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RESEARCH ARTICLE

Biodiversity and selection for scrapie resistance in sheep: genetic polymorphism in eight breeds of Algeria

AMAL DJAOUT[1,2]*, BARBARA CHIAPPINI[3], SEMIR-BECHIR-SUHEIL GAOUAR[4], FARIDA AFRI-BOUZEBDA[1], MICHELA CONTE[3], FAKHREDDINE CHEKKAL[5], RACHID EL-BOUYAHIAOUI[2], RACHID BOUKHARI[4], UMBERTO AGRIMI[3] and GABRIELE VACCARI[3]

[1] *Laboratoire de Production Animale, Biotechnologie et Santé, Institut des Sciences Agrovétérinaires (ISAV), Université Mohammed Cherif Messaadia, 41000 Souk-Ahras, Algeria*
[2] *Institut National de la Recherche Agronomique d'Algérie (INRAA), 19000 Sétif, Algeria*
[3] *Department of Food Safety, Nutrition and Veterinary Public Health, Istituto Superiore di Sanità, 00161 Rome, Italy*
[4] *Laboratoire de Physiopathologie et Biochimie de la Nutrition (PpBioNut), Université Abou Bekr Belkaid, 13000 Tlemcen, Algeria*
[5] *Centre de Recherche Scientifique et Technique sur les Régions Arides (CRSTRA), 07000 Biskra, Algeria*
*For correspondence. E-mail: djaout.amel08@gmail.com.

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Abstract. Scrapie is a prion disease that affects the sheep and goats. It belongs to the group of transmissible spongiform encephalopathies (TSE). TSEs are characterized by the accumulation of the pathological form (PrP^{Sc}) of the cellular prion protein (PrP^{C}). The susceptibility of sheep to scrapie is influenced by polymorphisms in the PrP gene (*PRNP*). The aim of this study was to identify the genetic variability of sheep *PRNP* in Algerian sheep. Two-hundred and thirteen Algerian sheep from eight breeds (Ouled Djellal, Rembi, Hamra, Berbere, Barbarine, Sidaou, Taadmit and Tazegzawt) with no clinical manifestation of scrapie were analysed. Sequencing of the entire coding sequence of *PRNP* showed four main alleles (ARQ, ARR, AHQ and ARH) based on codons 136, 154 and 171 with different frequencies among the investigated breeds. Moreover, 14 additional nonsynonymous polymorphisms (Q101R, N103K, M112T, A116P, M137I, L141F, I142M, H143R, N146S, R151G, Y172D, N176K, H180Y and S240P) as well as two synonymous polymorphisms at codons 231 and 237 were found in the *PRNP* gene. Interestingly, the N103K, M137I and I142M polymorphisms were not described in sheep. The ARQ, ARR and ARH haplotypes were present in all breeds with a highest frequency of ARQ in Barbarine. The ARH was absent in Barbarine breed and the VRQ haplotype was absent in all Algerian breeds studied. The ARQ and ARR alleles were the most common with frequencies ranging from 30 to 65% and from 8 to 26%, respectively, in different breeds. These results represent the first study on *PRNP* variability in Algerian sheep and may serve as a basis for the development of breeding programmes to render national sheep breeds resistant to scrapie.

Keywords. *PRNP* gene; sheep breeds; polymorphism; Algeria.

Introduction

The importance of sheep breeding in Algeria (26,880,000 heads (MADR 2013)) depend on the richness of genetic resources. Currently, this herd is composed of more than nine breeds (Ouled Djellal, Rembi, Hamra, Berbere, Barbarine, D'man, Sidaou, Taadmit and Tazegzawt) with various resistance characteristics, prolificacy, productivity of meat, milk and wool, also good adaptability to arid, steppe and saharan conditions. Algerian sheep farming is intended for the production of meat, which is considered as the principal source in Algeria. In fact, the religious and culinary habits target the consumption of sheep meat per year and per inhabitant above that of beef (2614092 versus 1321433) Quintal (MADR 2013).

