TRANSMISSION OF DRUG RESISTANCE FROM Escherichia coli TO DYSENTERY BACTERIA

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Transmission of resistance to chloramphenicol, tetracycline, and streptomycin was transmitted by conjugation in vitro from Escherichia coli, possessing multiple drug resistance, to Shigella flexneri and Shigella sonnei. Resistance to furazolidone could not be transmitted.

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During recent years importance has been ascribed to the episomal R-factor in the formation of drug-resistance strains of Escherichia coli [1-11].

The present investigation was carried out to determine whether antibiotic-resistant strains of dysentery bacteria can be formed by conjugation with E. coli.

EXPERIMENTAL METHOD

Of strains of E. coli isolated in Saratov and carrying "infectious" polyresistance to antibacterial drugs, 4 strains of different origin were used. Two of these strains, isolated from a patient with acute dysentery and from a child with staphylococcal enterocolitis, were resistant to chloramphenicol (Cm), tetracycline (Tc), and streptomycin (Sm). One strain of E. coli, resistant to Cm and Tc, was obtained from the stools of a healthy person. The fourth donor consisted of a reference strain of E. coli M-17, to which resistance to Cm and Tc have been transmitted by preliminary experiments in vitro by conjugation with an antiobiotic-resistant (Cm, Tc, Sm) strain of Shigella sonnei.

The recipients consisted of 12 strains of Shigella flexneri of all serologic types and three strains of Shigella sonnei obtained from the Museum of the L. A. Tarasevich State Control Institute and highly sensitive to the above-mentioned antibiotics.

The experiments on transmission of drug resistance were carried out by mixed cultivation (for 4 h) of the parent strains by the method described previously [3]. The recombinants of dysentery bacteria formed as a result were isolated on Ploskirev's medium with addition of the appropriate antibiotics (Tc 20μg, Cm 25μg, Sm 50μg/ml medium) or of their combinations.

The resistance of the individual recombinant colonies to antibiotics was determined by the serial dilution method. To detect linkage of the characteristics of resistance to antibiotics, Lederberg's replica method was used.

EXPERIMENTAL RESULTS

The experiments showed that frequency of formation of recombinants of dysentery bacteria carrying the factor of multiple resistance to antibiotics varied from $1 \cdot 10^{-5}$ to $9 \cdot 10^{-7}$ and depended on the individual properties of both donor and recipient strains. No dependence of the frequency of recombinant formation on the species of the recipient could be detected. Strain E. coli M-17, which transmitted resistance to all recipient strains used in the experiment (except one strain of Sh. flexneri), possessed the greatest donor capacity. These results are in agreement with those obtained by other investigators who found that strains of E. coli carrying "infectious" drug resistance can transmit it to cells not possessing this factor for several generations with a frequency exceeding the usual rate of transmission [6, 11].

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Determination of the level and spectrum of drug resistance of the recombinant colonies showed that these characteristics were similar in every case to those of the donor strains. If filtrates of 24-h donor cultures or of heat-fixed cultures of E. coli were mixed with recipient cells, resistance was not transmitted to the latter.

As in other investigations of experimentally obtained antibiotic-resistant recombinants of dysentery bacteria [8, 11] no significant differences were found in the biological properties of the recombinants and the initial recipient strains. Some investigated recombinants kept their ability to produce specific dysenteric keratoconjunctivitis in guinea pigs. Subcultures isolated in the course of the disease persistently maintained the characteristic of multiple resistance to antibiotics acquired during conjugation.

To determine whether strains of dysentery bacteria resistant to nitrofurans could be obtained by conjugation with E. coli, two strains of E. coli (EM and B) resistant to furazolidone (F) isolated from patients with infections of the urogenital tract were used. Besides being resistant to F, these strains also were resistant to Cm, Tc, and Sm. Because of observations [9] indicating that polyresistant strains of E. coli isolated from the urine of patients with urogenital infections exhibit an episomal type of resistance to antibiotics, it was decided to use these strains as donors. Besides E. coli strains EM and B, the reference strain E. coli M-17, in which resistance to F (63 μg /ml) had been obtained by subculture in broth with increasing concentrations of this compound, was also used as donor. The recipients were the same strains of Shigella as in the previous experiments. Selection of resistant recombinants was made on Ploskirev's medium containing antibiotics separately or in combination with F (10 and to F 20 μg/ml).

The results showed that resistance to F was not transmitted from E. coli EM, B and M-17 (F^+) to the dysentery bacteria by conjugation. Moreover, the presence of the factor of resistance to F in the donor strains completely inhibited the ability of the E. coli cells to transmit resistance to other antibacterial preparations to the dysentery bacteria.

There are reports in the literature [4, 5, 7] of the possibility of formation of "infectious" drug resistance by introduction of the factors responsible for transmission into a population carrying nontransmitted determinants of resistance to a particular antibacterial preparation. Accordingly, resistance to Tc and Cm was transmitted to a variant of E. coli M-17 which had acquired resistance to F in vitro during prolonged cultivation in the presence of furazolidone, by conjugation with antibiotic-resistant (Tc, Cm, Sm) strains of Sh. flexneri No. 16519 and Sh. sonnei No. 269.

However, even after acquisition of the R-factor responsible for transmission of resistance to Tc and Cm, variant of strain E. coli M-17 (F^+) was still unable to transmit resistance to F by conjugation with Sh. flexneri and Sh. sonnei. Meanwhile, E. coli M-17 (Tc, Cm, F), obtained by crossing, transmitted resistance to Tc and Cm at a varied frequency to recipient strains of dysentery bacteria sensitive to them.

The results do not completely rule out the possibility that with an increase in the number of furazolidone-resistant strains of E. coli circulating in nature, conditions will arise enabling "infectious" resistance to be formed to this chemotherapeutic preparation also.

LITERATURE CITED

1. T. M. Kokhanovskaya, Antibiotiki, No. 9, 789 (1966).
2. I. Tereshin, I. Belousova, and R. Él'gart, Abstracts of Proceedings of the 9th International Congress on Microbiology [in Russian], Moscow (1966), p. 30.
3. S. I. Sherishorina and B. A. Shenderov, Zh. Mikrobiol., No. 10, 90 (1968).
4. E. S. Anderson, Ann. Inst. Pasteur., 112, 547 (1967).
5. J. G. Baudens and Y. A. Chabbert, Ann. Inst. Pasteur., 112, 565 (1967).
6. D. H. Datta, A. M. Lawn, and E. Meynell, J. Gen. Microbiol., 45, 365 (1956).
7. S. Mitsuhashi, K. Harada, H. Hashimoto, et al., J. Bact., 84, 9 (1962).
8. D. H. Smith and S. Halls, Brit. Med. J., 1, 269 (1966).
9. M. Vondraskova and J. Starka, Zh. Gig. Épidemiol. (Prague), 10, 166 (1966).
10. F. Vymola, A. Heszler, A. Panokova, et al., Zh. Gig. Épidemiol., (Prague), 11, 361 (1967).
11. T. Watanabe, Bact. Rev., 27, 87 (1963).