Growth of Brown-Pearce Carcinoma in the Anterior Chamber of the Eyes of Tumor-Immune Rabbits*

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The anterior chamber of the eye has long been recognized as a valuable site for the study of the growth of transplanted tumors. Recently Greene (8) successfully grew human cancers in the eyes of lower animals despite the fact that all other tissues were resistant to such heterologous tumor transplantation. The explanation for this may lie in the fact that a "barrier" exists between the blood and aqueous humor (7) which prevents the passage of antibodies from the former to the latter. This assumption is in some respects supported by the work of Becht and Greer (2) and also that of Hektoen and Carlson (9) who studied the concentration of antibodies in the various body fluids of animals immunized to bacterial and other foreign proteins and found that the titer of these immune bodies was highest in the serum and that they were present in very low concentration or were absent from the aqueous fluid of the anterior chamber of the eye.

These facts suggested that tumor might be grown in the anterior chamber of the eyes of animals immunized to a particular tumor by intracutaneous transplantation with the same tumor (4, 5, 12), despite the fact that all other tissues of the host are resistant to the growth of this tumor. Besredka and Bardach (3) were unsuccessful in their attempts to grow Brown-Pearce carcinoma in the anterior chamber of the eyes of tumor-immune rabbits, although the same tumor grew readily in this location in normal rabbits. Nevertheless, in view of the other evidence mentioned above (2, 8, 9), it was thought advisable to make a detailed study of the fate of transplants of Brown-Pearce carcinoma in the anterior chamber of the eyes of rabbits immunized to this tumor.

**Experimental Procedure**

Thirteen mongrel brown-grey rabbits were used for this experiment. Transplantation into the skin covering the dorsal spine was carried out by the technic previously described (1). The tumors grew successfully in the skin of all animals receiving tumor transplants. They attained their maximum size varying from 2.2 × 1.8 cm. to 6.1 × 5.4 cm. within from 3 to 5 weeks and then began to regress. In all 13 rabbits the skin tumors were completely resorbed within 3 weeks after they had started to regress. Following the disappearance of the skin tumors, the Brown-Pearce carcinoma was reimplanted into the testes of all of these animals in order to verify their resistance to the growth of this tumor. None of these 13 rabbits showed any evidence of tumor growth following this second transplantation. For purposes of control, portions of the same tumor which was used for this second transplantation were also inoculated into the testes of 3 normal rabbits. All 3 of these control animals developed large testicular tumors.

The tissue used for intraocular transplantation was obtained from a metastatic nodule of the Brown-Pearce carcinoma in the liver of a rabbit. The tumor was removed aseptically, immersed in Tyrode's solution, and cut into fragments 0.5 to 1.0 cm. in diameter. The fragments used for transplantation were all obtained from the same metastatic nodule.

The eye was grasped with a forceps, rotated downwards so that the corneo-limbal junction was readily available, and an incision was made through the cornea at the limbus into the anterior chamber with a sharp keratome. After a small amount of aqueous fluid had escaped, a tumor fragment, grasped in a small curved forceps, was introduced into the anterior chamber. For purposes of control, portions of the same tumor tissue were transplanted into the anterior chambers of the eyes of 4 normal rabbits using identical technic to that described above.

**Results**

In the 4 normal animals used for controls the tumor fragments transplanted into the anterior chambers of the eyes began to grow within 7 days of the time of transplantation and within 3 weeks had completely filled the eyes.
The behavior of the tumor transplants in the tumor-immune animals was different. In 3 of the 13 rabbits the transplanted fragments became pale, the edges rounded, and the fragments gradually diminished in size and were resorbed within 3 weeks. Histologic examination of these eyes failed to reveal any tumor at the site of transplantation. In 10 of the 13 rabbits the transplanted fragments became vascularized within a period varying from 6 to 14 days. No increase in size of the transplants was observed, however, for at least 3 to 6 weeks following transplantation. In 2 of these 10 animals this latent period was prolonged to 8 weeks. Once instituted, growth of the transplants progressed at a much slower rate than in the control animals. Ultimately however, within a period varying from 2 to 4 weeks after the tumors started to grow, the eyes were filled with a mass of tumor similar in every respect to that in the anterior chambers of the eyes of the control rabbits.

