefore the largest detected QTL effect in this study. This value is probably an overestimation, and likely due to additional segregating QTL close to the putative position. A two-linked-QTL hypothesis should be tested for this trait. The amount of variance explained suggests that this chromosome segment could play an important role in improving fleece uniformity through MAS and should be further investigated. SDA has the lowest heritability of the quality traits included (Visser *et al.,* 2009) due to small additive variance component for this trait. Quantitative selection for decreased variation in fibre diameter along the staple has not been successful and MAS can make a significant contribution to this trait.

The putative QTL identified for CF on CHI 8 has a relatively large effect of 4.6 that could be due to the extensive variation for this trait (22% to 90%) present in family 5and possibly resulted in an

overestimation. On four chromosomes (CHI 8, 13, 18 and 20) two putative QTL influencing CF and SF respectively, were detected. Taking into account the high genetic correlation (-0.97) between these traits (Visser *et al.,* 2009) and the close proximity of the estimated positions, it is probable that these QTL are single QTL affecting both traits. Two additional QTL were however identified influencing only CF (one each on CHI 12 and 16), suggesting that the two traits are under different genetic control, and can not be considered as one. Spinning fineness and comfort factor are both unfavourably correlated with fleece weight. The identification of QTL which can be used to increase skin-comfort traits without a negative correlated response in yield, might lead to a significant improvement in selection efficiency.

Mapping of keratin (KRT) and keratin-associated protein (KAP) genes were performed in sheep, as these were expected to play an important role in wool traits. High glycine-tyrocine (HGT) KAPs as well as trichohyalin (THH, a wool follicle protein) were mapped to ovine chromosome 1, while the high sulphur KAP families were shown to reside on OAR 3 and 19 (McLaren *et al.,* 1997). A significant effect between high HGT loci and wool FD was observed by Parsons *et al.* (1994). Putative QTL for wool production and quality traits tend to cluster on OAR 1 ,3, 4, 11 and 25 (Allain *et al.,* 2006; Bidinost *et al.,* 2008; Roldan *et al.,* 2010), although a few have been reported on other chromosomes (Purvis & Franklin, 2005). The overall resemblance between the ovine and caprine linkage maps are good, with OAR 1 mapping to CHI 1 and 3; OAR 3 to CHI 5 and 11; OAR 4 to CHI 4, OAR 11 to CHI 19 and OAR 25 to CHI 28. Some of the KRT and KAP genes have been assigned to the goat genome, specifically CHI 1 and 5 (Cano *et al.,* 2007), and it was expected to find fibre-associated QTL on these chromosomes as well as the equivalents of the ovine chromosomes mentioned above. The results presented in this paper support the suggestion that KRT and KAP could be possible candidate genes for fibre yield and quality and should receive further attention in fibre-producing goats.

Molecular studies are performed at a high cost and using out-bred populations often limit the potential of having large numbers of offspring per sire with complete phenotypic data, especially OFDA data at an additional cost. Genotyping more offspring per family and larger family sizes will be advantageous for further studies. The large confidence intervals for the putative QTL were unfavourable, and could be improved by adding more markers and increased genome coverage. This study was quite robust and all but one QTL was verified at chromosome-wide significance level. The large QTL effect for SDA and variance estimate for CF (CHI8) appears to be over-estimations and could be further investigated in QTL verification and fine-mapping studies. When SNP arrays become available for goats, association studies could possibly be considered as an alternative to QTL studies. Their applicability in the South African goat industry will however depend on overcoming financial

and resource constraints. This study presented sufficient evidence for QTL for mohair traits to warrant further study that will focus on fine mapping of these candidate regions.

Conclusion

Identifying causative mutations for economically important traits is dependent on QTL identification and mapping. The results of this study indicated several QTL of medium effect influencing mohair production and quality, and explain some of the genetic variance (ranging between 6.9 & and 33.6%) in mohair traits. The first two QTL influencing FD in mohair was reported in this study. One of these QTL was not linked to a QTL affecting FW, posing an opportunity to improve one trait without a negative correlated response in the other. The unfavourable correlation between some quality traits and fleece yield can be addressed in the same way. Further studies should be conducted to fine-map these regions and detect favourable alleles, that can then be incorporated into selection strategies through marker-assisted selection.

As the goal of fine mohair production becomes more attainable, mohair quality will play a more important role in the industry. The opportunity of developing natural fibres with new properties through gene-introgression may also become a reality in the near future. To satisfy changing market demands, a better understanding of genes involved in fibre production is needed and should be addressed by future research.

