R FACTORS IN ENTEROBACTERIACEAE CAUSING URINARY-TRACT INFECTION IN GENERAL PRACTICE IN 1962-63 AND 1968-69

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SINCE the discovery of infectious drug resistance factors (R factors) in 1959 there has been great interest in their spread and incidence. In *Salmonella* and *Shigella*, a marked increase in resistance due to acquisition of R factors has been well documented (Anderson, 1968; Mitsuhashi, 1969; Davies, Farrant, and Uttley, 1970). The bacteria of the human bowel also carry R factors (Datta, 1969, Datta *et al.*, 1971) and represent a large pool of resistant organisms capable of causing infection in man. Non-enteropathogenic R+ bacteria are known to cause infections acquired within hospitals (Smith and Armour, 1966; Gardner and Smith, 1969; Anderson, Datta, and Shaw, 1972) but little is known of the incidence of infection with these organisms in the general community. Infections with organisms of these genera outside hospital are predominantly of the urinary tract, so the incidence of resistant organisms causing urinary infections was studied.

Brumfitt *et al.* (1971) reported resistance to one or more antibiotics in 7 of 37 (19 per cent.) of *Escherichia coli* causing urinary-tract infection in general practice. In the present study those figures are extended.

MATERIALS AND METHODS

Patients who had acquired urinary-tract infections outside hospital were studied at the Edgware General Hospital (Grüneberg, Leigh and Brumfitt, 1968). Cultures from 50 of the patients seen in 1962-63 and another 50 in 1968-69 were randomly selected for this study. The cultures had been stored either in stabs or slopes and were all examined together in 1970. The minimum inhibitory concentration (MIC) of the following drugs was determined: ampicillin (A), streptomycin (S), tetracycline (T), chloramphenicol (C), kanamycin (K), sulphafurazole (Su), nalidixic acid (NaI), and trimethoprim (Tr), by incorporating two-fold dilutions of the appropriate drug in Nutrient Agar plates (Oxoid No. 2) containing 4 per cent. lysed horse blood. Plates were inoculated with a multiple-point inoculator, the two inocula for each culture being 10^5 and 10 organisms. Plates were incubated overnight and the lowest concentration of drug which inhibited growth was recorded as the MIC. A control sensitive *E. coli* K12 strain was included in all tests. Results were reproducible on repetition with no more than two-fold variation. Except for sulphafurazole, the MIC for sensitive strains did

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not differ by more than two-fold from the control culture. Resistant strains were 8–128 times more resistant. For sulphafurazole, the control *E. coli* was inhibited by 8 µg per ml. However, a strain was recorded as resistant only if it grew in the presence of 100 µg per ml.

Cultures were tested for transfer of drug resistance to *E. coli* K12 in mixed culture (for methods see Datta, 1968). If resistance was not transferred directly, an attempt was made to "mobilise" it by introducing a de-repressed R factor (Anderson 1965; Meynell and Datta, 1967).

RESULTS

The results are shown in tables I and II. All organisms studied were *E. coli*. By the use of an exact test, a statistically significant increase ($p < 0.05$) was

TABLE I
Resistance of Escherichia coli isolates to antibiotics

Year	Total number of isolates	No. (and percentage) of isolates resistant to							
		A*	S	T	C	K	Su	Nal	Tr
1962–63	50	0	0	3 (6)	0	0	0	0	0
1968–69	50	1 (2)	4 (8)	5 (10)	0	0	8 (16)	0	0

* See *Methods* for key.

TABLE II
Resistance patterns of E. coli from patients with urinary-tract infections

Resistance pattern	Number of isolates with the stated resistance pattern in years	
	1962–63	1968–69
Fully sensitive	47	40
Resistant to Su*	...	2
Resistant to T	3	2
Resistant to SSu	...	3
Resistant to TSu	...	2
Resistant to ASTSu	...	1
Resistant to any agent	3 (6 per cent.)	10 (10 per cent.)

* See *Methods* for key.

detected in the incidence of resistance between the two series. All three of the tetracycline-resistant strains isolated in the 1962–63 series were able to transfer resistance directly to a recipient *E. coli* K12 strain. Of the second series, three strains transferred resistance (ASTSu, TSu and Su); in three strains the plasmid was mobilised (SSu, TSu, and T) and in four strains transfer could not be demonstrated (Su, SSu × 2, and T).

DISCUSSION

These small series show a definite but small increase in R-factor-mediated resistance in *E. coli* causing infections in the general community; but it was less than that seen in shigellae and salmonellae (Anderson, 1968; Davies *et al.*).

Studies on the changing pattern of antibiotic resistance in *E. coli* outside hospitals are few. In two series of observations made in different parts of this country (Essex: Robertson, 1971; Bristol: Gillespie *et al.*, 1971), the same general but slight trend of increasing resistance was seen. Other surveys have usually investigated only resistance to one or two drugs and these have shown variable changes in the frequency of resistance (Brooks, 1968; Brown *et al.*, 1968; Williams and Leigh, 1966).

It is not possible to be sure that R factors were not lost from the 1962-63 cultures during storage. Of 42 cultures of known R+ *E. coli* that had been stored in conditions similar to the urinary strains for 6 yr, R factors had been lost from six. A rate of loss of this order from the 1962-63 cultures would not negate the difference between the two series. Anderson (1968) also noted that R factors in *E. coli* were stable *in vitro* when stored in the original bacterial host.

It therefore appears that there is a general but not marked increase in infectious drug resistance in *E. coli* causing infection outside hospital in various parts of England.

SUMMARY

Antibiotic-resistance patterns of *Escherichia coli* causing urinary-tract infections contracted outside hospital were studied in 1962-63 and 1968-69. There was a slight but statistically significant increase in resistance over this period. The resistance was, in most instances, transferable.

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