Proc. Assoc. Advmt. Anim. Breed. Genet. 20:78-81

INCREASING PROLIFICACY OF AWASSI AND ASSAF BREEDS BY INTROGRESSION OF THE *FECB* (BOOROOLA) MUTATION: ACHIEVEMENTS AND CHALLENGES

E. Gootwine[1], A. Rosov[1], M. Abu Siam[2] and E. Seroussi[1]

[1]The Volcani Center, PO Box 6, Bet Dagan 50250, Israel
[2] Siam Veterinary Clinic, PO Box 519, Rahat 85357, Israel

SUMMARY

Awassi and Assaf are local Israeli sheep breeds with prolificacies of ~1.30 and ~1.65 lambs born/ewe lambing (LB/EL), respectively. Introgressing the *B* (Booroola) allele of the *FecB* locus into these breeds led to the formation of prolific strains designated 'Afec Awassi' and 'Afec Assaf', in which lamb production under both intensive and semi-intensive conditions is higher than in the respective local breeds by ~0.8 LB/EL and ~0.5 live lambs born/ewe lambing. Lower survival rate at birth in multifetal pregnancies reduces the ability to fully exploit the economic potential of the Afec strains. A genome-wide association study revealed QTLs on ovine chromosomes 1, 8, 10, 26 and X associated with lamb survival rate at birth as a maternal trait.

INTRODUCTION

Since the beginning of the last century, the Awassi—a low-prolificacy fat-tail sheep breed and the most common breed in the Middle East—has undergone consecutive genetic changes in Israel aimed at improving milk and lamb production (Gootwine 2011; Fig. 1). Within-breed selection for high milk production resulted in the formation of the Improved Awassi dairy strain. Later, crossing the Improved Awassi with the East Friesian breed led to the formation of the Assaf. Today, sheep production in Israel (about 0.5 million head) is managed under a wide range of conditions—from extensive production where the local Awassi is kept for lamb production to the highly intensive dairy and non-dairy flocks where Assaf and Awassi sheep are managed.

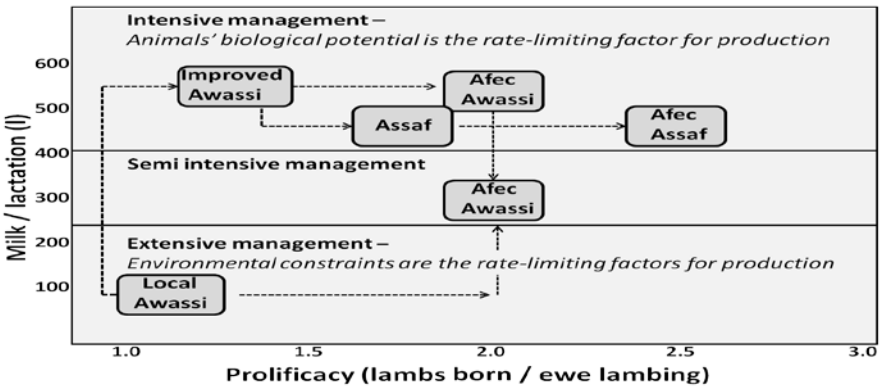

Figure 1. Schematic presentation of the breeding history of Awassi sheep in Israel

DEVELOPING THE AFEC STRAINS

Lamb production has been an important source of income in non-dairy and dairy flocks. The average prolificacy of the Awassi and Assaf is about 1.30 and 1.65 lamb born/ewe lambing (LB/EL), respectively. In 1986, the Volcani Center launched breeding programs to increase the prolificacy of the Improved Awassi and the Assaf by introgression of the *B* (Booroola) allele of the

FecB locus (Piper *et al.* 1985). Five homozygous *BB* Booroola Merino rams obtained from the Invermay Agricultural Centre, New Zealand, served as the source for the mutation. Through the backcrossing and intercrossing phases, selection for carriers of the Booroola mutation was carried out first by monitoring induced ovulation rate in ewe lambs and later, by direct genotyping for the *FecB* locus (Wilson *et al.* 2001). The breeding activity resulted in the formation of highly-prolific strains designated Afec Awassi and Afec Assaf, which carry the Booroola mutation and have average prolificacies of about 1.9 and 2.5 LB/EL, respectively (Gootwine *et al.* 2008).