References

Alan, M.F. & Smith, T.P.L., 2008. Present and future applications of DNA technologies to improve beef production. *Meat Science* 80, 79-85.

Allain, D. & Roguet, J.M., 2003. Genetic and non-genetic factors influencing mohair production traits within the national selection scheme of Angora goats in France. *Livest Prod Sci* 82, 129-137.

Allain, D., Schibler, L., Mura, L., Barillet, F., Sechi, T., Rupp, R., Casu, S., Cribiu, E. & Carta, A., 2006. QTL detection with DNA markers for wool traits in a sheep backcross Sarda x Lacaune resource population. In *Proc 8th World Congress on Genetics Aplied to Livestock Production. August 13-18, 2006. Belo Horizonte, Brazil* Communication 05-07.

Andersson, L., 2001. Genetic dissection of phenotypic diversity in farm animals. Nat Rev Genet 2, 130-138

Bidinost, F., Roldan, D.L., Dodero, A.M., Cano, E.M., Taddeo, H.R., Mueller, J.P. & Poli, M.A.,2006 Quantitative trait Loci related to Merino sheep wool quality. In *Proc 8th World Congress on Genetics Aplied to Livestock Production. August 13-18, 2006. Belo Horizonte, Brazil* Communication 05-11.

Bidinost, F., Roldan, D.L., Dodero, A.M., Cano, E.M., Taddeo, H.R., Mueller, J.P. & Poli, M.A., 2008. Wool quantitative loci in Merino sheep. *Small Rumin. Res.* 74, 113-118.

Cano, E.M., Daverio, S., Cáceres, M., Debenedetti, S., Costoya, S., Abad, M., Allain, D., Taddeo, H. & Poli, M.A., 2009. Detection of QTL affecting fleece traits on CHI 19 in Angora goats. *Tropical and Subtropical Agroecosystems* 11, 189-191.

Cano, E.M., Marrube, G., Roldan, D.L., Bidinost, F., Abad, M., Allain, D., Vaiman, D., Taddeo, H. & Poli, M.A., 2007. QTL affecting fleece traits in Angora goats. *Small Rumin. Res.* 71, 158-164.

Debenedetti, S., Cano, E.M., Abad, M., Allain, D., Taddeo, H. & Poli, M., 2010. Detection of QTL affecting fleece traits on CHI 5 in a backcross Angora x Creole goats in Argentina – Preliminary results. In *Proc 9th World Congress on Genetics Aplied to Livestock Production. August 1-6, 2010. Leipzig, Germany* Communication 0764.

Dekkers, J.C.M., 2004. Commercial application of marker- and gene-assisted selection in livestock: Strategies and lessons. *J. Anim. Sci.* 82 (E Suppl): E313-328.

Dodds, K.G., McEwan, J.C. & Davis, G.H., 2007. Integration of molecular and quantitative information in sheep and goat industry breeding programmes. *Small Rumin. Res*. 70, 32-41.

Goddard, M., 2009. Genomic selection: prediction of accuracy and maximisation of long term response. *Genetica* 136, 245-257.

Gong, H., Zhou, H. & Hickford, J.G.H., 2010. Diversity of the glycine/tyrosine rich keratin-associated protein 6 gene (KAP6) family in sheep. *Mol Biol Rep.* DOI 10.1007/s11033-010-0074-6.

Itenge-Mweza, T.O., Forrest, R.H.J., McKenzie, G.W., Hogan, A., Abbott, J., Amoafo, O. & Hickford, J.G.H., 2007. Polymorphism of the KAP1.1, KAP1.3 and K33 genes in Merino sheep. *Molecular and Cellular Probes.* 21, 338-342.

Jeon, J.T., Lee, J.H., Kim, K.S., Park, C.K. & Oh, S.J., 2006. Application of DNA markers in animal industries. *Aust J Exp Agric.* 46, 173-182.

Jin, M., Wang, L., Li, S., Xing, M.X. & Zhang, X., 2010. Characterization and expression analysis of KAP7.1, KAP8.2 gene in Liaoning new-breeding cashmere goat hair follicle. *Mol Biol Rep.* DOI 10/1007/s11033-010-9968-6.

Knott, S.A., Elsen, J.M. & Haley, C.S., 1996. Methods for multiple-mapping of quantitative trait loci in half-sib populations. *Theor. Appl. Genet.* 93, 71-80.

Marrube, G., Cano, E.M., Roldan, D.L., Bidinost, F., Abad, M., Allain, D., Vaiman, D., Taddeo, H. & Poli, M.A., 2007. QTL affecting conformation traits in Angora goats. *Small Rumin. Res.* 71, 255-263.

