MILK PROTEIN POLYMORPHISM AND ITS RELATIONSHIP TO MASTITIS IN FRIESLAND COWS

El polimorfismo en las proteínas de la leche y su relación con la mastitis en las vacas frisonas

Der Polymorphismus in den Milchproteinen und sein Zusammenhang mit der Mastitis der friesischen Kühe

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There is no better example of genetic variation which has already been demonstrated (1) but still awaits to be utilized, than that associated with mastitis in cattle. Mastitis is still the most costly disease of dairy cattle not under satisfactory control. Eradication of the various organisms causing septic mastitis is unlikely and hence the condition must be considered as enzoötic. Some cows are genetically able to resist mastitogenic infections. It would therefore seem sensible to increase the number of such animals. This has not been done in practice to any extent. The present study was undertaken in an attempt to obtain preliminary information on the possible relationship between milk protein types and the incidence of mastitis.

Material and methods

During the course of a mastitis control programme quarter milk samples were obtained from grade Friesland herds. In a preliminary study three herds comprised of 196 animals were examined. During the subsequent main investigation three herds with a total of 368 cows were tested. Sample aliquots were subjected to cytological, bacteriological and electrophoretical examination.

The diagnosis of mastitis was established according to international standards (2). Udders were classified as being affected with septic or aseptic mastitis if one or more quarters of the udder concerned exhibited somatic cell counts

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in excess of 500 000 cells/ml milk together with or without mastitogenic bacteria respectively.

During the pilot test preliminary study of the α-lact-albumin and β-lactoglobulin types of 196 samples was determined by methods described earlier (3). However, only the B-allele was found for the α-lactalbumin in the freshly collected samples. For the main investigation this technique was replaced by improved techniques (4) which allow simultaneous study of β-lactoglobulin, α_{s1}-casein, β-casein and K-casein in the same gel. With this method all four milk samples of each cow of the first two farms were analysed electrophoretically. Since the quarter samples from the same udder, did not differ qualitatively, a single milk sample from each cow in the remaining herd was typed. Although animals with both healthy and mastitic quarters, often displayed quantitative differences, typing never proved to be difficult.

Results

The results presented below emanate from 564 analyses of β-lactoglobulin from the preliminary and the main investigation. Data on the caseins only derive from the 368 cows in the main investigation. Throughout the results are tabulated indicating mastitis positive animals (one or more quarters affected) and mastitis negative animals (udder completely healthy).

Aseptic mastitis was recorded separately in the tables, since the affected cows were excluded from further analyses.

The phenotypes and gene frequencies of 564 β-lactoglobulins derived from the preliminary and main investigations are summarized in Table 1.

TABLE 1

Occurrence of β-lactoglobulin types in a total of 564 mastitis positive or negative Friesland cows

Mastitis diagnosis	Cows tested	β-lactoglobulin types			Gene frequencies	
		AA	*AB*	*BB*	β-Lg^A	β-Lg^B
Positive	199	78	86	35	0.608	0.392
Negative	269	64	154	51	0.524	0.476
Overall	468	142	240	86	0.560	0.440
Aseptic	96	33	47	16	0.588	0.412

The relatively large number of mastitis negative heterozygous animals are apparent. According to the Hardy-Weinberg equilibrium a total of 134 mastitis negative animals is expected; this gives for the results obtained an overall χ^2-value of 5.98^+ at the 0.05 level of statistical significance.

