

SHORT COMMUNICATION

NADH diaphorase polymorphism in goat erythrocytes

M. J. TUÑÓN & P. GONZALEZ *Departamento de Biología, Facultad de Veterinaria, Universidad de León, León, Spain*

M. VALLEJO *Departamento de Genética y Mejora, Facultad de Veterinaria, Universidad Complutense de Madrid, Madrid, Spain*

Summary. This paper describes for the first time polymorphism of the erythrocyte diaphorase in goats. Three diaphorase 1 phenotypes were observed in the red cells of goats. Breeding data indicated that polymorphism was controlled by two autosomal codominant alleles, *Dia^F* and *Dia^S*, the frequencies of which were determined in 14 different Spanish breeds of goat.

Keywords: goat, NADH diaphorase, polymorphism, erythrocytes, starch gel electrophoresis

Genetic polymorphism of NADH-diaphorase has been described in man and in various species of domestic animals (Hopkinson *et al.* 1970; Sandberg 1974; Manwell & Baker 1977; Tucker & Crowley 1978; Makavee 1979).

While carrying out a comparative study involving the different genera and species of the Caprinae family, including hybrids, Tucker & Clarke (1980) demonstrated the existence of two zones of diaphorase activity on starch gel in the domestic goat: Dia 1 and Dia 2, although the electrophoretic patterns in both zones were identical in all the animals they studied.

This genetic system of the blood has been shown to be monomorphic in the few goat populations so far studied (Nozawa *et al.* 1978a, b; Katsumata *et al.* 1981a, b; Di Stasio *et al.* 1984).

We analysed a total of 1368 blood samples taken from animals of the following breeds: Pirenaica (113), Verata (100), Guadarrama (101), Zamorana (110), Berciana (100), Granadina (101), Blanca Andaluza (100), Blanca Celtibérica (100), Murciana (100), Negra Serrana (100), Malagueña (100), Canaria (99), Palmera (36) and Retinta (108).

Red cell NADH diaphorase analyses were effected by starch gel electrophoresis according to the techniques described by Valenta *et al.* (1967) and Cepica & Stratil (1978) with slight alterations introduced by us: the electrophoretic run time of 5 h, at 6.8 V/cm and a 13% starch gel.

Correspondence: Dra M. J. Tuñón, Departamento de Biología, Facultad de Veterinaria, Universidad de León, Campus de la Vegazana, 24007 León, Spain.

