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THE BOOROOLA SHEEP BREED AS A GENETIC RESOURCE WORLDWIDE IN BULGARIA

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Abstract

The Booroola Merino Breed, developed in Australia, is a valuable genetic resource for increasing biological fecundity in sheep worldwide. The purpose of this study was to summarize information about the Booroola Merino breed and its use worldwide and in Bulgaria for increasing the reproductive potential of Merino sheep and other breeds. The positive effect of the introduction of the FecB gene for high fecundity in sheep has been confirmed by a number of authors over a period of over 30 years. It has been established that crosses with Booroola achieve up to 30% higher ovulation rates and 20% more lambs after weaning compared to the purebred animals. The positive effect of introducing the FecB gene from Booroola in the Bulgarian fine-fleece breeds of sheep was established in terms of increasing the ovulation rate and the number of lambs born, with an insignificant negative impact on weight development and wool productivity. Negative effects can be reduced by choosing appropriate crossing schemes. Achieving a balance between benefit and risk in the introduction of the FecB gene from Booroola is an important condition for increasing the economic effect of animal husbandry and creating sustainable production in sheep farms.

Key words: *Booroola Merino Breed, fecundity, FecB gene, sheep.*

INTRODUCTION

Booroola, the breed with high fecundity rates, was created in Australia. Under the conditions specific to this range, the sheep are reared year-round in large flocks, make long journeys, suffer losses from predator attacks, inseminate naturally, and under these realities, an average of 78.5 lambs are obtained from 100 ewes. In farms with better feeding and rearing conditions, 110-115% weaned lambs obtained at 120-130% biological fecundity are reached (Bojkovski et al., 2009). The breeding process of the Booroola breed began in 1919 in New South Wales - Australia, in the "Kumi" area. Farmer Berg Sayres found that one of his Merino sheep was giving birth to three lambs each year and decided to form a flock from her offspring. In 1944, brothers Dick and Jack Seares purchased two sheep from this flock for their Booroola farm. Through targeted selection of progeny from multiparous lambs, they created a flock with an average fecundity of 1.9 lambs born per ewe and this is the first flock of the Booroola breed. It is the only merino breed that is characterized by high biological fecundity. Initially, the Siares brothers teamed only female animals born from multiparous mothers or family selection. The breeders working in the multiparous flock are from other Australian Merino flocks with breed-normal fecundity. The history of the Booroola breed is based on a gradual increase in the frequency of individuals carrying a specific gene that largely determines the fecundity trait (Tzonev, 2014). Genetic analysis (Bindon, 1984) of sheep of this breed shows, that responsible for increasing the number of ovulated eggs and the number of lambs born in one birth is a major gene F (FecB). This was confirmed in subsequent scientific experiments when it was found that its occurrence was due to a mutation (BMPR-1B) in a chromosome no. 6. This mutation was later found in indigenous sheep breeds in India, China and Indonesia. Fogarty (2009) confirmed the hypothesis of the appearance of the Australian FecB gene after the importation of Garole (also known as Bengal) sheep from northeastern India in 1792 and 1793. Davis (2005) also found the presence of the gene for high fecundity in local Indian sheep breeds, found a high degree of similarity and considered the Booroola Merino to be a direct descendant of the Garole breed. The F (FecB) gene determines higher fecundity in other sheep breeds - Finnish Landrace, Romanovska, British Milk Sheep (Abraham & Thomas, 2012). In male Booroola breeders, the FecB gene can be detected after judging the fecundity of their daughters. In the Australian Merino, the birth of three lambs in one lamb is a very rare phenomenon (0.1%), therefore the registration of multiple births in daughters is considered as evidence that the father is a carrier of the FecB gene. The F (Fecundity) designation of this gene is analogous to publications related to mouse studies (Lyon, 1977). The same author found that in mice it determines fecundity and it is accepted to be marked with the letter "F", and the other type of alleles with the alternative symbol (+). According to Turner (1978), one of the problems with the use of this FecB gene is that, even in the heterozygous state, it can increase fecundity more than the desired level and this leads to delayed weight development and higher than average lamb mortality.

The purpose of this review is to summarize the information available in the scientific literature regarding the Booroola breed and the possibilities for the introduction of the FecB gene for high fecundity in other breeds worldwide and in Bulgaria, with the aim of increasing the reproductive potential and the economic effect in sheep breeding.

