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► **To cite this version:**

Assia Hadjazi, Fatima Zahra Belharfi, Fatima Zohra Mahammi, Stéphane Fabre, Semir Bechir Suheil Gaouar, et al.. Screening of FecLL prolific allele of the B4GALNT2 gene in Algerian sheep populations. *Revue d'Élevage et de Médecine Vétérinaire des Pays Tropicaux*, 2024, 77 (37433), pp.1-7. 10.19182/remvt.37433 . hal-04649905

**HAL Id: hal-04649905**

**<https://hal.inrae.fr/hal-04649905>**

Submitted on 17 Jul 2024

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# Screening of the *FecL<sup>L</sup>* prolific allele of the *B4GALNT2* gene in Algerian sheep populations

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## Keywords

Sheep, genotypes, genetic polymorphism, productivity, landraces, Algeria

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Submitted: 20 March 2024

Accepted: 17 June 2024

Online: 12 July 2024

DOI: 10.19182/remvt.37433

## Summary

**Background:** Research on the main genes controlling prolificacy in sheep, also known as fecundity genes (Fec genes), has highlighted the mechanisms involved in ewe fertility and the genetic improvement of prolificacy. The gene, *FecL/B4GALNT2* (beta-1,4-N-acetyl-galactosaminyl transferase 2) and its prolific allele *FecL<sup>L</sup>*, which segregate in French Lacaune sheep and some local sheep breeds in Morocco and Tunisia, were found to influence ovarian function. However, their action differs from that of other major genes discovered previously, such as: *FecX/BMP15* (bone morphogenetic protein 15); *FecG/GDF9* (growth differentiation factor 9); and *FecB/BMPRI1B* (bone morphogenetic protein receptor type 1B). The latter act in the bone morphogenetic protein (BMP) signalling pathway. **Aim:** The objective of this study is to investigate the segregation of the *FecL<sup>L</sup>* allele in Algerian sheep populations. **Methods:** A total of 338 animals from 12 breeds were genotyped using the PCR-RFLP technique. **Results:** Our results revealed the presence of *FecL<sup>L</sup>* in the Algerian D'man sheep population. Among the genotyped D'man sheep, 21% were carriers of the mutation in the heterozygous state. The frequency of the *FecL<sup>L</sup>* allele in the Algerian D'man population (0.11) is close to what is observed in Lacaune sheep and remains relatively low compared to Moroccan (0.58) and Tunisian (0.65) D'man sheep. **Conclusions:** The *FecL<sup>L</sup>* allele, which is shared by the French Lacaune population and the North African D'man populations, could indicate the ancestral origin of the mutation in *B4GALNT2* or the occurrence of an ancient introgression event to improve prolificacy. Managing this mutation in Algerian D'man flocks could help improve the numerical productivity of D'man sheep in Algeria.

■ How to cite this article: Hadjazi A., Belharfi F.Z., Mahammi F.Z., Fabre S., Gaouar S.B.S., Tabet-Aoul N., 2024. Screening of the *FecL<sup>L</sup>* prolific allele of the *B4GALNT2* gene in Algerian sheep populations. *Rev. Elev. Med. Vet. Pays Trop.*, 77: 37433, doi: 10.19182/remvt.37433

## ■ INTRODUCTION

The numerical productivity of sheep is of major economic importance. It is primarily linked to the prolificacy of ewes (Fathallah, 2015). Therefore, as a key determinant of flock productivity, prolificacy is an important feature of selection programmes. Prolificacy, i.e.

the number of lambs born per lambing (LB/L), varies considerably between breeds. Some breeds typically produce a single lamb per birth, occasionally twins. In contrast, more prolific breeds frequently have triplets or quadruplets, with an average of 2 or more lambs per birth (Juengel et al., 2013). The number of lambs born/lambing is an essential aspect of the profitability of sheep farming. It results from a complex interaction between breeding management, and environmental and genetic factors (Fathallah, 2015). Prolificacy is a polygenic trait with low heritability ( $h^2 = 0.05$  to  $0.2$ ) (Bradford, 1985), which complicates genetic progress through selection. Conventional methods of herd improvement, based solely on phenotypic evaluation, are slow and involve many generations for limited gains. However, the discovery of major fecundity genes (Fec genes), which have significant effects on ovulation rate and litter size, could be a breakthrough. These Fec genes could be used as markers to identify individuals carrying beneficial mutations, thus paving the way for marker-assisted selection (MAS). This method could enhance and accelerate genetic improvement to optimize sheep production (Abdoli et al., 2016). As a result, there is growing interest in research on mutations

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that influence prolificacy. The *B4GALNT2* gene (beta-1,4-N-acetyl-galactosaminyl transferase 2), is the most recently discovered major gene. Its *FecL<sup>L</sup>* mutation was found to be associated with increased prolificacy in Lacaune and Noire du Velay sheep in France, as well as in North African D'man sheep (Drouilhet et al., 2010; Drouilhet et al., 2013; Chantepie et al., 2018; Ben Jemaa et al., 2019).

