THE RELATION OF THE HYPOPHYSIS TO THE GROWTH OF MALIGNANT TUMORS

I. THE EFFECT OF HYPOPHYSECTOMY ON TRANSPLANTED MAMMARY CARCINOMA IN THE WHITE RAT

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INTRODUCTION

In spite of the fact that we know that the anterior lobe of the hypophysis affects body growth (1, 5) and basal metabolism (6) and therefore cellular growth and development, relatively little has been done with regard to direct observations on these cellular effects or their bearing on the problem of malignancy. Sechi (13) obtained a small increase in oxidative activity of embryonic tissue and Rous sarcoma with the addition of "hypophysin." Baker and Carrel (2) showed that peptic digests of the anterior pituitary promoted proliferation of sarcoma cells of a rat tumor in tissue culture to the same degree as embryonic extract. Semura (14) reported that extracts of the anterior pituitary remarkably increased the growth of chick embryo ventricle in plasma containing embryonic extract or diluted with Ringer's solution only. Rohdenburg (12) observed an inhibition of the division energy of Paramoecia by the introduction of dried whole pituitary substance into the medium. He finds an inhibition with pituitrin alone, however, which may account for the findings with the whole gland.

Because of the interrelations of the gonads and the hypophysis, Hofbauer (8, 9) began a series of studies on the effect of anterior hypophysis transplants and extracts upon the epithelium of the uterus. He reports a proliferation of the epithelial cells of the uterine cervix into the sub-epithelial tissues which he considers analogous to a precancerous lesion. We also have the observation of Zondek (16) that cases of uterine carcinoma give positive tests for "Prolan."

The authors have undertaken, therefore, a series of studies
concerning the effect of the hypophysis on malignancy. The primary attack consists in studying the effect of the removal of the whole gland upon the development of transplanted and autogenous neoplasms. This paper deals with the effect of hypophysectomy on the growth of a transplanted mammary carcinoma in a series of albino rats.¹

**Experimental Procedure**

A large number of rats, most of which were between eleven and thirteen weeks old, were divided into the following groups:

*Group A:* These rats were hypophysectomized by the method of P. E. Smith (15). A number died of postoperative pneumonia, either before the inoculation of the tumor or shortly thereafter. Only those that lived at least twenty-six days after inoculation are considered in the study.

*Group A':* These were animals from Group A which on autopsy were found to have had incomplete removal of the gland. They thus become the most valid control group of the experiment, since everything that was done to group A was also done to them, including the removal of a large portion of the hypophysis.

*Group B:* A small number of hypophysectomized animals were selected from group A but were not inoculated with tumor. These were used as controls in certain chemical studies and do not require further consideration in the present report.

*Group D:* This group served as a control on the effect of the operation aside from the actual removal of the gland itself. Eleven animals were operated upon in the same manner as group A, except that the hypophysis was simply visualized, the bone button then returned, and the wound closed. Nine animals survived.

*Group E:* These animals were not operated upon. They were inoculated with the tumor at the same time as the other groups.

*Group F:* Animals in this group were selected from those in group E after the inoculated tumors had grown for a period of about three weeks, and were hypophysectomized according to the original procedure. Animals bearing nearly exactly duplicate tumors were retained in group E and constitute group E'. Group F is considered as a distinct group; while for all calculations of the E group, the E' animals are included.

¹Since the completion of the work included in this report, Bischoff, Maxwell and Ullmann (4) have reported retardation of tumor growth in rats whose heads were irradiated with radon or sub-lethal doses of x-rays. They assume that this retardation is secondary to an interference with the growth principle of the hypophysis since the body weights of the animals were also affected.
Operations on groups A and D were performed between the 19th and 29th of March, 1931. On April 8 all animals except those of group B were inoculated with mammary carcinoma cultivated by Dr. Geo. Walker of Baltimore and designated as No. 256. This tumor gives a high percentage of takes in albino rats; has few regressions, particularly after the tumor has reached a diameter of 20 mm., and grows to large size. In transplanting the tumor a single donor was placed under "Dial" anesthesia, the tumor carefully exposed, and by means of a small punch constructed for the purpose from a large hypodermic needle, a cylinder of the actively growing tissue was cut, of suitable diameter to enter the trocars. This was divided into grafts of similar size which were deposited subcutaneously on the left side about midway between the fore and hind legs.

Takes in this series of 78 transplants were 94.5 per cent with two regressions, both of which occurred early. The tumor was allowed to grow in the animal until death supervened. Semi-weekly measurements of tumor length and breadth were used to chart the rate of neoplastic increase. The onset and progress of necrosis were also recorded. The animals were weighed on the same days that tumor measurements were taken. When death occurred the tumor was dissected away and its volume measured by displacement. In the case of ulcerated tumors the defect was obliterated by the use of paraffin and the volume determined. The size of the tumor was also recorded by a plaster cast as a matter of permanent record.

The completeness of the hypophysectomies was determined indirectly by observations on the degree of genital atrophy; and directly by microdissection of the animals' heads. Thus the original group A was divided into the group A with complete removal and group A' with incomplete removal. In this latter group regeneration had occurred in all instances.

In the final compilation 47 males and 8 females are considered.

