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Selection for scrapie resistance in the French dairy sheep populations: Breeding strategy and main results since 1995

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SUMMARY – Studies of natural and experimentally induced scrapie in sheep have shown that genetic susceptibility to the disease is modulated by allelic variation in three different codons in the sheep *PrP* gene (136, 154 and 171). Besides the wildtype $A_{136}R_{154}Q_{171}$ allele, four mutually exclusive allelic variants are found in sheep: ARR, AHQ, ARH and VRQ. Studies of *PrP* genotypes linked to scrapie, carried out since 1995 in French dairy sheep populations, were in agreement with the abundant literature in other sheep breeds: it confirmed that only the homozygous ARR/ARR animals are clinically resistant. Moreover demonstration action, based on the use of ARR/ARR rams in 15 highly affected flocks, showed a rapid decrease of scrapie incidence in two years. Therefore, the selection of ARR/ARR genotypes may be a strategy to control scrapie at the population level, provided that the genetically resistant sheep are not healthy carriers of infectivity. The monitoring implemented since 1999 in these 15 dairy sheep affected flocks, based on tonsil samples of culled reproducers to detect PrP^{sc}, so far reject this hypothesis in agreement with previous publications. The *PrP* genotype distribution of the 5 French dairy sheep breeds is now well known since 6443 AI rams and 3149 elite ewes have been genotyped between 1995 and 2001. Large differences between these 5 breeds were revealed, and were taken into account when conceiving a selection program on scrapie resistance using *PrP* genotyping. This selection started between 1995 and 2001 according to the considered breed. From 2002 about 15,000 to 20,000 genotypings of animals per year during 5 years are planned, mainly candidate rams for the AI centres and elite ewes of the nucleus flocks to account for the *PrP* genotypes in the assortive matings. Main objectives related to the *PrP* genotype of the rams are presented.

Key words: Dairy sheep, scrapie, *PrP* genotype, healthy carrier, breeding.

RESUME – *"Sélection pour la résistance à la tremblante dans les populations ovines laitières françaises : Stratégie d'amélioration et principaux résultats depuis 1995". Les études de tremblante naturelle et expérimentale en ovins ont montré que la susceptibité génétique à la maladie est modulée par le polymorphisme allélique en 3 codons (136, 154 et 171) du gène* PrP *ovin. Outre l'allèle sauvage $A_{136}R_{154}Q_{171}$, quatre autres allèles mutuellement exclusifs sont trouvés chez le mouton : ARR, AHQ, ARH et VRQ. Les études d'association entre génotypes* PrP *des ovins aux 3 codons et incidence de la tremblante naturelle, conduites depuis 1995 dans les populations françaises ovines laitières, sont cohérentes avec l'abondante littérature disponible dans d'autres races ovines : elles confirment que seuls les ovins ARR/ARR sont cliniquement résistants. De plus, une action de démonstration, fondée sur l'utilisation de béliers ARR/ARR dans 15 élevages très infectés, a montré une rapide diminution de l'incidence de la tremblante en deux ans. En conséquence, la sélection en faveur du génotype ARR/ARR peut constituer une stratégie pour contrôler la tremblante à l'échelle d'une population, à condition toutefois que les ovins résistants ne soient pas des porteurs sains de l'agent infectieux. La surveillance mise en place depuis 1999 dans ces 15 élevages ovins laitiers atteints de tremblante, fondée sur des prélèvements d'amygdales de brebis de réforme pour la détection de PrP^{sc}, rejette pour l'instant l'hypothèse de porteurs sains conformément aux publications déjà disponibles. La structure génétique au gène* PrP *des 5 races de brebis laitières françaises est maintenant bien connue, puisque 6443 béliers d'IA et 3149 mères à béliers ont été génotypés de 1995 à 2001. Des différences importantes existent entre ces 5 races, dont il faut tenir compte pour concevoir un programme de sélection pour la résistance à la tremblante fondé sur le génotypage au gène* PrP. *Cette sélection a débuté entre 1995 et 2001 selon chaque race considérée. A partir de 2002, celle-ci suppose le* *génotypage de 15 000 à 20 000 animaux par an durant 5 ans, principalement les béliers candidats à l'entrée en centres d'insémination artificielle et les mères à béliers dans les noyaux de sélection, pour tenir compte des génotypes* PrP *dans les accouplements raisonnés. Les principaux objectifs concernant le génotype* PrP *des béliers sont présentés.*

