

***Escherichia coli* serotypes in the faeces of healthy adults over a period of several months**

By R. A. SHOOTER, K. A. BETTELHEIM, S. M. J. LENNOX-KING
AND S. O'FARRELL

*Department of Medical Microbiology, St Bartholomew's Hospital,
West Smithfield, London EC1A 7BE*

(Received 22 July 1976)

SUMMARY

The faeces of nine subjects eating mainly at home were collected at regular intervals over periods ranging from 2–5 months. Although a large number of serotypes of *E. coli* were isolated, the variety per subject was lower than is usually found. In most subjects only a limited number of serotypes persisted over most of the periods of study while many serotypes were only isolated on single occasions.

INTRODUCTION

There have been a number of reports that acquisition of new serotypes of *Escherichia coli* may be related to the development of diarrhoea (Rowe, Taylor & Bettelheim, 1970), particularly when related to travel or some other changes in diet. These studies have been complicated by the fact that little is known about the distribution of faecal *E. coli* serotypes in healthy subjects eating at home over an extended period of time, which might be used as a baseline for such studies. In the present study the faecal *E. coli* of nine people eating at home were examined over a period of months. This is an extension of studies on the effect of diet on the changing faecal *E. coli* flora of man (Cooke *et al.* 1970; Shooter, Cooke, Rousseau & Breadon, 1970).

MATERIALS AND METHODS

The subjects examined were eight women varying in age from 42 to 56 years and the husband of one of them aged 61 years. They all lived in a North London suburb and ate at home, although four went on holiday during the period of study and one also ate in a canteen. They regularly attended their general practitioner for reasons unrelated to their bowels. Regular faecal specimens, obtained from them over periods varying from 2 to 5 months, were cultivated directly on MacConkey's medium.

All plates were incubated overnight and from each ten colonies of *E. coli* were selected to represent each colonial type present. Only those that produced indole, were urease negative, did not utilize citrate, produced acid from lactose and acid and gas from glucose, were O serotyped by the methods of Bettelheim *et al.* (1975) using 154 O antisera. Representatives of each O group found in each specimen were also H typed by the method of Chandler & Bettelheim (1974).

Table 1. *Faecal specimens studied and numbers of serotypes isolated*

Index no. of person studied	No. of specimens studied	Period of study (months)	Total no. of different serotypes found
2	11	5	12
3	12	3	7
4	12	4	23
5	12	3	21
6	7	1½	4
7	11	2½	24
8	12	2½	3
9	12	3	1
11	12	3½	27

RESULTS

For the purpose of presenting the results, each different combination of O and H antigens found in a specimen is considered to represent a different serotype. The total number of specimens studied and the serotypes found are listed in Table 1. Subject 5 was the husband of 3 and those that went on holiday during the study were 2, 3, 5 and 8. Only subject 11 also ate in a canteen.

A large number of different OH serotypes were identified. Only those serotypes isolated from more than one stool specimen are listed in Table 2 which also gives the frequency of their isolation and the subjects from whom they were isolated.

In all, by means of 154 different O sera, 36 O types have been identified and these comprised 55 different OH serotypes. Nineteen different serotypes were found whose O antigen, though smooth, could not be typed and 13 rough types with different H antigens.

Some single specimens yielded strains with complex O antigens such as O19/O36.H33 and O19/O36/O88.H33 in subject 4 and O40(O15).H30 in subject 7. From a number of subjects were isolated apparently related strains; thus subject 4 yielded O18ab.H14 a week before O18ab.H — and subject 5 yielded O147.H30 and R.H30 from the same specimen and from one specimen of patient 8 were isolated O15.H30, O40(O15).H30 and O40.H30 but a week later she excreted Ont.H30.

A large number of serotypes only appeared in single specimens. The remaining serotypes are listed in Table 2 with the frequency of their isolation and the subjects from whom they were isolated.

DISCUSSION

Despite the variety of serotypes of *E. coli* that can be found in a single faecal specimen (Bettelheim, Faiers & Shooter, 1972) it is reassuring that some of these subjects yielded so few serotypes. Thus, over a period of study of three months 12 specimens from subject 9 yielded only *E. coli* O2.H6 although 120 strains were serotyped. Also, subjects 8, 6 and 3 only yielded 3, 4 or 7 different serotypes each.