The diversity of sheep breeds in Algeria reflects the country's varied climatic zones. The national sheep population is about 31.1 million, according to the Ministry of Agriculture and Rural Development (MADR, 2021). They are divided into twelve populations, and among them four are large populations (Ouled-Djellal, Hamra, Ifilène and Sidaou) and eight are small populations (Rembi, D'man, Taâdmit, Berbère, Barbarine, Tazegzawt, Srandi and Darâa). In general, the latter are rustic and have different adaptation and reproduction characteristics (Djaout et al., 2017).

The D'man sheep population is known for its high prolificacy, with rates of 150 to 250% compared to other Algerian sheep populations, whose prolificacy rate does not exceed 120% (Chekkal et al., 2015). D'man ewes have 1.88 lambs on average, with litter sizes ranging from 1 to 4 lambs at a frequency of 33.3% for singles, 49.1% for twins, 13.8% for triplets and 3.8% for quadruplets. In comparison, average litter size is lower for local breeds, such as Ouled-Djellal (1.13 lambs), Taâdmit (1.02 lambs), Hamra (1.06 lambs), and Rembi (1.10 lambs) (Boubekeur et al., 2019).

Little is known about the specific genetic foundations of prolificacy in Algerian sheep populations. The molecular basis of prolificacy in D'man sheep, in particular, the segregation of the prolific allele *FecL<sup>L</sup>* of the *B4GALNT2* gene, has not been explored in Algeria, despite its potential for breed improvement. This study aims to investigate the segregation of the prolific allele *FecL<sup>L</sup>* and its prevalence across various Algerian sheep populations, using the PCR-RFLP technique. This new research seeks to fill the knowledge gap regarding the genetic foundations of sheep prolificacy in Algeria. It also aims to contribute to the optimization of selection practices in order to enhance the productivity and efficiency of sheep farming in the country.

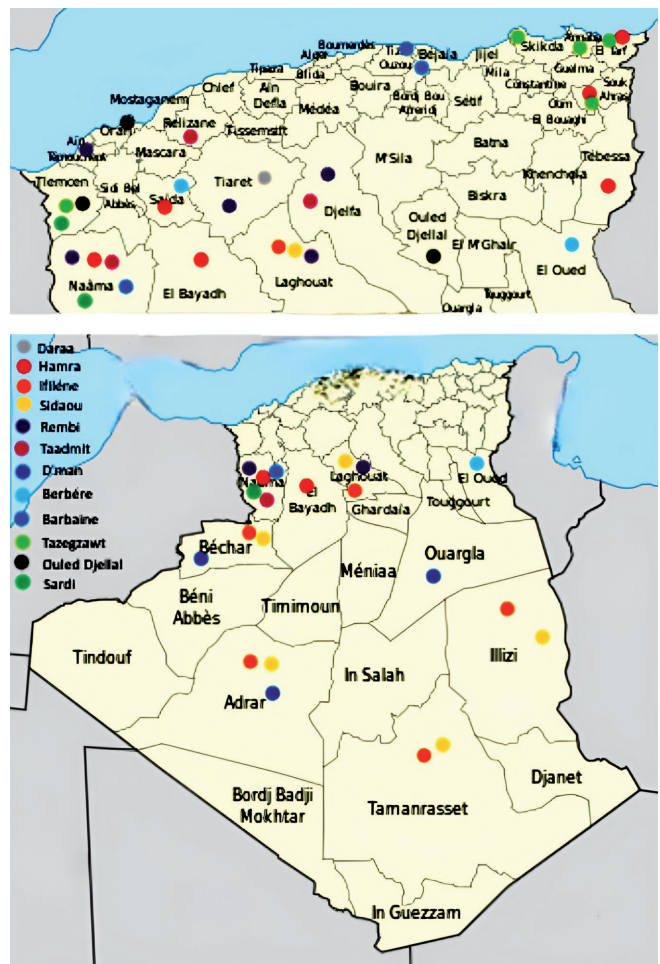
## ■ MATERIAL AND METHODS

### *Animals and blood sampling*

The present study was conducted on 338 unrelated animals from 12 sheep populations reared in Algeria. Their distribution was as follows: Sidaou (n=60), Hamra (n=49), Sardi (n=10), Ifilène (n=5), Darâa (n=4), Ouled Djellal (n=10), Taâdmit (n=44), D'man (n=42), Tazegzawt (n=30), Barbarine (n=23), Rembi (n=52) and Berbère (n=9). Sampling took place across different regions of the country between 1999 and 2019. Figure 1 presents the geographical distribution of the sampling sites for the breeds studied. Whole blood samples (about 5 ml) were collected aseptically, by venipuncture from the jugular vein, in collection tubes containing ethylene diamine tetra-acetic acid (EDTA). All sheep were sampled during routine animal health checks by an authorized veterinarian. The blood samples were transported to the laboratory at a low temperature and then frozen until they were used.

