may occasionally occur even when 200 μg of testosterone per day are absorbed[2] they do not usually occur with an absorption of only 40 μg per day (see Table I). The difference in results with Δ^4-androstenedione and testosterone were not due to differences in their masculinizing faculty. Though the capon unit of androstenedione is about 8 times that of testosterone[8] the quantities of androstenedione used in the present work were so considerable that the typical penis-like organ[9] was present in all animals. The corpus cavernosum reached in one case (with 500 μg per day) a length of 5 mm and the horny styles a length of 2 mm.

Sixty to 158 μg per day of cholestenone (M.P. = 78-80°) were absorbed. This is 10 times more than the antifibromatogenic threshold of progesterone. Abdominal fibroids were present in all 6 animals, and the average F.T.E. was not less than with estradiol alone.

As judged by the average F.T.E. androstenedione and cholestenone were not antifibromatogenic. Other criteria also supported this conclusion: the percentage of animals reaching the average F.T.E. of the estradiol group was slightly if at all influenced by androstenedione or cholestenone. The same is true concerning the average number of tumoral marks of classes 2 and 3 (tumors with a diameter of not less than 3 millimeters).

Uterine weight was smaller than with estradiol alone. Because of great variations we cannot say whether the difference is significant, but this may be an indication that with quantities still greater some antiestrogenic action might be obtained with the two substances.

Summary. Quantities of Δ^4-androstene-3-17-dione up to 40 times greater than the antifibromatogenic threshold of progesterone were not sufficient to prevent abdominal fibroids induced by alpha-estradiol. These quantities of androstenedione were also many times greater than the antifibromatogenic quantities of testosterone. The masculinizing action on the clitoris of the guinea pig was so pronounced as with testosterone. Quantities of cholestenone 10 times those of progesterone revealed no antifibromatogenic activity.

8 Goldberg, M. W., *Ergebn. d. Vitamin u. Hormonforsch.*, 1938, **1**, 37, 403, 405.

9 Lipschütz, A., *The Internal Secretions of the Sex Glands*, Cambridge (England), Baltimore, 1924, 301.

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Differential Behaviour of the Liver Towards Natural and Artificial Estrogens.*

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It has been known since the work of Biskind and associates that estrogens absorbed from intrasplenic pellets are inactivated in the liver.[1] The same has been stated concerning intrasplenic injections of estrogens.[2,3] These statements have been applied also to the study of abdominal fibroids induced by

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1 Biskind, G. R., and Mark, J., *Bull. Johns Hopkins Hosp.*, 1939, 212. Quoted from Biskind, G. R., and Meyer, M. A., *Endocrinol.*, 1941, **28**, 217.

2 Segaloff, A., and Nelson, W. O., PROC. SOC. EXP. BIOL. AND MED., 1941, **48**, 33.

3 Fels, E., *Medicina* (Buenos Aires), 1942, **2**.

estrogens in the female guinea pig. Quantities of estradiol or its esters which induce abdominal fibroids when absorbed from subcutaneously implanted tablets or pellets do not induce fibroids when absorbed from the spleen.[4] When the same quantities are absorbed from pellets implanted into the liver, large abdominal fibroids may be produced also, but their incidence diminishes greatly.[5] The difference between results with intrasplenically and intrahepatically implanted pellets has been tentatively explained by the assumption that with intrahepatic pellets the whole quantity absorbed was put into contact with but a small fraction of the liver and that the inactivating faculty of the liver was a *limited* one. It was thought worth while to study the problem of a quantitative limitation of the inactivating faculty of the liver by using different *artificial* estrogens, as diethylstilbestrol and hexestrol. These artificial estrogens are known to be relatively more potent by oral application than the natural ones; up to 20% or more of the quantity administered by injection may appear in the urine as against 2 to 3% with alpha-estradiol.[6] Following intravenous injection diethylstilbestrol behaves differently from estrone and alpha-estradiol also concerning excretion in bile.[7] When given to female guinea pigs by subcutaneous injection in oil the artificial estrogens were found to be more likely to produce abdominal fibroids than free alpha-estradiol or estrone.[8]

Experiments. Pellets of stilbestrol or of hexestrol[9] were implanted subcutaneously, intrasplenically, or intrahepatically into 152 castrated female guinea pigs.† Necropsy was performed 70 to 100 days later according to the group; the fibrous tumoral reaction was classified in units as in our former work[10] (see also the foregoing paper). The results with diethylstilbestrol in an experimental series of 70 days duration are given in Fig. 1. In a number of cases adhesions between the spleen and the abdominal wall were found. These adhesions consisted sometimes of thin and unconspicuous strands between the spleen and the wall, or between the epiploon and pancreas and the wall ("without adhesions," in the diagram), the epiploon being normally in contact with the under pole of the spleen. In other cases the strands between the spleen