McLaren,R.J., Rogers, G.R., Davies, K.P., Maddox, J.F. & Montgomery, G.W., 1997. Linkage mapping of wool keratin and keratin-associated protein genes in sheep. *Mamm Genome* 8, 938-940.

Mohammed Abadi, M.R., Askari, N., Baghizadeh, A. & Esmailizadeh, A.K., 2009. A directed search around caprine candidate loci provided evidence for microsatellites linkage to growth and cashmere yield in Rayini goats. *Small Rumin. Res.* 81, 146-151.

Parsons, Y.M., Cooper, D.W. & Piper, L.R., 1994. Evidence of linkage between high-glycine-tyrosine keratin gene loci and wool fibre diameter in a Merino half-sib family. *Anim Genet* 25, 105-108.

Pollak, E.J., 2005. Application and impact of new genetic technologies on beef cattle breeding: a "real world" perspective. *Aust J Exp Agric.* 45, 739-748.

Purvis, I.W. & Franklin, I.R., 2005. Major genes and QTL influencing wool production and quality: a review. *Genet. Sel. Evol.* 37 (Suppl 1), S97-S107.

Purvis, I.W. & Jeffery, N., 2007. Genetics of fibre production in sheep and goats. *Small Rumin. Res.* 70, 42-47.

Qi, K., Lupton, C.J., Pfeiffer, F.A., Minikhiem, D.L., 1994. Evaluation of the Optical Fibre Diameter Analyser (OFDA) for measuring fiber diameter parameters of sheep and goats. *J. Anim. Sci.* 72, 1675-1679.

Roldan, D.L., Dodero, A.M., Bidinost, F., Taddeo, H.R., Allain, D., Poli, M.A. & Elsen, J.M., 2010. Merino sheep: a further look at quantitative trait loci for wool production. *Animal* 4:8, 1330-1340.

Seaton G., Hernandez J., Grunchec J.A., White I., Allen J., De Koning D.J., Wei W., Berry D., Haley C. & Knott S., 2006. GridQTL: A Grid Portal for QTL Mapping of Compute Intensive Datasets. *Proceedings of the 8th World Congress on Genetics Applied to Livestock Production, August 13-18, 2006. Belo Horizonte, Brazil.*

Smith, J.L., Purvis, I.W., Lee, G.J., 2006. Fibre diameter profiles – potential applications for improving fine-wool quality. *International Journal of Sheep and Wool Science* 54, 54-61.

Snyman, M.A. & Olivier, J.J., 1996. Genetic parameters for body weight, fleece weight and fibre diameter in South African Angora goats. *Livest. Prod. Sci.* 47, 1-6.

Snyman, M.A. & Olivier, J.J., 1999. Repeatability and heritability of objective and subjective fleece traits and body weight in South African Angora goats. *Small Rumin. Res*. 34 103-109.

Snyman, M.A., 2002. Evaluation of a genetically fine mohair producing herd. *Small Rumin. Res*. 43, 105-113.

Van der Werf, J.H.J., 2007. Marker assisted selection in sheep and goats. In: Marker-Assisted Selection: Current status and future perspectives in crops, livestock, forestry and fish. Eds. Guimarães, E., Ruane, J., Scherf, B., Sonnino, A. & Dargie, J. FAO, Rome, Italy. pp. 230-247.

Van der Werf, J.H.J., Marshall, K. & Lee, S., 2007. Methods and experimental designs for detection of QTL in sheep and goats. *Small Rumin Res.* 70, 21-31.

Visser, C., Snyman, M.A., Van Marle-Köster, E. & Bovenhuis, H., 2009. Genetic parameters for physical and quality traits of mohair in South African Angora goats. *Small Rumin Res* 87, 27-32.

Visser, C. & Van Marle-Köster, E., 2009. Genetic variation of the reference population for QTL research in SA Angora goats. *AGRI* 45, 113-119.

Visser, C., Crooijmans R.P.M.A. & Van Marle Köster, E., 2010. A genetic linkage map for the South African Angora goat. *Small Rumin Res*. 93, 171-179.

Weller, J.I., Kashi, Y. & Soller, M., 1990. Power of daughter and granddaughter designs for determining linkage between marker loci and quantitative trait loci in dairy cattle. *J. Dairy Sci* 73: 2525-2537.