### *DNA extraction*

Genomic DNA was extracted from white blood cells using the conventional salt-based method (Miller et al., 1988) in two separate laboratories: the Laboratory of Physiopathology and Biochemistry of Nutrition (PpBioNut) at Abou Bekr Belkaid University of Tlemcen; and the Laboratory of Molecular and Cellular Genetics (LGMC) at the University of Science and Technology of Oran Mohamed Boudiaf in Algeria. The DNA samples were then stored at -20 °C for later use.



**Figure 1:** Geographical distribution of sampling sites in Algeria, for the breeds studied // Répartition géographique des sites d'échantillonnage en Algérie, pour les races étudiées

### *FecL<sup>L</sup> genotyping*

To detect the segregation of the *FecL<sup>L</sup>* allele (Oar v3.1, chr11:36938224T> A) of the *B4GALNT2* gene in each breed, a total of 338 animals were genotyped, using PCR amplification, followed by the forced restriction fragment length polymorphism method (PCR-RFLP) at the GenPhySE laboratory, INRAE-Toulouse, France. The forward primer was designed to generate a forced Hph1 restriction site in PCR products from carriers of the *FecL<sup>L</sup>* allele (chr11:36938224A). In contrast, PCR products from individuals without the *FecL<sup>L</sup>* allele (chr11:36938224T) will be devoid of the Hph1 restriction site. The *B4GALNT2* gene fragment (354bp) was amplified. The PCR reaction was conducted in a final volume of 20 µl containing 50 ng of genomic DNA, 0.5 U of GoTaq DNA polymerase (Promega), 1X GoTaq PCR buffer, 0.2 mM of dNTPs, and 0.5 µM of each primer (forward: TGCAAGAAGCTGCGGGTG; reverse: CCATGGCTTGCTCTTGGTT), following 35 cycles of denaturation at 95°C (30 sec), annealing at 58°C (30 sec) and elongation at 72°C (30 sec). Digestion of 5 µl of the PCR product with 0.3 units of Hph1 enzyme (New England Biolabs) was performed overnight (16h) at 37°C in 15 µl, followed by an inactivation step of 20 min at 65°C. All reactions included a heterozygous control sample.

The amplification and digestion products were stained with GelRed (Biotium, Dutscher) on 2% agarose gel, pictured and analysed. Based on the Lacaune control DNA (Figure 2), a unique 354 bp band is expected for homozygous non-carriers (+/+). Two bands at 327 bp and

27 bp were expected for homozygous *FecL<sup>L</sup>* carriers (L/L), and three bands at 354 bp, 327 bp and 27 bp for heterozygous carriers (L/+). Although the band at 27 bp was barely visible, it did not hinder the interpretation of genotypes (Figure 3).

### Statistical analysis

The genotype and allele frequencies of the *FecL* locus were determined by direct counting. Allele frequencies were estimated from counts of alleles in DNA genotypes. The number of homozygotes and heterozygotes was computed for observed genotype frequencies. Considering the two-allele case, the Hardy-Weinberg theorem was used to calculate the expected genotype frequencies. The Pearson's

chi-square ( $\chi^2$ ) (p-value <0.05) was used to check whether or not the population is in Hardy-Weinberg Equilibrium (HWE).

### RESULTS

The PCR-RFLP technique was applied to detect the *FecL<sup>L</sup>* mutation of the *B4GALNT2* gene in 338 individuals from 12 different breeds (Figure 3, Table I).

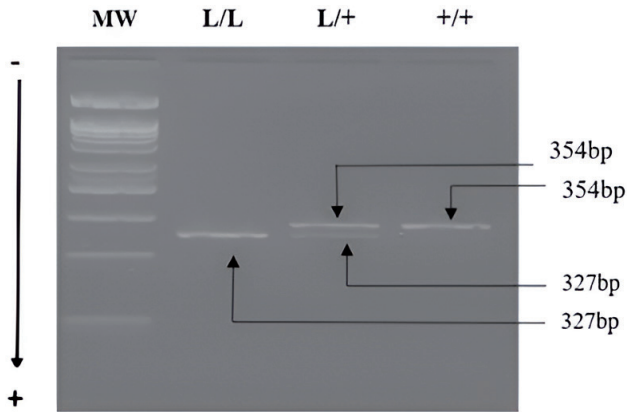
Following DNA amplification, electrophoresis on 2% agarose gel revealed clear bands of approximately 354 bp. This is consistent with the expected size, according to the 100 bp size marker, for all sheep DNA amplifications using the same primer pair.

The segregation of the *FecL<sup>L</sup>* allele of the *B4GALNT2* gene was observed in the D'man sheep population. Among the 42 genotyped D'man sheep, 9 were heterozygous carriers of *FecL<sup>L</sup>* (L/+, 21%), with no observation of homozygous carriers. Individuals from all other sheep populations genotyped in our study exhibited the wild-type allele (+/+) of the *B4GALNT2* gene (Table I).