INTRODUCTION OF THE F GENE (FecB) FROM BOOROOLA

In 1973 in New Zealand the Ministry of Agriculture bought two rams of the Booroola breed. In 1974 and 1977 Heldon Station, MacKenzie Province made large imports of multifertile Merino rams and ewes from Australia and established an elite flock of 200 ewes with an average fecundity of 3.6 born and 2.9 weaned lambs a sheep. Since 1981, the Heldon station has been actively producing and exporting rams, ewes, embryos and deep-frozen semen of the multi-fertile Booroola breed to Brazil, Chile, Hungary, Uruguay, and later to England, the USA, Canada, the Czech Republic, Scotland, Bulgaria.

Fecundity of F1 crosses with Booroola have been well studied by Robertson (1974), Allison et al. (1978). Daughters of homozygous Booroola (BB) rams can produce twice as many lambs as Merino ewes in Australia and New Zealand, where they produce an average of 0.85 lambs per ewe. In Hungary, two programs are being worked on to obtain thin-fleece sheep with longer wool and higher fecundity and milk yield based on the Hungarian Worsted Merino. Fine fleece ewes are crossed with Booroola and F1 crosses with Suffolk or German Blackheads (Veress, 1983). The same scientist reports that when crossing the Hungarian fine-fleece breeds with homozygous Booroola (BB) rams, fecundity increases to 150-170%. When transferring the FecB gene from the Booroola breed in the Czech Republic, they were able to increase the fecundity of fine-fleece sheep to about 150% (Horak et al., 1989). Davis et al. (1981) found that 88% of the progeny of a ram (BB) homozygous for the fecundity gene ovulated 3 or more eggs, but only 56% gave birth to 3 lambs. In the heterozygous (B+) ram progeny, 62% had 3 or more ovulated eggs and 61% lambed with three or more lambs. Bindon (1984) found that 30% of Booroola ewes ovulated up to 3 ovules and the remainder ovulated up to 9 ovules, with an average number of 4.65. In the control group, Australian Merino ewes in 98.6% of them ovulated up to 2 eggs with an average number of 1.62. Maguer et al. (1999) found of 21 Booroola rams imported from New Zealand to Hungary that one ram lacked the FecB gene for high fecundity, 8 were homozygous (BB), 10 were heterozygous (B+), and 2 have not been identified. According to Davis et al. (1998) the animals were considered carriers of the gene, in which at least once an ovulation rate of 3 or more eggs was recorded. Davis and Hinch (1998) reported the successful transfer of the FecB gene in various semi-fine fleece breeds - Dorset, Romney-marsh, Border Leicester etc.). The authors found that the presence of the FecB gene increased the fecundity of the mothers by 0.7 lambs. This suggests that using a homozygous Booroola (BB) ram, all would receive the FecB gene and the mean fecundity of the daughters would increase by 0.7 lambs. If the father is heterozygous for the gene (B+) it is transmitted to half the offspring and the increase in fecundity of the daughters is on average 0.35 lambs. If the ram is homozygous for the gene for normal fecundity (++) there will be no changes in the fecundity of his daughters. The authors recommend the appropriate phenotypic criteria for judging the genotype of rams with respect to the FecB gene. Davis & Hinch (1981) recommended breeding crosses with rams carrying the F gene in order to increase the fecundity of various sheep breeds, including stock flocks. The productive direction of these breeds can be maintained if the Booroola blood is reduced by backcrossing to 12.5% or 6.25%. When applying the selection schemes, care must be taken to preserve the FecB gene in the crosses. The creation of an intrabreed type Awassi "+", with high fecundity in Israel, is an example of the transfer of the FecB gene from the multifertile merino Booroola in a very different productive direction, exterior and interior Awassi breed (Mihailova, 1998). In this case, homozygous Booroola rams (BB) are used only in the first cross. In the following generations, a reverse cross is made with Awassi and after 11 years, as a result of the applied selection schemes, the flock has 90% Awassi-blooded and only 10% Booroola-blooded. Heterozygous carriers of the F gene (B+) predominate, but there are also homozygous rams and sheep (BB) in the nucleus. Fecundity from 1.1-1.2 increases to 2 lambs born and 1.7 lambs weaned per ewe. The average milk yield of 500 liters in the starting flock of Awassi in F1 is almost halved. It is likely that the reduction in milk yield is the result of the influence of other Booroola merino genes. After backcrossing the final form with 90% Awassi, she reaches 400 liters per lactation, with almost double the fecundity. The most successful transfer of the FecB gene from the Booroola breed is obtained in merino and fine fleece breeds. Selection problems and negative effects are insignificant in sheep from one productive direction. A slight decrease in live weight and wool yield of the crosses was reported.