The improved Awassi and Assaf fat-tail dairy breeds diverge a great deal from the non-dairy, small-body-size Booroola Merino breed. The genetic backgrounds of Afec sheep and Awassi and Assaf sheep (n = 176) were compared in 2012, using the 50K ovine single-nucleotide polymorphism (SNP) beadchip (Illumina). Results showed that throughout the introgression process, the original genetic background of the local breeds was retained almost completely with a main selection signature on ovine chromosome 6, where the *FecB* locus is mapped (Fig. 2).

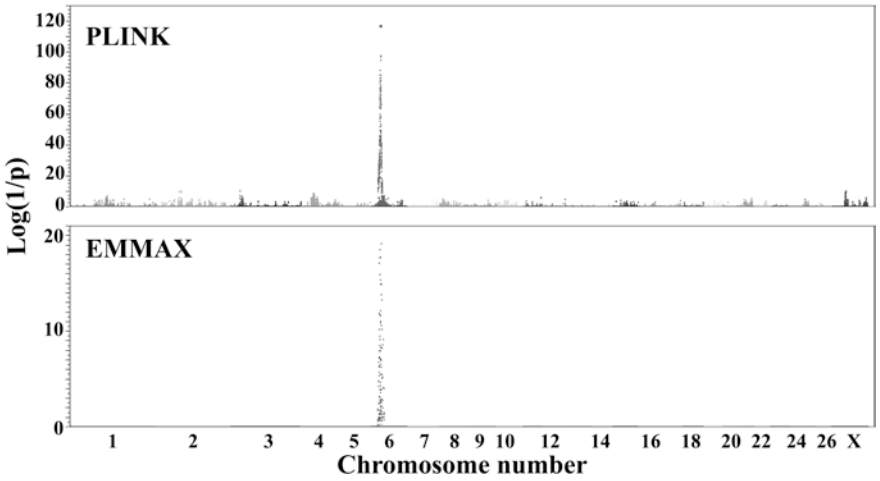

Figure 2. Association analysis between SNP haplotypes and carrying the Booroola mutation in local Israeli sheep breeds. Using the 50K ovine SNP beadchip (Illumina), a selection signature was observed by haplotype analysis of a 10-SNP sliding window using the PLINK package (Purcell *et al.* 2007) and by SNP association using EMMAX software (Kang *et al.* 2010), the latter correcting for population relatedness. While the most significant association ($p < 10^{-116}$) detected by the PLINK analysis pointed to the haplotype spanning the *FecB* locus, EMMAX detected the most significant ($p < 10^{-23}$) SNP 2.3 Mb telomeric to the mutation.

Dissemination of the Afec sheep from the breeding nuclei at the Volcani Center and the Kibbutz Ein Harod Awassi flock has been achieved mainly by selling homozygous *BB* rams to mostly non-dairy intensive commercial flocks where animals are fed to meet all of their metabolic needs. The desired genotype at the *FecB* locus for Afec ewes is *B+*, as homozygous *BB* ewes bear some disadvantages in terms of prolificacy and growth (Gootwine *et al.* 2008). Selection of replacements in the commercial flocks is based on genotyping for the *FecB* locus and in recent years, about 4,000 genotypings for *FecB* have been carried out annually with 0.51 of the genotypes being *B+* (unpublished results). Introduction of the Afec strains to commercial flocks was followed by implementation of managerial means to support the maintenance of highly prolific sheep, including a new treatment for pregnancy toxemia (Zamir *et al.* 2009).