Chapter 6: Critical review and recommendations

6.1. Critical Review

Quantitative theory has been the basis for genetic improvement of livestock for many decades and animal breeders world-wide moved from the subjective determination of genetic merit to objectively measuring phenotypes and ultimately selection based on BLUP EBVs. Selection based on recorded data for specifically chosen criteria was a major advantage and lead to genetic progress in many economically important traits. The application of standard EBVs can however be a relatively slow and costly procedure (Allan & Smith, 2008). Moreover, the additive model of inheritance used in BLUP analysis has been shown to have inherent limitations, and it has been acknowledged that individual genes with noticeable effects on quantitative traits can be identified (Dodds *et al.,* 2007). DNA technology can be applied in genetic improvement programs in various ways, from parentage verification and traceability to the identification of QTL and whole-genome selection. The major aim of these practices is to improve EBV accuracies and selection efficiency, resulting in faster genetic progress and reduced cost of altering genetics (Allan & Smith, 2008).

The South African Angora goat industry has relied on mass selection to genetically improve mohair-producing goats over the past few decades. Selection practices have evolved over time from an over-emphasis on fibre diameter (resulting in small, unthrifty animals) to a more balanced selection index approach. Currently three traits (fleece weight, fibre diameter and body weight) are included in the index with moderate success in the national herd. An evaluation of the goats subjected to selection based on this index has indicated that fibre diameter can be decreased without a decrease in body weight (Snyman, 2002). In order to maintain the high quality clip that is known as a reliable source of mohair worldwide, quality traits are currently investigated for possible inclusion in the selection strategy. The use of quantitative information has progressed over time from mass selection to the application of BLUP breeding value estimation in the stud industry. The South African Angora goat stud breeders have however very limited participation in the national small-stock recording scheme and little use is made of EBVs.

Although the global mohair industry has a relatively stable niche market, natural fibres are under severe pressure from competing synthetic textiles. The mohair industry needs to align its production profile to the market demands, which include a greater focus on quality and uniformity of fibres. In order to maintain their global stronghold, the South African mohair industry assigned priority to projects for investigation and application of new technology (including molecular technology) which resulted in the current research project.

Even though the advantages of molecular technology have been proven in other species, small stock and especially goats have received limited interest from researchers. The absence of a densely populated caprine linkage map is an indicator of how far this species is lagging behind the more economically important livestock breeds and species (e.g. dairy and beef cattle, swine and poultry). The microsatellites that are available are currently used mostly in small-scale (local or national) projects for parentage verification and genetic diversity studies. The dearth of information on fibre goats - both on quantitative and molecular level - has contributed to the challenges of this study. The Angora goat is the only mohair producing animal, and research on this breed is limited to France, Argentina and South Africa. Very little data was available for comparison, with much variation between traits investigated, population sizes and statistical models used in the few published studies available.

It was realized very early on during the planning of this project that the absence of a bio-bank and reference population would be a severe limitation to the study. At that stage no DNA samples were routinely kept for any livestock species in the country, and pedigree and performance records were only available for performance recorded animals. Although fleece testing has been practised in South Africa from as early as 1934 and a formal fleece testing facility was established in 1965 (Schoeman *et al.,* 2010), participation in the National Small Stock Improvement Scheme (NSIS) is very limited in the Angora goat industry. Fleece traits recorded in the NSIS include fleece weight, fibre diameter and staple length, but no OFDA-measured traits. Farmers need to pay an additional fee for the measurement of these quality traits. Only four Angora goat breeders participate in the NSIS, with an average of 140 does per breeder (Schoeman *et al.,* 2010). With the QTL detection phase of the study in mind, a reference population was established. As a paternal half-sib design was followed, a specific population structure needed to be generated. The challenge of QTL design in out-bred populations lies therein that on the one hand there is a force towards a small number of sire families with large progeny groups, while on the other hand there is the probability that the sires used in the project is not heterozygous for the QTL (Weller, 2001). The size of a QTL experiment is however limited by the high costs of genotyping large numbers of animals. Therefore, the number of reference families, their sizes and the number of microsatellite markers that could be used were optimized within these constraints.

Firstly stud breeders were selected who were known to keep accurate records. These breeders had to use a number of specific sires over a three year period, in order to generate sufficient lines with the required number of offspring. Natural mortalities of some of the sires occurred, as well as less than expected progeny per kidding season for some sires. In these cases, alternative sires had to be selected and mated with an increased number of does during the next mating period. These problems associated

with natural populations resulted in a three year waiting period before genotyping of the selected families could commence. Blood was collected after each kidding season, and a positive spin-off was the creation of a small-stock bio-bank at the National Department of Agriculture, Forestry and Fisheries' facilities in Middelburg, Eastern Cape. Blood and DNA samples, as well as phenotypic and pedigree records are now routinely kept, and will greatly assist future molecular projects.

