ONCOLOGY

Walker 256 Tumor Growth in Rats with Hereditary Defect of Vasopressin Synthesis

I. I. Khegai, N. A. Popova*, L. A. Zakharova, and L. N. Ivanova**

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Stable deceleration of Walker 256 tumor growth was detected in Brattleboro rats with vasopressin synthesis defect in comparison with normal WAG rats. In contrast to continuous tumor growth typical of rats, the growth of this tumor in Brattleboro rats was negligible and was observed during the first 15-18 days after transplantation, after which the tumor regressed and disappeared. The effect was age-dependent and was more pronounced in old animals. Repeated injection of Walker 256 cells does not lead to tumor development, which attested to direct involvement of the immune system in the detected phenomenon.

Key Words: *vasopressin; diabetes insipidus; Walker 256 carcinosarcoma; immunity*

Non-strain-specific Walker 256 carcinosarcoma is widely used for simulation of solid tumor growth in rats and evaluation of the therapeutic efficiency of antitumor drugs and technologies [6,8,10]. Walker 256 strain originates from rat mammary tumor and is characterized by hormone sensitivity [7]. A specific feature of Brattleboro rats is hormone imbalance because of vasopressin (VP) gene inactivation. The absence of VP primarily affects the water-electrolyte balance and leads to the development of severe diabetes insipidus [3]. It was also proven that parameters of the immune system are also modified in Brattleboro rats [1,4].

We studied the dynamics of Walker 256 tumor in Brattleboro and WAG rats during different periods of ontogeny.

Laboratory of Physiological Genetics, Institute of Cytology and Genetics, Siberian Division of Russian Academy of Sciences, Novosibirsk; *Novosibirsk State University; **N. K. Koltsov Institute of Developmental Biology, Russian Academy of Sciences, Moscow. ***Address for correspondence:*** khegay@bionet.nsc.ru. I. I. Khegai

MATERIALS AND METHODS

We used cell suspension of Walker 256 tumor strain stored in liquid nitrogen. After defrosting the suspension was injected into the thigh to WAG rats in a dose of 8×10^5 cells [9] and the strain was then maintained in the solid form in rats of this strain. WAG rats, serving as the control of normal VP gene expression, and Brattleboro rats have at least 95% locuses with identical set of fixed alleles [2]. Cell suspension was transplanted to WAG and Brattleboro rats aged 3 months (n=10; 150-180 g), 8 months (n=10; 220-300 g), and 12 months (n=10; 250-350 g). The tumors were measured with a slide gage and their volumes were calculated (product of 3 perpendicular dimensions). The significance of differences was evaluated using Student's t test.

RESULTS

Solid tumor nodes on the external side of the thigh were palpated in all animals several days after intra-

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muscular transplantation of Walker 256 cells. At the age of 3 months the rate of tumor growth was similar in rats of both strains during the first 2.5 weeks after injection of 5×10^5 cells in suspension (Fig. 1, *a*). However, starting from day 18 the tumor growth ceased in Brattleboro rats; the tumor node decreased in size and completely disappeared by day 30. In WAG rats, the tumor continued to grow and led to animal death when its size reached about 25% animal body weight.

Rats aged 8 and 12 months were injected with 8×10^5 cells. In 8-month-old rats, the differences in the dynamics of tumor growth manifested virtually from the very beginning of the tumor appearance (Fig. 1, *b*). Tumor growth in Brattleboro rats was observed during the first 2 weeks, after which the tumor gradually regressed and disappeared by day 30. In WAG rats the tumors continued to grow until animal death. At the age of 12 months the pattern of changes in the transplanted tumor detected in younger rats was the same, quantitative differences between the two strains were still more pronounced (Fig. 1, *c*).