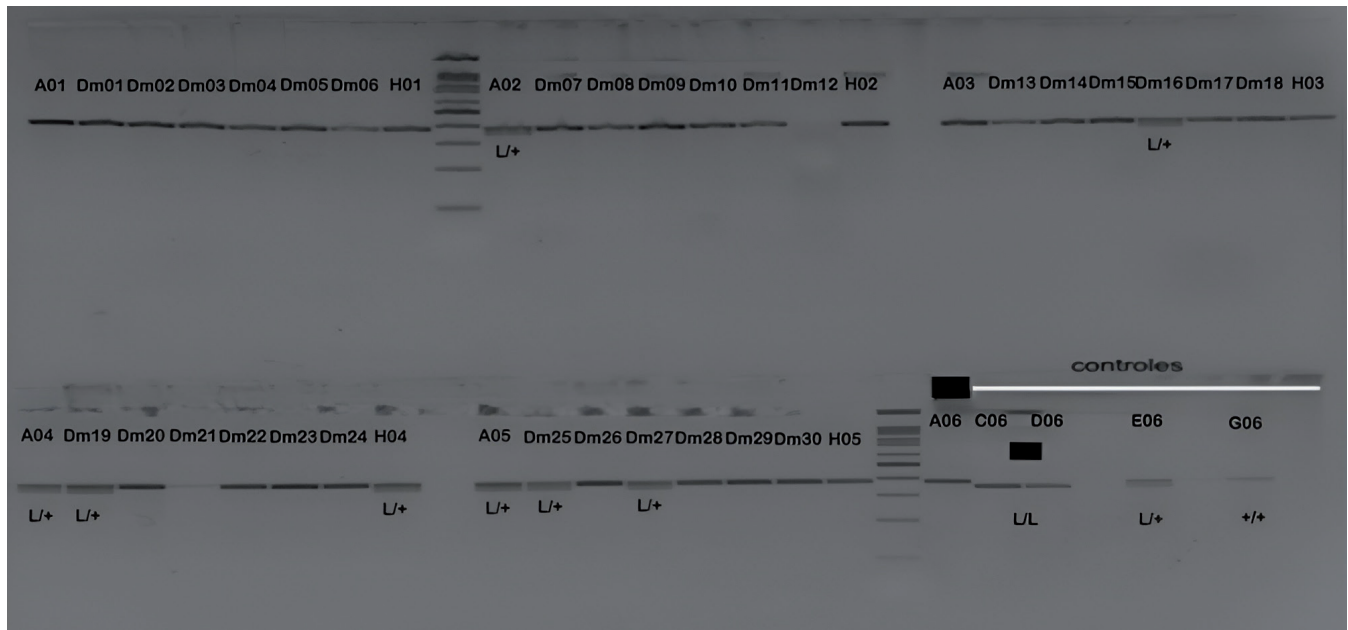
Based on this sampling, the *FecL<sup>L</sup>* allele frequency was estimated at 0.11 (95% confidence interval [CI]: [0.057 - 0.191]), and the distribution of genotypes did not deviate from the Hardy-Weinberg equilibrium ( $\chi^2 = 0.58$ ,  $p = 0.45$ ).

### DISCUSSION

Improving sheep productivity is a crucial challenge for the agricultural sector. By increasing the supply of red meat, it could make a significant contribution to food security and economic development.



**Figure 2:** Electrophoretic profile of the Lacaune control DNA // Profil électrophorétique de l'ADN de contrôle Lacaune



**Figure 3:** Electrophoretic profile of the different genotypes detected: the enzymatic digestion of PCR products of the *FecL<sup>L</sup>* allele was performed using the *Hph1* enzyme. The D'man individuals studied (Dm01, Dm02, Dm03, etc.) and the Lacaune individuals used as controls (A01, H01, A02, H02, etc.) are classified into three categories, based on the DNA fragments generated after digestion: homozygous non-carrier individuals (+/+) showed a single 354 bp band pattern; heterozygotes (L/+) showed three bands, 354, 327 and 27 bp; and homozygous carriers (L/L) showed 2 fragments, 327 and 27 bp. Bands were resolved on 2% agarose gel. (MW: molecular weight marker of 100-1000 bp) // Profil électrophorétique des différents génotypes détectés : La digestion enzymatique des produits PCR de l'allèle *FecL<sup>L</sup>* a été réalisée avec l'enzyme *Hph1*. Les individus D'man étudiés (Dm01, Dm02, Dm03, etc.) et les individus Lacaune utilisés comme témoins (A01, H01, A02, H02, etc.) sont classés en trois catégories en fonction des fragments d'ADN générés après digestion : Les individus homozygotes non porteurs (+/+) ont montré une seule bande de 354 pb, les hétérozygotes (L/+) ont montré trois bandes : 354, 327 et 27 pb et les porteurs homozygotes (L/L) ont montré 2 fragments de 327 et 27 pb. Les bandes ont été résolues sur un gel d'agarose à 2 %. (MW : marqueur de poids moléculaire de 100-1000 pb)