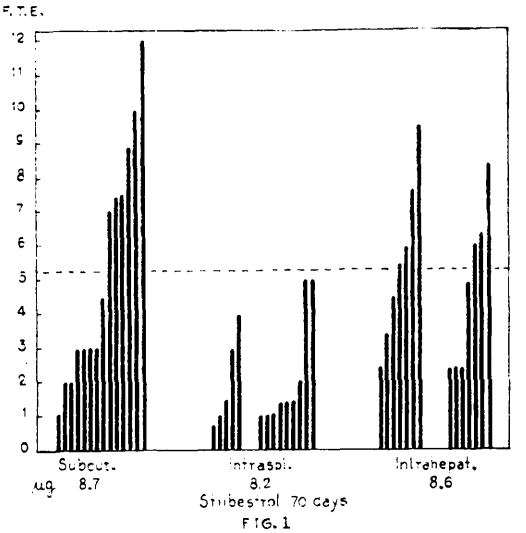

FIG. 1

Fibrous tumoral effect (F.T.E.) in 42 castrated female guinea pigs with subcutaneous, intrasplenic, or intrahepatic pellets of diethylstilbestrol, without adhesions (first group of columns) or with adhesions (second group of columns). Duration of the experiment—70 days. F.T.E. in units as explained in the foregoing paper. Horizontal line—average of the subcutaneous group. Note the enormous difference between the subcutaneous and intrasplenic groups; small or no difference between the subcutaneous and intrahepatic groups.

[4] Acuña, L., Tesis Universidad de Chile, 1942, *Public. Med. Exp. Chile,* No. 12; Lipschütz, A., and Acuña, L., *Rev. Med. Alim.* (Chile), 1943, **5**, 310.

[5] Carrasco, R., Tesis Universidad de Chile, 1943, *Public. Med. Exp., Chile,* No. 18; Lipschütz, A., and Carrasco, R., *Rev. Med. y Alim.* (Chile), to be published.

[6] Stroud, S. W., *J. Endocrinol.*, 1939, **1**, 201.

[7] Cantarow, A., Rakoff, A. E., Paschkis, K. E., Hansen, L. P., and Walking, A. A., *Endocrinology,* 1942, **31**, 515.

[8] Lipschütz, A., Vargas, L., Egaña, E., and Bruzzone, S., PROC. SOC. EXP. BIOL. AND MED., 1941, **48**, 467.

[9] Campbell, N. R., Dodds, E. C., and Lawson, W., *Nature,* 1938, **142**, 1121.

† The greater part of these experiments will be described in U. Quintana, Tesis Universidad de Chile, 1943 (*Public. Med. Exp.* (Chile), No. 22).

[10] Lipschütz, A., Bellolio, P., Chaume, J., and Vargas, L., PROC. SOC. EXP. BIOL. AND MED., 1941, **46**, 164.

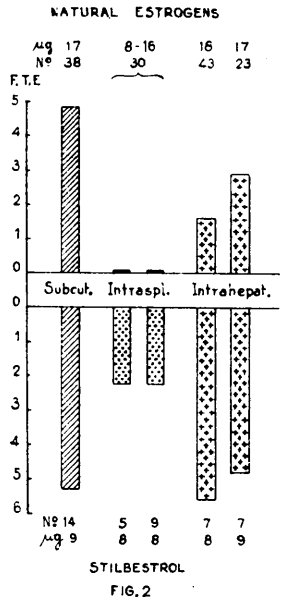

FIG. 2

Average fibrous tumoral effect of 42 animals with pellets of stilbestrol and 143 animals with pellets of natural estrogens as estrone, estradiol, and different esters.[4,5] Duration—70 days. The first intrasplenic and intrahepatic columns—without adhesions; the second intrasplenic and intrahepatic columns—with adhesions. See also the number of animals (No.) in each group and the average quantities absorbed per day. The greater resistance of the artificial estrogens against hepatic inactivation is evident.

and the abdominal wall were more extensive; in rare cases the spleen may adhere directly to the abdominal wall ("with adhesions" in the diagram). As seen in Fig. 1, there was some fibrous reaction also with intrasplenic pellets of stilbestrol. This was remarkable since a fibrous reaction was never observed with intrasplenically implanted alpha-estradiol and its esters.[4] The fibrous reaction was no less pronounced with intrahepatic pellets of stilbestrol than with subcutaneous pellets (Fig. 1) where as with intrahepatic pellets of natural estrogens the incidence of large fibroids was greatly diminished. Comparative results with natural and artificial estrogens are given in Fig. 2. Results with 19 animals with pellets of hexestrol with an absorption of 5 to 6 µg per day were similar to those with stilbestrol. These experiments afford evidence of the greater resistance of artificial estrogens to inactivation in the liver.

The evidence of greater resistance of artificial estrogens to inactivation in the liver was corroborated by the fact that other toxic actions of prolonged treatment with estrogens were present with intrasplenic pellets of stilbestrol and hexestrol such as genital bleeding, monstrous uterine growth, uterine polyps, great development of the nipples and of the mammary glands. Most of these toxic actions were absent with intrasplenic pellets of natural estrogens or they were present only exceptionally and transiently.

Summary. Experiments with intrasplenic and intrahepatic pellets of diethylstilbestrol and of hexestrol showed that these artificial estrogens are more resistant against hepatic inactivation than the natural ones. Abdominal fibroids could not be induced with intrasplenic pellets of natural estrogens, free or esterified; they were induced with intrasplenic pellets of diethylstilbestrol and of hexestrol though the incidence of tumors of a certain size was greatly diminished when compared with subcutaneously implanted pellets. Other toxic actions characteristic of prolonged treatment with estrogens also were present with intrasplenic pellets of artificial estrogens, whereas they were mostly absent or only transient with intrasplenic pellets of natural estrogens.