In fact, the genetic characterization is the best way to study the conservation and improvement of herd productivity. In this case, we studied the genetic polymorphisms of the prion protein gene (*PRNP*) involved in the susceptibility of sheep to scrapie in eight sheep breeds of Algeria.

Transmissible spongiform encephalopathies (TSEs) or prion are neurodegenerative diseases of the central nervous system (CNS) affecting humans and animals. Member of this family are in humans (Creutzfeldt–Jakob disease) and its variant (vCJD) linked with the bovine spongiform encephalopathy (BSE) of cattle, classical and atypical scrapie (Nor98) in sheep and goats, and chronic wasting disease (CWD) in deer. They are mainly characterized by the accumulation of a pathological isoform (PrP^{Sc}) of the cellular prion protein (PrP^{C}) in the CNS (Prusiner 1993; Palmer and Collinge 1997).

In sheep, the susceptibility of an individual to develop the disease is determined by its genetic heritage. Definitely, the resistance of sheep to scrapie is strongly controlled by polymorphisms of the PrP gene (*PRNP*) and modulated by the strain of the agent (for a review, see Goldmann 2008). In particular, the *PRNP* polymorphisms at codons 136 (A/V), 154 (R/H) and 171 (Q/H/R/K) are the main determinants of susceptibility/resistance of sheep to classical scrapie. They are combined with six main variants of the wild-type allele: ARQ, VRQ, AHQ, ARH, ARK and ARR. The genotypes resulting from the combination of these alleles are grouped into five categories according to the level of resistance to scrapie (Dawson *et al.* 2008). It is well established that genotypes of the ARR allele are associated with resistance to classical scrapie with the exception of the ARR/VRQ genotype (Goldmann *et al.* 1994; Hunter *et al.* 1994, 1996; Baylis *et al.* 2002a, b). Certainly, the VRQ allele confers high susceptibility in all its genotype combinations instead, the ARQ/ARQ genotype is associated with high to moderate susceptibility to classical scrapie depending on the scrapie strain (Goldmann *et al.* 1994; Hunter *et al.* 1994, 1996; Baylis *et al.* 2002a, b). The remaining genotypes with AHQ or ARH alleles are associated with an intermediate susceptibility (Vaccari *et al.* 2007).

Additional variations at codons 136, 154 and 171, such as TRQ, ALQ, ARK, VHQ, AHR and VRR (Kutzer *et al.* 2002; De Silva *et al.* 2003; Billinis *et al.* 2004; Alvarez *et al.* 2006) were described at very low frequencies in some sheep breed but their association with susceptibility is still unknown. However, the variability of sheep PRNP is greater than that of these three codons and an additional 24 polymorphic codons were described to date, giving rise to 43 allelic variants mainly derived from variations of the ARQ allele (Goldmann 2008).

For some of these alleles, a clear effect influencing susceptibility to classical or atypical scrapie was observed. In particular, the allele $AF_{141}RQ$ with A at codon 136, F at codon 141, R at codon 154 and Q at codon 171, was associated along with the allele AHQ from high susceptibility to the atypical form of scrapie named Nor98 (Moum *et al.* 2005; Goldmann 2008). Interestingly, additional alleles such as $AT_{137}RQ$ and $ARQK_{176}$ were associated with resistance to classical scrapie (Vaccari *et al.* 2007, 2009a, b).

Based on the evidences, susceptibility of sheep to scrapie is greatly influenced by the host genotype at the *PRNP* with the EU Decision 2003/100/EC. It was required to each European Member State to introduce breeding programmes to increase the frequency of the resistant allele in sheep populations.

Actually, in some European countries, a statistically significant decrease in the prevalence of the disease was observed (EFSA BIOHAZ Panel 2014). Although no scrapie case was documented yet in any breed of Algerian sheep (Kalai *et al.* 2017), identifying the PRNP polymorphism spectrum in Algerian sheep may be useful for the implementation of future breeding plans in terms of scrapie resistance.

