RELATION BETWEEN LITTER SIZE AND KAPPA CASEIN GENOTYPE IN INRA RABBIT LINES

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INTRODUCTION

Milk composition varies between as well as within livestock species and dairy animals have been improved both for milk quality and quantity. Even though rabbit milk composition is well characterized, rabbits have never been selected for such traits (Baranyi *et al*., 1995). Among casein genes, κ-casein gene has been cloned and the polymorphism (two alleles A and B) in its non-coding region has been described (Hiripi *et al*., 1998). It has been shown that the mRNA from both alleles accumulates at similar levels and are translated into identical κ-casein. The frequency of the κ-casein A allele has been found to be higher in 11 European breeds (Bolet *et al*., 2000). Preliminary data obtained on a reduced number of New Zealand White rabbits pointed to the possible relation between doe genotype at the κ-casein locus and reproduction traits (Bõsze *et al*., 2000). The aim of this study was to verify and amplify these first results about the relation between κ-casein genotype and litter size.

MATERIAL AND METHODS

Animals. The reproductive performances were recorded on an intercross generation of two synthetic INRA lines in 2000 and 2001 : the INRA 1077 line originating from New Zealand White rabbits and the INRA 2066, originating from Californian and Himalayan rabbits. Both have been selected for 30 generations for litter size. INRA 1077 does from generations 29 and 30 were inseminated with semen from INRA 2066 males in three groups in April, May and October 2000. Their genotype for κ-casein gene was determined. All INRA 2066 males were AA, whereas INRA 1077 females were AA, AB and BB with a frequency of allele A of around 50%. Only daughters from AB dams were selected for this experience, in order to compare the reproduction of does differing by their κ-casein genotype but originating from the same sires and dams. These crossbred females were first inseminated when they were 18 weeks old, and every 6 weeks. For all litters, litter size at birth (born alive and stillborn) was recorded. At birth of the first three parities, litter size was standardized to 9 (first litter) or 10 (next litters) young, by removing or adding young, and litter size at weaning, 35 days later, was recorded.

Determination of κ casein genotype. Genomic DNA was isolated from blood samples by standard protocol and PCR was performed on genomic DNA. κ-casein genotypes were determined by the size difference in the first intron. (Bõsze *et al*., 2000).

Statistical analysis. The effect of kappa-casein genotype on different traits was analysed by analysis of variance (GLM procedure of SAS software). The model included the fixed effects of κ-casein genotype (AA or AB), parity (4 levels : 1, 2, 3 or 4 and more), group (3 levels) and the random effect of female within group and genotype. The effect of genotype was contrasted to residual variance within does.

RESULTS

Data from 809 litters were recorded on 223 females. They were daughters of 58 AB dams and 18 AA sires, 83 of them were AA and 140 AB. This ratio was unbalanced, whereas the expected ratio was 1:1 and expected allele frequency was 0.75 A and 0.25 B, but only females were genotyped in litters of AB dams.

All effects except the random effect of females were significant. Especially there was a significant effect of κ-casein genotype of does on litter size at birth, in favour of AB females. The total number of young was higher by 0.75 ($P<0.016$) and the number of young born alive was higher by 0.82 ($P<0.008$) (table 1). After standardisation of litter size at birth, there was no significant effect of κ-casein genotype on litter size at weaning.

Table 1. Influence of κ-casein genotype of females on their litter size

	Number of litters	κ-casein		Residual S.D.	P*
		AA	AB		
Litter size at birth (total)	809	11.14	11.89	3.15	0.016
Litter size at birth (alive)	809	9.96	10.78	3.87	0.008
**Litter size at weaning	433	9.92	10.06	1.17	0.271

* Contrasted to residual variance within doe

**Parities 1 to 3 only, standardized at birth at 9 or 10 young.

DISCUSSION

Our results clearly show that there is a significant relationship between κ-casein genotype of females and their litter size at birth. As AA and AB females are issued from the same AB dams, their genetic background is the same, and their difference in the prolificacy is clearly related to their genotype at κ-casein locus or nearby loci. As there is no effect of genotype on κ-casein content (Hiripi *et al.*, 1998), the absence of effect on litter size at weaning after standardisation at birth is coherent. Further investigations are needed, on one hand to compare also the reproductive performances of BB females (by comparing the progeny of ABXAB parents), on the other hand to understand this relation.