Martyniuk et al. (2009) summarized the results of the import and use of 121 Booroola embryos, homozygous carriers of FecB (BB) in Poland. The average ovulation rate for 5 years was 4.25 /ranging from 2 to 8/, average fecundity - 228% and up to 5 lambs per lambing were obtained. Despite the increased fecundity of crosses with Polish Merino and Suffolk in the second stage, the introduction of the FecB gene did not gain popularity in Poland, due to the lower level of the traits birth and weaning live weight, growth intensity, average daily gain and carcass conformation compared to parent breeds. Gootwine (2009) published results using homozygous (BB) Booroola Merino rams in Israel, where lamb production is the main source of income from non-dairy flocks and contributes about half of the gross income of dairy flocks. Crosses with Awassi and Assaf are named Afec - Awassi and Afec - Assaf. Fecundity of heterozygous B+ and homozygous BB ewes was 1.90 and 1.92 lambs per ewe, respectively, in Afec - Awassi and 2.40 and 2.55 lambs per ewe, respectively, in Afec-Assaf. Compared to the baseline fecundity of Awassi and Assaf, which is about 1.3 and 1.7 lambs, respectively, the increase is significant. Introduction of the FecB gene, however, had some adverse effects on lamb birth weight and survival, as well as on body weight and milk production. Van der Werf (2009) investigated the genetic aspects of the introduction of the FecB gene from Booroola into a commercial Australian sheep breed and found that the results obtained depended on the effect of the gene, the genetic distances between the two breeds and the specific interaction of the two genotypes. In developing breeding improvement strategies, the author recommends the use of gene markers to achieve rapid genetic progress. Walkden-Brown et al. (2009) reported that the use of the direct DNA test for FecB genotyping since 2001 created great opportunities for research deployment in Australia. The scientific interest of the authors is also focused on the negative effects observed in homozygous variants carrying FecB, which are expressed in low lamb survival and are associated with increased ovulation frequency. Further studies of genetic mechanisms and other factors modulating the effect of FecB are needed.

Fogarty (2009) published a large-scale study on the topic of the effects of the introduction of the FecB gene in different breeds, geographies and production systems. The author summarizes 45 reports with information on crosses with different types of breeds and studies of the different genotypes (homozygous BB, heterozygous B+ and non-carriers ++) and their reproductive outcomes. A positive effect of FecB (B+) on Ovulation Rate (HO - number of corpora lutea) +1.3 (range +0.8 to +2.0) and + 0.7 (range +0.4 to +1.3) for number of lambs born was found. According to Fogarty (2009), the effect of FecB (BB) homozygous genotypes is manifested by a greater increase in the Ovulation Rate, but without a significant change in the number of lambs born. His summarized data show that heterozygous variants are characterized by better growth intensity, number of lambs weaned and total weight of the animals weaned. The majority of studies have shown lower birth weight and less growth intensity in crosses with Booroola Merino and their progeny compared to parent forms. Fogarty (2009) argued that it is difficult to differentiate to what extent this is due to the effects of the introduction of the FecB gene or the general potential of Booroola Merino for these traits, as well as correlations with lamb size and dependencies with feeding and rearing conditions. Regarding other selection traits with economic significance, Fogarty (2009) found no significant and reliable effects. The introduction of the F (FecB) gene did not significantly affect seasonal estrogenic activity, carcass and meat quality, and breeding productivity.

With the idea of elucidating the mechanism of action of FecB, Abraham & Thomas (2012) investigated and found increased plasma levels of follicle-stimulating hormone (FSH) during the oestrous cycle and a higher concentration of FSH in the urine of Booroola Merino sheep of reproductive age. The authors found some differences in the structure of the ovaries and follicles compared to the Australian Merino control group. Of interest is the comparison of the reproductive characteristics of male Booroola and Australian Merino breeders. Abraham & Thomas (2012) found no significant differences in terms of testis size, quantitative and qualitative characteristics of the semen in the two Merino breeds. The results for hormonal status are also similar, which gives the authors reason to conclude that the phenotypic expression of the F (FecB) gene is limited by sex (Abraham & Thomas, 2012). Adkinson et al. (2013) analyzed the realities of sheep production in Turkey and the opportunities presented by the use of the FecB gene in breeding schemes to meet the increasing market demand for sheep meat. The authors report the absence of this gene in genetic studies of 6 local breeds, but consider that its introduction into the genotype of adapted breeds, through appropriate cross schemes, is a better option for increasing fecundity than importing homozygous introduced animals. The production of milk and meat from sheep has been significantly reduced in the last 20 years in Turkey, and the demand is increasing, and the use of the FecB gene has great potential and prospects for increasing productivity and economic efficiency in sheep farming (Adkinson et al., 2013).

