Table 1. Immunochromatographic Separation of Breast Carcinoma Antigens on a Column of Cross-linked Horse Anti-normal (Human) γ-Globulin

Fraction	Titre in single gel-diffusion Against *NHGG*	Against *HTUGG*	Protein concentration g%	No. of lines in double gel diffusion (Fig. 1) Against *NHGG*	Against *HTUGG*
Unabsorbed tumour antigens	2^{-7}	2^{-5}	3·5	2	8
0 (1st 100 ml.)	0	0	0·0	0	0
1 (2nd 100 ml.)	0	2^{-3}	0·070	0	1
2 (3rd 100 ml.)	0	2^{-3}	0·062	0	1
3 (4th 100 ml.)	0	2^{-2}	0·051	0	1
4 (5th 100 ml.)	0	2^{-4}	0·480	0	2
5 (6th 100 ml.)	0	2^{-4}	1·680	0	6
6 (7th 100 ml.)	0	2^{-5}	0·600	0	4

NHGG, equine hyperimmune anti-normal γ-globulin; *HTUGG*, equine hyperimmune anti-tumour γ-globulin.

extract (that is, 8) which react with anti-tumour globulin (Fig. 1, No. 1 and *B*) minus the antigens in the same (that is, 2) reacting with antinormal globulin (Fig. 1, No. 1 and *A*). Analysis of the protein complement of breast tumour tissue reveals only antigenic acquisition but no antigenic deletion or deviation, 70 per cent of the protein being of normal antigenic nature.

These results seem to indicate that this method resulted in the resolution and large-scale preparation of specific antigens from human adenocarcinoma of the breast.

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Effect of Early Thymectomy on Development of Mammary Tumours in Mice

It is now well established that the thymus plays an important part in the development of immunological competence in mammals. For example, in the mouse, removal of the thymus within the first 24 h after birth results in a serious impairment of the capacity to produce circulating antibody and the ability to reject allogeneic grafts of normal or neoplastic tissues. In addition, these mice also develop a wasting syndrome ending in early death[1–7]. Surgical ablation of the thymus at 6 and up to 35 days of age also produces a state of immunological impairment, which is more severe when thymectomy is performed earlier in life. These animals do not develop the wasting syndrome and have a life-span comparable with that of non-thymectomized controls[8–10].

Since development of mammary tumours in susceptible strains of mice depends at least in part on the presence of an infectious agent transmitted by the mother to the progeny via the milk (Bittner's virus), it was considered of interest to ascertain whether or not thymectomy performed at 6 days of age in mice of a high cancerous strain would affect the spontaneous development of mammary tumours in these animals.

Two groups of female mice of the *C3H/Bi* strain were used. One group of animals was thymectomized 6 days after birth by the technique routinely used in this laboratory[11], and the other left as non-thymectomized controls. After surgery, mice were raised by their own mothers and weaned at 30 days of age, at which time the thymectomized and non-thymectomized mice were housed in plastic cages in groups of 4–5 female mice per cage. One normal male of the same strain was introduced into each cage and the mice were allowed to breed. Breeding behaviour was assessed by the number of litters born from each mother in both groups (Table 1). The mice at all times had free access to 'Purina Laboratory Chow' and tap water.

The incidence of spontaneous mammary tumours in both groups was determined by weekly inspection of each individual mouse. The age of the mouse was recorded at the time a tumour appeared.

Table 1. Incidence of Spontaneous Mammary Adenocarcinoma in Thymectomized and Non-thymectomized Female Mice of the *C3H/Bi* Strain kept as Breeders

Group	No. of mice	Breeding behaviour. No. of litters born (mean)	No. of mice with cancer	%	Mean cancer age (days ± *S.D.*)
C3H normal	73	4·4	69	94·5	276 ± 5·04
C3H thymect. at 6 days of age	26	4·6	15	57·6	335 ± 16·2

The results of these experiments (Table 1) demonstrate that, in the group of *C3H* mice thymectomized at 6 days of age, 57·6 per cent of the females developed spontaneous mammary tumours as compared to 94·5 per cent in the group of non-thymectomized controls. This difference is statistically significant at the 1 per cent level. Furthermore, thymectomized mice developed tumours significantly later than non-thymectomized controls. In the former group the average cancer age was 335 ± 16·2 and in the latter 276 ± 5·04 days.

Although the interpretation of these results must remain a matter of conjecture at the present time, a few pertinent comments are in order. According to Bittner[12] development of breast cancer in mice is dependent on the concurrent effect of three factors, namely, inherited susceptibility, proper hormonal stimulation, and the presence of the mammary tumour 'agent' or virus which is transmitted from the mother to the offspring by the milk. Since the inherited susceptibility for tumour development is genetically controlled and presumably does not change after thymectomy, it might be that the mechanism operating in cancer prevention by early thymectomy, as reported herein, could be related to an alteration of the hormonal stimulation for tumour development. Another possibility is that the thymus may be essential early in life for proper growth and multiplication of the mammary tumour virus. Finally, the development of mammary tumours could be related to the immunological responsiveness of the host animal to the virus. It has been shown recently that neonatal thymectomy in mice greatly reduces the mortality resulting from inoculation with lymphocytic choriomeningitis virus (LCM), and this reduction in mortality is the result of decreasing the host response to the virus[13,14]. Perhaps the development of spontaneous mammary tumours in mice is also in part dependent on a similar mechanism.

Experiments designed to investigate these and other possibilities are in progress.

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