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Establishing a National Cryo Bank for Ovine Breeds with Semen Collected *post mortem*

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Objective

To eradicate BSE is one of the major aims of the veterinary administration in the EU. To avoid covered distribution of BSE in the European sheep populations Scrapie is now be combated. Genetic based resistance against TSE is therefore a major selection criterion for the favourite PrP genotype in all ovine breeds in the EU. *ARR*-homozygous animals are most resistant, while *VRQ* is the most susceptible out of five known alleles. Despite the very different frequencies among breeds, strong selection for *ARR* is carried out on the basis of EU-decision 2003/100/EC.

All over Europe the ovine breeds are now checked for their specific genetic resistance based on their frequencies of the relevant five alleles. Some breeds have high frequencies of the proposed allele *ARR*, whereas others have such low occurrences that any selection on *ARR* will result in a strong genetic bottle neck with severe loss of genetic diversity.

Situation in German ovine breeds

Allele frequencies in German breeds vary from 6.2 % for *ARR* and 17.8 % for *VRQ* in the Bentheim breed (Bentheimer Landschaf) as the worst case up to more than 70 % *ARR* and no *VRQ* in the German Whitehead and Blackhead sheep (Deutsches Weißköpfiges und Schwarzköpfiges Fleischschaf). 11 breeds have frequencies of *ARR* around or below 25 % that will result in strong bottle necks, if high selection pressure is put on *ARR*. Typically have the more meaty types of sheep higher frequencies of *ARR* than the extensive used land races. But in the group of extensive used land races are the most endangered populations as well. In the mentioned Bentheim breed only about 100 rams are in use at all and we can expect only about one homozygous *ARR* ram at all. Therefore specially in those breeds which have as well small numbers of breeding animals and low ARR-frequencies severe genetic bottle necks can be expected by the promoted selection strategy to increase TSE resistance. And the sheep breeders are forcing that by themselves more than really demanded by the EU, which prospect exceptions for rare breeds with very hopeless genetic basic, and despite other possible ways of careful selection for *ARR (BRANDT, et al. 2004).*

In table 1 the 31 relevant ovine breeds registered in Germany are listed with their results for the representative investigations in accordance to the regulation (EC) no. 2002/1003 for a monitoring of the all national breeds in Europe:

Table *1:* *Frequencies of the PrP-alleles (in %) and genotype classes (in %) of the sheep breeds in Germany (status: 21st of July 2003)*
(Result of the representative investigations concerning decision 2003/100/EC in the context of the TSE-combat based on regulation (EC) no. 999/2001)

ARR-Frequenz	Breed	n	ARR	AHQ	ARH	ARQ	VRQ	G1	G2	G3	G4	G5
Very bad	Kamerunschaf	50	**0.0**	5.0	0.0	95.0	0.0	**0.0**	**0.0**	100.0	0.0	0.0
	Bentheimer Landschaf	73	**6.2**	11.6	11.0	53.4	17.8	**0.0**	**11.0**	58.9	1.4	28.8
	Ostfr. Milchschaf weiß	83	**10.8**	33.7	0.0	54.8	0.6	**1.2**	**19.3**	78.3	0.0	1.2
	Kärntner Brillenschaf	55	**11.8**	20.0	37.3	21.8	9.1	**1.8**	**12.7**	67.3	7.3	11.0
	Romanovschaf	50	**12.0**	0.0	0.0	82.0	6.0	**0.0**	**24.0**	64.0	0.0	12.0
	Merinolandschaf	57	**14.0**	8.8	0.0	77.2	0.0	**1.8**	**24.6**	73.7	0.0	0.0
	Graue Geh. Heidschnucke	79	**15.2**	12.7	0.0	72.2	0.0	**3.8**	**22.8**	73.4	0.0	0.0
	Bergschaf weiß	50	**16.0**	18.0	8.0	58.0	0.0	**0.0**	**32.0**	68.0	0.0	0.0
	Ostfr. Milchschaf braun	59	**18.6**	33.9	0.0	47.5	0.0	**1.7**	**33.9**	64.4	0.0	0.0
Medium - unconvenient	Weiße Hornl. Heidschnucke	56	**20.5**	2.7	0.0	76.8	0.0	**0.0**	**41.1**	58.9	0.0	0.0
	Bergschaf braun	53	**21.7**	3.8	0.0	73.6	0.9	**5.7**	**30.2**	62.3	2.0	0.0
	Shropshire	60	**24.2**	24.2	0.0	51.7	0.0	**8.3**	**31.7**	60.0	0.0	0.0
	Rauhw.Pomm. Landschaf	53	**26.4**	24.5	0.0	48.1	0.9	**5.7**	**41.5**	50.9	0.0	1.9
	Waldschaf	50	**29.0**	0.0	0.0	68.0	3.0	**10.0**	**36.0**	48.0	2.0	4.0
	Merinolangwollschaf	51	**29.4**	2.0	2.9	62.7	2.9	**5.9**	**45.1**	43.1	2.0	3.9
	Alpines Steinschaf	50	**33.0**	5.0	13.0	39.0	10.0	**10.0**	**44.0**	28.0	2.0	16.0
	Skudde	64	**35.2**	6.3	0.0	58.6	0.0	**15.6**	**39.1**	45.3	0.0	0.0
	Texel	93	**36.6**	1.6	9.1	41.4	11.3	**11.8**	**40.9**	25.8	8.6	12.9
	Weiße Geh. Heidschnucke	50	**39.0**	0.0	0.0	61.0	0.0	**18.0**	**42.0**	40.0	0.0	0.0
	Merinofleischschaf	51	**45.1**	4.9	0.0	50.0	0.0	**21.6**	**47.1**	31.4	0.0	0.0
	Dorper	50	**49.0**	0.0	0.0	50.0	1.0	**24.0**	**50.0**	24.0	0.0	2.0
Convenient	Blauköpfiges Fleischschaf	50	**52.0**	0.0	0.0	32.0	16.0	**28.0**	**34.0**	8.0	14.0	16.0
	Leineschaf	50	**53.0**	5.0	3.0	39.0	0.0	**32.0**	**42.0**	26.0	0.0	0.0
	Suffolk	73	**58.9**	0.0	0.7	39.0	1.4	**32.9**	**50.7**	13.7	1.4	1.4
	Coburger Fuchsschaf	51	**60.8**	0.0	0.0	38.2	1.0	**35.3**	**49.0**	13.7	2.0	0.0
	Nolana	53	**61.3**	1.9	0.0	16.0	20.8	**37.7**	**18.9**	7.6	28.3	7.6
	Ile de France	50	**66.0**	0.0	0.0	24.0	10.0	**50.0**	**22.0**	8.0	10.0	10.0
	Rhönschaf	51	**67.6**	2.0	7.8	21.6	1.0	**49.0**	**35.3**	13.7	2.0	0.0
	Weißköpfiges Fleischschaf	54	**73.1**	0.0	0.0	26.9	0.0	**57.4**	**31.5**	11.1	0.0	0.0
	Schwarzköpf. Fleischschaf	50	**78.0**	0.0	0.0	22.0	0.0	**60.0**	**36.0**	4.0	0.0	0.0
	Berrichon du Cher	23	**84.8**	4.3	0.0	4.3	6.5	**69.9**	**17.4**	0.0	13.0	0.0