Qi et al. (2020) found that between homozygous, heterozygous carriers of the FecB allele and those without it, there was no significant difference in frequency and duration of oestrus. The results show that carriers of this allele in both variants have a larger litter size, after oestrus synchronization and artificial insemination. Lamb survival rates at weaning did not differ between groups, but higher fecundity in ewes carrying the B allele was associated with lower offspring body weight at birth and weaning. Wang et al. (2021) investigated the effects of FecB gene on oestrus, ovulation and endocrine characteristics in Small Tail Han Sheep in China. The authors recommend that FecB allele heterozygous animals (B+) showing moderate ovulation and lamb size and a shorter oestrous cycle be widely used in sheep crossbreeding systems for commercial lamb production.

INTRODUCTION OF THE F GENE (FecB) FROM BOOROOLA IN BULGARIA

Economic analyzes of management in sheep farming in our country show that the main share of income, between 50% and 80%, depending on the breed, comes from the sale of lambs for meat. In this aspect, an important factor for achieving profitable production in the modern sheep farm is obtaining a greater number of born and weaned lambs, respectively the level of the fecundity characteristic. The fecundity of the Ovis species varies widely - from 1 to 6-7 lambs at one birth. Reproductive traits are characterized by a low coefficient of heritability (h^2). For most reproductive performance traits, heritability values are low and variable, around 0.1. This is due to the relatively small part of the additive genetic diversity in terms of reproductive traits. For this reason, it is considered that selection for these characters is not sufficiently effective. A certain increase in fecundity is achieved when cross-breeding methods are applied, as a result of the heterosis effect, and this is used in appropriate selection schemes, with the aim of consolidating the increased level of the trait in subsequent generations. The greatest successes in terms of increasing fecundity are achieved through genetic improvement. The number of lambs born at one birth was until recently considered a quantitative trait determined by a large number of genes. For most breeds and lines, this explanation is considered correct even now, but for some of them, such as the Booroola breed in Australia, some Icelandic, Pacific and new breeds, in recent years new hypotheses have been built and theoretical clarifications have been made (Venev & Stoykov, 2002). The most significant progress in genetic improvement of fecundity has been achieved in Australia, where the Booroola breed was created. Studies on the gene for high fecundity in sheep of this breed are of great interest, motivated by the idea of a relatively quick and effective way to increase fecundity.

In Bulgaria in 1988, 1351 doses of deep-frozen seminal fluid from two male breeders of the Booroola breed - No. 61 and No. 377, originating from the "Booroola" - Haldon flock in New Zealand (Boykovski et al., 2018) were imported. In the progeny evaluation carried out at the Heldon station, it was found that 80% of the daughters of breeder No. 61 had an ovulation rate of 3 and more eggs, and for ram No. 377 - 85% of the female offspring.

The process of creating inbred structures (lines) with high fecundity begins with the introduction of the FecB gene from Booroola in sheep from the North East Bulgarian Merino breed (NEBM) - Shumen type, bred at the Research Center for Agriculture in Targovishte. The idea of the breeders is that the predicted genetic progress on this trait will subsequently be transferred to the entire population of the NEBM breed. For the successful introduction of genetic variability for high fecundity, intrauterine insemination with cryopreserved sperm was performed by laparoscopy after pre-synchronization of oestrus. Inseminated were 274 sheep according to the scheme "♀ NEBM x ♂ Booroola" and 35 sheep according to "♀F1 (♀NEBM x ♂ Bo) x ♂Bo". The realized biological fecundity of the sheep was 152.21 lambs from 100 ewes obtained according to the scheme "♀ NEBM x ♂ Bo" and 220.00 from "♀F1 x ♂Bo" (Boykovski et al., 2002). Different levels of trait and line studies have been reported in the offspring of individual breeders. The ewes inseminated with ram No. 377 on both maternal bases (♀ NEBM) and ♀ (NEBM x Bo) realized higher fecundity 164.8 and 240.00, and those inseminated with material from ram No. 61 respectively 134.80 and 213, 33 lambs from 100 ewes. As a result of the introduction of the FecB gene from the multi-fruited Booroola breed into the genetic structure of the flock in Research Center for Agriculture - Targovishte, two breeding lines were formed, originating from the homozygous for this gene (BB) breeders No. 61 and No. 377. By using appropriate methods of breeding is aimed at increasing the fecundity of sheep and preserving the FecB gene, reducing the adverse impact of the introduction of the Booroola gene on the live weight and wool production of the animals in the following generations. The rearing of lambs from multiparous ewes creates technological problems, due to lower birth weight and reduced vitality, but this does not reduce the importance of high fecundity, as the main productive trait with great economic weight (Boykovski et al., 2018).