During the preliminary investigation animals with only one or two healthy quarters were included in the analysis, the other quarters being atrophied and without secretion due to some previous udder damage. Since the exact history of these cases could not be established and to avoid inaccurate classification it became essential to disregard the data of the preliminary investigation to consider only the data from cows with four functional quarters examined during the main investigation. The occurrence of β-lactoglobulins in the milk of 368 cows during the main investigation is summarized in Table 2:

TABLE 2

OCCURRENCE OF β-LACTOGLOBULIN TYPES IN 368 MASTITIS POSITIVE OR NEGATIVE FRIESIAN COWS WITH FOUR FUNCTIONAL QUARTERS EACH

Mastitis diagnosis	Cows tested	β-lactoglobulin types			Gene frequencies	
		AA	*AB*	*BB*	β-Lg^A	β-Lg^B
Positive	126	53	54	19	0.635	0.365
Negative	169	44	100	25	0.556	0.444
Overall	295	97	154	44	0.590	0.410
Aseptic	73	24	37	12	0.582	0.418

From Table 2 it is apparent that there is again a larger number of heterozygotes in the mastitis negative group. A total of 83 mastitis negative animals is expected in this group giving for the results obtained on overall χ^2-value of 7.09$^+$

TABLE 3

β-LACTOGLOBULIN TYPES AND BACTERIA IN MILK OF UDDERS WITH ONE OR MORE MASTITIC QUARTERS

Bact. type	β-lactoglobulin types		
	AA	*AB*	*BB*
Sc. * *agalactiae*	13	11	5
S. ** *epidermidis*...	34	34	12
Sc. uberis	3	0	2
Sc. dysgalactiae	3	8	2
Sc. faecalis	1	3	3
S. aureus	33	43	18
	89	99	42

* Sc. = *Streptococcus.*
** S. = *Staphylococcus.*

at the 0.05 level of significance. Results summarized in Table 2 strongly suggest that heterozygous Friesian dairy cows are less frequently infected with mastitogenic micro-organisms than homozygous animals.

The relationship between β-lactoglobulin types and various bacteria isolated from mastitic udders is illustrated in Table 3.

The total number of β-lactoglobulin types presented in Table 3 amounts to 230 animals in contrast to 199 mastitis positive cows in Table 1 because several animals were infected with more than one type of bacteria i. e. mixed infections in same or different quarters were present.

From Table 3 there is clearly no significant difference between the incidence of mastitis caused by a specific microorganism and the β-lactoglobulin type secreted.

The occurrence of α_{s1}-casein, β-casein and K-casein types was distributed almost at random among mastitis positive or negative Friesland cows, and is therefore not further discussed.

Discussion

It would appear that there is a positive correlation between the biosynthesis of certain milk proteins and susceptibility or resistance to septic mastitis. The investigation also indicates the extremely important role which genetic typing of dairy cows could play as supplementary measure in the control of mastitis and possibly other stock diseases. Genetic typing could be put to increased practical use if it were possible to develop a system of typing heifers, calves and bulls to allow of early elimination of animals which are genetically predisposed to infectious mastitis.

Since it was the primary objective of these investigations to obtain information on the possible correlation between the function of the bovine mammary gland and its susceptibility to septic mastitis, a wide range of variables have been disregarded. It is intented to examine a larger number of animals from different herds with subdivision into age groups, since animals with higher lactation numbers are likely to have a higher incidence of mastitis. Because a positive correlation has been shown to exist between high milk production and mastitis suceptibility (5, 6) production levels should also be taken into account. However, this correlation does not appear to be constant and other factors such as morphological features of the udder, teats and teat canal have also to be considered when determining those genetic factors which cause an udder to be more susceptible or resistant towards septic mastitis (7, 8).

RESUMEN

Parece existir una correlación positiva entre la biosíntesis de ciertas proteínas de la leche y la susceptibilidad o resistencia a la mastitis séptica. La investigación indica asimismo la excepcionalmente importante parte que la tipificación genética de las vacas lecheras podría representar como medida suplementaria en el control de la mastitis y de posiblemente otras enfermedades del ganado. La tipificación genética podría incrementar su empleo práctico, dentro de lo

posible, en novillas, terneras y toros, a fin de lograr la precoz eliminación de los que genéticamente estén predispuestos a la mastitis infecciosa.