Mots-clés : *Ovins laitiers, tremblante, génotype* PrP*, porteur sain, sélection.*

Introduction

Scrapie, the ovine prion disease, belongs to the Transmissible Spongiform Encephalopathies (TSE), which also include Creutzfeldt-Jakob disease in humans and Bovine Spongiform Encephalopathy (BSE) in cattle. An increasing amount of information about natural scrapie has been accumulated and it is now well known that genetic susceptibility to scrapie is modulated by allelic variations at three codons (136, 154 and 171) in the sheep *PrP* gene encoding for protein PrP (Hunter, 1997). On the other hand, the nature of the scrapie agent and its mode of transmission are still under debate. Following Prusiner hypothesis (1982), this agent is an infectious protein, the PrP itself (PrP for prion – proteinaceous infectious particle – protein). Moreover it has been proven that different scrapie strains co-exist: until now the strain typing is costly and time consuming since it is based on mouse bioassay as developed in Edinburgh (Bruce *et al.*, 1994). Under these conditions, breeding for scrapie resistance may be consider as a sound strategy (Dawson *et al.*, 1998; Smits *et al.*, 2000) only if the two following critical points are well documented: (i) the resistant sheep must be clinically resistant against any scrapie strain and also against BSE strain; and (ii) the possibility that resistant animals may be healthy carriers of infectivity must be carefully verified.

In French dairy sheep populations, research contributing to give answers to the above questions has started in 1995. In a first step, the *PrP* genotype structure of the 5 French dairy sheep breeds has been described by *PrP* genotyping each year the AI rams (6443 rams between 1995 and 2001). Then epidemiological studies were implemented respectively in 10 and 32 Lacaune and Manech blond faced flocks, to carry out a case-control study at the population level, whose main results have already been presented (Barillet *et al.*, 2002). Third, demonstration action using resistant AI rams in 15 affected flocks of the Manech blond faced breed has been in progress since 1999, as an alternative solution to the usual slaughtering of the flock according to the current policy. Finally, the selection for scrapie resistance has been gradually intensified in the French dairy sheep populations, as the answers to the above critical questions have been favourably accumulated in France and Europe for the last five years.

The purpose of this paper is to present the main results of the demonstration action in progress in 15 affected Manech blond faced flocks. Then the implementation of the selection for scrapie resistance at the population level within the framework of the existing breeding programmes is described.

PrP genotypes and scrapie susceptibility

PrP genotyping at the 3 codons 136, 154 and 171 was performed using a PCR-RFLP method which did not distinguish the alleles H and Q at the codon 171, so that only 4 alleles were described at the 3 codons: ARR, AHQ, ARQ and VRQ. A monitoring of 15 affected Manech blond faced flocks has been implemented in 1999, all the sheep of these flocks being *PrP* genotyped (7346 until now). Most of these flocks had a high incidence of scrapie, often over 10% of the exposed animals, before starting a demonstration action based on the use by AI of resistant ARR/ARR rams to eradicate scrapie: as described in Table 1, this action started in 1998 for one of these 15 flocks, in 1999 for three others, in 2000 for 3 new flocks. Finally all the 15 flocks have been involved in this action in 2001 at least for one year (Table 1).

