

## EXPERIMENTAL OVARIAN TUMORS IN RATS

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Several methods of obtaining ovarian tumors in laboratory animals have been described in the literature. Tumors have been observed to develop after irradiation of the ovaries of mice [3, 9, 15] and rats [2] with roentgen rays, after transplantation of the ovary into the spleen of castrated mice [5, 13, 16, 19], rats [1, 6, 10], and guinea pigs [14, 20], after ligation of the vascular bundle of the ovary in rats [11], and also after subtotal castration of mice [18]. As a result of these procedures luteomas, thecomas, tubular adenomas, granulosa-cell tumors and mixed tumors have developed. Hypophysectomy is known to prevent the appearance of experimental ovarian tumors [8, 12, 17] while an increase in the level of gonadotropins in the body, conversely, increases the frequency [21] and shortens the latent period [7, 22] of development of these tumors.

The object of the present investigation was to verify the hypothesis that an essential prerequisite for the development of ovarian tumors is a temporary depression of their function, leading initially to a lowering of the level of ovarian estrogens in the body, followed by a compensatory increase in gonadotropin production. The decisive importance of the latter to the development of this type of tumor is not doubted, even by clinicians [4].

### EXPERIMENTAL METHODS

A temporary depression of ovarian function was attained by irradiation with roentgen rays, transplantation of the ovary into muscles, or subtotal resection of the organ. Observations were made on 435 female rats, taking part in the experiments at the age of 2-3 months. The animals were divided into 4 groups.

The 1st group consisted of 138 control rats. The 2nd group contained 196 animals irradiated in the lumbar region with doses of between 100 and 1000 R at a single exposure or 300 R daily for 4 days (total 1200 R). The rats of the 3rd group underwent homotransplantation of the ovary, previously irradiated in a dose of 600 R (16 animals) or unirradiated (44 animals), into the thigh muscles, accompanied at the same time by ovariectomy. Subtotal castration was performed on the rats of the 4th group in which one ovary was removed in toto and the greater part of the other ovary was resected.

### EXPERIMENTAL RESULTS

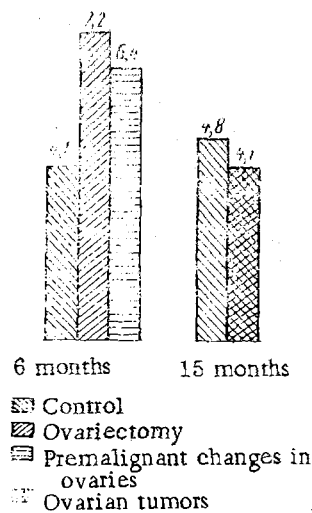
The results given in the table show that no ovarian tumors were present in the control rats. The incidence of tumors in the rats of group 2 depended on the dose of irradiation: their highest yield (64%) together with the shortest latent period (11 months) were observed after irradiation in a dose of 1200 R, when 32% of rats developed bilateral tumors. In the 3rd group, tumors appeared in 75% of rats following transplantation of the irradiated ovary into muscles and in 41% of rats following transplantation of the unirradiated ovary; the tumors first appeared in this group after 9.5 months. The incidence of tumors in the 4th group was 32% and their minimal latent period of development was 12 months.

In 79% of rats mixed tumors – thecaluteomas and granulosa cell-thealuteomas – developed. Granulosa-cell tumors, luteomas, and fibrosarcomas of the ovary developed less frequently. The various histological types of tumor structure were found in roughly the same frequency after the various procedures.

# Results of Experiments to Induce Ovarian Tumors in Rats

Group of animals	Dose (in R)	Total	Number of rats				Time of appearance of first tumor (in months)	Type of rats				
			with pemeco- plastic changes	with tumors		with bilateral tumors		granulosa cell	thecalureoma	granulosa cell- thecalureoma	luteoma	fibrosarcoma
				absolute	%							
1st	-	138	0	0	-	-	-	-	-	-	-	-
2nd	100	18	1	0	-	-	-	-	-	-	-	-
	200	50	2	0	-	-	-	-	-	-	-	-
	300	48	2	7	15	0	15.5	-	2	3	-	2
	600	22	3	8	36	2	12.5	5	4	1	-	-
	1000	30	8	16	53	5	11.5	1	10	6	2	2
	1200	28	3	18	64	6	11.0	2	11	10	1	-
3rd	600	16	3	12	75	-	9.5	2	3	6	1	-
	-	44	5	18	41	-	9.5	3	6	9	-	-
4th	-	41	5	9	22	-	12.0	1	4	4	-	-
Total	-	435	32	88	-	13	-	14	40	39	4	4

\* Some figures are taken from a previously published paper [2].