Accepted 4 December 1986

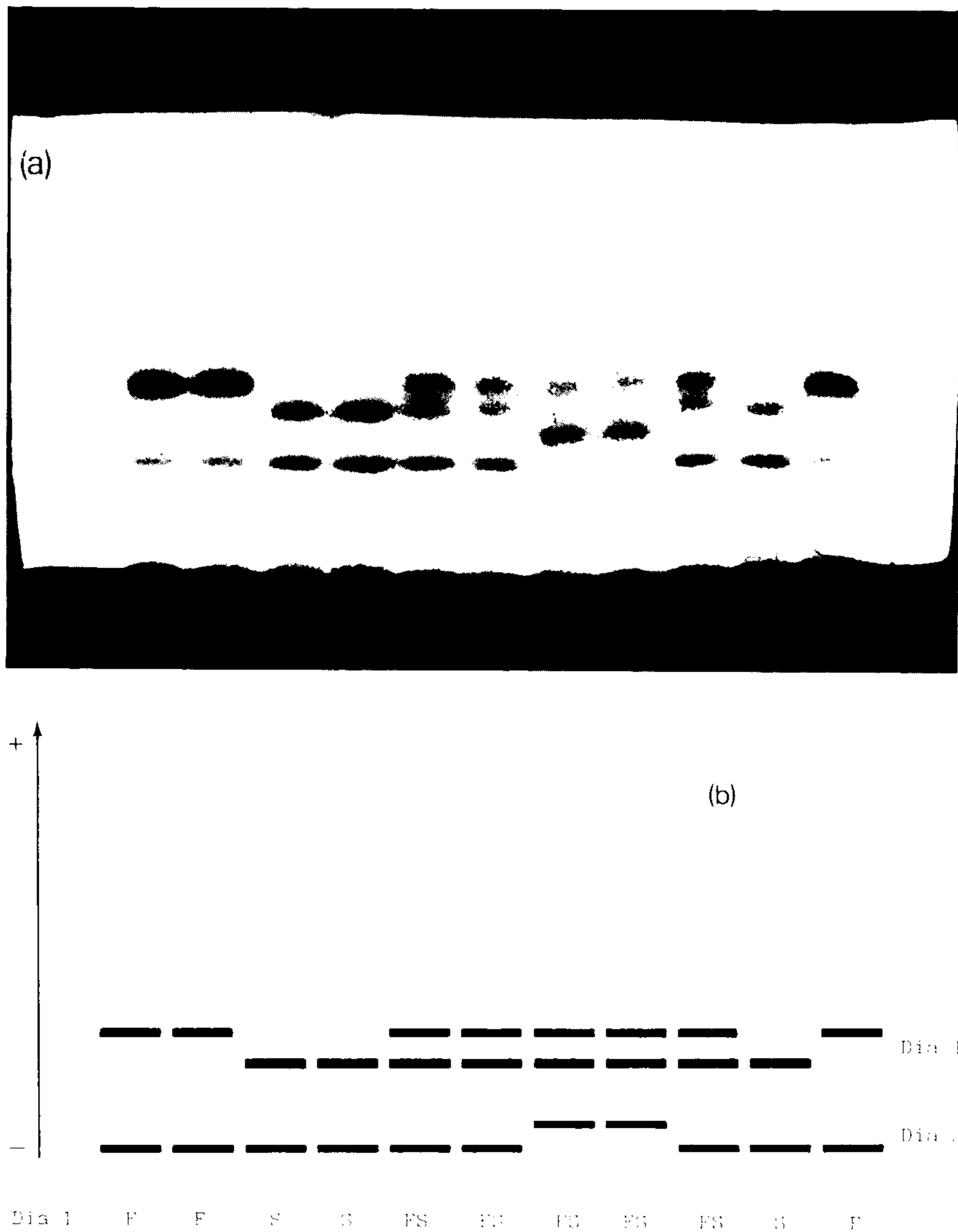


Figure 1. Photograph and diagram showing red cell NADH Dia 1 phenotypes. A variant of Dia 2 is also shown in two of the samples — see text for comment.

Two regions of diaphorase activity were observed on the starch gel (Fig. 1). Region 2 consisted of a single band of activity in the most cathodal area of the starch gel. This region was present in all the samples analysed and did not vary, except in the case of six animals, where a band of greater mobility was present instead of the common one (see Fig. 1). Segregation studies did not enable us to establish whether the variations were of a genetic nature, as the animals concerned lacked any kind of individual identification. Although, as Cepica & Stratil (1978) pointed out for

ovines, this could be due to changes brought about by the storing of red cell lysates, it should be mentioned that in none of the populations we studied did more than 3–5 days pass between the taking of samples and their analysis for this marker.

In region 1 one or two bands appear in a more anodal position than region 2. Figure 1 shows the three activity patterns observed in this region on starch gels. In some samples the band was relatively anodal in mobility (phenotype F), whereas in others there was a clearly more cathodal migration of the band (phenotype S) and in others there was a pair comprising each type of band (phenotype FS). We should point out that the band corresponding to phenotype Dia F was thicker, and stained more intensely than that corresponding to phenotype Dia S. These differences are probably due to variations in their enzymatic activities, as is the case in ovines, but the other way round, that is, a lower enzymatic activity has been suggested for phenotype Dia F (Tucker & Crowley 1979).

Regions 1 and 2 as described in this paper probably correspond to types 1 and 2 as described by Tucker & Clarke (1980), and, like those authors, we did not find genetic polymorphism in the more cathodal region, Dia 2.

Red cell lysates from different goats were analysed according to the hypothesis that the NADH diaphorase activity in region 1 was under the control of two codominant alleles, band F being the product of gene *Dia*^F and band S of gene *Dia*^S. The inheritance data are in agreement with the hypothesis proposed (Table 1).

The mode of inheritance which we propose for this system in caprines is similar to the one put forward by Cepica & Stratil (1978), Tucker & Crowley (1978) and Shimaoka *et al.* (1980) in sheep.

Table 2 gives the frequencies of NADH diaphorase phenotypes in the 14 breeds of goat studied. With the exception of the Palmeras, all of which had the Dia F phenotype (perhaps because it was possible only to analyse 36 animals of that breed), all the breeds studied showed variability, although phenotype Dia F was the most common in all of them.

Gene frequencies of the Dia types for the 14 breeds are shown in Table 2. The highest frequencies for all breeds are of allele *Dia*^F. The highest frequency of *Dia*^S was found in Guadarrama goats.

