Intrasplenic Ovarian Grafts in Syrian Hamsters and Peritoneal Fluid Cellular Distribution (34823)

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The influence of ovarian neoplasms on peritoneal fluid cellular distribution is of interest because of the need for methods to detect developing ovarian neoplasms in women. We have previously studied the effect of radiation-induced and spontaneous ovarian tumors on cellular patterns of peritoneal fluid (1, 2). Further information as to the possible hormonal factors involved in ovarian tumor development and their effect on peritoneal fluid cellular distribution may be obtained by studying Syrian hamsters with intrasplenic ovarian grafts. Ovaries transplanted into the spleens of gonadectomized rats, mice, or rabbits transformed into neoplasms (3–5); however, tumors did not occur in hypophysectomized or estrogen-treated animals (3, 6–8). The estrogen secreted by the intrasplenic ovary becomes inactivated by the liver (9–14) so that the lack of estrogen feed-back action on the pituitary–hypothalamus produces an increased secretion of gonadotropin. This increased secretion causes tumor development in splenic-transplanted ovaries. Spontaneous ovarian tumors rarely form in Syrian hamsters (15). In spayed hamsters, follicular growth and luteinization were inhibited in ovaries transplanted to spleens over a 6-week period. Thus, the excess endogenous gonadotropin appeared incapable of stimulating hamster follicular growth (16). Since our previous experience indicated that the peritoneal fluid cellular pattern of hamsters was comparable to that in women (17), we decided to determine if a splenic ovarian tumor could be developed in a Syrian hamster over a long period of time and to determine its effect, if any, on the distribution of peritoneal fluid cells.

Materials and Methods. We ovariectomized adult female (golden) Syrian hamsters, 14 weeks old; 6–7 animals/group; Lakeview Colony, Newfield, New Jersey and transplanted the right ovary into the spleen (18). Normal control animals were included in the study as were sham-operated animals. At 48 weeks, we aspirated abdominal fluid from each animal by a 27-gauge needle through the ventral surface of the hamster, placed the aspirated cytologic specimen on an albumin-coated slide, and fixed and stained it according to Papanicolaou's technique (19). Two hundred consecutive cells were counted and grouped as mesothelial cells, lymphocytes, polymorphonuclear leukocytes, histiocytes, mast cells, bare nuclei, and daisy cells. Bare nuclei are cells without cytoplasm and we called a cell a daisy cell when the nucleus bulged in a pattern resembling a daisy.

The animals were killed by cervical dislocation, and each animal was weighed as well as the uteri and the right ovaries of the control, sham-operated animals, and intrasplenic ovarian grafts. All tissues were fixed in 10% buffered formalin and stained with hematoxylin and eosin.

We calculated the standard error for the mean cellular counts as well as for each mean ovarian and uterine weight for all animals by using the formula, $SE \sqrt{\Sigma d^2/N(N-1)}$. Student's t test was used to obtain probability values (p) for significant differences between means. By dividing the average cellular count by 2, the percentage distribution of each individual mean count was obtained.

Results. The mean weight of the 48-week-old intrasplenic ovaries was five times heavier ($p<.01$) than corresponding ovaries of ani-

TABLE I. Peritoneal Fluid Cellular Pattern of Ovariectomized Adult Female Syrian Hamsters Containing 48-Week-Old Ovarian Splenic Transplants.

	Control	Sham-operated	Ovarian splenic transplants
No. of animals	7	6	6
Body weight (g)	146.0 ± 7.5	137.4 ± 11.6	149.5 ± 7.3
Right ovarian weight (mg/100 g)	15.9 ± 4.7	15.9 ± 1.9	85.4 ± 17.4
Uterine weight (mg/100 g)	325.6 ± 41.4	399.9 ± 51.6	303.0 ± 46.6
Cell type		% Distribution of cells	
Mesothelial cells	46.9 ± 1.4[a]	55.1 ± 2.5	61.9 ± 2.8
Lymphocytes	17.8 ± 1.7	17.1 ± 2.6	18.3 ± 2.9
Polymorphonuclear leukocytes	6.9 ± 1.0	3.7 ± 4.3	3.9 ± 0.6
Histiocytes	2.4 ± 0.3	2.6 ± 0.5	2.4 ± 0.6
Mast cells	8.1 ± 2.6	7.5 ± 2.6	3.9 ± 1.5
Bare nuclei	18.0 ± 3.7	14.1 ± 3.7	9.0 ± 3.6
Daisy cells	0.0 ± —	0.0 ± —	0.3 ± —

[a] Standard error.