Changes in the gonadotropic activity of the hypophysis in rats during the formation and development of ovarian tumors.

Meanwhile, in the ovaries of 32 experimental rats of all three groups, foci of proliferation of the follicular epithelium and stroma (thecal cells) were observed and were regarded as preneoplastic changes. They were first noticed after 4.5 months in the animals irradiated in a dose of 1000 R.

The ovarian tumors which developed possessed estrogenic activity, as demonstrated by the presence of prolonged estrus in most of the animals with a tumor, and also by an increase in the weight of the uterus in castrated rats with a tumor arising from the ovary after transplantation into the muscles. The hormonal activity of the resulting tumors was also shown by their frequency of association (in 80% of cases) with the development of hyperplastic processes and tumors of the mammary gland, uterus, and hypophysis, characteristically found after long exposure to estrogens.

To determine whether the hormonal activity of the developing tumors was maintained during successive transplantations, 5 tumors developing after irradiation (all thecaluteomas) were transplanted into a total of 66 rats. A piece of tumor tissue was transplanted intramuscularly, beneath the kidney capsule, or subcutaneously into intact male and intact and spayed female animals. The transplantation was successful only in the spayed females (in 21 of 35 rats), in which from 2 to 5 passages were obtained. All the tumors studied retained their hormonal activity after transplantation, as shown by the increase in the weight of the uterus of the castrated rats with the transplanted tumor.

The observations showed that after various procedures known to cause a temporary depression of ovarian function, in many rats morphologically identical tumors develop, possessing estrogenic activity and maintaining it through serial passages. Consequently, the hypothesis that a depression of ovarian function is an important factor in the creation of the necessary conditions for development of experimental ovarian tumors was confirmed.

To study the changes in the gonadotropic function of the hypophysis in rats during the formation of ovarian tumors, investigations were made of the hypophyses of 10 rats of the 2nd group in which the presence of preneoplastic changes in the ovaries was demonstrated histologically, and of the hypophyses of 5 rats with fully developed tumors (6 and 15 months after irradiation in a dose of 1000 r). The hypophyses of 12 intact female rats of the same age, and of 5 young, spayed rats (2 months after ovariectomy), acted as controls.

The activity of the hypophyses was tested biologically in infantile female mice weighing 7-9 g. Daily for 3 days the mice received  $\frac{1}{3}$  of a hypophysis, in the form of a suspension in physiological saline, by subcutaneous injection and they were sacrificed 96 h after the beginning of the experiment. The results were evaluated by the use of an index representing the ratio between the weight of the uterus together with the ovaries, multiplied by 1000, and the body weight of the mouse.

The investigations showed that the gonadotropic activity of the hypophysis of the rats (see figure) with preneoplastic changes in the ovaries was considerably higher than normal (indices 6.4 and 4.1 respectively), and approached the activity of the hypophysis of castrated animals (index 7.2). In the rats with fully developed tumors the activity of the hypophysis was lower than in the control intact females (indices 4.1 and 4.8 respectively). Hence it was found that during the development of ovarian tumors the gonadotropic activity of the hypophysis of the rats changed, being increased before the tumors appeared and lowered after they were fully formed.

#### SUMMARY

Estrogen-active tumors of the ovaries, differing in histological structure, were obtained in rats as a result of depression of ovarian function by X-irradiation, by transplantation of the ovary into the muscles of spayed female, or by subtotal ovariectomy. In rats with pre-tumor changes there was a rise in the gonadotropic activity of the hypophysis, while animals with developed tumors presented a reduction of hypophyseal activity. Induced tumors were successfully transplanted to spayed females with retention of estrogenic activity following transplantation.

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