**Table I:** Genotype frequencies of the 338 genotyped sheep /// *Fréquences génotypiques des 338 moutons génotypés*

Sheep populations	Genotyped	+/+	L/+	Percentage of wild-type homozygous +/+	Percentage of heterozygous L/+	95% CI of <i>FecL<sup>L</sup></i> allele frequency
Sidaou	60	60	0	100%	0%	[0 - 0.031]
Hamra	49	49	0	100%	0%	[0 - 0.037]
Sardi	10	10	0	100%	0%	[0-0.161]
Ifilène	5	5	0	100%	0%	[0-0.277]
Darâa	4	4	0	100%	0%	[0 - 0.324]
Ouled Djellal	10	10	0	100%	0%	[0-0.161]
Tâadmit	44	44	0	100%	0%	[0 - 0.041]
D'man	42	33	9	79%	21%	[0.057 - 0.191]
Tazegzawt	30	30	0	100%	0%	[0 - 0.060]
Barbarine	23	23	0	100%	0%	[0 - 0.077]
Rembi	52	52	0	100%	0%	[0 - 0.035]
Berbère	9	9	0	100%	0%	[0 - 0.175]
Total	338					

Note. The confidence interval was calculated using the Wilson score formula /// L'intervalle de confiance a été calculé à l'aide de la formule du score de Wilson

Some countries, like New Zealand and Australia, have implemented genetic improvement programmes focusing on prolificacy to increase sheep flock productivity. In Algeria, a similar initiative was launched to improve the productivity of the national flock. It focused on two local Algerian sheep breeds: the Ouled Djellal, known for its exceptional meat production qualities; and the D'man, known for its exceptional reproductive qualities (Adaouri et al., 2017; Sebkhil et al., 2024). However, it is important to note that these programmes did not take into account genetic parameters that significantly influence prolificacy. In the context of genetic crossbreeding, in particular, a thorough analysis of genetic factors is required to ensure the success of improvement programmes.

Although prolificacy in sheep is thought to be a polygenic trait with low heritability, the variations in ovulation rate and litter size observed in certain sheep populations are genetically controlled by the effect of four major genes. These so-called fecundity genes or *Fec* genes (Fabre et al., 2006) include: *FecB/BMP1B* (bone morphogenetic protein receptor type 1B); *FecX/BMP15* (bone morphogenetic protein 15); and *FecG/GDF9* (growth differentiation factor 9). These three genes are part of the same *BMP* signalling pathway, which controls ovarian activity and ovulation (Mishra, 2014; Abdoli et al., 2016). The fourth and most recently identified fecundity gene is the *FecL/B4GALNT2* gene (beta-1,4-N-acetyl-galactosaminyl transferase 2) in the French Lacaune sheep population. This gene is located on chromosome 11. It has been the subject of several studies leading to the discovery of a new pathway involved in regulating ovarian function in sheep (Drouilhet et al., 2013; Demars et al., 2013)..

Numerous studies have demonstrated the segregation of various point mutations in these major genes, distributed across different populations worldwide (Table II). Some of these mutations are associated with increased prolificacy in heterozygous and homozygous carriers, while some prolific mutations are associated with infertility when homozygous (Galloway et al., 2000; Hanrahan et al., 2004; Bodin et al., 2007; Lassoued et al., 2017).

Domestic ewes exhibit tremendous genetic diversity, as shown by their numerous productive and reproductive traits, including prolificacy rate. Some breeds are known for their low prolificacy, with ewes typically giving birth to a single lamb, sometimes twins. Conversely, some breeds are more prolific, such as Romanov, with an average

litter size of 3.1 lambs, Chios with 2.5 lambs, Han with 2.9 lambs and Barbados Blackbelly with 2.3 lambs (Davis et al., 2006). The genetic variability in prolificacy is a key factor to consider in breeding and selection programmes (Juengel et al., 2013). In Algeria, the D'man sheep population is renowned for its exceptional prolificacy. This breed is widespread in the oases of southwestern Algeria and southern Morocco, due to close historical ties between these regions. The D'man population, originating from Morocco, was introduced to Algeria by Moroccan tribes, who were expelled by invaders during the Moroccan Rif War (1921–1926) (Boubekeur, 2017). In 1994, the D'man population was also introduced in Tunisia (Rekik et al., 2005).

The major genes associated with sheep prolificacy have generated significant interest among researchers and sheep breeders because of the potential for rapid improvements in lamb production. However, breeding ewes homozygous for major mutations is not recommended in commercial flocks due to high lamb losses in prolific ewes homozygous for *BMP1B* and *B4GALNT2*, or the sterility of ewes homozygous for *BMP15* or *GDF9* (Hanrahan et al., 2004). Rather than creating homozygous females, a more judicious strategy is to select and manage heterozygous ewes through molecular genotyping. This would leverage their increased prolificacy without the drawbacks of homozygotes. This approach entails managing mating and gene diffusion (Raoul et al., 2018). The effects of prolificacy should also be assessed in the context of the farming systems. High prolificacy can be counterproductive in extensive systems in challenging environments, where food availability and climatic conditions may limit lamb production (Menéndez Buxadera et al., 2004). In contrast, in semi-intensive or intensive systems, the reproductive potential of ewes may be advantageous. Indeed, when the nutrient availability is optimum and adequate veterinary care is provided, rearing large litters can generate additional income through increased lamb sales (Byrne et al., 2012). Therefore, optimal prolificacy depends on the production system and the farming context. The risks of foetal and lamb mortality should also be taken into account.