Materials and methods

Samples

Two-hundred and thirteen blood samples from eight sheep breeds, Ouled Djellal (35), Hamra (27), Rembi (40), Berbere (20), Barbarine (20), Tazegzawt (31), Taadmit (10) and Sidaou (30) were collected. All selected sheep were identified and their history, age, breed and sex were obtained. Sampling was done choosing each animal from a different flock and from all 16 provinces of Algeria (table 1). Blood samples were collected from the jugular vein into EDTA tubes and stored at 20°C until genomic DNA extraction.

DNA extraction

The DNA was extracted from the total blood using the standard protocol of NaCl method (Miller *et al.* 1988) and stored at −20°C until further processing.

Sequence analysis

The complete sequence of the *PrP* coding region of all the samples was performed following the protocol described by Vaccari *et al.* (2009a). Briefly, the amplification reactions were set up in a 50 μL reaction volume using 5 μL of extracted DNA and 45 μL of reaction mix as follows: 1× Ampli Taq Gold 360 PCR Buffer, 2.5 mM $MgCl_2$, 1× 360 GC Enhancer, 200 μM dNTPs, 0.25 μM of primer F1 (5′-CATTTATGACCTAGAATGTTTATAGCTGATGCCA-3′) and primer R1 (5′-TTGAATGAATATTATGTGGCC TCCTTCCAGAC-3′), 0.5 μL of Ampli Taq Gold 360 (Life Technologies) according to the following amplification protocol (5 min at 96°C; 30 s at 96°C, 15 s at 57°C, 90 s at 72°C for 35 cycles, 4 min at 72°C).

Primers and dNTPs were removed enzymatically, incubating 15 μL of PCR product with 1.7 μL of illustra Exo Pro Star (GE Healthcare Life Sciences) according

Table 1. Sampling, localities, sample size (*n*) and sex of Algerian sheep breeds studied.

Breed	Abbreviation	Localities	Number of samples analyzed	
			n	Sex
Ouled-Djellal	ODJ (35)	Biskra	13	5 ♀+ 8 ♂
		OumElbouaghi	7	5 ♀+ 2♂
		Setif	5	2 ♀+ 3 ♂
		Tlemcen	9	4 ♀+ 5 ♂
		Tiaret	1	1 ♀
Hamra	HAM (27)	Naama	6	6 ♀
		AïnTimouchent	1	1 ♀
		Tlemcen	1	1 ♀
		Saïda	19	11♀+ 8 ♂
Rembi	REM (40)	Ain Timouchent	11	2 ♀+ 9 ♂
		Tiaret	25	14 ♀+ 11 ♂
		Setif	2	2 ♀
		Naama	1	1 ♀
		El-Tarf	1	1 ♂
Berbere	BER (20)	El-Tarf	17	13 ♀+ 4 ♂
		Skikda	3	3 ♀
Barbarine	BAR (20)	OuedSouf	13	13 ♀
		Saïda	7	4 ♀+ 3 ♂
Tazegzawt	TAZ (31)	Souk-Ahras	2	2 ♀
		Biskra	1	1 ♀
		Naama	3	1 ♀+ 2 ♂
		Bejaia	25	18 ♀+ 7 ♂
Taadmit	TAA (10)	Djelfa	10	10 ♀
Sidaoun	SID (30)	Tamanrasset	25	25 ♀
		Illizi	5	2 ♀+ 3 ♂
Total			213	148 ♀+ 65 ♂

BAR, Barbarine; BER, Berbere; HAM, Hamra; ODJ, Ouled Djellal; REM, Rembi; SID, Sidaou; TAA, Taadmit; TAZ, Tazegzawt.

to the following protocol (15 min at 37°C; 15 min at 80°C). Forward and reverse sequencing reactions were carried out in a final volume of 10 μL using 1 μL of DNA and 9 μL of mix reaction as follows: 0.32 μM of primer T3 (5′-TTTACGTGGGCATTTGATGC-3′) and T4 (5′-GGCTGCAGGTAGACACTCC-3′) using Big Dye Terminator Cycle sequencing kit ver. 1.1 (Life Technologies), following a standard thermal profile (10 s at 96°C; 5 s at 50°C; 4 min at 60°C). Sequences were purified with the Big Dye × Terminator and detected with ABI PRISM3130 apparatus (Applied Biosystems).