**DISCUSSION**

Davson and Quilliam (7) have stated that "there is a great deal of evidence that the aqueous humor is a filtrate from the blood plasma, molecules of the size of serum albumen or larger being retained while crystalloids are distributed between the two fluids in a manner characteristic of ultra-filtrates in vivo." These authors referred to a "barrier" between the blood and the aqueous humor. It is conceivable that this "barrier" is capable of preventing the passage of antibodies from the blood plasma to the aqueous fluid resulting in the failure of tumor transplants to grow in this fluid when all other tissues are resistant to their growth. This view is supported by the experiments of Becht and Greer (2) and Hektoen and Carlson (9), mentioned above.

It has frequently been stated that humoral antibodies have never been demonstrated in rabbits immune to Brown-Pearce carcinoma (6). However according to Lumsden (11) regression of tumors in rats and subsequent immunity was associated with a rise in the titer of agglutinins in the serum for this particular tumor protein. Kidd (1o) has described a complement-fixing antibody in the blood of rabbits bearing the Brown-Pearce carcinoma and also in rabbits in which the growth had regressed. It is not unlikely therefore that other immune bodies for the Brown-Pearce carcinoma appear in the blood of immunized animals even though serologic tests employed at the present time are unable conclusively to demonstrate them. It is also possible that, associated with this rise in the titer of immune bodies for this particular tumor protein in the blood, they also appear in markedly reduced concentration in the aqueous fluid. While the titer of these immune bodies in the aqueous humor of tumor-immune animals may be sufficient to retard the growth of the tumor transplants in the anterior chamber of the eye, they are apparently not present in sufficient concentration to suppress completely the growth of these transplants as occurs in all other tissues of the body in such refractory animals. In this connection it should be mentioned that in 3 of our 13 rabbits the tumor transplants did not grow at all in the aqueous fluid of the anterior chamber. This is probably due to the fact that sufficient antibodies were able to pass from the plasma to the aqueous fluid to suppress the growth of the tumor in the anterior chambers of the eyes in these animals.

In a previously well controlled study (13) it was shown that not only can rabbits be rendered refractory to the growth of the Brown-Pearce carcinoma by regression of intracutaneous tumors, but also that intracutaneous growth and regression of this tumor causes disappearance of pre- and co-existing testicular tumors and their metastases. In these studies we observed that occasional metastases within the eye also regressed following regression of the skin tumor. If it is true that the aqueous humor of the anterior chamber of the eye in the tumor-immune animal contains no antibodies for the Brown-Pearce carcinoma, or a very low concentration of them, the regression of pre-existing eye tumors in the refractory animal is difficult to explain. One would expect that following active immunization against this tumor, regression of the carcinoma would occur in every tissue except in the anterior chamber of the eye because of the supposed lack of immune bodies in this location. It is of course possible that the regression of pre-existing eye metastases in the immunized animal may have been a spontaneous coincidental regression in no way whatsoever related to, or dependent upon, the regression of the skin tumor. However we must emphasize that in our experience with the Brown-Pearce carcinoma over a period of 6 years, spontaneous regression was only rarely encountered.

The failure of Besredka and Bardach (3) to obtain successful intraocular growth in previously immunized animals may have been due to the fact that insufficient time was allowed to elapse from the time of transplantation to the termination of the experiment. As pointed out above, the latent period from the time of transplantation into the anterior chamber until the appearance of growth is markedly prolonged in the immune animals. In 2 animals, 8 weeks elapsed before the transplanted fragment showed any increase in size in contrast to the control animals where growth began usually after a latent period of 4 to 7 days.

**SUMMARY AND CONCLUSIONS**

Immunization of rabbits to the Brown-Pearce carcinoma by intracutaneous transplantation does not
confer protection against the growth of this tumor in the anterior chamber of the eye, despite the fact that all other tissues tested are resistant to its growth following such intracutaneous transplantation. However the growth of the tumor transplants in the eyes of such tumor-immune rabbits is considerably delayed.

REFERENCES
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