The dynamics of solid tumor growth in WAG rats after injection of Walker 256 carcinosarcoma is typical of rats [9]. The data obtained on Brattleboro rats are absolutely new. It seems that the absence of VP is an important factor, but it is also obvious that in this case we observe not a rapid hormonal effect, but the effect mediated through activation or inactivation of gene expression and cell proliferation [12]. By the terms and rate of formation, the process is similar to suppression of immune reactions observed in Brattleboro rats [1]. The regulatory processes with participation of Rho family guanosintriphosphate-binding proteins and Rho-associated kinase can be involved in rats with Walker 256 carcinosarcoma [5,7], as well as the phosphatidylinositol-3-kinase pathway [11]. Since tumor cells used in experiment do not express the main tissue compatibility genes, weak histocompatibility genes seem to be expressed; the tumor overcomes incompatibility by these genes. Although the tumor in rats of both strains develops under conditions of immune response to transplantation tumor antigens, the differences in the level of this response form a sufficient base for the specific features of tumor growth observed in Brattleboro rats.

In order to check this hypothesis, we repeatedly injected Walker 256 cells to all Brattleboro survivors of the three studied age groups in which the first tumor regressed completely. Repeated transplantation did not lead to the development of tumor nodes, which confirms direct participation of the immune system in the realization of the detected phenomenon. Further experiments are needed for more detailed analysis.

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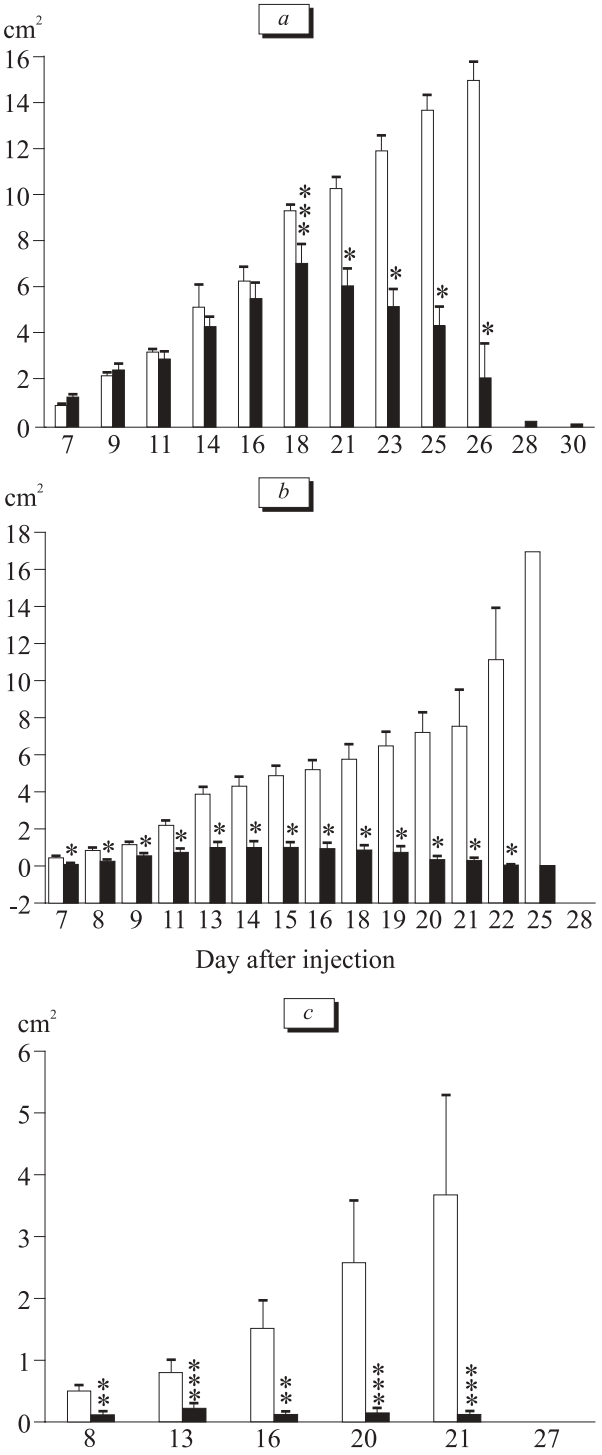

Fig. 1. Development of solid tumor after transplantation of Walker 256 carcinosarcoma cells in WAG (light bars) and Brattleboro rats (dark bars) aged 3 (*a*), 8 (*b*), and 12 (*c*) months. *$p<0.001$, **$p<0.01$, ***$p<0.05$ compared to WAG rats.

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