Antitumor Activity of Antiserum Nerve Growth Factor (anti-NGF) (36031)

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A protein in mouse sarcoma (1) when isolated (4) greatly enhances the growth and differentiation of sympathetic ganglia in chicken embryo (7) and in neonatal mice (6). Similar protein is also present in snake venom (2, 3) and in the submaxillary gland of mice. Due to its highly specific growth properties it is referred to as the nerve growth factor (NGF). The antiserum to protein (anti-NGF), when injected in newborn mice or rats, inhibits the growth and differentiation of sympathetic ganglia and results in almost total sympathectomy (5).

Many electronmicroscopic studies *in vivo* and *in vitro* have shown the cytotoxic effects of anti-NGF (thought to be of antigen–antibody type) on ganglionic cells, particularly their nuclei (10, 7). We have previously reported briefly on similar but rapid cytotoxic and cytocidal effects of anti-NGF on cultured human glioblastoma cells (8, 9).

As the presence of NGF in mouse sarcoma is well established, and as the anti-NGF has the effect opposite to NGF on the cells, the present study was undertaken to determine whether or not antinerve growth factor can affect the growth of chemically induced tumors in mice.

Materials and Methods. Newborn CF-1 mice (Carworth Farm) were injected subcutaneously with "Wellcome" antiserum to nerve growth factor for the first 5 days of life in increasing doses as follows: 0.07 ml, 0.10 ml, 0.10 ml, 0.12 ml and 0.15 ml; thus each mouse received just over 0.5 ml. This dose produced complete immunosympathectomy. Other newborn mice received an equivalent volume of horse serum. On day 27 after their birth, all animals were injected subcutaneously with 3 mg of benzo[*a*]pyrene (dissolved in olive oil at 37°). Three days later, half of the mice which had received horse serum were given daily 3 injections of nicotine (0.5 mg/kg; subcutaneously) while the other half (serving as control), which had been immunosympathectomized, received an equivalent volume of saline.

Injections of benzo[*a*]pyrene were repeated every 30 days to mice having no lesions. A total of three injections were administered. These animals were observed periodically for 170 days after the first injection of chemical carcinogen and the first appearance of tumor was recorded.

Results and Discussion. The present study indicates that pretreatment of animals with anti-NGF delayed the appearance of tumors induced by benzo[*a*]pyrene (Fig. 1). While in the control mice, tumors began to appear from day 27 of the first injection of benzo [*a*]pyrene, their appearance occurred only at day 90 in immunosympathectomized mice. Likewise, nicotine pretreatment also delayed the appearance of tumor; it appeared on day 100.

Both these treatments, *i.e.*, anti-NGF and nicotine, not only delayed the appearance of tumors but also caused lower incidence of tumors. The percentage of incidence of tumor in immunosympathectomized mice, nicotine-treated mice, and control mice, was 40.0, 53.3, and 77.4, respectively. While nicotine treatment delayed the first appearance of tumor and reduced the percentage of incidence of tumors, it caused significant increase in the average weight of tumor. Average of tumor weight in nicotine-treated animals was 8 g; in control mice 4.5 g; and in immunosympathectomized mice 3.1 g.

Thus, pretreatment of mice with anti-NGF caused: (i) a delay in the formation of tu-

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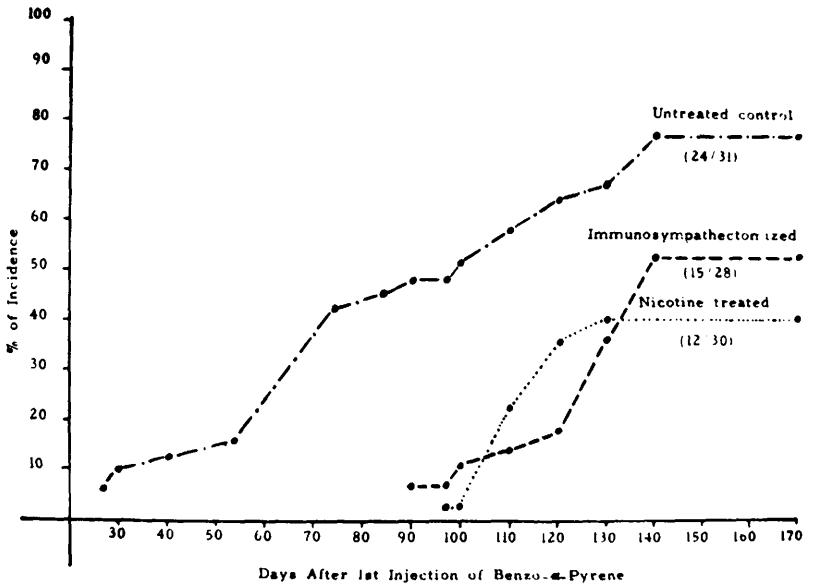

FIG. 1. The cumulative percentage incidence of benzo[*a*]pyrene tumors: There is a marked delay in both immunosympathetic and nicotine-treated animals.

mors; (ii) a reduction in the occurrence of the tumors; and (iii) a decrease in the rate of growth of tumors that do develop. These results suggest that antiserum to nerve growth factor has an antitumor activity. This is further confirmed by experiments using tumor cell culture stained cover slip preparations (unpublished data).

The present finding represents the first clear description of the prevention of and the inhibition of the growth of tumors by anti-NGF. This unexpected and dramatic finding naturally directs our attention to a heretofore unrecognized feature of the anti-nerve growth factor.

Summary and Conclusion. Pretreatment of newborn mice with antiserum to nerve growth factor or daily three injections of nicotine to mice delayed the formation of tumors induced by benzo[*a*]pyrene and reduced the occurrence. While anti-NGF reduced significantly the rate of growth of the tumor developed, nicotine treatment increased it.

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