A genetic map forms the foundation for DNA research for any species, detailing the specific DNA marker positions on the genome. The relatively under-developed goat linkage map thus poses a limitation for most molecular studies on goats, including the current project. One of the aims of the study was to improve the status of the map, and to verify previously reported discrepancies with the sheep map. The use of a relatively large number of animals for this study resulted in a significant improvement in the number of informative meiosis and shorter mapping distances. This lead to the generation of an improved linkage map which provides the basis for an advanced linkage map. Large marker intervals between successive microsatellite markers still exist and the main challenge to incorporating more markers in this study was a financial constraint. Funding was obtained for the study, but South Africa is still a developing country with limited financial resources especially for scientific research. Another limitation to the improvement of the goat linkage map is the absence of a published SNP panel. No results of SNP discovery projects in goats have yet been reported, limiting many molecular applications in goats.

Genetic parameter estimation was crucial as newly measured OFDA traits have not yet been included in previous studies. These traits are becoming more important as the focus on mohair moves from quantity to quality, and should be considered for possible inclusion into the breed selection index. The quality traits have only been measured in South Africa since the late 1990s and relatively few records are available. Another reason for the limited number of records is that measuring of these is not compulsory in the performance testing scheme and farmers have to pay an additional fee to have them measured. Even though some farmers paid for the extra traits to be measured, they did not use consequent numbering for the fleeces, and thus sample data could not be matched to original individual animals. The loss of data impacted on both the estimation of variance components and the QTL identification study. Despite these challenges, genetic parameters for OFDA-measured quality traits were estimated for the first time in South Africa. Some of these quality traits should receive consideration for inclusion in a selection index for increased genetic progress in South African Angora goats.

The QTL identification phase of the study required the merging of phenotypic and genotypic data. The use of natural out-bred populations generated by breeders as a reference posed a limitation

as there was no control over available phenotypic records. Some offspring were sold or slaughtered at a young age, before traits were expressed and thus have a genotype without any fleece records. Although birth and weaning records were complete, later measured growth traits may also be incomplete. Fortunately all the growth records can still be used in the next phase of the study, which will focus on identification of QTL linked to growth traits. An increase in animal numbers will increase the statistical power of the study significantly and should result in the detection of QTL with smaller effects.

The International Goat Genome Consortium (IGGC) is currently busy with a goat genome mapping project and it is expected that a large number of caprine SNPs will be available in the near future. A draft reference genome has been completed and twenty five breeds worldwide are now sought for re-sequencing to complete the project (Wenguang Zhang, 2010. Personal communication). A complete SNP panel will allow high density genome scans and will improve the statistical power of QTL identification studies significantly. Fine-mapping of regions of interest identified in recent studies will also become a more attainable prospect, with much denser coverage. A complete genome scan using SNP might even be more cost effective than the current approach of initial scanning, linkage analyses and subsequent fine-mapping (Van der Werf *et al.,* 2007).

6.2. Conclusion

In this study a molecular approach was followed for genetic improvement in South African Angora goats, with emphasis on mohair traits. This was the first attempt to apply molecular techniques in South African Angoras and despite the limitations of using a natural out-bred population, the objectives of the study were met. The reference population established proved to have adequate genetic variation and the accuracy of the goat linkage map was improved by adding new markers and verifying inter-chromosomal re-arrangements on specific segments. Genetic parameters were also calculated for OFDA-measured mohair traits, which were not previously available. QTL for a number of fibre traits could be identified and some of these show promise for MAS, especially as unfavourable genetic correlations could possibly be overcome. Fine-mapping of the regions of interest detected in this study should now form the next phase, in order to identify favourable alleles that can be used for MAS. The outcomes of this study make a valuable contribution to the scientific knowledge of Angora goats, and can be applied in the South African goat industry.

Research areas which should receive attention in future include a QTL identification study for growth traits (which is in planning) and an investigation into the reproductive inefficiency of Angora goats. Due to limited recording of fitness traits, QTL identification for reproduction traits is challenging. The economic impact of the high mortality rate of ewes however warrants attention and

breeders are encouraged to record abortions, miscarriages and stillborn lambs. As the reproduction data is accumulated, it is envisaged that a future study identifying these QTL would be possible. The continued recording of phenotypic data and storage of DNA samples will assist future molecular studies. The sampling of specific phenotypic outliers (e.g. unthrifty animals, does that tend to abort) and DNA analyses of these should also lead to an increased knowledge of the underlying molecular mechanisms.

