

## HETEROGENOUS ANTIGENS IN VACCINE PREPARATIONS OF *Vibrio cholerae*

N. N. Zhukov-Verezhnikov,\* P. I. Anisimov,  
N. S. Goncharova, G. M. Bochko,  
Z. N. Karaseva, L. N. Shanina,  
and S. N. Fomin

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Serological similarity was found between antigens of the human small intestine, stomach, and liver and antigens of various fractions of cholera vibrios. An antigenic similarity was found on testing the heart and kidney. Heterogenous antigen was found not only in somatic antigen of *V. cholerae* strain 569 (B), but also in the cholerogen, the toxoid which is the most widely used prophylactic preparation in use at the present time, obtained from it.

KEY WORDS: *Vibrio cholerae*; human tissues and organs; heterogenous antigens.

The problem of the presence of heterogenous antigens of *Vibrio cholerae* has started to be studied only comparatively recently [2, 3]. *Vibrio cholerae* induces weak immunity. Successful specific prophylaxis in the control of infectious diseases is directly dependent on the existence of epidemiologically highly effective preparations. The existence of heterogenous antigens common with antigens of human tissue cells in vaccine preparations of *V. cholerae* reduces their allogenicity. The discovery of protective antigens in *V. cholerae* cells is thus a very important practical task. It is important to consider weak and heterogenous antigens, which possibly play the principal role in the induction of antibodies depressing the viability of cholera vibrios [1], when tackling this problem.

In this investigation the presence of heterogenous antigens similar to antigens of certain human organs and tissues, was studied in several strains of *V. cholerae*, including vaccine preparations.

### EXPERIMENTAL METHOD

Virulent strain *V. cholerae* 569 (B), vaccine strain *V. cholerae* El-Tor T-4, and V. NAG 2423 and 19306 were investigated. Suspensions of living cholera vibrios in physiological saline in a concentration of  $8 \cdot 10^{10}$  cells/ml and their subcellular fractions — flagellae, cytoplasm, membranes, and cell walls — were used as the antigen. Subcellular fractions were obtained by centrifugation after disintegration of the living bacterial suspension of vibrios in the UZD-N-1 ultrasonic disintegrator at 35 kHz for 5-20 min (acoustic power 50 W/cm<sup>2</sup>).

Besides normal rabbit serum, Hottinger's broth in which the nutrient agar for cultivation of the bacteria were prepared, was used as the control. The tested antisera and normal rabbit serum with Hottinger's broth formed no precipitation lines.

Antisera against tissues of the human small intestine, stomach, liver, kidney, and heart were obtained by immunizing rabbits as described previously [2]. The antigen for immunization consisted of tissues of a stillborn infant. Saline extracts were tested by seeding for sterility. The precipitation test in agar gel was carried out with these sera and antigens.

\*Academician of the Academy of Medical Sciences of the USSR.

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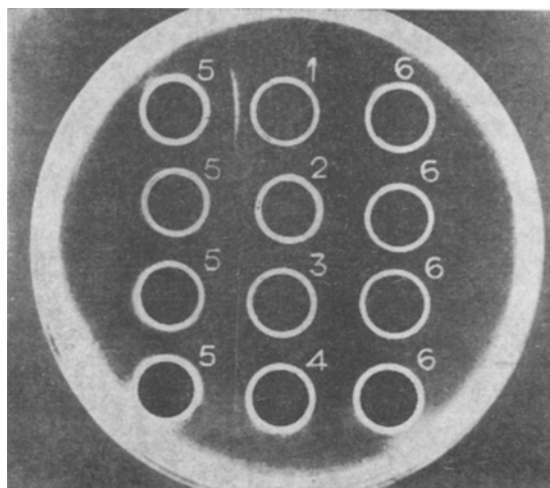


Fig. 1. Antigenic similarity between various fractions of *V. cholerae* 569 (B); and human stomach tissue: 1) flagellar fraction of *V. cholerae* 569 (B); 2) cytoplasm of *V. cholerae* 569 (B); 3) cell walls of *V. cholerae* 569 (B); 4) whole cells of *V. NAG* 19306; 5) anti-serum against human stomach; 6) anti-serum against human kidney.

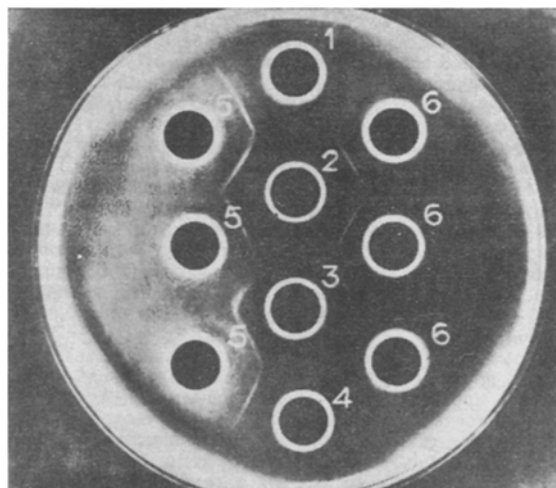


Fig. 2. Precipitation test with antisera against human small intestine and stomach and antitoxic and agglutinating O serum with the components of *V. cholerae* cells: 1) antitoxic serum; 2) agglutinating O serum; 3) antiserum against human stomach; 4) antiserum against human small intestine; 5) cell walls obtained from living *V. cholerae* 569 (B) cells; 6) cell walls obtained from *V. cholerae* 569 (B) cells heated to 100°C for 1 h.

## EXPERIMENTAL RESULTS

The results of the precipitation test in agar gel with antiserum against human small intestine showed that the virulent strain *V. cholerae* 569 (B) differed from the vaccine strain *V. cholerae* El-Tor T-4 and *V. NAG* 2423 and 19306 by forming two additional precipitation lines. Distinct lines also were obtained with antiserum against human stomach and the flagellar fraction of *V. cholerae* 569 (B). It will be clear from Fig. 1 that the highest content of heterogenous antigen was present in the flagellar fraction of *V. cholerae* 569 (B) and there was less in the cytoplasm and cell walls. No antigenic similarity was found with antiserum against human kidney. Antigenic similarity also was found between lithium spheroplasts of *V. cholerae* 569 (B) and stomach tissue. Antiserum against human liver reacted with the flagellar fraction of *V. cholerae* 569 (B). Meanwhile no reaction took place with the flagellar fraction of *V. NAG* 19306. No antigenic similarity was found with human heart tissue.

The writers showed previously that precipitation lines of a heterogenous antigen similar to the antigen of human small intestine tissue are identical with one of the precipitation lines of the somatic antigen of *V. cholerae* 569 (B). Subsequent experiments showed that the precipitation line of the cell wall antigen of *V. cholerae* 569 (B), which is similar not only to human small intestine, but also to human stomach tissue, is identical with the precipitation lines both of the somatic antigen and of the exotoxin, cholergen (Fig. 2).

The results thus indicate the existence of antigenic similarity between tissues of the human small intestine, stomach, and liver and various fractions of cholera vibrios. No antigenic similarity was discovered with heart or kidney tissue. Antigenic similarity was found not only with the somatic antigen of *V. cholerae* strain 569 (B), but also with the toxoid obtained from it, i.e., cholergen, the most widely used prophylactic preparation at the present time. This fact could detract from its protective properties.

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