The present study analyses the segregation of the *FecL<sup>L</sup>* prolific allele of the *B4GALNT2* gene. Rather than focusing on the mechanism of action of the *B4GALNT2* gene or its effect on the level of prolificacy in the studied flocks, our main objective was to determine the presence of the *FecL<sup>L</sup>* allele in Algerian sheep populations, particularly the D'man population.

**Table II:** The major alleles of sheep prolificacy and their geographical distribution // *Allèles majeurs de prolificité des ovins et leur répartition géographique*

Gene	Allele	Sheep populations	Distribution
<i>BMPR1B</i>	<i>FecB<sup>B</sup></i>	Booroola Merino/Garole/ Javanese/Hu/Han	Australia/Asia
<i>BMP15</i>	<i>FecX<sup>C</sup></i>	Belclare/Cambridge/Lley	Ireland and UK
	<i>FecX<sup>B</sup></i>	Belclare	Ireland and UK
	<i>FecX<sup>I</sup></i>	Romney	New Zealand
	<i>FecX<sup>H</sup></i>	Romney	New Zealand
	<i>FecX<sup>L</sup></i>	Lacaune	France
	<i>FecX<sup>N</sup></i>	Noire du Velay/BMC/ Lacaune	France
	<i>FecX<sup>R</sup></i>	Rasa Aragonesa	Spain
	<i>FecX<sup>O</sup></i>	Olkuska	Poland
	<i>FecX<sup>Gr</sup></i>	Grivette/ Mouton Vendéen	France
	<i>FecX<sup>Bar</sup></i>	Barbarine	Tunisia
<i>GDF9</i>	<i>FecG<sup>T</sup></i>	Icelandic	Northern Europe
	<i>FecG<sup>WN</sup></i>	White Norwegian	Northern Europe
		Belclare/Fynnsheep	Europe
	<i>FecG<sup>H</sup></i>	Belclare/Cambridge/Lley	Ireland and UK
	<i>FecG<sup>E</sup></i>	Santa Inês	Brazil
<i>FecG<sup>L</sup></i>	Loa	Iceland	
<i>B4GALNT2</i>	<i>FecL<sup>L</sup></i>	Lacaune, Noire du Velay/D'man	France, Morocco, Tunisia

(Ben Jemaa et al., 2019 ; Abdoli et al., 2016)

Our results confirmed the segregation of the *FecL<sup>L</sup>* allele of the *B4GALNT2* gene in the D'man sheep population, with an estimated allelic frequency of 0.11. The allele was not detected in the other breeds. The allelic frequency observed in the Algerian D'man population studied is close to observations in French Lacaune (0.12) and similar to those in the Noire du Velay population (0.11). However, it is low compared to the Moroccan and Tunisian D'man populations of 0.58 and 0.65, respectively (Ben Jemaa et al., 2019). This difference could be linked to various factors, such as genetic drift, which is a random change in allelic frequencies over time due to chance or different national breeding practices. For instance, the high frequency of the *FecL<sup>L</sup>* allele with 43% homozygous carriers among Tunisian D'man sheep might be explained by: the initial importation from Morocco of 25 rams for 200 ewes, possibly selected for their high prolificacy; and subsequent inbreeding (Rekik et al., 2011; Ben Jemaa et al., 2019). Conversely, the *FecL<sup>L</sup>* allele, known to be associated with increased lamb mortality, may not have been subject to selection in the Algerian D'man sheep population given the harsh environmental conditions. This could explain its low frequency. Further research is needed to understand the reasons for the differences in allelic frequency, which may have implications for the conservation and management of these sheep populations. In addition, the association of the *FecL<sup>L</sup>* allele with increased prolificacy in Algerian D'man ewes has yet to be established. Ben Jemaa et al. (2019) also highlighted the existence of this mutation in other Moroccan sheep populations, such as the Sardi and Beni Guil (apparent frequency less than 0.16), as well as in other local sheep populations, which were genotyped as part of the NextGen

European project. The present study on Algerian breeds did not show segregation of the *FecL<sup>L</sup>* allele in Sardi or Hamra sheep, which are the Algerian equivalents of the Moroccan Beni Guil breed. Although two heterozygotes out of six animals were observed in the Beni Guil and four heterozygotes out of 27 animals were observed in the Sardi in Morocco, the differences observed in the Hamra and Beni Guil are more likely to be the result of uncontrolled crossbreeding between the D'man and other breeds sampled in Morocco. However, the small number of genotyped Sardi animals (n=10) could still be a limiting factor in detecting the *FecL<sup>L</sup>* allele in this breed.