Introgression of the Booroola mutation into local Awassi flocks. About half of the national sheep flock in Israel belongs to the local Awassi and is kept by Bedouin farmers in the Negev—the arid southern part of the country—under traditional semi-extensive management, where animals rely for about half the year on seasonal pasture. Decreases in recent years in the availability of grazing land have forced Bedouin growers to spend more on feeding their animals by purchasing costly grains and fodder, making sheep production nearly unprofitable. To overcome the new economic constraints, we investigated improving local Awassi flocks' productivity by introducing the Afec-Awassi strain. The question arose as to how the Bedouin farmers would be able to change their traditional management to support highly prolific ewes. Since 2007, controlled dissemination of the *FecB* mutation in Bedouins' Awassi flocks has been carried out by distributing *BB* Afec-Awassi rams. It is estimated that in 2013, about 20,000 Afec-Awassi sheep will be successfully bred by Bedouin farmers who appreciate the economic advantage of the genotype (Fig. 3).

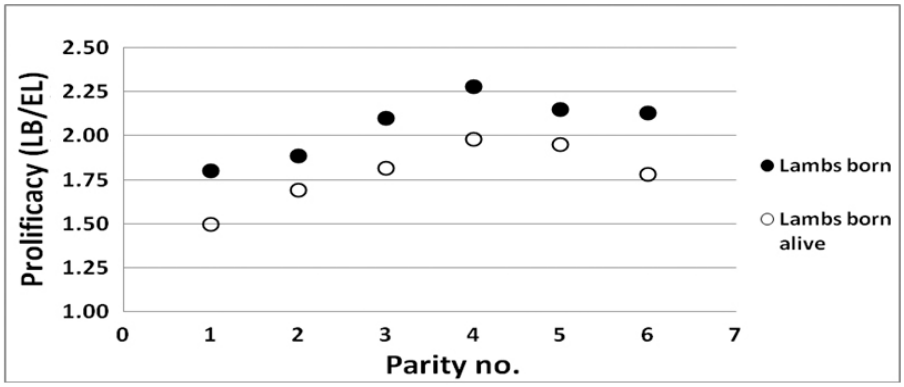

Figure 3. Prolificacy of *B*+ Afec-Awassi ewes (n = 930) in Bedouin flocks in the Negev, Israel, according to parity number. Average prolificacy of mature local Awassi ewes was estimated to be 1.14 lambs born per ewe lambing (LB/EL) based on 4,692 lambing records.

RESEARCH INTO LAMB SURVIVAL RATE AT BIRTH

Larger litter size is associated with lower lamb survival rate at birth (LSRAB), which drops from about 0.95 in the case of singletons to about 0.5 in the birth of sixplets (Gootwine *et al.* 2008). The full economic advantage of high litter size at birth is not captured because of lower LSRAB in the Afec strains, as litters of 4 or more lambs comprise about 15% of the litters in the Afec Assaf (Gootwine *et al*. 2008).

Multifetal pregnancy affects maternal metabolism (Moallem *et al*., 2012) and fetal body weight in a manner comparable to the adverse effects of severe experimental protocols aimed at restricting fetal growth such as maternal undernutrition or carunclectomy (Gootwine 2013). Research into morphometric parameters of newborn Afec-Assaf lambs (n = 957) which account for the effects of crop, sire, litter size, parity number, sex and lamb viability at birth showed that while liveborn and stillborn lambs were similar in their crown rump length, being on average 51.4 ± 0.4 cm, stillborns were significantly (p<0.0001) lower in birth weight (4.1 ± 0.1 and 3.5 ± 0.1 kg for liveborns and stillborns, respectively). This indicates that fetal death in multifetal pregnancies occurs on average some 7–10 days before lambing.