Bioinformatics tools

The sequences were aligned by Seq Scape 2.5.0 (Applied Biosystems) software. Analysis of Hardy–Weinberg equilibrium (HWE) and χ^2 test were calculated using STATISTICA 7.

Results

This study provides data about polymorphisms of the sheep PRNP locus in eight Algerian breeds. In total, 18 polymorphic sites (table 2) combined in 44 different genotypes (table 3) were observed. All genotypes were in HWE. Four alleles (ARQ, ARR, $AR_{143}RQ$ and $ARQK_{176}$) were detected in all the sheep breeds (table 2). The ARQ is the most frequent allele in all the breeds with higher frequency in Barbarine (65%) and the lowest in Taadmit (30%) followed by ARR allele; $AR_{143}RQ$ and $ARQK_{176}$ alleles were detected at low frequencies (table 2).

The resistant ARR allele frequencies vary between 26 and 8%, respectively, in Hamra and both in Sidaou and Tazegzawt, it is the second most frequent allele with the exception of Sidaou and Tazegzawt breeds. The $AR_{143}RQ$ is the second most frequent allele in Sidaou with a 17% and it varies between 20% in Taadmit and 5% in Tazegzawt. The $ARQK_{176}$ allele is also present in all the breeds and the frequencies vary between 15 and 5%. Other important alleles, ARH and AHQ are present respectively, in five or six of eight analysed breeds. Interestingly, the VRQ was not observed in the entire analysed breed.

Twelve additional alleles ($R_{101}ARQ$, $K_{103}ARQ$ T_{112} ARQ, $P_{116}ARQ$, $AI_{137}RQ$, $AF_{141}RQ$, $AM_{142}RQ$, AS_{146} RQ, $AG_{151}RQ$, $ARQD_{172}$, $ARQY_{180}$ and $ARQP_{240}$) were detected with low frequencies in the breeds analysed.

The $K_{103}ARQ$, $AI_{137}RQ$ and $AM_{142}RQ$ alleles are described for the first time in sheep. The $K_{103}ARQ$ allele

Table 2. PRNP haplotypes frequencies (%) of Algerian sheep breeds.

Alleles	BAR ($n = 20$)	BER ($n = 20$)	HAM ($n = 27$)	ODJ ($n = 35$)	REM ($n = 40$)	SID ($n = 30$)	TAA ($n = 10$)	TAZ ($n = 31$)
ARQ	65	48	41	31	43	45	30	47
ARR	15	18	11	26	24	8	30	8
AHQ	–	3	7	3	3	2	10	3
ARH	–	10	9	3	1	–	–	18
R_{101}ARQ	–	–	2	–	1	–	–	–
K_{103}ARQ	–	3	–	–	–	–	–	–
T_{112}ARQ	–	–	–	–	3	–	–	–
P_{116}ARQ	–	–	–	–	–	7	–	–
AI_{137}RQ	–	–	–	3	–	–	–	–
AF_{141}RQ	–	–	–	–	1	–	5	2
AM_{142}RQ	–	–	–	1	–	–	–	–
AR_{143}RQ	13	10	9	18	13	17	20	5
AS_{146}RQ	–	–	9	1	–	–	–	2
AG_{151}RQ	–	3	–	–	–	3	–	–
ARQD_{172}	–	–	–	–	–	3	–	–
ARQK_{176}	5	5	11	7	10	15	5	7
ARQY_{180}	3	3	–	4	3	–	–	10
ARQP_{240}	–	–	–	3	–	–	–	–