The absence of the *FecL<sup>L</sup>* mutation in the other Algerian sheep populations that we studied could be explained by the small number of samples and the random selection of animals.

Livestock farming in North Africa has evolved traditionally over the centuries. The region's arid mountainous ecosystems have led to the emergence of highly diverse sheep populations adapted to various dry environments. Thus, it is home to unique sheep populations, in terms of their genetic diversity (Gaouar et al., 2017). The Food and Agriculture Organization of the United Nations (FAO, 2011) indicates that a quarter of the world's biodiversity hotspots are located in these highly heterogeneous environments. A recent study to characterize the genomic diversity of the major Moroccan sheep breeds, uses comprehensive genomic data to reveal that local Moroccan sheep breeds exhibit both high genetic diversity and a wide range of adaptive variations. Thus, they constitute a valuable genetic resource for sheep conservation in the context of climate change. The study also revealed that although these populations are not highly genetically differentiated, they exhibit numerous population-specific variants and very large population sizes (Ouhrouch et al., 2021). For example, the D'man sheep population is well adapted to arid and semi-arid environments and has an exceptional reproductive capacity. Studies on the genetic diversity and population structure of D'man sheep reveal the population's pronounced homogeneity and high level of inbreeding. This may result from their geographical isolation in restricted areas (oases), where flocks are small. Management plans should take this into account to mitigate the impact of inbreeding on the sustainability of the D'man population (Simm et al., 2023). In Algeria, the genetic uniqueness of D'man sheep has been preserved better than that of other sheep populations, such as the Berber and Barbarine breeds. The latter have undergone genetic dilution due to uncontrolled breeding with Ouled-Djellal sheep (Gaouar et al., 2017) According to Ben Jemaa et al. (2019), the *FecL<sup>L</sup>* allele was discovered in the French Lacaune population, North African D'man populations, and more recently in the Noire du Velay population. This discovery suggests that the breeds concerned share a common ancestral origin, where the mutation may have occurred before the populations were separated geographically. This type of ancient mutation could then have been transmitted through reproduction across generations. The practice of selecting sheep with improved characteristics is thought to have started in Southwest Asia, before spreading to Europe, Africa and the rest of Asia (Chessa et al., 2009). Additionally, it is possible that the *FecL<sup>L</sup>* allele appeared in these populations independently. However, it is difficult to determine the exact origin without more genetic information. Further studies to investigate the mutation in other populations could shed light on the question.

Despite its high prolificacy, the D'man population is considered to be one of the minority sheep populations in Algeria. Given its small population, it is important to develop further conservation and improvement programmes. Additional studies, involving a large number of animals and sequencing technologies, are recommended to improve our understanding of the gene polymorphism linked to prolificacy in different Algerian sheep populations. The identification of the prolific *FecL<sup>L</sup>* allele is an asset for genetic selection programmes, which target the LB/L trait in order to improve the numerical productivity of sheep in Algeria.

## ■ CONCLUSION

In this study, we identified the presence of segregation of the *FecL<sup>L</sup>* prolific allele of the *B4GALNT2* gene in the Algerian D'man population. This explains the genetic basis of its high prolificacy. The discovery opens new avenues for increasing sheep productivity in Algeria. We suggest that marker-assisted selection can be used to optimize prolificacy, especially in semi-intensive or intensive farming systems. One interesting approach would be to introduce the desired *FecL<sup>L</sup>* mutation into other local breeds through crossbreeding. This could significantly enhance prolificacy and, therefore, lamb production in a relatively short timeframe. However, it is crucial to strike a balance between improving prolificacy through crossbreeding and maintaining the genetic integrity of well-adapted local breeds in order to preserve long-term genetic diversity. Genotyping individuals early on to identify the desired phenotype could help improve the profitability of sheep farming. This method could pave the way for faster and more effective genetic selection.

This breeding strategy would conserve the adaptability and production characteristics of the local breeds, while increasing prolificacy. It has the potential to rapidly improve sheep production in Algeria, without resorting to methods, such as sponges and hormonal treatment. In parallel, a pure breed conservation programme should be launched to guarantee sustainable genetic diversity.

## Acknowledgments

We would like to thank all the members of the GenPhySE Laboratory, National Institute for Agriculture, Food and Environment Research (INRAE), Occitanie Centre, Toulouse, France for conducting the experiments in the framework of this research project.

## Funding

This research was funded by the Ministry of Higher Education through the FNR (Formation Nationale de la Recherche) for the two Algerian laboratories, the Laboratory of Molecular and Cellular Genetics (LGMC) and the Laboratory of Physiopathology and Biochemistry of Nutrition (PpBioNut). Authors received also additional funding from the GenPhySE laboratory, INRAE-Toulouse, France.

## Conflicts of interest

The authors declare that there is no conflict of interest.

## Ethics approval

All procedures were carried out in accordance with the ethical standards of the relevant institutional and national guidelines for the care and use of animals, as stipulated by the Ministry of Agriculture and Rural Development (MADR) of Algeria, under law number 88-08 concerning veterinary activities.