mals that were sham-operated or nonoperated hamsters (Table I). The ovaries placed in the spleens of hamsters were all grossly and histologically cystic (Fig. 1). All ovaries were approximately 1 × 1 cm, with multiple thin-walled cysts containing clear to straw-colored fluid. The lining of the benign cysts was made up of single to several layers depending on the amount of intracystic distention of cuboidal or columnar epithelium. The ovarian stroma was compressed between the cysts. The uterine weight of the animals with ovarian splenic transplants was usually less than control and sham-operated ($p > .1$) ani-

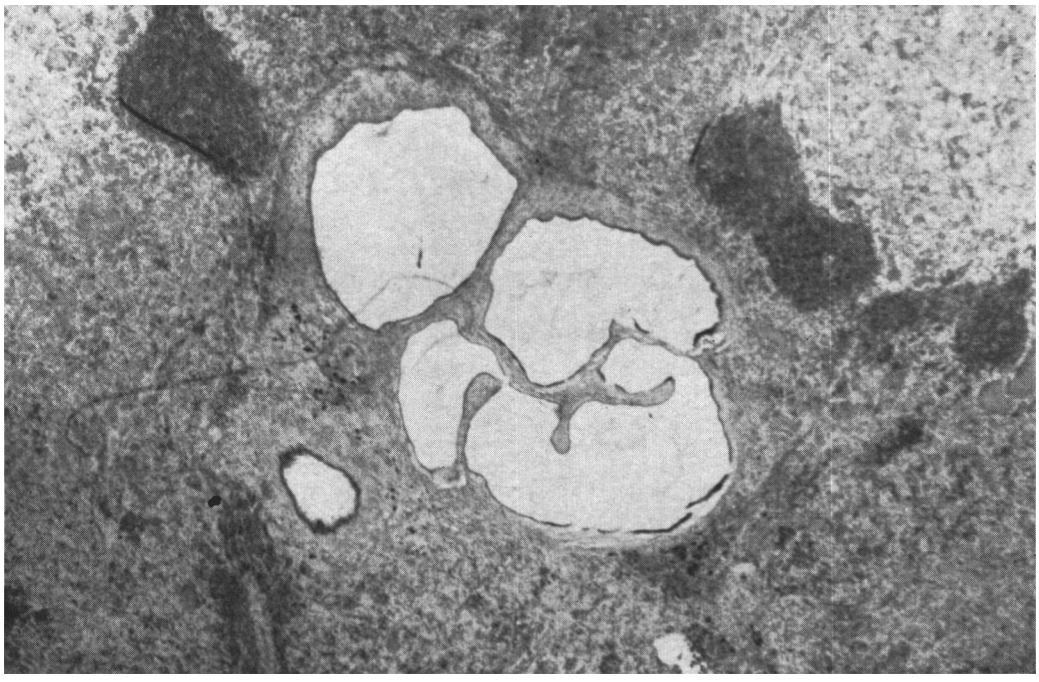

FIG. 1. A 48-week intrasplenic ovarian graft in a Syrian hamster showing epithelial-lined cysts (×40).

mals and did not reflect excessive estrogenic secretion which could not be metabolized by the liver. The percentage distribution of mesothelial cells in the peritoneal fluid of animals with ovarian cysts was higher than that in control and sham-operated animals. The percentage distribution of polymorphonuclear leukocytes, mast cells, and bare nuclei appeared to be consistently less in the peritoneal fluid of animals with intrasplenic cystic ovaries than control or sham-operated animals. Daisy cells were seen only in aspirates from animals with intransplenic grafts.

Discussion. We can assume that the anterior pituitary, free from ovarian inhibition, was capable of stimulating the growth of intrasplenic hamster ovaries. They contained cystic ovaries and were significantly heavier than ovaries of control and sham-operated animals. The ovariectomized adult female Syrian hamsters most likely exhibited excessive gonadotropin blood levels with associated low amounts of estrogen. Our previous work has suggested that, in the presence of elevated estrogen levels, relative mesothelial cellular counts are low whereas polymorphonuclear leukocytes are high (20–29). Possibly, the altered intrasplenic ovarian hemodynamics contributes to ovarian cyst formation. Peritoneal fluid cellular patterns of ovariectomized adult female Syrian hamsters containing 48-week-old ovarian splenic transplants appears to be influenced by the associated hormonal changes and the resulting cystic ovarian tumors.

Summary. We ovariectomized adult female Syrian hamsters and transplanted the right ovaries into the spleens where they remained for 48 weeks. The resulting excessive gonadotropin appeared to be the major stimulating factor in the production of hamster cystic ovaries. The uterine weight and characteristic distribution of peritoneal fluid cells reflected low circulatory estrogen levels.

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