The exterior of the Booroola rams and ewes does not differ from the typical Australian Merino, of the "Medium" type, with a medium fine fleece (19-23 μ), an average staple length of 9 cm, an average live weight of 50-55 kg and a shear of unwashed wool 5.5 kg (Boykovski et al., 2009). Desired fecundity, according to Boykovski et al. (2009) is 2-3 lambs. Intensive rearing systems in the US and Israel produce more than 3 lambs. In New Zealand, they apply a scheme to combine high fecundity with good precociousness and meat-producing qualities in the Dorset breed. 1/8 blood ewes are obtained from homozygous rams (BB) and a backcross is used (Boykovski et al., 2009). An important condition for successful work in this direction is the discovery of animals that carry the gene for multiple fecundity. To a large extent, the number of lambs born from an ewe is determined by the number of ovulated eggs. These can practically be established by enumeration of the corpora lutea after slaughter or in vivo by laparoscopy.

Dimitrov (1997) studied the effect of the implemented schemes and presented results for the fecundity of the sheep from the flock in the Research Center for Agriculture - Targovishte (Table 1). The highest fecundity is the ½ Bo crosses at 2.5, 3.5 and 4.5 years of age - 202% on average, and the lowest - purebreds from the NEBM breed - 114%. The increase in fecundity is 88%. Animals with ¼ Bo occupy an intermediate position in this regard.

Table 1. Fecundity of different Booroola-blooded ewes (Dimitrov, 1997)

Groups	Age of ewes, years		
	2.5	3.5	4.5
	x	X	x
NEBM-purebred	1.125	1.303	1.000
½ Bo-blooded	2.000	1.932	2.125
¼ Bo-blooded	1.622	1.690	1.825

Data on the live weight of the studied animals are shown in Table 2. At weaning, the half-breeds with the lowest live weight were 26.74 kg, and the purebreds with the highest - 29.89 kg. At 18 months, half-breeds have 10.56 kg lower live weight than purebreds. This trend in live weight differences was maintained at 2.5 and 3.5 years of age. Animals ¼ Bo-blooded occupy an intermediate position, but at all ages studied, they have a significantly lower live weight than purebreds. Kumar et al. (2008) found that genotypes with the FecB allele had a significant genetic effect ($P < 0.01$) on live weight of lambs from birth to 12 months of age, in crossbred Garole x Malpura sheep.

Table 2. Live weight (kg) of different-blooded ewes at different ages (Dimitrov, 1997)

Groups	Age at			
	Weaning	18 months	2.5 years	3.5 years
	x	x	x	x
NEBM-purebred	29.89	61.95	67.60	71.32
½ Bo-blooded	26.74	51.39	55.97	61.55
¼ Bo-blooded	27.84	54.04	62.67	64.54

As a result of the crossbreeding, Dimitrov (1997) found changes in the levels of the wool production trait (Table 3). Half-breed crossbreds at 1.5 years of age have 2.127 kg lower wool production compared to purebreds from the NEBM breed. This difference is also confirmed at later ages. At 2.5 years of age, the wool production of half-breed crosses was 1.656 kg lower, and at 3.5 years of age by 1.008 kg, compared to that of purebred sheep of the NEBM breed. Quarter blood crosses occupy an intermediate position in this respect at the three studied ages. The clean wool yield of half-breeds is 3.31% higher than that of purebreds. In terms of clean fiber, the results were opposite, purebreds outperforming half-breeds by 0.635 kg. This is a logical consequence of the decrease in wool yield with the introduction of FecB.