## Author contributions

All the authors were involved in defining the aim of this article and took part in sampling, genotyping and data analysis. The manuscript was written by A.H. and the review was conducted by N.T.A., S.B.S.G., F.M. and S.F.

## Data availability

The data were not deposited in an official repository. The data that support the study findings are available from the authors upon request.

## Declaration of Generative AI and AI-assisted technologies in the writing process

The authors did not use any artificial-intelligence assisted technologies when writing the article.

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## Resumé

**Hadjazi A., Belharfi F.Z., Mahammi F.Z., Fabre S., Gaouar S.B.S., Tabet-Aoul N.** Dépistage de l'allèle prolifique *FecL* du gène *B4GALNT2* chez les populations ovines algériennes

Les principaux gènes contrôlant la prolificité chez les moutons, également connus sous le nom de gènes de fécondité (gènes Fec), sont depuis longtemps utilisés comme outil intéressant pour comprendre les mécanismes impliqués dans la fertilité femelle et dans l'amélioration génétique de la prolificité des troupeaux de moutons. Parmi ces gènes, *FecL/B4GALNT2* et son allèle prolifique *FecL*, ségrégant dans la population ovine française Lacaune et certaines populations locales marocaines et tunisiennes, influencent la fonction ovarienne d'une manière différente des autres gènes majeurs précédemment découverts tels que *FecX/BMP15*, *FecG/GDF9* et *FecB/BMPR1B*, agissant tous dans la voie de signalisation de la protéine morphogénétique osseuse BMP. Afin d'étudier la ségrégation de l'allèle *FecL* dans les populations de moutons algériens, le génotypage de 338 animaux issus de 12 races a été réalisé à l'aide de PCR-RFLP. Nos résultats ont montré la présence de *FecL* uniquement dans la population ovine algérienne D'man. Parmi les moutons D'man génotypés, 21 % étaient porteurs de la mutation à l'état hétérozygote. La fréquence de l'allèle *FecL* dans la population algérienne de D'man (0,11) est proche de ce qui est observé chez les Lacaune et reste relativement faible comparée aux D'man marocains (0,58) et tunisiens (0,65). Le partage de l'allèle *FecL* entre les populations françaises Lacaune et les populations D'man du Nord d'Afrique pourrait indiquer soit une origine ancestrale de la mutation dans *B4GALNT2*, soit un événement d'introgession ancien visant à améliorer la prolificité. Quoi qu'il en soit, la gestion de cette mutation au sein des troupeaux D'man algériens pourrait fournir un outil de départ pour l'amélioration de la productivité numérique des moutons D'man en Algérie.

**Mots-clés :** Ovin, génotype, polymorphisme génétique, productivité, race indigène, Algérie

## Resumen

**Hadjazi A., Belharfi F.Z., Mahammi F.Z., Fabre S., Gaouar S.B.S., Tabet-Aoul N.** Detección del alelo prolífico *FecL* del gen *B4GALNT2* en poblaciones ovinas argelinas

Los principales genes que controlan la prolificidad de las ovejas, también conocidos con el nombre de genes de fecundidad (genes Fec), se utilizan desde hace tiempo como herramienta interesante para comprender los mecanismos implicados en la fertilidad de las hembras y en la mejora genética de la prolificidad de los rebaños de ovejas. Entre estos genes, el *FecL/B4GALNT2* y su alelo prolífico *FecL*, segregados en la población ovina francesa Lacaune y algunas poblaciones locales marroquíes y tunecinas, influyen en la función ovárica de una forma diferente a otros genes importantes descubiertos anteriormente, como el *FecX/BMP15*, el *FecG/GDF9* y el *FecB/BMPR1B*, todos ellos actúan en la vía de señalización de la proteína morfogenética ósea BMP. Con la finalidad de estudiar la segregación del alelo *FecL* en las poblaciones de ovejas argelinas, se realizó el genotipado de 338 animales procedentes de 12 razas utilizando PCR-RFLP. Nuestros resultados mostraron la presencia de *FecL* únicamente en la población ovina argelina D'man. Entre las ovejas D'man genotipadas, el 21 % eran portadoras de la mutación en el estado heterocigótico. La frecuencia del alelo *FecL* en la población argelina de D'man (0,11) es parecida a lo que se observa en las Lacaune y resulta relativamente baja comparada con las D'man marroquíes (0,58) y tunecinas (0,65). La compartición del alelo *FecL* entre las poblaciones francesas Lacaune y las poblaciones D'man del norte de África podría indicar o bien un origen ancestral de la mutación en *B4GALNT2*, o bien un acontecimiento de introgresión antiguo para mejorar la prolificidad. Sea como sea, la gestión de esta mutación en el seno de los rebaños D'man argelinos podría proporcionar una herramienta de partida para la mejora de la productividad numérica de las ovejas D'man en Argelia.

**Palabras clave:** Ovinos, genotipos, polimorfismo genético, productividad, razas indígenas, Argelia