LSRAB can be considered both a maternal and a fetal trait. To investigate the effect of the maternal genome on LSRAB, a whole-genome association analysis utilizing the ovine 50K beadchip (Illumina) was performed on 71 ewes with an average prolificacy of 3.04 LB/EL (4–8 parity records) and with LSRAB values ranging from 0.00 to 0.95. EMMAX (Kang *et al.* 2010) and PLINK (Purcell *et al*. 2007) haplotype analyses indicated a total of 14 regions on chromosomes 1, 8, 10, 26 and X associated ($p < 0.05$) with LSRAB (Fig. 4).

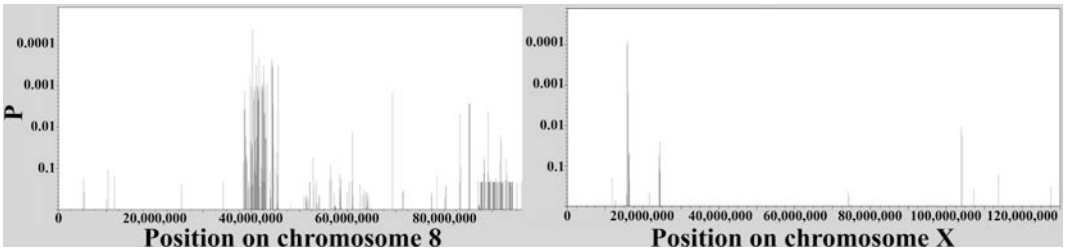

Figure 4. Chromosomal regions associated with lamb survival rate at birth in Afec-Assaf ewes. Association analysis between 10 SNP haplotypes and LSRAB was carried out using the PLINK package (Purcell *et al.* 2007). Probabilities were corrected for multiple comparisons following Bonferroni.

CONCLUSIONS

Introgression of the Booroola mutation into Awassi and Assaf breeds is a relatively fast way to increase lamb production while retaining phenotypic characteristics and important production traits, such as high milk production, large body size and adaptability to local conditions. Further research into the genetic control of LSRAB in sheep as either a maternal or fetal trait may contribute to an improvement in the economic benefits of breeding prolific sheep.

ACKNOWLEDGEMENTS

Invermay Agricultural Research Centre, New Zealand is acknowledged for supplying Booroola rams. Prof. A. Valle Zárate and Dr. A. Al Baqain, University of Hohenheim, Germany, are acknowledged for their support of the Afec-Awassi work in Bedouin flocks, and Alon Lam is acknowledged for his contribution to the genome-wide association study on lamb survival.

REFERENCES

Gootwine E. (2011) *Trop. Anim. Health Prod.* **43**: 1289.

Gootwine E. (2013) *J. Anim. Sci.* **91**: 111.

Gootwine E., Reicher S. and Rozov A. (2008) *Anim. Reprod. Sci.* **108**: 402.

Kang H.M., Sul J.H., Service S.K., Zaitlen N.A., Kong S., Freimer N.B., Sabatti C. and Eskin E. (2010) *Nat. Genet.* **42**: 348.

Moallem U., Rozov A., Gootwine E. and Honig H. (2012) *J. Anim. Sci.* **90**: 318.

Piper L.R., Bindon B.M. and Davis G.H. (1985) In 'Genetics of reproduction in sheep'. pp. 115-125, editors R.B. Land and D.W. Robinson, Butterworths, London.

Purcell S., Neale B., Todd-Brown K., Thomas L., Ferreira M.A.R., Bender D., Maller J., Sklar P., de Bakker P.I.W., Daly M.J. and Sham P.C. (2007) *Am. J. Hum. Genet.* **81**: 559.

Wilson T., Wu X.Y., Juengel J.L., Ross I.K., Lumsden J.M., Lord E.A., Dodds K.G., Walling G.A., McEwan J.C., O'Connell A.R., McNatty K.P. and Montgomery G.W. (2001) *Biol. Reprod.* **64**, 1225.

Zamir S., Rozov A. and Gootwine E. (2009) *Vet. Rec.***165**: 265.