In order for the QTL identified in this study to have an impact on Angora goat genetics in South Africa, fine-mapping of the identified regions of interest should follow. As the QTL were detected on chromosome-wide, and not genome-wide, significance levels some of the putative QTL might turn out as false positives. Other identified regions should however be confirmed as regions contributing to phenotypic variation. Fine-mapping will firstly focus on CHI 1, where a putative QTL for variation along the fibre (SDA) has been identified. This was the QTL with the largest contribution to phenotypic variation, and also the trait with the lowest estimated heritability. Genetic progress using quantitative selection has been slow as environmental influences play an important role in within-fibre variation. The trait is however of economic importance as it influences processing ability due to mainly its influence on staple strength and hauteur. It is envisaged that fine-mapping will be performed with SNPs, which will contribute to a large extent to improving the density of markers, and more accurate fine-mapping.

The generation of publicly available SNP panels for whole genome selection could further contribute to the future of selection in Angora goats. The SNP assays and development of prediction equations would however require vast amounts of commitment and funding from both industry and funding bodies. This is not expected to become a reality for a historically under-researched species in the near future. South African Angora goat farmers are facing many challenges, including farming under harsh, extensive conditions, unfavourable current economic environment and changing land-use patterns. Support for an expensive selection technique is thus expected to be limited, at least until its economic value has been proven. In this scenario, farmers might be more willing to implement MAS or GAS where a direct improvement can be expected. This will surely only be the case for QTL with considerable effects on economically important traits.

The future for South African Angora goat genetics should be based on a comprehensive, integrated approach where both quantitative and molecular tools should be applied for genetic improvement of South African Angora goats.

References

Allan, M.F. & Smith, T.P.L., 2008. Present and future applications of DNA technologies to improve beef production. *Meat Science* 80, 79-85.

Dodds, K.G., McEwan, J.C. & Davis, G.H., 2007. Integration of molecular and quantitative information in sheep and goat industry breeding programmes. *Small Rumin Res.* 70, 32-41.

Schoeman, S.J., Cloete, S.W.P. & Olivier, J.J., 2010. Returns on investment in sheep and goat breeding in South Africa. *Livest. Sci.* 130, 70-82.

Snyman, M.A., 2002. Evaluation of a genetically fine mohair producing herd. *Small Rumin Res.* 43, 105-113.

Van der Werf, J.H.J., Marshall, K. & Lee, S., 2007. Methods and experimental designs for detection of QTL in sheep and goats. *Small Rumin Res.*70, 21-31.

Weller, J.I., 2001. Quantitative Trait Loci Analysis in Animals. CABI Publishing, UK.

ADDENDUM A: MOHAIR CLASSING STANDARDS

(http://www.mohair.co.za/index.php/home/growers-producers/classing-standards)

1. CLASSING STANDARDS

To achieve uniformity with the classing of mohair, it must be classed according to regulations laid down by law. (Act on Agricultural Product Standards 1990 - Act no 119).

The object is to class each lot as evenly as possible according to the physical characteristics of mohair, with the correct content marks on bales and bags, thereby creating the necessary confidence.

2. PHYSICAL CHARACTERISTICS OF MOHAIR

The following physical characteristics play an important role in the classing of mohair:

2.1 Fineness
2.2 Length
2.3 Style-and-character
2.4 General appearance

2.1 FINENESS

Fineness is the most important characteristic which must be taken into account in the classing of mohair. To define fineness is difficult if the classer does not have the necessary knowledge. The finest hair is obtained from Kids shorn for the first time at the age of six months. As the animal grows older, the hair becomes stronger.

Within each age group fineness can be determined by the following method: The thinner the staple and the fibre and the softer the handle, the finer the hair. Open, webbed fleeces of very soft handle are usually the finest hair.

2.2 LENGTH

The ideal length for mohair is from 125 mm to 150 mm. The trade prefers mohair to be not too short or too long. Short hair is unacceptable.

Length is a characteristic that can be measured and the different lengths with the corresponding symbol, are the following:

SYMBOL	LENGTH
A =	+150 MM
B =	125MM - 150 MM
C =	100 MM - 125 MM
D =	75MM - 100MM
E =	50MM - 75MM

In each class the length should not differ with more than 25 mm.

2.3 STYLE-AND-CHARACTER

Style is the twist of the staple and character the crimp or wave of the staple. The ideal is a combination of twist and even character within a soft but nevertheless firm staple. Too much character results in spongy mohair which is an undesirable type of hair. Older Angora goats of good style-and-character continue producing hair of good quality, whilst excess style causes the goats to produce hair of a poor quality, even at a young age.