In 13 of the 14 breeds studied, the diaphorase was shown to be polymorphic, allele *Dia*^S being absent from Palmera goats. On the other hand, in Japanese and

Table 1. Inheritance data for NADH diaphorase types

	Mating type		Number and type of offspring		
	Sire	Dam	F	FS	S
52	F	F	52		
1	F	FS	1		
2	F	S		2	

Table 2. Frequency of NADH diaphorase types in different Spanish goat breeds

Breed	n	Number of phenotypes			Gene frequency	
		F	FS	S	Dia ^F	Dia ^S
Pirenaica	113	79	33	1	0.845	0.155
Verata	100	70	27	3	0.835	0.165
Guadarrama	101	67	32	2	0.822	0.178
Zamorana	110	109	1	0	0.995	0.005
Berciana	100	97	3	0	0.985	0.015
Granadina	101	87	13	1	0.926	0.074
B. Andaluza	100	87	11	2	0.925	0.075
B. Celtibérica	100	98	2	0	0.990	0.010
Murciana	100	68	30	2	0.830	0.170
N. Serrana	100	99	1	0	0.995	0.005
Malagueña	100	82	18	0	0.910	0.090
Canaria	99	89	10	0	0.949	0.051
Palmera	36	36	0	0	1.000	0.000
Retinta	108	104	4	0	0.981	0.019

'Shiba' populations (Nozawa *et al.* 1978a, b), in the Japanese Saanen breed and native populations of Indonesia (Katsumata *et al.* 1981b), in native Korean populations (Katsumata *et al.* 1981a) and in native goats of Italy (Di Stasio *et al.* 1984), authors point to its monomorphic character in all cases. We may therefore consider that diaphorase is an important genetic marker for distinguishing Spanish breeds of goat throughout the world.

References

- Cepica S. & Stratil A. (1978) Further studies on sheep polymorphic erythrocyte diaphorase. *Animal Blood Groups and Biochemical Genetics*, **9**, 239–44.
- Di Stasio L., Rasero R. & Sartore G. (1984) Blood biochemical polymorphisms in Somali sheep and goats. *XIXth International Conference on Animal Blood Groups and Biochemical Polymorphisms, Göttingen*, 24.
- Hopkinson D.A., Corney G., Cook P.J.L., Robson E.B. & Harris H. (1970) Genetically determined electrophoretic variants of human red cell NADH diaphorase. *Annals of Human Genetics* **43**, 1–10.
- Katsumata M., Amano T., Suzuki S., Nozawa K., Martojo H., Abdulgani I.K. & Nadjib H. (1981a) Morphological characters and blood protein gene constitution of Indonesian goats. In: *The Origin and Phylogeny of Indonesian Native Livestock. Part II.* (Report by Grant-in Aid for Overseas Scientific Survey, 504353), 55–68.
- Katsumata M., Nozawa K., Amano T., Shinjo A. & Abe T. (1981b) Blood protein gene constitution of the Japanese Saanen breed of goat. *Japanese Journal of Zootechnical Science* **52**, 553–61.
- Makavee T. (1979) On the genetic polymorphism of NAD.H methemoglobin reductase in the erythrocytes of sheep, cattle and swine. *XVIth International Conference on Animal Blood Groups and Biochemical Polymorphisms, Leningrad* **13**, 42–5.
- Manwell C. & Baker C.M.A. (1977) Genetic distance between the Australian Merino and the Poll Dorset sheep. *Genetical Research* **29**, 239–53.

- Nozawa K., Kano Y., Sawasaky T., Nishida T., Abe T., Shotake T. & Matsuda Y. (1978a) Gene constitution of miniature 'Shiba' goats. *Experimental Animals* **27**, 413-22.
- Nozawa K., Shinjo A. & Shotake T. (1978b) Population genetics of farm animals. III. Blood protein variations in the meat goats in Okinawa Islands of Japan. *Zeitschrift für Tierzüchtung und Züchtungsbiologie* **95**, 60-77.
- Sandberg K. (1974) Genetically controlled variants of NADH-diaphorase from horse red cells. *Animal Blood Groups and Biochemical Genetics* **5**, 23-4.
- Shimaoka T., Tanaka K., Tsunoda K. & Suzuki S. (1980) Polymorphism of haemoglobin and, X-protein and NADH diaphorase in red cells and alkaline phosphatase and transferrin in blood plasma of sheep. *Journal of Agricultural Science* **25**, 145-54.
- Tucker E.M. & Clarke S.W. (1980) Comparative aspects of biochemical polymorphism in the blood of Caprinae species and their hybrids. *Animal Blood Groups and Biochemical Genetics* **11**, 163-83.
- Tucker E.M. & Crowley C. (1978) NADH diaphorase a genetic marker for sheep red cells. *Animal Blood Groups and Biochemical Genetics* **9**, 161-8.
- Valenta M.J., Hyldgaard-Jensen J. & Moustgaard J. (1967) Three lactic dehydrogenase isoenzyme systems in pig spermatozoa and the polymorphism of sub-units controlled by third locus C. *Nature* **216**, 505-7.