(nucleotide change AAC to AAG at codon 103) and AM_{142}RQ (nucleotide change ATA to ATG at codon 142) were observed in heterozygote animals, while the AI_{137}RQ (nucleotide change ATG to ATA at codon 137) was observed in one homozygote animal (figures 1, 2 and 3). Further, two silent nucleotides polymorphisms at codons 231 (AGG/CGG) and 237 (CTC/CTG) were detected in all the breeds (data not shown).

Within the National Scrapie Plan (Dawson *et al.* 2008), the sheep are classified on the basis of PrP genotype at codons 136, 154 and 171 as five risk groups (R1–R5) according to susceptibility to scrapie. Based on such classification in Algeria, the most resistant animals of group R1 (those with the ARR/ARR genotype) were observed in Taadmit, Rembi, Ouled Djellal and Barbarine breeds with low frequencies ranging from 20 to 8%, however, such genotype is absent in Barbarine, Hamra, Sidaou and Tazegzawt breeds.

The resistant R2 group comprehending ARR/ARQ, ARR/AHQ and ARR/ARH genotypes (table 3) is the second most represented group.

The ARR/ARQ is present in all Algerian sheep breeds and it is the most frequent genotype of the group ranging from 20% in Barbarine, Berbere and Rembi to 3% in Sidaou. The ARR/AHQ was observed only in Taadmit and Rembi breeds with low frequencies (10 and 3%).

The third genotype of this group, ARR/ARH was absent in Algerian sheep breeds studied. Sheep of this type are genetically resistant to scrapie but will need careful selection when used for further breeding.

The group R3 comprehending genotypes of animals susceptible to scrapie (ARQ/ARQ, AHQ/ARQ, ARH/ARQ, AHQ/AHQ, AHQ/ARH and ARH/ARH) is the most represented group. The ARQ/ARQ was observed in all the breeds and represented the most frequent genotype with a mean of 20% ranging from 40% in Barbarine to 11% in both Hamra and Ouled Djellal. The rest of the genotypes (table 3) were observed in low frequencies. Interestingly, no animal was classified in group R4 or R5, those that are highly susceptible to scrapie.

Discussion

Scrapie of sheep and goats is an inevitably fatal disease for which the tools used for the management of 'conventional' infectious disease exhibited ineffective results. Definitely, control policies based on selecting rams of resistant genotype for breeding showed positive results in the contest of both outbreak control in classical scrapie-affected sheep flocks and disease prevention showing a decrease of scrapie prevalence in some European country (EFSA BIOHAZ Panel 2014).

In Algeria, although no scrapie case was described, it would be important to evaluate the genetic status of sheep with respect to susceptibility to classical scrapie

Table 3. PRNP genotypes and scrapie risk group frequencies (in %) of Algerian sheep.

