EXPERIMENTAL GENETICS

TRANSMISSION OF GENETIC DETERMINANTS

OF A HETEROGENEIC ANTIGEN SIMILAR TO HUMAN

TYPE O (H) ANTIGEN FROM Escherichia coli O55 TO

E. coli K12 IN MICE

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The possibility of transmission of genetic material from <u>Escherichia coli</u> O55 to <u>E. coli</u> AB1157 (K12) by their conjugation in the intestine of these animals was demonstrated in experiments on sterile CC57W mice. Transmission of determinants controlling the synthesis of a heterogeneic antigen similar to human type O(H) antigen also was discovered.

A study of crossing of enteropathogenic strains of \underline{E} , \underline{coli} containing heterogeneic antigens similar to the isoantigens of the human ABO blood group system with \underline{E} , \underline{coli} K12 showed that the genetic determinants of these antigens are localized in the region of the his* genetic locus on the chromosome of \underline{E} , \underline{coli} [4].

The object of this investigation was to study the possibility of transmission of genetic determinants of heterogeneic antigens from bacteria of serologically typed strains of \underline{E} , \underline{coli} O55 to \underline{E} , \underline{coli} K12 in the mouse intestine.

EXPERIMENTAL METHOD

Enteropathogenic strains of <u>E. coli</u> O55 sensitive to streptomycin and containing a heterogeneic antigen similar to human type O(H) antigen were used as donors, and a semiauxotrophic strain of <u>E. coli</u> AB1157, originating from E. coli K12 (thr⁻, leu⁻, pro⁻, his⁻, arg⁻, lac⁻, S^r) was used as the recipient.

The bacteria of these strains were crossed in mice. Altogether 40 CC57W mice weighing 18-20 g were used in the experiments. The intestine of the animals was first sterilized by administration of streptomycin (50,000 units), erythromycin (5 mg), and nystatin (4 mg) simultaneously to the animals by mouth [2, 3]. About 10^8 cells of an 18-h broth culture of <u>E. coli</u> AB1157 was given by mouth to the mice 24 h after the last dose of antibiotics, and the same dose of <u>E. coli</u> O55 cells was given 24 h later. The feces were collected daily thereafter for 1 week. The material was suspended in warm physiological saline in the ratio of 1:10 and seeded on selective media permitting growth of thr⁺, leu⁺, Sr; pro⁺, Sr; his⁺, Sr; arg⁺, Sr, and lac⁺, Sr recombinants.

Intact mice receiving antibiotics and also mice receiving bacteria of only one of the parental strains acted as the control.

*Abbreviations used: his + (his -) ability (inability) to synthesize histidine, thr + (thr -) ditto for threonine, leu + (leu -) for leucine, pro + (pro -) for proline, arg + (arg -) for arginine, and lac + (lac -) for lactose; Sr denotes resistance to streptomycin.

Research Laboratory of Experimental Immunobiology, Academy of Medical Sciences of the USSR. Patrice Lumumba University, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR N. N. Zhukov-Verezhnikov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 76, No. 12, pp. 90-91, December, 1973. Original article submitted March 12, 1973.

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TABLE 1. Isolation of Genetic Recombinants of E. coli from Intestine of Mice in Relation to Time of Taking Material

Crossing of serologically typed and un- typed strains	No. of ex- perimental mouse	Mean number of recombi- nants isolated (per dish)					
		1st day	2nd day	3rd day	4th day	5th day	6th day
	1	230	0	0	0	0	0
I—O55 × AB1157	2	231	0	111	0	0	0
	3	202	35	10	0	0	0
2—O55 × ×AB1157	4	26	6	0	0	0	0

The isolated genetic recombinants were studied for the presence of heterogeneic antigens by a test based on adsorption of specific isoagglutinins by heterogeneic bacterial antigens [1] and also for their content of somatic antigen by the usual technique of bacterial agglutination in tubes.

EXPERIMENTAL RESULTS

The results of the separate experiments given in Table 1 show that the number of isolated recombinants depended on the time when the feces was taken and was highest on the 1st day, falling considerably on the 2nd and 3rd days. Starting from the 4th day no recombinants were isolated in any experiments.

A study of 113 isolated recombinants showed that all were prototrophs; i.e., they inherited from E. coli

O55 the genes controlling the synthesis of threonine, leucine, proline, histidine, and arginine. At the same time they remained unable to ferment lactose. The overwhelming majority of the recombinants preserved the type of sensitivity to specific "female" phage φII .

Of the total number of recombinants tested, a heterogeneic antigen of the human O(H) type was found in 22.

The study of recombinants containing heterogeneic antigen showed that, unlike cells of the original strain \underline{E} , $\underline{\operatorname{coli}}$ K12 and recombinants without the heterogeneic antigen, they were agglutinated by OB serum against \underline{E} , $\underline{\operatorname{coli}}$ O55 to titers of 1:1600-1:3200; this result demonstrates that they contained a somatic antigen of the enteropathogenic bacteria of the donor strain. The results thus indicate that determinants of a heterogeneic antigen, similar to human type O(H) antigen, can be transmitted during conjugation of serologically typed and untyped strains of \underline{E} , $\underline{\operatorname{coli}}$ in the mouse intestine.

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