Reproductive traits are of economical interest in farm animals because they play important role in livestock production. Marker assisted selection (MAS) could potentially be employed in conjunction with traditional selection methods to accelerate the rate of change in economically important traits. Litter size is a typical quantitative trait controlled by both genetic and environmental factors. Candidate gene markers for litter size have been identified in sheep (Davis *et al*., 1991 ; Montgomery *et al*., 1994.) and in pig, including estrogens receptor locus

(Rothschild *et al*., 1996), prolactin receptor gene (Vincent *et al*., 1998) follicle stimulating hormone beta (Li *et al*., 1998) and retinol-binding protein 4 gene (Rotschild *et al*., 2000), although no strong associations could be evidenced (Linville *et al*., 2001). The Booroola fecundity gene (FecB) maps to chromosome 6 in sheep and it was shown to be linked to αs1-casein (CSN1S1) at 12 cM (Montgomery *et al*., 1994). The casein locus maps to chromosome 8 in pigs on which a large ovulation rate/litter size QTL was mapped to the region which is synthenic to the Booroola fecundity gene in sheep (Rothschild, 1998). The casein genes are located close to each others and have been recently assigned to chromosome 15 in rabbits (Pauloin *et al*., 2002). Since it is not very probable that κ-casein per se does influence prolificacy, comparative genome mapping experiments are in progress to identify a gene(s), which is located in a nearby locus.

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REFERENCES

Baranyi, M., Brignon, G., Anlage, P. and Ribadeau-Dumas, B. (1995) New data on the proteins of rabbit (*Oryctogalus cuniculus*) milk. *Comp. Biochem. Physiol*. **B.111** : 407-415.

Bolet, G., Brun, J.M., Monnerot, M., Abeni, F., Arnal, C., Arnold, J., Bell, D., Bergoglio, G., Besenfelder, U., Bõsze, S., Boucher, S., Chanteloup, N., Ducourouble, M.C., Durand-Tardif, M., Esteves, P.J., Ferrand, N., Gautier, A., Haas, C., Hewitt, G., Jehl, N., Joly, T, Koehl, P.F., Laube, T., Lechevestrier, S., Lopez, M., Masoero, G., Menigoz, J.J., Piccinin, R., Queney, G., Saleil, G., Surridge, A., Van Der Loo, W., Vicente, J.S., Viudes de Castro, M.P., Virag, G. and Zimmermann, J.M. (2000) *World Rabbit Sci*. **8**, supp.1, vol. A : 281-315.

Bõsze, Zs., Hiripi, L., Virág, Gy., Tóth, Sz., Makovics, F., Fontaine, M.L. and Devinoy, E. (2000) *Pflügers Archiv -Eur. J. Physiol* **439** (suppl. 3) : R 2-3.

Hiripi, L., Devinoy, E., Rat, P., Baranyi, M., Fontaine, M.L. and Bősze, Zs. (1998) *Gene* **213** : 23-30.

Davis, G.H., McEwan, J.C., Fennessy, P.F., Dodds, K.G. and Farquhar, P.A. (1991) *Biol. Reprod.* **44** : 620-624.

Li, N., Zhao, Y.F., Xiao, L., Zhang, F.J., Chen, Y.Z., Dai, R.J., Zhang, J.S., Shen, S.Q., Chen, Y.F. and Wu, C.X. (1998) *Proc. 6th WCGALP* **26 :** 183-190.

Linville, R.C., Pomp, D., Johnson, R.K. and Rothschild, M.F. (2001) *J. Anim. Sci.* **79** : 60-67.

Montgomery, G.W., Lord, E.A., Penty, J.M., Dodds, K.G., Broad, T.E., Cambridge, L., Sunden, S.L., Stone, R.T. and Crawford, A.M. (1994) *Genomics* **22** : 148-153.

Pauloin, A., Rogel-Gaillard, C., Piumi, F., Hayes, H., Fontaine, M.L., Chanat, E., Chardon, P. and Devinoy, E (2002) *Gene* (in press).

Rotschild, M.F. (1998) Identification of quantitative trait loci and interesting candidate genes in the pig: progress and prospects. *Proc. 6th WCGALP* **26** : 403-409.

Rothschild, M., Jacobson, C., Vaske, D., Tuggle, C., Wang, L., Short, T., Eckhardt, G., Sasaki, S., Vincent, A., McLaren, D., Southwood, O., van der Steen, H., Mileham, A. and Plastow, G. (1996) *Proc. Natl. Acad. Sci*. **93** : 201-205.

Rotschild, M. F., Messer, L., Day, A., Wales, R., Short, R., Southwood, O. and Plastow, G. (2000) *Mammalian Genome* **11** : 75-77.

Vincent, A.L., Evans, G., Short, T.H., Southwood, O., Plastow, G.S., Tuggle, C.K. and Rotschild, M.F. (1998) *Proc. 6th WCGALP* **27 :** 15-18.