Scrapie risk	Genotype	BAR ($n = 20$)	BER ($n = 20$)	HAM ($n = 27$)	ODJ ($n = 35$)	REM ($n = 40$)	SID ($n = 30$)	TAA ($n = 10$)	TAZ ($n = 31$)
Scrapie risk R1	ARR/ARR	–	5	–	8	8	–	20	–
Total scrapie risk R1		–	5	–	8	8	–	20	–
Scrapie risk R2	ARR/ARQ	20	20	19	17	20	3	10	10
	ARR/AF_{141}RQ	–	–	–	–	–	–	–	3
	ARR/AG_{151}RQ	–	5	–	–	–	–	–	–
	ARR/AR_{143}RQ	–	–	–	11	3	10	–	–
	ARR/$ARQK_{176}$	10	–	4	6	3	3	–	–
	ARR/$ARQY_{180}$	–	–	–	3	–	–	–	3
	ARR/AT_{112}RQ	–	–	–	–	5	–	–	–
	ARR/AHQ	–	–	–	–	3	–	10	–
Total scrapie risk R2		30	25	23	37	34	16	20	16
Scrapie risk R3	ARQ/ARQ	40	20	11	11	18	23	10	23
	AF_{141}RQ/AR_{143}RQ	–	–	–	–	–	–	10	–
	AI_{137}RQ/AI_{137}RQ	–	–	–	3	–	–	–	–
	AR_{143}RQ/AR_{143}RQ	–	5	4	3	3	–	–	–
	AR_{143}RQ/$ARQK_{176}$	–	–	–	3	3	7	10	–
	AR_{143}RQ/$ARQY_{180}$	–	–	–	–	–	–	–	3
	AR_{143}RQ/AS_{146}RQ	–	–	4	3	–	–	–	3
	ARQ/AG_{151}RQ	–	–	–	–	–	7	–	–
	ARQ/AM_{142}RQ	–	–	–	3	–	–	–	–
	ARQ/AR_{143}RQ	25	5	7	8	15	13	20	–
	ARQ/$ARQD_{172}$	–	–	–	–	–	7	–	–
	ARQ/$ARQK_{176}$	–	10	11	–	8	7	–	10
	ARQ/$ARQP_{240}$	–	–	–	6	–	–	–	–
	ARQ/$ARQY_{180}$	5	5	–	–	3	–	–	6
	ARQ/AS_{146}RQ	–	–	4	–	–	–	–	–
	ARQ/K_{103}ARQ	–	5	–	–	–	–	–	–
	ARQ/P_{116}ARQ	–	–	–	–	–	3	–	–
	ARQ/R_{101}ARQ	–	–	4	–	–	–	–	–
	$ARQK_{176}$/$ARQY_{180}$	–	–	–	–	3	–	–	–
	$ARQK_{176}$/$ARQK_{176}$	–	–	–	3	3	3	–	–
	$ARQY_{180}$/$ARQY_{180}$	–	–	–	3	–	–	–	–
	AS_{146}RQ/$ARQK_{176}$	–	–	7	–	–	–	–	–
	P_{116}ARQ/AR_{143}RQ	–	–	–	–	–	3	–	–
	P_{116}ARQ/$ARQK_{176}$	–	–	–	–	–	7	–	–
	R_{101}ARQ/AF_{141}RQ	–	–	–	–	3	–	–	–

Table 3 (*contd*)

Scrapie risk	Genotype	BAR ($n = 20$)	BER ($n = 20$)	HAM ($n = 27$)	ODJ ($n = 35$)	REM ($n = 40$)	SID ($n = 30$)	TAA ($n = 10$)	TAZ ($n = 31$)
	AHQ/ARQ	–	–	4	–	3	3	10	3
	AHQ/AR$_{143}$RQ	–	–	–	6	–	–	–	3
	ARH/ARQ	–	10	11	6	3	–	–	19
	ARH/AS$_{146}$RQ	–	–	4	–	–	–	–	–
	ARH/AR$_{143}$RQ	–	5	–	–	–	–	–	–
	ARH/ARQK$_{176}$	–	–	–	–	–	–	–	3
	ARH/ARQY$_{180}$	–	–	–	–	–	–	–	6
	AHQ/AHQ	–	–	4	–	–	–	–	–
	AHQ/ARH	–	5	4	–	–	–	–	–
	ARH/ARH	–	–	–	–	–	–	–	3
Total scrapie risk R3		70	70	79	58	65	83	60	82

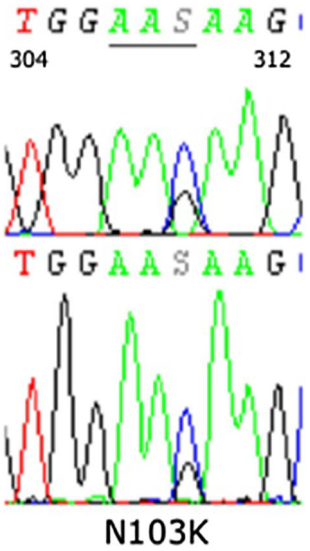

Figure 1. Electropherograms of the new *PRNP* allele variants (AK$_{103}$RQ) observed on Algerian sheep (nucleotides are numbered from the first ATG of the *PRNP* ORF).