Como el primer objetivo de estas investigaciones fue el de obtener información sobre la correlación posible entre la función de la glándula mamaria bovina y su susceptibilidad a las mastitis sépticas, se ha descartado una amplia gama de variables. Se ha tratado de examinar el mayor número posible de animales en diferentes establos, con una subdivisión en grupos de edad, ya que los animales con elevado número de lactaciones están propensos a una mayor frecuencia de mastitis. Como se demostró una correlación positiva entre la alta producción lechera y la susceptibilidad a la mastitis (5, 6), los niveles de producción deberían ser también tomados en cuenta. Sin embargo, esta correlación parece no ser constante, debiéndose tomar en consideración también otros factores, tales como la morfología de la ubre, pezones y conductos galactoforos, que tienen que interpretarse cuando están determinados por factores genéticos que hacen a la mama más o menos susceptible a las mastitis sépticas (7, 8).

ZUSAMMENFASSUNG

Es scheint, als ob es eine positive Korrelation zwischen der Biosintesis von bestimmten Proteinen und der Empfindlichkeit oder Widerstandsfähigkeit gegen die septische Mastitis gibt. Die Forschung weist uns auch auf die außerordentliche Wichtigkeit der genetischen Typifikation der Milchkühe für die Kontrolle der Mastitis und anderer Viehkrankheiten hin. Die genetische Typifikation könnte ihre praktische Anwendung, innerhalb ihrer Möglichkeiten, bei den Kälbern, Stieren und Färsen vergrößern, um die frühzeitig auszuschließen, die eine genetische Anfälligkeit für infektiose Mastitis mitbringen.

Da die ersten Forschungen darauf ausgerichtet waren Information über die mögliche Korrelation zwischen der Rindmilchdrüsenfunktion und ihrer Empfindlichkeit für die septische Mastitis zu erhalten, hat man eine große Reihe verschiedener Möglichkeiten ausgeschlossen. Man ist bemüht eine möglichst große Anzahl von Tieren, in verschiedenen Ställen zu prüfen, und mit einer Unterteilung in Altersgruppen, wegen der größeren Anfälligkeit für Mastitis bei einer größeren Anzahl von Nährungen. Da man eine positive Korrelation zwischen der hohen Milchproduktion und der Bereitschaft für Mastitis (5,6) bewies, müsste das Produktionsniveau beachtet werden. Aber diese Korrelation scheint nicht konstant zu sein, und man muß noch anderen Faktoren Rechnung tragen, wie z. B. die Form des Euters, die Zitzen und die Zitzenkanäle, die man berücksichtigen muß, da sie durch genetische Faktoren bestimmt werden, die das Euter mehr oder weniger anfällig für die septische Mastitis machen.

REFERENCES

1. King, J. O. L. (1972): Mastitis as a production disease. *Vet. Rec.*, 91:325.
2. Kästli, P. (1967): Definition of mastitis. *Ann. Bull. I. D. F.*, Part. III:1.
3. Osterhoff, D. R., and Pretorius, A. M. G. (1966): Inherited biochemical polymorphism in milk proteins. *Proc. S. Afr. Soc. Anim. Prod.*, 5:166.
4. Michalak, W. (1967): Anomalous electrophoretic pattern of milk proteins. *J. Dairy Sci.*, 50:1319.

5. Boge, A. (1965): Undersuchungen über verschiedene prädisponierende Faktoren für die Entstehung von Mastitiden. *Vet. Med. Dis.,* Hannover.
6. Brodauf, F. (1960): Über die Weschselbeziehungen von Milchleistung und Mastitisanfälligkeit. *Züchtungskd.,* 35:8.
7. Legates, J. E., and Grinnels, C. D. (1952): Genetic relationships in resistance to mastitis in dairy cattle. *J. Dairy Sci.,* 35:820.
8. Murphy, J. M., and Stuart, O. M. (1955): Teat canal length in the bovine and its relation to susceptibility to swabinduced infection with *Sterptococcus agalactiae. Cornell Vet.,* 45:112.