For 3803 ewes born between 1997-2000 and exposed in 12 of these 15 flocks, an analysis of risk factors was performed using a logistic regression model. The fixed effects included in the model, i.e. potential risk factors for scrapie, were year of birth of the ewes (1997 to 2000), type of replacement (born in the flock or bought ewe), flock effect and *PrP* genotype of the ewes. Odds Ratio (OR) measures how much more (or less) likely the outcome is among observations with a reference level of risk factor. The reference levels were ewes born in 1997, animals bought from another flock for replacement, ewes bred in the flock H and genotyped ARQ/ARQ. All the 4 effects were highly significant (Table 2). The risk to be scrapie affected decreased logically dramatically according to year of birth, the ewes born in 2000 being 5 times less risky than those born in 1997 in agreement with the increase over time of the use of AI resistant sires in these 12 flocks (Table 2). Compared to ARQ/ARQ ewes, the risk to be scrapie affected increased significantly by 2 times for ARQ/VRQ or VRQ/VRQ sheep, by 3 times for not genotyped ewes suggesting that they were mostly ARQ/VRQ or VRQ/VRQ sheep. Conversely the risk was extremely low for ARR/ARQ animals (OR = 0.02) while ARR/ARR sheep were never affected. In agreement with the literature reviewed in Hunter (1997) and Smits *et al*. (2000), most of the scrapie cases appeared as associated with the genotype VRQ/VRQ, ARQ/VRQ and ARQ/ARQ. The ARR allele is nearly dominant over the other alleles since most of the ARR/ARQ animals appear to be resistant.

Table 1. Demonstration action in 15 affected flocks (Manech blond faced breed): percentage of ewes procreated by AI rams genotyped ARR/ARR according to the year of birth of the females (born in the flock or bought)

Flock	Year of birth of the ewes				
	1997	1998	1999	2000	2001
A	0	1	0	1	72
B	2	0	2	14	95
C	3	5	100	100	100
D	0	9	3	64	62
E	10	0	2	0	51
F	1	4	0	1	98
G	11	5	5	34	100
H	4	2	4	63	100
I	16	56	100	98	100
J	0	2	2	100	94
K	5	16	58	0	63
L	9	0	6	24	64
M	1	0	1	62	100
N	0	0	0	6	38
O	0	0	0	14	89

PrP^{sc} carrying and *PrP* genotypes

Transmissible spongiform encephalopathies are characterised by the accumulation in the brain of an abnormal isoform (PrP^{Sc}) of the protein prion, the normal one (PrP^{c}) being present in the neurones and in different organs. This abnormal isoform (PrP^{Sc}) is the only known molecular marker of scrapie in sheep. In agreement with physiopathological knowledge, the accumulation of PrP^{Sc} was investigated by immunohistochemistry as described in Andréoletti *et al*. (2000) in palatine tonsils from 654 adult healthy ewes culled in 2000 and 2001 in the 15 affected flocks (Manech blond faced breed). These culled ewes were between 2 and 6 years old.

Only ewes genotyped ARQ/ARQ, ARQ/VRQ or VRQ/VRQ were found positive, e.g. exhibited PrP^{Sc} in tonsils (Table 3). Moreover, quite all positive sheep were found only in the 4 flocks for which the scrapie incidence in 2000 and 2001 was higher than 3%: they represented about 2% of ARQ/ARQ exposed sheep and 14% of ARQ/VRQ or VRQ/VRQ sheep in these 4 flocks. Conversely, it means that none of the ARR/ARR sheep exposed in the 15 affected flocks showed PrP^{Sc} accumulation in their tonsils, in agreement with kinetics studies suggesting the absence of healthy carrier (van Keulen *et al*., 1999; Andréoletti *et al*., 2000).

Demonstration of selection efficiency in affected flocks

The demonstration was based on the use of AI rams genotyped ARR/ARR in the 15 affected Manech blond faced flocks, for which the annual scrapie incidence was often over 10% during the last 3 years before starting the action.