The persistence of only a few serotypes is demonstrated in these three subjects,

Table 2. *The serotypes of E. coli isolated from more than one specimen*

Serotype	Subjects from whom isolated (Index No.)		Serotype	Subjects from whom isolated (Index No.)	
	No. of specimens yielding serotypes	No. of specimens yielding serotypes		No. of specimens yielding serotypes	No. of specimens yielding serotypes
O1.H7	3	6	O147.H-	2	1
	5	8		7	1
O1.H-	6	4	Ont.H4	4	1
	7	1	—	5	1
O2.H6	9	12	—	11	7
O3.H4	4	1	Ont.H7	2	1
	7	1	—	7	1
O4.H1	3	11	—	11	1
O4.H5	3	1	Ont.H9	2	2
	5	2	Ont.H10	2	2
O4.H-	4	2	Ont.H12	2	1
O6.H1	6	6	—	11	1
O8.H9	2	2	Ont.H21	5	1
O11.H-	5	2	—	7	1
	7	1	—	11	1
O15.H30	4	1	Ont.H30	7	1
	7	1	—	11	2
O16.H-	4	5	Ont.H34	7	1
O18ab.H14	4	1	—	11	1
	6	1	Ont.H40	4	1
	7	3	—	7	1
O19.H-	7	2	—	11	1
O21.H10	2	7	Ont.Hnt	4	2
O22.H1	8	8	Ont.H-	2	1
O25.H12	11	2	—	3	1
O29.H10	4	3	—	4	2
O40.H30	4	1	—	5	3
	7	1	—	7	2
O40 (O15).H30	7	2	—	11	4
O75.H55	3	1	R.H1	8	4
	4	1	R.H4	11	4
	11	1	R.H30	5	1
O75.H-	5	2	—	11	1
O79.H40	4	1	R.H-	4	8
	11	1	—	5	4
O87.H2	11	2	—	6	1
O136.H40	2	2	—	11	5
O147.H30	4	1	—	—	—
	5	1	—	—	—

because in subject 8 seven of the twelve specimens yielded O22.H1 and of the remaining two serotypes R.H1 was probably derived from O22.H1 and it is noteworthy that the other (O106.H1) also carried the H1 antigen. The antigen O106 is a complex O antigen whose serum is particularly difficult to purify (Bettelheim, 1968). In the two subjects 3 and 6 two serotypes persisted in each, O6.H1 and O1.H- in subject 6 and O1.H7 and O4.H1 in subject 3. The remaining serotypes were only isolated from these subjects on single occasions.

Of the remaining five subjects the persistence of certain serotypes is again indicated in three; thus O21.H10 was isolated from subject 2 on seven occasions, O1.H7 from subject 5 on eight occasions and O16.H- from subject 4 on six occasions. Subject 11 who also ate in canteens yielded the greatest variety of serotypes, confirming the suggestion that it is through food that humans acquire new serotypes of *E. coli* (Cooke *et al.* 1970; Shooter *et al.* 1970).

It is also noteworthy that of the only married couple in this study, subjects 5 and 3, both carried O1.H7 for the same period of time but the husband (subject 5) had a greater variety of serotypes than the wife.

This study again reveals the great variety of *E. coli* serotypes that can be isolated from even a limited number of human subjects and that as in previous studies food is the most probable source. Related serotypes were noted, suggesting that antigenic variation occurs *in vivo*.

We are indebted to the Wellcome Trust and to the Band of Governors of St Bartholomew's Hospital for a grant in support of this work.

REFERENCES

- BETTELHEIM, K. A. (1968). Investigations into the pathogenicity of *Escherichia coli* in human infections. Ph.D. thesis, University of London.
- BETTELHEIM, K. A., BUSHROD, F. M., CHANDLER, M. E., TROTMAN, R. E. & BYRNE, K. L. (1975). An automatic method for serotyping *Escherichia coli*. *Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene* (Abt. I, Orig. A) **230**, 443-51.
- BETTELHEIM, K. A., FAIERS, M. & SHOOTER, R. A. (1972). Serotypes of *Escherichia coli* in normal stools. *Lancet* *ii*, 1224-6.
- CHANDLER, M. E. & BETTELHEIM, K. A. (1974). A rapid method of identifying *Escherichia coli* H antigens. *Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene* (Abt. I, Orig. A) **229**, 74-9.
- COOKE, E. M., SHOOTER, R. A., KUMAR, P. J., ROUSSEAU, S. A. & FOULKES, A. L. (1970). Hospital food as a possible source of *Escherichia coli* in patients. *Lancet* *i*, 436-7.
- ROWE, B., TAYLOR, J. & BETTELHEIM, K. A. (1970). An investigation of travellers' diarrhoea. *Lancet* *i*, 1-5.
- SHOOTER, R. A., COOKE, E. M., ROUSSEAU, S. A. & BREADON, A. L. (1970). Animal sources of common serotypes of *Escherichia coli* in the food of hospital patients. *Lancet* *ii*, 226-8.