2.4 GENERAL APPEARANCE

General appearance is determined by the following:

2.4.1 Lustre.
2.4.2 Absence of foreign fibres.
2.4.3 Condition of hair.
2.4.4 Dust, stain and seed.

2.4.1 LUSTRE

Mohair must have a bright lustre and not be dull in appearance. This lustre is very important in the processing of mohair as it accentuates the colour the manufacturer requires. This is one of the reasons why mohair is so much sought after as textile fibre. 2.4.2 Absence of foreign fibres

Mohair must be free of kemp, black and brown fibres or any other foreign fibres. Foreign fibres can harm the end-product considerably and are easily discernable once the hair has been washed and combed. Kemp does not absorb dyes and can therefore easily be noticed after the dyeing process because it appears as lighter uncoloured fibres in the end-product.

2.4.2 CONDITION OF HAIR

Mohair must contain enough natural oil to be hardly noticeable. This natural oil protects the fibre against weathering and ensures healthy fibres for processing.

2.4.3 DUST, STAIN & SEED

Dust - must be limited to the minimum. Goats must therefore not be driven on dusty roads or into kraals that have not been dampened. Dust can, however, be washed out reasonably effectively by dipping the goats prior to shearing.

Stain - Avoid stained hair by not herding flock while the veld is wet with dew. Stained hair can also be reduced by crutching the goats at 3 months growth.

Seed - can be avoided by keeping the goats in clean camps till after shearing; goats with long hair should not be kept in spared camps or in old lands. After general rains seed can appear which can be detrimental to the value and quality of the clip.

3. COMPOSITION OF THE FLOCK

A flock of Angora goats consists of the following:

3.1 Adult goats - Goats of 24 months and older.
3.2 Young goats - Goats of 18 months (third shear).
3.3 Kids - Can be sub-divided as follows:
(i) Six months old (First shear)
(ii) Twelve months old (Second shear)

The composition of the flock determines, to a large degree, the fineness (quality) of the clip. The younger the goats and by restricting the wethers and over strong ewes in the flock to a minimum, the finer the clipping will be. The object must always be to produce fine quality hair which is always in demand.

4. UNIFORMITY IN THE FLOCK

By classing the flock regularly, the undesirable goats such as those with overstrong hair, too much kemp, spongy hair, excessive oily hair, etc. can be culled out, enabling a uniform flock being built up. It can also, to a large extent, help by using the same type of ram every season.

A uniform flock simplifies the classing of the clip, enabling larger lines to be made of every type.

5. LENGTH OF HAIR

The trade prefers hair not to be too short nor too long. By shearing every six months the desired length is obtained. In some areas where the vegetation is too dense or in the Noorsveld of Jansenville, goats may be shorn at 4 months, if the hair tends to comb out. It is, however, not a practice that is recommended, unless circumstances justify it. If at all possible, even in these areas longer hair should be grown, as a glut of short hair can have a negative effect on sale prices.

6. PRODUCTION CONDITIONS

Feeding is the only factor having the greatest influence on the fineness of the hair. Under very good grazing conditions, the hair is inclined to grow stronger, whereas during periods of drought, the hair is of a finer quality and more dusty. After general rains, the clip will contain more seed and stain.

To enhance the attractive appearance of the clip, it is recommended that mohair from different farms or even different camps, eg mountain veld and plains, which give the hair a different colour, be packed separately.

7. HEALTH OF THE FLOCK

To shear an attractive clip of hair, the goats must be free of any internal or external parasites. Dose the goats to a program and dip before lice are noticed. Neglecting to do this can result in great financial loss.

8. SHEARING-SHED AND REQUIREMENTS.

Light: To class mohair properly, a spacious shearing-shed with adequate lighting is required, to be able to define the characteristics and faults of the hair clearly.

Floor: The floor must be clean and care must be taken that there is no oil, skin salt, dust or rubbish which could contaminate the hair. Tables: It is very important that sufficient tables be available and at

least three are recommended - one for the fleeces, one for the bellies and pieces and one for the stained hair and Lox. The height of the table is determined by the height of the classer and the top must be the approximate height of the classer's waist (belt) and must be made of mesh wire 2 x 2 cm. The perimeter of the table must be approximately 215 x 105 cm.