Table 2. Risk factors for scrapie expressed as Odds Ratio (OR) relative to ewes born in 1997, bought from another flock (not born within the flock), bred in the flock H and genotyped ARQ/ARQ (3803 animals born between 1997-2000 and exposed in 12 flocks)

Risk factor	Level	Number (n)	Scrapie affected animal	
			P††††	OR
Year of birth			0.0001	
	1997	980		1.000
	1998	1018		0.975*
	1999	945		0.856*
	2000	860		0.184*
Type of replacement			0.0001	
	Born in the flock	477		5.961*
	Bought ewe	3326		1.000
Flock effect		346	0.0001	
	A	359		0.195*
	B	304		0.270*
	C	201		1.358*
	D	253		0.305*
	G	233		0.225*
	H	287		1.000
	I	495		1.873*
	J	251		1.095
	K	425		0.258*
	L	315		0.928*
	M	334		1.276*
	N	287		0.477*
PrP genotype of the ewe			0.0001	
	ARR/ARR			No cases
	ARR/ARQ†	1541		0.021*
	Unknown	682		2.958*
	ARQ/ARQ††	1431		1.000
	ARQ/VRQ†††	149		2.140*

†or ARQ/AHQ if any.
††or ARQ/AHQ if any.
†††or VRQ/VRQ if any.
††††P = global significance (Wald statistics).
*OR significantly different from 1.000 ($P < 0.05$).

Table 3. Number of PrP^{sc} affected ewes (in palatine tonsil) according to their *PrP* genotype and scrapie incidence of the flock

Scrapie incidence of the flocks (No. flocks)	*PrP* genotype of the (culled) ewes					
	ARR/ARR	ARR/ARQ†	ARR/VRQ	ARQ/ARQ††	ARQ/VRQ	VRQ/VRQ
<1% (4)	0/4	0/37		1/70	0/7	
1-2% (3)	0/2	0/25	0/2	0/53	0/3	
2-3% (4)	0/23	0/93	0/7	0/99	2/15	
>3% (4)	0/7	0/91	0/9	2/85	2/21	1/1
Total	0/36	0/246	0/18	3/303	4/46	1/1

†and ARR/AHQ (6 animals).
††and ARQ/AHQ (8 animals).

As already described in Table 1, this action started in 1998 for one of these 15 flocks, in 1999 for three others, in 2000 for 3 new flocks. Finally all the 15 flocks have been involved in this demonstration in 2001 at least for one year (Table 1). The presentation will focus first on the flock C whose breeder entered the project in 1999, which gives enough time to describe in 2002 the trend on 4 years.

Use of ARR/ARR rams and evolution of *PrP* structure of the flock C

The cohorts of ewes born between 1997 and 2000 in the flock C are presented in Table 4. Since 1999 all the alive animals of the flock C were blood sampled for *PrP* genotyping as in the 14 other affected flocks involved in this demonstration: until now 7346 animals have been *PrP* genotyped in these 15 flocks. Thus *PrP* genotypes are available for most of the ewes born since the year 1999 in the flock C. In agreement with the *PrP* structure of the Manech blond faced breed, before breeding for scrapie resistance, only 3 to 5% of the ewes of the cohorts 1997 and 1998 were born from ARR/ARR rams: as a consequence, about 40 to 50% of these ewes were susceptible animals (ARQ/ARQ or ARQ/VRQ), 32 to 35% heterozygous ARR/ARQ, and only 2 to 7% homozygous resistant ewes (ARR/ARR). After the use of only AI resistant rams since 1999 in the flock C (Table 4), the *PrP* genetic structure of these new cohorts of ewes born in 1999 and 2000 changed dramatically: since the cohort of birth 1999, there were no more homozygous susceptible sheep, and the most frequent genotypes were ARR/ARQ ewes (64 to 68%) followed by ARR/ARR ewes (23 to 20%).