Project

An efficient semen preservation method to maintain the current genetic diversity had previously been developed in Mariensee, employing deep frozen epididymal semen. All those breeds with a main origin in Germany were included in the programme where remarkable bottle neck effects can be expected caused by their low frequencies of *ARR* and/or low numbers of registered animals (table 1 and DGFZ 2003). The aim was to store the semen of 15 rams in each of these 20 most endangered breeds, according to the National Scrapie Plan in UK (NSP 2003) and the guidelines for cryopreservation in European countries (ERFP 2003).

Main results

Semen was successfully collected from 95 rams out of 16 German breeds after slaughter. A maximum of 400 straws were frozen per ram, filled in 0.25 ml straws and extended to 50 million spermatozoa per straw. Motility of spermatozoa and percentage of intact acrosomes (in brackets) are on average for fresh and thawed semen: 79.1 (93.5) % and 59.8 (72.9) %, respectively. Subsequent laparoscopic test inseminations resulted in a pregnancy rate of 87.5 %, 7 out of 8 inseminated ewes have given birth to 14 lamps. Less than 7 % of all rams (7 out of 102) could not be used for semen collection at all (e.g. atrophies of the testes) or missed the semen quality requirements. Some effects of season and breed could be noticed. All carcasses of the rams could sold for marketing after slaughter, despite of their higher age in some cases, maximum was 10 years. Depending on the breed the live weight ranged from 23 to 180 kg, slaughter weights from 9.1 to 104.5 kg. To fulfil the required altogether 300 rams out of 20 breeds this programme should be complemented and the missing rams in the design should be filled.

Finally it can be noticed that cryo-preservation of epididymal semen is an easy, quick, and low cost method for the long-term preservation of an endangered mammal breed. This method has to be strongly preferred if registered AI stations are not available or the existing AI stations are not able to fulfil the semen collection in that time and efficiency what is necessary in specific cases.

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References

BRANDT, H., T.A. SCHMIDT, and H. WAGNER (2004): Erfolgreiche Zuchtstrategie gegen TSE-Empfänglichkeit. Deutsche Schafzucht 96 (12), 4-7.

DGFZ 2003: TSE-Resistenzzucht bei Schafen. Deutsche Gesellschaft für Züchtungskunde DGfZ, report no. 29, 2003, ISSN 0949-8842

ERFP 2003: Guidelines for the Constitution of National Cryopreservation Programmes for Farm Animals. Publ. No. 1 of the European Focal Point on Animal Genetic Resources. Hiemstra, S.J. (ed.)

NATIONALES FACHPROGRAMM 2003: Nationaler Bericht Deutschlands mit einem Nationalen Fachprogramm zur Erhaltung und nachhaltigen Nutzung tiergenetischer Ressourcen in Deutschland. www.genres.de/tgr/nationales_fachprogramm

NSP 2003: National Scrapie Plan (NSP) Semen Archive, DEFRA, www.defra.gov.uk/animalh/ bse/bse-science/scrapie/nsp/nsp.html