Table 3. Influence of Booroola blood on wool production, clean wool yield and clean fiber (Dimitrov,1997)

Groups	Wool production at, kg			Clean wool yield, % at 2.5	Clean fiber, kg at 2.5
	1.5 years	2.5 years	3.5 years		
	x	x	x	X	x
NEBM-purebred	8.436	8.007	7.587	49.86	3.978
½ Bo-blooded	6.309	6.351	6.579	53.17	3.343
¼ Bo-blooded	7.430	6.849	7.177	51.67	3.439

Detailed results on the effects of crossing North-Eastern Bulgarian Thin Fleece sheep with Booroola rams have been published by Dimitrov (2001). He found that with an increase in the percentage of blood from the Booroola breed, the level of the biological fecundity trait increased significantly, with a slight negative effect on wool yield and the amount of pure fiber. The conclusions drawn by the author are in line with the findings of Young et al. (1991), that sheep obtained from a cross with Booroola realized about 5% lower live weight, produced on average 6% less clean fiber, but gave with 20% more weaned lambs compared to the starting forms.

Dimitrov (2001) considers that not only purebred breeders of the Booroola breed can be included in schemes for increasing biological fecundity, but also heterozygous variants (B+), judged by offspring and carriers of this gene. In the period 1989-1997, a number of studies were carried out in the Research Center for Agriculture - Targovishte, with the aim of making a comparative analysis of the crossbreeds ½ Booroola and ¼ Booroola with the purebred sheep of the NEBM breed. The obtained results regarding productive traits with greater economic weight and the degree of undesirable effects in some schemes outline the direction of breeding work with the Booroola breed. Analogous results regarding the experience with the introduction of the FecB gene in Hungary were reported by Laszlo et al. (1987). They use 13 Booroola rams to cross with the local Merino sheep. F1 crosses have a lower live weight at 100 days of age by 6 to 28%, at one year of age by 4 to 10% and at wool cut by 1 to 8% compared to the parent breed. At 22 months of age, 501 daughters of 3 imported rams realized 131.5% fecundity, 20% higher than that of the source breed - Hungarian Merino. Cenkova (1990) reported similar results in our country. These studies confirm that a substantial and sustained increase in the biological fecundity trait can be achieved through genetic improvement with the introduction of FecB from Booroola. According to most researchers of this process, the high fecundity rates give a significant negative effect on live weight, wool production and clean fiber in animals, the product of cross compared to purebred sheep.

Dimitrov (2001) investigated the effect of the inclusion of FecB for high fecundity in the genetic structures of the NEBM breed on the growth intensity of lambs and their fattening abilities (Table 4). He found that the average daily gain of purebred lambs of the Shumen inbred type of NEBM, fattened up to 40 kg of live weight was 2.55% higher than that of ½ blooded crosses with the Booroola breed. The age at final live weight of the ½ blooded crosses was 1.19% higher. The same tendency was found in terms of consumption of nutrients in feed units and digestible protein per 1 kg of growth. In crossbred animals, it was greater by 4.44% and 1.18% compared to purebreds. In ¼ Bo-blooded crossbreeds, the levels of the studied traits were close to purebreds and occupy an intermediate position.

Table 4. Influence of the FecB gene on growth intensity and fattening capacity (Dimitrov, 2001)

Groups	n	Average daily gain, g	Age up to 40 kg live weight, day	Consumption for 1 kg gain	
				Feed units	Digestible protein, g
NEBM	16	241	173.1	3.38	399
NEBM x Bo	16	235	176.4	3.53	417
NEBM x ½ Bo	16	240	174.0	3.42	403

Boykovski et al. (2002) provided data on the average increase in the number of offspring obtained when applying each individual breeding scheme (Table 5). The authors make a comparison between the theoretically expected effects and the actually obtained results, differentiated by schemes.

Rams No. 61 and No. 377 are homozygous for the FecB gene (BB) with 80.00% and 85.00% of their breeding value daughters respectively having an ovulation rate of 3 or more eggs, or an average of 82.50%. Theoretically, in a cross with sheep of the Shumen inbred type of the North East Bulgarian Merino breed, without this gene (++), all the offspring obtained (½ Bo) are carriers of one copy of FecB (B+) and the average increase in biological fecundity is hypothetically expected in them to be 82.50%.