Table 4. Use of AI resistant ARR/ARR rams within an affected flock (flock C): consequences on *PrP* genotype of the ewes

Year of birth	No. ewes	% ewes born from ARR/ARR rams	*PrP* genotypes of the ewes (%)					
			ARR/ ARR	ARR/ ARQ†	ARR/ VRQ	ARQ/ ARQ††	ARQ/ VRQ†††	Unknown
1997	98	3.1	2.1	34.7	2.0	41.8	2.1	17.3
1998	92	5.4	7.6	32.6	1.0	36.9	7.6	14.3
1999	92	100.0	22.8	68.4	6.5	0.0	0.0	2.3
2000	73	100.0	28.7	64.3	5.4	0.0	0.0	1.6

†and ARR/AHQ if any.
††and ARQ/AHQ if any.
†††and VRQ/VRQ if any.

Use of ARR/ARR rams and evolution of scrapie incidence in the flock C

Since the average incubation period was about 2.6 years in these affected flocks, it was needed to wait at least until the year 2002 to draw up a first report of the evolution of the scrapie incidence in the flock C. When describing the results according to the year of birth of ewes, the trend was very clear (Table 5): no more scrapie cases for the cohorts born in 1999 and 2000 in the flock C versus a scrapie incidence between 22 and 28% for the two last susceptible cohorts of ewes born in 1997 and 1998 in this flock. As a consequence, the annual scrapie incidence which ranged between 7.4 and 11.9% in the years 1997-1998, decreased to 3.3% in 2000 and fell down to 0.9% in 2001 (Table 6).

Table 5. Use of AI resistant ARR/ARR rams within an affected flock (flock C): consequences on scrapie incidence according to the year of birth of the ewes

Year of birth	No. ewes	% ewes born from ARR/ARR rams	No. scrapie cases	Scrapie incidence (%)
1997	98	3.1	27	27.5
1998	92	5.4	20	21.7
1999	92	100.0	0	0.0
2000	73	100.0	0	0.0

Table 6. Use of AI resistant ARR/ARR rams in 4 affected flocks: consequences on annual scrapie incidence

Flock	First year of use of ARR/ARR rams	Annual scrapie incidence (%)				
		1997	1998	1999	2000	2001
I	1997	7.7	5.7	3.9	3.7	0.7
C	1998	11.9	7.4	7.8	3.3	0.9
J	1999	8.3	7.6	8.9	5.4	2.5
M	1999	14.5	9.8	5.3	8.6	4.1

Use of ARR/ARR rams and evolution of scrapie incidence in the 4 flocks C, I, J and M

When considering the 4 flocks (C, I, J and M) involved in the demonstration since several years (Table 1), the same trend than the one previously described for the flock C was also observed for the other flocks: as soon as most of the ewes were born from ARR/ARR rams (between 56 to 100% for the 4 flocks), the scrapie incidence of the corresponding cohorts ranged between null (if 100% ewes were born from resistant rams) to very low and close to 2% (when about 60% ewes were born from ARR/ARR rams as in flock I for cohort 1998 or in flock M for cohort 2000), while the scrapie incidence was usually over 20% in the same flocks for the 2 last cohorts born before the use of AI resistant rams (Table 7).

Table 7. Use of AI resistant ARR/ARR rams in 4 affected flocks: consequences on *PrP* genotype of the ewes and on scrapie incidence according to the year of birth of the ewes

Flock	Year of birth					
	1996	1997	1998	1999	2000	2001
I						
% ewes born from ARR/ARR rams	0.0	16.0	56.0	100.0	98.0	100.0
Scrapie incidence (%)	20.5	28.9	2.5	0.0	0.0	0.0
C						
% ewes born from ARR/ARR rams	0.0	3.0	5.0	100.0	100.0	100.0
Scrapie incidence (%)	36.8	27.5	21.7	0.0	0.0	0.0
J						
% ewes born from ARR/ARR rams		0.0	2.0	2.0	100.0	94.0
Scrapie incidence (%)		34.9	18.2	11.3	0.0	0.0
M						
% ewes born from ARR/ARR rams		1.0	0.0	1.0	62.0	100.0
Scrapie incidence (%)		24.8	19.3	21.4	2.1	0.0