Bins: Sufficient bins must be available. They should be placed so to have sufficient natural light to be able to compare easily the contents, (fineness, etc) of the different bins (lines). It is recommended that the bins be made from loose, movable frames, covered with wire mesh, which are ideal.

ADDENDUM B: GENOTYPING SETS

Primer	Label	TA	Sire Range	Set
BMS2258	6FAM	56	125-149	1
INRABERN192	PET	56	178-199	1
BM0719	NED	56	133-141	1
BM143	NED	56	93-101	1
OarVH098	6FAM	56	158-169	1
OarHH64	PET	56	123-131	1
BMS0712	VIC	56	167-182	1
BM6526	NED	56	155-186	1
OarAE129	VIC	54	136-156	1
IL2RA	6FAM	50	183-195	1
MAF214	NED	54	233-302	1
OARFCB005	PET	56	90-120	1
MCM527	NED	56	152-168	2
INRA177	NED	56	171-191	2
BM4630	VIC	54	133-135	2
INRA210	NED	54	140-147	2
TGLA304	6FAM	56	91-96	2
OarCP26	VIC	56	146-155	2
INRA005	6FAM	54	135-138	2
OarFCB48	PET	60	154-167	2
HEL11	VIC	56	158-172	2
CSSM43	PET	56	230-241	2
BM415	6FAM	50	114-128	2
BP28	NED	50	209-223	2
LSCV25	6FAM	56	140-180	2
BM1312	PET	56	100-140	2
BM3205	VIC	55	210-230	2
OarCP34	6FAM	56	110-119	3
BMS2782	NED	54	183-191	3
BMC1222	NED	56	276-284	3
INRA206	NED	56	132-140	3
OarCP73	PET	56	162-207	3
OarFCB11	VIC	56	131-146	3
BM0121	6FAM	56	142-162	3
McM210	VIC	54	148-152	3
TGLA040	PET	56	100-120	3
CSSM 54	6FAM	54	125-140	3
MAF 050	NED	60	150-174	3
BM1329	VIC	54	168-176	3
BMS1248	PET	54	135-160	3
BM4621	VIC	56	132-143	4
BM4208	NED	54	163-188	4
ILSTS058	PET	56	141-189	4
BMS1332	6FAM	56	135-149	4
MCM 58	VIC	54	159-219	4
OarHH 35	PET	50	111-136	4
BMC 1009	VIC	54	274-296	4
CSRD287	NED	60	136-143	4

OLA-DRB	PET	60	269-293	4
CSSM19	NED	54	150-159	4
BM2830	VIC	56	100-120	4
CSSM47	VIC	56	132-141	5
CSSM32	VIC	54	210-220	5
INRA003	VIC	56	132-184	5
ILSTS034	VIC	55	141-180	5
BM7160	6FAM	55	160-190	5
BM1225	6FAM	55	210-250	5
BM3517	PET	55	100-120	5
BM8125	VIC	55	105-125	5
BMC8012	6FAM	55	189-191	5
BMS1714	NED	55	110-116	5
BMS1788	6FAM	55	90-115	5
BMS2252	NED	55	140-165	5
INRA006	PET	55	106-126	6
MAF70	PET	58	150-156	6
CSRD247	PET	58	234-245	6
MAF64	VIC	58	134-147	6
INRA063	VIC	55	160-166	6
BM1818	VIC	55	252-265	6
DRBP1	6FAM	55	113-135	6
ILSTS087	6FAM	58	139-145	6
INRABERN172	6FAM	55	224-244	6
ILSTS011	6FAM	58	278-280	6
SRCRSP09	NED	58	120-134	6
BM1258	NED	55	100-120	7
BMS0745	PET	55	120-124	7
ILSTS033	PET	55	159-172	7
INRA011	VIC	55	223-261	7
MCM136	VIC	55	126-128	7
SRCRSP10	6FAM	55	264-275	7
LSCV52	VIC	55	96-103	7
TGLA179	6FAM	55	85-88	7
BM0321	6FAM	58	118-148	7
INRA040	PET	55	226-246	7
MCM64	NED	58	137-147	7
SRCRSP05	6FAM	58	160-167	7
ILSTS045	NED	55	162-168	8
LSCV36	6FAM	55	100-112	8
BMS2526	NED	55	148-158	8
HUJ614	VIC	55	175-177	8
ILSTS059	6FAM	55	151-155	8
LSCV46	NED	55	82-100	8
MCM104	PET	55	112-120	8
OARFCB193	VIC	55	117-129	8
SRCRSP24	VIC	58	152-168	8
SRCRSP08	PET	58	214-242	8
BMS1332	PET	55	135-149	8