¹ The heads of these animals were fixed in formalin at the time of autopsy and some weeks later were dissected under the same magnifier as was used in performing the hypophysectomy. This was a working magnification of ten times. By clipping off the entire vertex of the skull with scissors, it was possible to lift the brain in the frontal region and readily visualize the optic chiasm and the pituitary fossa. When hypophysis tissue was present it was saved for microscopic record, but in the nine cases in which it was completely absent it could obviously not be subjected to microscopic study. Because of the difficulty encountered in a trial method of subjecting the entire brain or skull to serial section, it is felt that the method employed was distinctly more feasible and certainly as complete.
PRESENTATION OF FINDINGS

The semi-weekly measurements of the transplanted tumors were used in computing tumor area. Instead of figuring the exact cross-sectional area of an ellipsoid bisected at its maximum diameter, it was obviously easier to determine the area of a quadrilateral figure within which the tumor could be placed. This, of course, bears the same relation to the growth rate of the tumor as the more complex determination. The charts bearing tumor area data are understood, therefore, to be derived in this manner.

**Chart I. Tumor Area Data for Groups A, A', E, D**

Broken lines indicate additional tumor area of axillary metastases.
way, and the apparent discrepancy between these figures and the directly measured tumor volumes at autopsy is explained.

Chart I presents tumor growth rate data for groups A (completely hypophysectomized), A' (partially hypophysectomized with regeneration), E (unoperated controls), and D ("fake-hypophysectomy" controls). It will be observed that the group-average tumor area for the control animals at the end of five weeks' growth is not quite three times that of the hypophysectomized group (A). Groups A' and D approach or slightly exceed, respectively, the group-average tumor area of the unoperated controls (E). The broken lines include the area of metastatic growths. It is interesting that no metastases occurred in the small series of animals comprising groups A and F.

Chart II depicts similar data for groups F and E'. This latter group consisted of picked animals from group E that were strictly comparable on the basis of tumor size with those of group F after
two and one-half weeks of tumor growth, at which time group F were submitted to hypophysectomy. It will be seen from this chart that the effect of hypophysectomy on tumor growth rate takes place at once. Two days following the operation a distinct group-average tumor area difference is observable. Two weeks following operation the group-average area of the operated animals

**Chart III**

Group A completely hypophysectomized............................9 animals

" A' partially hypophysectomized..............................13 "

" F hypophysectomized three weeks after tumor growth........9 "

" D exposure of hypophysis without removal....................8 "

" E unoperated controls........................................16 "

Column height indicates average total weight at death. Column position indicates average duration of life. Shaded column indicates proportion of total weight attributable to tumor tissue.


only slightly exceeds one-half that of the unoperated controls (E'). This is a change of about the same order in proportion to the period of observation as that observed in Chart I.

Chart III presents the weight curves for various groups, the average duration of life after tumor inoculation, and information concerning the proportion of total weight attributable to the growth of tumor tissue.
It is apparent that the completely hypophysectomized group (A) never later exceeded their weight prior to operation, even with the weight of the tumor growths included. All other groups show about the same gradient of increase in total body weight, including tumors. The gradient of group A' shows an initial delay caused by the partial removal of the hypophysis. The end-results, however, are not distinctly different from the other control groups (E, D).

There is observable a marked difference in the proportion of tumor tissue weight to body weight exclusive of tumor in the various groups at death. These are: 1:3.2 (E); 1:2.9 (D); and 1:3 (A'); while on the other hand group A is 1:14.5, even though the body weight is less; and group F is 1:6.7, being a composite of two and one-half weeks' growth of tumor prior to hypophyseal removal and two weeks' growth thereafter. The ratios of tumor weight at death in the various groups is as follows, using group A as unity: 1:5.9 (A'), 1:6 (E), and 1:7 (D) for the control groups; and 1:2.4 for the composite F group. Thus there is a tumor weight difference between group A and the controls of well over six times. This weight difference is even more significant than the tumor area data presented.

It should be noted that the hypophysectomized groups (A, F) had a shorter average duration of life than any of the control groups. The possible bearing of hypophysectomy on general body resistance should be investigated.

Attention is called to the plateau in the central portion of the weight curves. During this period a large number of rats suffered from what appeared to be a fermentative diarrhea brought on by the substitution of a local calf meal with a somewhat different composition than that previously employed. At point Z on the graph the diet was changed to a cornmeal and powdered milk ration, with rapid subsidence of the intestinal disorder and marked increases in weight for all groups except A.

**Discussion**

It is plain from these data that hypophysectomy is responsible for a delay in the usual growth rate of this tumor in rats. Whether this effect is direct or indirect, whether it is an inhibition or a deficiency, is at present undetermined. It seems reasonable to assume that the absence of the growth principle may have a direct bearing on the problem. The fact that the physiological effects of hypophyseal removal, as well as the effect on tumor
growth rate, occurs so directly after the operation suggests a hormonal deficiency as a basis of explanation. It would seem at least to be evidence against a very indirect effect where the time factor would be expected to be more apparent.

An additional point which must be clarified is whether these findings hold true for other tumors or are limited to tumors of mammary origin similar to the one used by us. We feel that the effect of the ovarian hormone as shown by Loeb (10, 11) can be disregarded for this series because of the small number of females included and their complete absence from groups A and E.

That the effect could be due to a poor general condition of the animals seems doubtful in view of the work reported by Bischoff, Maxwell and Ullmann (3), in which poisoning and starvation of animals bearing this tumor did not appreciably change the tumor growth rate, even when the animals became markedly emaciated.

The observed retardation of tumor growth rate in our series should lead to more fundamental questions concerning cellular proliferation both in its relation to tumor growth and in the field of general tissue repair.

SUMMARY

1. In a group of hypophysectomized albino rats largely composed of males, the growth rate of a transplantable mammary carcinoma was greatly reduced.

2. If hypophysectomy was performed after development of the tumor, there was an immediate retardation of the rate of neoplastic growth.

3. In this series hypophysectomy did not cause complete cessation of tumor growth.

The number of completely hypophysectomized animals was, of course, small, and for that reason the findings cannot be considered as conclusive.

BIBLIOGRAPHY