As a consequence, when considering now the scrapie incidence by year (and not by cohort of birth), it can be noticed that one year after the use of resistant rams a first significant decrease in scrapie incidence was observed, the scrapie cases being divided about by twice. Then 2 years after the use of ARR/ARR rams, the scrapie incidence within a flock fell down under 1% (Table 6). It is in agreement with in progress physiopathological studies showing that two conditions are needed for the contamination of placenta at lambing: both dams affected by scrapie and lambs *PrP* susceptible. In other words, if the lamb is heterozygous or homozygous resistant, the placenta is negative even if the dam is scrapie positive (results to be published). Under these conditions, the use of ARR/ARR rams appears to be a successful approach to reduce dramatically the scrapie incidence in two years within a high affected flock (Table 6).

Breeding for scrapie resistance in the French dairy sheep breeds

In France, breeding for scrapie resistance started in the end of the 90's. In a first step, breeding was based on *PrP* genotyping of the AI rams, since 1995 for the Lacaune breed, since 1997 for the Basco-Bearnaise and Manech breeds, and 1999 for the Corsica breed. The results of *PrP* genotypings showed that fortunately the frequency of the VRQ allele (which is the most susceptible one) was very low whatever the breed, while 4 dairy sheep breeds had an initial ARR frequency high enough, between 40 and 54%, and one breed, the Manech blond faced, a low ARR frequency 17% (Table 8).

Table 8. Initial *PrP* allele frequencies (ARR and VRQ alleles) in the 5 French dairy sheep breeds (before breeding for scrapie resistance)

Breed	ARR frequency	VRQ frequency
Basco-Bearnaise	0.399	0
Corsica	0.470	0.003
Lacaune (milk)	0.545	0.012
Manech black faced	0.496	0.004
Manech blond faced	0.167	0.019

In the years 1995-1999, the main objective was more to eliminate the VRQ allele and to carry out demonstration action using ARR/ARR rams in affected flocks (as described above) than to select for ARR allele at the population level. It was an illustration that more knowledge was needed (as explained in the introduction) to take the decision to select for ARR allele at the population level. On the other hand, since only AI rams were *PrP* genotyped during these first years, the selection was more or less limited (specially for a susceptible breed) and occurred only after the progeny test for milk traits to choose the proven rams and specially the elite rams.

The situation has changed since 1999 when those responsible for the dairy sheep breeding programs have decided to intensify the selection for scrapie resistance. The first decision was to *PrP* genotype the candidate rams before entering the breeding centres of young rams and the AI centres to be able to increase the selection pressure on the *PrP* gene. Moreover the number of candidate rams has been doubled or tripled (according to the breed) in the aim to eliminate the rams not carrier of the ARR allele (depending however on the management of the families to reduce inbreeding). This important modification of the initial selection scheme has been implemented for the first time between 2000 and 2002, according to the situation of each breed. So far the ARR allelic frequency of the AI rams born between 1997 and 2001 has increased from 0.54 to 0.68 in dairy Lacaune breed, and from 0.17 to 0.39 in Manech blond faced breed (Fig. 1).