Table 5. Average increase in number of offspring obtained from the used mating schemes (Boykovski et al., 2002)

№	Mating scheme ♀ ♂	Lambings	Biological fecundity, n	Increase in the number of offspring, n	Increase in biological fecundity	
					Theoretically, %	Practically, %
1	(NEBM x NEBM)	Average from 4	1.177	-	-	-
		including 1st lambing	1.138	-	-	-
2	(NEBM x Bo) (½ of Bo)	Average from 4	2.045	0.868	82.50	86.80
		including 1st lambing	1.962	0.824	82.50	82.24
3	(NEBM x ½ Bo) x (NEBM x ½ Bo) (½ of Bo)	Average from 4	1.863	0.686	74.25	68.60
		including 1st lambing	1.838	0.700	7425	70.00
4	(NEBM x ½ Bo) (¼ of Bo)	Average from 4	1.721	0.544	41.25	54.40
		including 1st lambing	1.648	0.510	41.25	51.00
5	¼ Bo x ½ Bo (⅜ of Bo)	Average from 4	1.938	0.761	61.87	76.10
		including 1st lambing	1.750	0.612	61.87	61.20
6	(½Bo x Bo) (¾ of Bo)	Average from 4	1.864	0.687	123.75	68.70
		including 1st lambing	1.942	0.804	123.75	80.40

The results obtained show a real increase of 86.80% on average from four years, incl. on the 1st - with 82.24%. Using heterozygous Booroola rams (B+) on the same foundation (NEBM) or (++), theoretically only half of the progeny are heterozygous carriers of FecB (B+), and average productivity should increase by 41.25%, but in practice, an increase of 54.40% was realized.
After about 3 generational intervals, Dimitrov (2006) carried out a new study on the influence of the Booroola bloodline in the genetic structure of the North East Bulgarian Merino sheep breed - Shumen type, raised in the Research Center for Agriculture - Targovishte. The object of the study was the weight development of the animals from weaning to 2.5 years and the biological fecundity of the 1st and 2nd lambings. Live weight measured at different ages was higher in purebred ewes by 3.04% to 10.05% compared to crosses carrying FecB. A tendency for a gradual reduction of differences in live weight with increasing age of the animals was established, and at 2.5 years the advantage in favor of purebreds was only 3.04% (Table 6).

Table 6. Influence of Booroola blood on live weight (kg) (Dimitrov, 2006a)

Groups	Age at							
	100 days		9 months		1.5 years		2.5 years	
	n	x	n	x	n	x	n	x
NEBM-purebred	453	28.68	406	39.97	345	55.47	206	58.26
NEBM x Bo	271	26.06	251	37.05	200	53.72	134	56.54

The fecundity of purebred sheep at 2.5 and 3.5 years was 117.1% and 111.4%, respectively, and for animals with Booroola blood - 141.5% and 137.5% (Table 7). In another study, Dimitrov (2006) investigated the influence of the Booroola-blooded breed on the wool production and natural wool length at 1.5 and 2.5 years of age of sheep of the Shumen inbred type of the NEBM breed. Similar to the previous studies, purebred sheep were found to have a higher amount of unwashed wool and clean fiber than those with Booroola blood at 1.5 and 2.5 years of age (Tables 8, 9). Clean wool yield was higher by 3.82% in Booroola crosses compared to purebreds (Table 9).

Table 7. Influence of Booroola blood on fecundity (Dimitrov, 2006a)

Groups	Age at			
	2,5 years		3,5 years	
	n	x	n	x
NEBM-purebred	228	1.171	158	1.114
NEBM x Bo	142	1.415	96	1.375

Table 8. Influence of Booroola blood on wool productivity and natural wool length (Dimitrov, 2006b)

Groups	Wool productivity at, kg				Natural wool length at, cm			
	1,5 years		2,5 years		1,5 years		2,5 years	
	n	x	n	x	n	x	n	x
NEBM	389	8.846	288	7.741	392	10.29	309	9.61
NEBM x Bo	231	7.647	152	6.624	233	10.41	180	9.59

Table 9. Influence of Booroola blood on clean wool yield and clean fiber (Dimitrov, 2006b)

Groups	Clean wool yield at, %				Clean fiber at, kg			
	1,5 years		2,5 years		1,5 years		2,5 years	
	n	x	n	x	n	x	n	x
NEBM	237	48.32	233	49.1	237	4.286	233	3.798
NEBM x Bo	130	52.01	125	52.9	130	3.889	125	3.462

