

**The distribution of serotypes of
Escherichia coli in cow-pats and other animal material
compared with serotypes of *E. coli* isolated
from human sources**

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SUMMARY

The serotypes of 13,139 strains of *Escherichia coli* isolated from humans were compared with the serotypes of 1076 strains isolated from animals. 689 of these strains were isolated from fresh cow-pats on 22 sites in England and Wales. 708 different O/H combinations were found. Of these, 520 were found in human strains only, 130 from animal strains only and 58 O/H serotypes from humans and animals. Approximately half of the animal strains could not be typed with the full set of sera used.

INTRODUCTION

In a previous paper (Bettelheim *et al.* 1974) we concluded that there were considerable differences in serotype distribution between man and animals. The number of strains of *Escherichia coli* examined however, was not great, and most of the animal strains were obtained from cattle and pigs slaughtered in an abattoir and from chickens in a poultry packing station on only a few days. These animals, therefore, are likely to have been from a limited environment. Also, the human faecal specimens were obtained from only 55 persons – 7 from hospital staff and 48 from normal people outside the hospital in the London area. Because so little is known of the distribution in nature of *E. coli* serotypes, we have re-examined this problem, using all the animal strains and human strains, both faecal and urinary, reported in the past with other human strains not yet reported and with strains of *E. coli* collected from cow-pats on 22 sites in England and Wales.

MATERIALS AND METHODS

The main sources of our cattle strains were 85 fresh cow-pats from 22 sites in England and Wales. These were in the counties of Yorkshire, Cheshire, Staffordshire, Shropshire, Warwickshire, Denbighshire, Herefordshire, Pembrokeshire, Cornwall, Devon, Somerset, Hampshire, Sussex, Surrey, Hertfordshire, Suffolk and Norfolk, with one further collection from imported French cows held in

quarantine. From these were isolated 689 strains. Other animal strains were those reported previously (Bettelheim *et al.* 1974), comprising 387 strains.

Human strains were those 116 reported by Bettelheim *et al.* (1974) with an additional 99 strains isolated from urinary infections, 150 from introital swabs and 2659 from faeces of nurses, 2788 from faeces of mothers and their babies in hospital and 5078 from students, mainly from overseas, working in Dublin.

All these 11,966 strains were typed in this laboratory by tube agglutination (Bettelheim *et al.* 1975; Chandler & Bettelheim, 1974) using 154 O and 53 H anti-sera, including the recently described O162, O163 and H56 isolated at St Bartholomew's Hospital (Ørskov, Ørskov, Bettelheim & Chandler, 1975).

A further source of human strains were those collected between 1961 and 1967 – 1570 from baby stools and 679 from infected urine, described by Bettelheim (1968). At that time O antigens were only available up to O147 and H antigens up to H49.

RESULTS

Serotypes from cow pats. From the cow-pats in 22 sites in England and Wales 689 different strains of *E. coli* were isolated and serotyped. Of these, 366 could be O and H typed, 75 O typed only and 216 H typed only, 227 strains, although not rough, could not be typed with 154 O antisera, and 21 strains were rough. 72 strains, although motile, could not be typed with the 53 H sera, and 35 strains were non-motile.

To show more clearly the geographical distribution of serotypes, each O/H combination was counted once only from each site where it was isolated. In this way, the 22 sites yielded 78 identifiable combinations of O and H antigens in strains of *E. coli*, with 49 different O antigens. Some of these O antigens were also associated with non-typable flagellar antigens or the strains were non-motile, and 4 additional O antigens were associated with non-typable flagellar antigens. Rough strains and strains where O antigens could not be determined were also found with typable H antigens. Thus, it is probable that considerably more than the 78 O/H combinations we found were present.

Regarding each identified O and H combination as a single serotype, there was a wide scatter across the country. Of the 78 combinations, no less than 65 were found once only. Twelve serotypes were found twice and one serotype, O2.H5, was found at 5 sites. Forty-seven of these O and H combinations were unique to cow-pat specimens among all the 14,215 strains studied.

Serotypes from humans and animals. It is difficult to determine exactly the number of strains which were rough or O and H non-typable, as data for all these are not available any more. The results quoted were mainly concerned with identified serotypes.

When we put together all our human and animal strains that could be fully serotyped, we found that we had 708 different O/H combinations. Of these, 520 were found in human strains only, 130 from animal strains only and 58 O/H serotypes from humans and animals.

Serotypes associated with infantile diarrhoea in babies found in animals included O18.H7, O112ab.H8 and O114.H19.

O serotypes from humans and animals. Taking fully serotyped human and animal strains, representatives of 139 O antigens were found, strains belonging to groups O34, O49, O56, O70, O93, O95, O130, O136, O137, O143, O144, O151, O155, O156 and O158 being the only ones missing. Only ten O groups, O43, O64, O65, O97, O98, O105, O109, O116, O132 and O157 were found in animals alone.

However, both human and animal strains included many where O antigens could be determined but flagellar antigens were absent or could not be typed. These included some complex O groups such as O12/O15, O12/O141, O13/O147, O34/O41, O9/O104 and groups O95 and O137 listed among those not present in the O/H combinations.

H serotypes from humans and animals. With the exception of H22 and H44, all antigens were found in our total material, although some were found only associated with rough or non-typable O strains. Apart from these two, the only H serotype not found from humans was H43. From the fully typed animal strains, serotypes H3, H17, H23, H29, H35, H36, H46, H47, H49, H51, H52, and H56 were missing although H17, H29, H46, and H49 were present among rough or O non-typable strains.

DISCUSSION

We have had available 154 different O antigens and 53 different H antigens. Therefore, in theory, 8,162 combinations could have been found and without doubt more O and H antigens wait to receive official recognition if they become significant in a medical or veterinary context or for some other reason. Many of these as yet undescribed antigens probably exist in animals. As is usual with strains of *E. coli* derived from animals, even with a full set of typing sera, 227 of the strains from cow-pats, although not rough, could not be typed with O sera and, similarly, 72 strains, although motile, could not be typed with H antisera.

As 65 of the 78 identified O/H combinations from cow-pats were found only once, it is not possible to discuss geographical variations of serotypes. For such results to be significant, a much larger sample would have to be taken. Samples also varied between containing only one serotype among all the 10 *E. coli* colonies picked and containing up to 7 serotypes. Presumably the type of variation of distribution observed with human faeces (Bettleheim, Faiers & Shooter, 1972) also occurs among cow pats.

At first glance the results described in this paper would indeed support the view that human and animal strains of *E. coli* are largely distinct. Second thoughts, however, suggest a little caution in accepting the opinion too firmly.

However, thoroughly human or animal stools are examined, only a minute fraction of the total bacterial content is examined, and inevitably strains recorded as being isolated tend to be those that predominate. It is always probable that if examination is continued, further strains may be isolated but after an amount of work that is impracticable in any ordinary investigation. If this is so, it is possible that many of the strains recorded as coming from humans only or from animals only might, with more diligent examination, be recorded as present in both man and animals. It is, too, possible, in view of the very wide scatter of serotypes in

cow-pats from different parts of the country, that if more sites had been examined, more strains corresponding to those found in humans would be isolated.

When using O and H antigens to type *E. coli*, there is a tendency to assume that each serotype so determined is distinct and an organism in itself. If this is so, and if O and H antigens do not change, the prospect of investigating the full natural history of *E. coli* is a daunting one, in view of the enormous number of different strains.

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