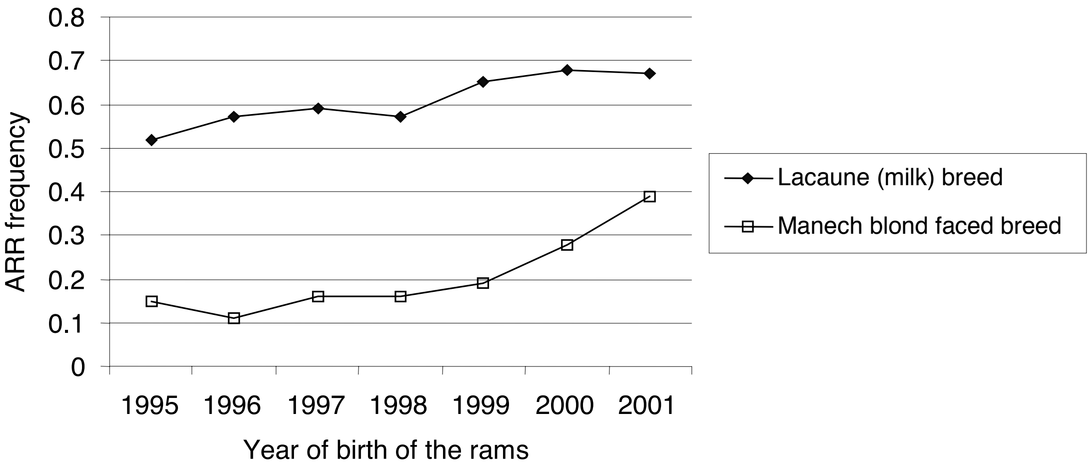

Fig. 1. Evolution of the ARR frequency of the AI rams in Lacaune (milk) and Manech blond faced breeds.

On the other hand, a specific modification of the breeding scheme of the Manech blond faced breed has been conceived in 1999, to face the need for ARR/ARR rams in affected flocks of this breed, which was initially the more susceptible of the 5 French dairy sheep breeds. It consisted in the rapid procreation in 2 years (1999-2000) of a second AI stock of rams, which must be homozygous ARR/ARR rams to mate by AI with the ewes of the affected flocks (see demonstration action described above): 5800 AI in 2000, 9800 in 2001 and 20,000 planned in 2002. These rams were born from ARR carrier sires and dams (of the nucleus flocks) themselves born from ARR carrier sires. These "protective" AI rams are used only in affected flocks and their breeding value for milk production is less favourable than the one of the usual AI progeny tested rams.

Finally those responsible for the dairy sheep breeding programs have intensified again in 2002 the selection for scrapie resistance, now within the framework of a national scrapie plan supported by the Ministry of Agriculture and Fisheries (Elsen *et al.*, 2002): besides the AI rams, all the rams used in natural mating in the nucleus flocks have been *PrP* genotyped in 2002; the elite dams of the nucleus flocks of all the dairy sheep breeds (except Lacaune breed) have also been genotyped to optimise the assortive matings for *PrP* selection and production traits. The objective is that in 2004 all the rams in the AI centres and nucleus flocks will be ARR/ARR in the (dairy) Lacaune breed and at least heterozygous ARR (excluding ARR/VRQ) in the 4 other French dairy sheep breeds. Then the same goal is defined later for the rams of the commercial flocks. To reach this objective, it is planned that each year during 5 years 15,000 to 20,000 animals of the nucleus flocks of the 5 dairy sheep breeds will be *PrP* genotyped to provide the nucleus and commercial flocks (by AI and natural mating) with *PrP* selected rams.

Conclusion

Breeding for scrapie resistance has been now considered as an attractive solution to control this disease: many results (including those of this paper) have been accumulated for the last 5 years to validate that the resistant ARR/ARR sheep are fully resistant against any scrapie strain and that they are not healthy carriers. The monitoring of the 15 affected flocks involved in a demonstration using *PrP* selection to eradicate scrapie will be continuing for the next years to confirm the present successful results. Moreover this resistance appears to be extended to BSE strain after experimental contamination with this TSE strain (Goldmann *et al.*, 1994; Foster *et al.*, 2001; Jeffrey *et al.*, 2001) which is of course the main result to improve the safety of the sheep food chain.

On the other hand, it is necessary to verify that increasing the ARR frequency will not have reverse effect on the selection for other traits as production or functional traits included in the breeding objectives. This could be caused either by a direct effect of the *PrP* gene on these other traits or by a linkage between the *PrP* locus and a Quantitative Trait Locus. A first analyse was not able to detect such effects on milk production traits (Barillet *et al.*, 2002). While selecting the French dairy sheep breeds for scrapie resistance, such studies will be continuing to add knowledge to the possible genetic relationships between the *PrP* locus and production or functional traits, and resistance to other diseases.

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