SEARCH FOR ANTITUMOR ANTIBODIES IN THE BLOOD SERUM OF CANCER PATIENTS BY THE INDIRECT HEMAGGLUTINATION TEST

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Sera of patients with carcinoma of the breast, rectum, and stomach were investigated by the indirect hemagglutination test. When saline extracts of the corresponding tumors were used as antigen, no antitumor antibodies could be detected in sufficiently high titers in the patients' sera.

Data concerning the presence or absence of an immunologic reaction of the body to a tumor developing in it have been published in the literature [1-4, 6-8]. The discovery of antibodies in the sera of animals with tumors induced by carcinogens and viruses can be explained by the formation of antigens induced during the process of artificial carcinogenesis or by the presence of introduced virus antigens [2, 6, 8].

In the investigation described below an attempt was made to discover specific antibodies in the sera of patients against tumors developing in them.

EXPERIMENTAL METHOD

The indirect hemagglutination test was carried out with tanninized sheep's erythrocytes by Boyden's method. The Soviet pharmacopoeial preparation of acidum tannicum was used.

Sheep's erythrocytes, washed with physiological saline, were treated for 30 min with tannin solution in dilutions of between 1:20,000 and 1:40,000 (the solution was chosen each time depending on the reaction), after which they were washed three times with buffered physiological saline. One portion of erythrocytes was treated with a saline extract from the patient's tumor in one of three dilutions (1:5, 1:10, or 1:50), another portion with saline extract from the tissue of the same organ as that in which the tumor developed and in the same dilution as the tumor extract. Next, each of the portions, together with control tanninized erythrocytes, was washed three times with buffered physiological saline containing normal rabbit serum in a dilution of 1:200, and a 2% suspension of erythrocytes was made up in buffered physiological saline containing 1:200 normal rabbit serum. Successive dilutions were made of the test serum, and to tubes containing 0.2 ml of a diluted serum, 0.2 ml of 2% suspension of sheep's erythrocytes was added. One row of tubes contained erythrocytes sensitized by tumor extract, another row contained erythrocytes sensitized with extract from healthy tissue, while the third row (control–serum) contained tanninized erythrocytes. In addition, an antigen control was set up: 0.2 ml of 2% sheep's erythrocytes sensitized with antigen was added to 0.2 ml of buffered physiological saline containing normal rabbit serum in a dilution of 1:200. The results were read after the tubes had been kept in a refrigerator for 16-20 h and were verified under the microscope.

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EXPERIMENTAL RESULTS

Material for the investigations was taken from 31 patients with carcinoma of clinical group 2a and in stages II-III during operation. There were 15 patients with carcinoma of the breast, 10 with carcinoma of the rectum, and 6 with carcinoma of the stomach. Blood, tumor tissue, and macroscopically healthy tissue of the affected organ were taken from each of these patients, and the reaction was set up in the manner described above with the serum, tumor, and healthy tissue of the same patient. None of the sera gave a positive reaction in a titer higher than 1:12.

In the indirect hemagglutination test by Boyden's method using tanninized erythrocytes, between patient's blood serum and proteins obtained from the patient's tumor by salting out with saturated ammonium sulfate from a saline extract of the tumor at 30 and 50% saturation, with subsequent dialysis, a positive result likewise could not be obtained in titers higher than 1:12.

In addition, cross tests were carried out: 1) tumor and healthy tissue of one patient, serum of another patient with carcinoma of the same blood group as the first patient (12 tests); 2) tumor and healthy tissue of one patient, serum of a person of the same blood group as the patient, but free from cancer (9 tests). These tests likewise failed to give a positive result (the titer of the sera did not exceed 1:16).

Hence, no antibodies reacting sufficiently clearly with extracts of the tumor itself could be found in the sera of patients with carcinoma of the breast, rectum, and stomach, because, in view of the high sensitivity of Boyden's test, a serum titer lower than 1:50 cannot be regarded as positive.

Another possible explanation of the negative reactions in these experiments could be that whole extracts were used to sensitize the erythrocytes, and not purified antigens, against which antibodies could be present because the sensitivity of Boyden's test is known to be determined by the concentration of sensitizing antigen.

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