In practice on farms, it is difficult to calculate the ovulation rate in sheep, so it is recommended to use the data on the number of lambs born in the lambing to judge breeders by progeny. The number of offspring is a more widely used and accessible method, but as a criterion it has a significantly lower reliability for proving the bearing of FecB for fecundity compared to the ovulation rate. It is characteristic of the action of the FecB gene that with respect to the trait ovulation rate there is an additive, and for the trait number of offspring born - a dominant action (Laleva et al., 2000). Laleva et al. (2000) found the presence of FecB in ewes of the Thracian thin-wool breed, daughters of rams crossed with Booroola. The rate of ovulation and number of lambs born at 1.5 and 2.5 years of age of the studied sample was determined. The ovulation rate at 1.5 years in crossbreds is significantly higher - 2.875 than in purebreds (1.500), with statistically significant differences. These differences are also reliably confirmed at 2.5 years of age. The data show that the ovulation rate (NO - number of corpora lutea) is 1.3 higher in sheep, the product of a cross of the two ages studied. Laleva et al. (2000) found no significant differences in live weight at birth between crossbred and purebred ewes. In a similar study involving live birth weight and growth intensity of Booroola × Merinos d̓Arles crosses, Abella et al. (2005) concluded that these traits were not affected by genotype effects, respectively by the presence of the FecB gene. Similar data were published by Fogarty et al. (1995) in Poll Dorset × Booroola crosses.

Slavov et al. (2008) found an increase in genetic variance when including genetic components from the Booroola, Australian Merino and Ile de France breeds in the NEBM-Dobrudzha type population. Laleva et al. (2014) published the results of a comparative study of the traits ovulation rate, fecundity and live weight at birth in ewes of the Thracian fine-fleece breed and its crosses with Booroola. Daughters of FecB (B+) heterozygous rams No. 4012 and No. 509 were found to outperform purebreds in terms of ovulation rate. At 1.5 years, the ovulation rate in the former is 2.875 and 2.560, respectively, and in purebred animals of the Thracian fine-fleece breed it is 1.526. The results obtained for the other studied age are similar. The sheep, a product of the cross, also realized a greater number of lambs born, compared to their purebred counterparts, except for the daughters of ram 509 at 4.5 years. High variation is characteristic of this trait. Laleva et al. (2014) reported C = 46.51% variation in fecundity of

Thracian fine-fleece sheep at 4.5 years. The results obtained and the fact that the authors did not find a significant negative effect on the live weight at birth feature allowed them to confirm the positive effect of the introduction of FecB from Booroola to increase the biological fecundity of sheep from the same productive line. Stancheva et al. (2020a) found that the genetic components of the Booroola breed had a negative effect on live weight at weaning, at 9 months and at 18 months in sheep of the North east Bulgarian Merino breed. The main part of the genetic variation of the traits related to wool productivity is due to the individual characteristics of the individuals (Stancheva et al., 2020b). The same authors claim that the variability of traits caused by the combination of genes from another breed in the genotype and the heterosis effect were very low.

CONCLUSIONS

The Australian Booroola Merino (BoM) is a valuable genetic resource for increasing the biological fecundity trait in fine-wool and other sheep breeds worldwide. The economic effect of the introduction of the FecB gene for high fecundity in sheep has been confirmed by a number of authors over a period of over 30 years. It has been established that crosses with Booroola achieve up to 30% higher ovulation rates and 20% more lambs after weaning compared to the purebred animals.

The positive effect of the introduction of genetic variability from Booroola in the Bulgarian fine-fleece breeds of sheep was established in terms of increasing the ovulation rate and the number of lambs born, with insignificant negative effects on live weight and wool production.

The analysis of the genetic variability of the intrabreed linear structures, carriers of the FecB gene in the only nucleus flock of sheep from the North East Bulgarian Merino breed - Shumen type, bred in the Research Center for Agriculture - Targovishte shows that the real increase in the biological fecundity of the F1 crosses NEBM x Bo is with 86.80%, with a theoretically expected 82.50%. Optimal reproductive performance in the nucleus of the breed is a basic condition for the extension of genetic progress for the fecundity trait throughout the population.

Achieving a balance between benefit and risk in the introduction of the FecB gene from Booroola is an important condition for increasing the economic effect of animal husbandry and creating sustainable production in sheep farms.

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