SEROTYPING AND BACTERIOPHAGE TYPING OF HUMAN AND BOVINE GROUP-B STREPTOCOCCI

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SUMMARY. Serotypes and phage types of group-B streptococci isolated from healthy and diseased human subjects and from cattle in different parts of Norway have been compared. The phage set used was found to be suitable for the study of strains from cows as well as from man.

INTRODUCTION

Streptococcus agalactiae (Lancefield-group B) is recognised as an important cause of bovine mastitis, a disease of economic importance (Wilson and Salt, 1978). In addition, the organism causes neonatal meningitis and sepsis (Parker, 1977). Neonatal infections by group-B streptococci generally follow maternal carriage of the organism. Reports of carriage rate show wide variation, but Bevanger (1974) found it to be 18% in Norwegian pregnant women. The amount of transmission of this organism between cattle and man is still uncertain. Haug, Bakken and Berdal (1979) found no difference in udder pathogenicity of human and bovine strains. In Denmark, infection in cattle has been attributed to human sources (Jensen, 1976). Furthermore, the prevalence of infections with group-B streptococci was higher in persons drinking unpasteurised milk than in those consuming heat-treated milk (Tolle, 1975).

The subdivision of serogroup B is usually achieved by biotyping or serotyping or both. Bacteriophages that lyse group-B streptococci are known (Russell, Norcross and Kahn, 1969), and Stringer (1980) has described a phage-typing system with phages isolated from cultures of human origin. The aim of the present study was to compare the serotypes and phage types of strains of *S. agalactiae* isolated from healthy and diseased human subjects with strains isolated from cattle in different parts of Norway.

MATERIALS AND METHODS

Strains. A total of 89 strains of human origin were examined. Dr J. Jelinková, Prague and Dr J. Stringer, London donated 17 and 8 strains respectively. Thirty strains, isolated from a variety of human infections, were obtained from the National Institute of Public Health, Oslo; 18 strains originated from routine vaginal swabs taken in Numedal, and 16 strains from the Rikshospitalet, University of Oslo and had been isolated from patients in the gynaecological ward. The bovine group-B streptococci, 125 strains, were isolated from bulk milk and quarter milk as part of a mastitis study. The quarter milk came from clinically affected as well as normal cows.

Bacteriological examinations. The strains were grown on horse-blood (5% v/v) agar, and were grouped and typed by conventional methods (Lancefield, 1933, Rotta *et al.*, 1971). Type antisera were prepared by absorption of antisera made against the following reference strains: O90R (Ia), H36B (Ib), A909 (Ic), 18RS21 (II) and D136C (III), kindly provided by the late Dr Rebecca C. Lancefield of The Rockefeller University, New York and the strains 9828 (R) and 9829 (X) from the National Collection of Type Cultures, Colindale, London. Antiserum for recognition of the proposed new type, 1169, was generously sent to us by Dr Beate Perch of the

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State Serum Institute, Copenhagen. The phage typing was performed by the procedure and with the phages described by Stringer (1980). The shortened set of 24 phages was used. The strains were considered to be typable if they were lysed strongly, i.e., semiconfluent lysis was seen, by one or more phages.

RESULTS

Strains of group-B streptococci, whether of human or of bovine origin, were lysed by phages propagated on human group-B streptococci at the same frequency, 78%. The results are summarised in the table. Among the human strains, members of serotypes Ia, Ib, Ic and III were most frequently lysed. A total of 18 human strains were lysed by only one phage of which seven were lysed by phage 28 and three by phage 12. The latter strains belonged to serotypes III and III-R respectively. Bovine strains belonging to serotypes Ic, Ic protein, III-R and R were more often lysed than strains of the serotype II or 1169 Perch and those not reacting with any serum. The 31 bovine strains lysed by only one phage were distributed particularly among phage type 8 and phage type 14. Of these 31 strains, 12 belonged to serotype R and phage type 8 and 5 strains to serotype Ic protein and phage type 26. Lysis by both phages 26 and 28 was found in serotypes Ic, III, 1169 Perch and among the untypable strains. Bovine strains of the serotypes III and III-R were often lysed by both of the phages 12 and 20.

The strains of human origin isolated in Norway and Czechoslovakia belonged to various serotypes and phage types but 23 strains were lysed by phage 28. The combinations of serotype III-R and lysis by phages 12 and 20 and the association of R protein and phage type 8 predominated among the bovine strains from Numedal. These strains came from herds that were located either in the same region or had been near each other on summer pastures. The majority of bovine strains from other districts either could not be serotyped or belonged to serotypes Ic, Ic protein, II or 1169 Perch. Many of these bovine strains were not lysed by any phage.

TABLE
Phage typability of human and bovine strains of group-B streptococci

Serotype	Origin	Number of strains	Number phage typable	Number of phage patterns
Ia	Human	10	8	5
	Bovine	0	...	...
Ib	Human	9	7	5
	Bovine	2	1	1
Ic	Human	15	14	10
	Bovine	11	11	7
Ic protein	Human	2	2	2
	Bovine	14	14	7
II, II-Ic protein	Human	16	9	8
	Bovine	10	5	5
III, III-R	Human	22	19	12
	Bovine	33	26	13
R	Human	5	3	3
	Bovine	18	18	5
X	Human	4	3	3
	Bovine	1	1	1
Perch 1169	Human	0	...	...
	Bovine	11	6	4
Not typable	Human	6	4	4
	Bovine	25	15	12
All	Human	89	69	52
	Bovine	125	97	55

When human and bovine strains from the same district were available for comparison, the same strain was not detected in all instances except one; but on one occasion, an isolate from one cow and the wife of a dairyman, were serologically not typable but were lysed by the phages 17 and 26. One year later an isolate from a cow in the same herd was distinguishable serologically and by sensitivity to phages.

When strains were isolated from the same cows or herds at different times, the serotype and pattern of phage sensitivity were generally constant. If they were not the same then, there was a change in both, suggesting the isolation of a different strain.

DISCUSSION

The phage set that we used comprised 24 phages isolated from human group-B streptococci. It was found, however, to be suitable for the study of strains isolated from cows as well as from man. From the first, Stringer (1980) had noted that there was no specificity of phage pattern to serotype but that there was a predominance of particular patterns of lysis. In the present study most strains were lysed by two or three phages. Some phage patterns occurred more frequently than others. Of these, combinations of the phages 12, 20, 26 or 28 were notable. Phage 8 was most often associated with bovine strains belonging to serotype R. No single pattern seemed, however, to be specific for any of the serotypes. Different serotypes, human as well as bovine, were lysed by the same phages.

One group-B streptococcus isolated from the wife of a dairyman was found to belong to the same phage type (17/26) as a culture isolated at the same time from quarter samples collected from one of their cows suffering from mastitis. Both strains were serologically untypable. Apart from this observation, no relationship was demonstrated between human and bovine strains. According to Parker (1977) group-B streptococci considered to be of human origin, by his own objective criteria, are not found in animal sources to any significant extent.

Variation in the antigenic structure, especially of the X antigen has been reported by Jensen (1980). In the present study, a change in the serotype or phage type pattern was an exception. However, the X antigen was demonstrated infrequently among the strains examined.

Phage typing has been used successfully in tracing the spread of group-B streptococci in hospitals (Stringer 1980). One conclusion of the present study is that the method can provide useful information, when combined with serotyping, on the ecology of the organism and the epidemiology of diseases that it causes.

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