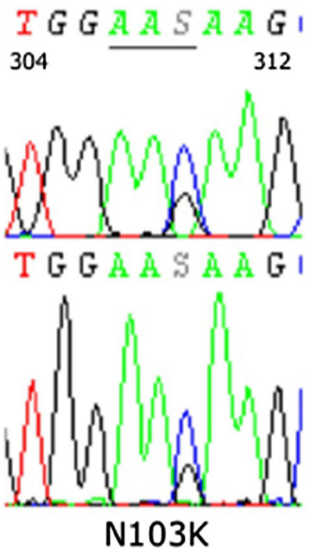

Figure 2. Electropherograms of the new *PRNP* allele variants (AI$_{137}$RQ) observed on Algerian sheep (nucleotides are numbered from the first ATG of the *PRNP* ORF).

and Nor98. A total of 213 samples from the eight most important sheep breeds of the Algerian population were analysed for the evaluation of the potential genetic susceptibility of sheep to scrapie. Based on the absence of identification of the genotypes at more risk of scrapie, those with the VRQ allele and classified as R5 or R4 (Dawson *et al.* 2008), should be, if present in Algerian breed, at very low frequencies

However, the susceptible genotypes belonging to the R3 group are present with a variable but with high frequencies between 83 and 58%. Conversely, resistant or semi

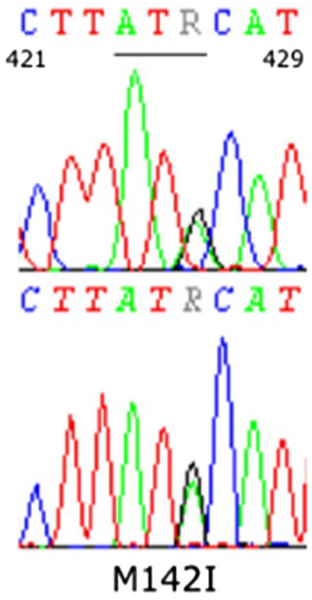

Figure 3. Electropherograms of the new *PRNP* allele variants ($AI_{142}RQ$) observed on Algerian sheep (nucleotides are numbered from the first ATG of the *PRNP* ORF).

resistant genotypes belonging to the risk class 1 (R1) or 2 (R2) are present in low frequencies ranging from 16 to 37%. The variability at the PRNP locus is very high in Algerian sheep. A total of 18 different allelic variants were observed. In Algeria, we have observed the most represented worldwide alleles (ARQ, ARR, AHQ, ARH, $AF_{141}RQ$, $AT_{112}RQ$ and $AR_{143}RQ$) that were observed in Europe (for a review, see Goldmann 2008), Asia (Saunders *et al.* 2009; Guan *et al.* 2011; Lan *et al.* 2014), Africa (Serrano *et al.* 2007; Kdidi *et al.* 2014), America (Harrington *et al.* 2010) and Australia.

The $ARQK_{176}$ allele is highly represented in Algerian sheep. Interestingly, it was associated with scrapie resistance in case–control studies, in *in vivo* transmission studies and *in vitro* conversion studies (Vaccari *et al.* 2007; Vaccari *et al.* 2009a; Bucalossi *et al.* 2011). It is important to note that this allele was observed in all the Algerian breeds with a frequency ranging from 5 to 15% and it is present in some Italian, Spanish and west African breeds, and in all north African sheep breeds analysed so far (Serrano *et al.* 2007; Kdidi *et al.* 2014).

Another potential resistant allele, $AS_{146}RQ$ is the homologous of a goat allele indicated as resistant in Cyprus (Papasavva-Stylianou *et al.* 2011) and is present in three Algerian sheep breeds and it was already observed in Turkish, Iranian, Asian (Ün *et al.* 2008; Alvarez *et al.* 2011; Karami *et al.* 2011; Meydan *et al.* 2013) and Tunisian sheep (Barbarine and Western Thin Tail) (Kdidi *et al.* 2014).

Other alleles already observed in African sheep breeds, $AR_{101}RQ$, $P_{116}ARQ$, $AG_{151}RQ$, $ARQD_{172}$, $ARQY_{180}$ and $ARQP_{240}$ were also found in some of the Algerian sheep breeds analysed at very low frequencies. The $AR_{101}RQ$ allele observed in the Hamra and Rembi breeds was observed in two Tunisian (Kdidi *et al.* 2014), in several Spanish (Acín *et al.* 2004), in a few Chinese (Lan *et al.* 2006; Guan *et al.* 2011), one Italian (Curcio *et al.* 2015) and Turkish (Meydan *et al.* 2013) sheep breeds. The $P_{116}ARQ$ allele was observed in Sidaou breed and previously described only in west African sheep (Traoré *et al.* 2012) and sheep in the USA in the St Croix White breed (Seabury and Derr 2003), a breed developed from sheep of west Africa brought to the Americas in the 1600. The $ARQD_{172}$ allele observed only in Sidaou breed in Algeria was already described in African breed in west Africa and Tunisia (Traoré *et al.* 2012; Kdidi *et al.* 2014), but it was also observed in Turkey, Spain and Iran (Acín *et al.* 2004; Alvarez *et al.* 2011; Karami *et al.* 2011). The $ARQY_{180}$ was already described in Tunisia (Kdidi *et al.* 2014), Italy (Acutis *et al.* 2004; Curcio *et al.* 2015) and the USA (De Silva *et al.* 2003).

Interestingly, the $ARQP_{240}$, i.e. homologous of one of the two wild-type alleles of goats (Vaccari *et al.* 2009b), was reported here in two animals of the Ouled Djellal and was already observed only in sheep of Moussi breed in Burkina Faso (Traoré *et al.* 2012).

Polymorphisms of M137I and I142M were already observed in goats (Goldmann *et al.* 1990; Acutis *et al.* 2004) and are found here for the first time in sheep. Of interest, the $AI_{137}RQ$ and $AM_{142}RQ$ alleles reported here are in linkage with Serine (S) at codon 240, while in goats I137 was first described in linkage with Proline (P) at codon 240 (Goldmann *et al.* 1996; Acutis *et al.* 2004) and also subsequently observed in linkage with Serine (S) (Serrano *et al.* 2007; Windig *et al.* 2016).

Finally, the $AG_{151}RQ$ allele was observed in Africa, it was also observed in Berbere and Sidaou, Tunisian Sicilo Sarde (Kdidi *et al.* 2014) and west African sheep (Traoré *et al.* 2012).

In conclusion, this work represents the first report on Algerian sheep's *PRNP* gene variability. Our results showed the presence of relatively high frequencies of the ARQ haplotype, but also of other haplotypes possibly associated with scrapie resistance, such as $AS_{146}RQ$ and $ARQK_{176}$. The VRQ haplotype, associated with higher susceptibility to scrapie, was absent in all the Algerian breeds studied. Overall, our results indicated that the sheep population in Algeria could be susceptible to both classical and atypical scrapie. These results will eventually help the development of breeding programmes in Algeria to render sheep resistant to scrapie. This is the fourth study of PRNP polymorphisms in African sheep breeds, after the study from Morrocco, Tunisia and west African countries. These preliminary results indicate that the Algerian indigenous sheep have a moderate frequencies of genotypes associated with scrapie resistance, but a considerable high genetic variability at the *PRNP* locus.

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