TOXIC ACTIONS OF OESTROGENS ON THE LIVER

BY JUDITH KULCSÁR-GERGELY AND ANDRÓS KULCSÁR

From the Institute of Pharmacology and Clinic I of Internal Medicine,† Medical University of Debrecen, Hungary*

Received October 19, 1959

The endogenous oestrogen level rises after hepatic damage. With its rise, the condition of the damaged liver deteriorates. This is supported by experiments in which mature ovariectomised animals were loaded with oestrogen. The introduction of oestrogen increases the strain on the damaged liver because of its reduced inactivating capacity. On comparing the results with clinical data in the literature, it is concluded that oestrogen therapy is contra-indicated when hepatic damage is present.

THE oestrogens may be grouped into three categories: (i) human, natural oestrogens, (ii) other natural oestrogens, (iii) synthetic oestrogens.

Oestrogens in man are steroids which have an important role in ensuring the periodicity of hypophyseal function, especially in developing and secreting trophic hormones produced by the basophil cells of the anterior lobe. Oestrogens have become more and more widely used in therapy, and such popular use calls for the elucidation of previously unknown interrelations of these hormones.

The liver has been considered as the principal site for oestrogen inactivation. Talbot[1] produced severe lesions in female rats with carbon tetrachloride and ethanol. At death the uteri of these animals were removed and weighed, and were found to be double that of the uteri of control, untreated animals. In contrast, there were no increases of weight in previously ovariectomised rats after carbon tetrachloride poisoning. This excludes a direct effect of this substance, and supports the view that the inactivation of endogenous oestrogen is a function of the liver.

Experimenting in dogs, Israel and others[2] investigated the effects of transfused oestrogen in heart-lung preparations. They found no oestrogen inactivation in the blood *in vitro*, nor in the heart-lung preparation, but it was rapid in a heart-lung-liver perfusion. Riegel[3] mixed homogenised rat-liver fractions with oestrogen and observed that the inactivating enzymic system is in the microsomatic fraction.

The part played by the liver in oestrogen inactivation as investigated *in vivo* in man was conflicting, especially when a liver lesion was involved. While testing for the oestrogen activity of human urine in rats, Beretervide[4] did not observe any decrease of oestrogen activity, whereas Enzinger[5] recorded a significant and regular rise in the quantity of oestrogens. In *in vitro* examinations of tissues of human liver by a colorimetric method, Tagnon[6] did not observe that oestradiol breakdown was affected by a basic lesion of the liver, even when structural changes pointed to hepatic damage.

* Director, T. Vályi Nagy, M.D. † Director, B. Fornet, M.D.

As is evident, there are not only details but fundamental problems to be solved about the liver-oestrogen relation. The purpose of our investigations was to examine the intricate function of inactivation and particularly to look for possible changes in the liver loaded with exogenous oestrogen.

Experimental and Results

In our experiments, liver lesions were produced by Kaufmann's[7] method, using carbon tetrachloride. Most of the deaths were found to occur after 3 to 6 doses of carbon tetrachloride (0·05 ml./100 g.). The lighter the animal, the sooner it was affected. On dissection, even after a few doses of carbon tetrachloride, macroscopic changes were visible in the liver, the surface of which was dotted with yellow globules, some as large as 3–5 mm. in diameter, and striped with yellow bands. Animals weighing

TABLE I

Development of the endogenous oestrogen level following liver lesion

Phase	Average duration in	
	treated animals	controls
Oestrus	32·00 hr.	32·40 hr.
Metoestrus	23·06 ,,	30·48 ,,
Dioestrus	28·04 ,,	29·52 ,,
1(*a*)	21·00 ,,	33·80 ,,
Prooestrus	10·00 ,,	23·08 ,,
2(*a*)	9·30 ,,	24·00 ,,
Total	123·40 hr. 5·14 days	173·28 hr. 7·22 days

180–220 g. gave better results, and the death rate disappeared when the treatment was maintained for a longer period but with smaller doses (0·03 ml./100 g.). Of the 89 test rats on the 0·05 ml./100 g. dosage, 12 normal and 8 ovariectomised ones died.

Our basic observations were on the modification of the oestrus cycle of the group with liver lesions compared with the untreated control group. These tests were made in 28 fully grown animals treated with carbon tetrachloride, and 10 controls. On comparing the cycle of treated and control animals, it can be seen (Table I) that in the treated animals this averaged 2·08 days less than the control value. This is a significant difference. It is striking that the shortening of the cycle occurred in phase 1(*a*), in prooestrus and the following 2(*a*) transitional phase, while oestrus and dioestrus remained unchanged.

In addition to the data in Table I, the following changes were observed. Under normal circumstances, epithelial plugs occur in the 2(*a*), oestrus and metoestrus phases only. After liver lesions epithelial plugs were also atypically found in the vaginal smear in 82 per cent of the examined cycles. It was also atypical in that the phases did not always follow each other regularly.

Consideration of the atypical and frequently appearing epithelial plugs and the fact that oestrus always sets in sooner in the group with liver

lesions than in the control group suggests the probability that the endogenous oestrogen level rose after liver lesions.

We then investigated how normal and ovariectomised animals with loaded livers reacted to the introduction of oestrogen. If mature animals are given oestrogen the oestrus cycle of the animals with normal livers is prolonged. The question then arises: how can a liver with decreased

TABLE II

EFFECT OF OESTROGEN LOADING OF MATURE ANIMALS AFTER LIVER LESION ON PROLONGATION OF OESTRUS

Treatment	Oestrone dose	Average duration of oestrus	Prolongation
CCl_4	4 U	67 hr.	39 hr.
Control	4 U	28 ,,	
CCl_4	8 U	132 ,,	95 ,,
Control	8 U	37 ,,	
CCl_4	12 U	194 ,,	138 ,,
Control	12 U	56 ,,	

function assimilate introduced oestrogen, and how will this loading influence liver function? To study this, 28 animals with liver lesions and 10 control animals were used. The animals received a single injection of oestrone acetate, 4, 8 or 12 U/100 g. (0·4, 0·8, 1·2 μg./100 g.), during dioestrus. Oestrus started 28 hours later in the group with liver lesions, and lasted 67 hours. In control animals with normal livers the oestrus appeared also after 28 hours, but lasted only 28 hours. The difference

TABLE III

EFFECT OF OESTROGEN LOADING OF OVARIECTOMISED ANIMALS AFTER LIVER LESION ON PROLONGATION OF OESTRUS

Treatment	Oestrone dose	Average duration of Oestrus	Prolongation
CCl_4	1 U	not measurable	
Control	1 U		
CCl_4	2 U	24 hr.	18 hr.
Control	2 U	6 ,,	
CCl_4	4 U	48 ,,	21·5 ,,
Control	4 U	26·5 hr.	
CCl_4	8 U	94 hr.	52 ,,
Control	8 U	42 ,,	

between the two groups therefore was 39 hours. In fact, 15 animals in the group with liver lesion attained the criterion of "lasting oestrus," a reaction of 72 hours. None of the control animals did this.

With 8 U/100 g. of oestrone, the reaction in both groups occurred in 28 hours; in the group with liver lesions it lasted 132 hours, and in the control group 37 hours, a difference of 95 hours. This difference is even more striking if we consider that in the controls the double dose of oestrogen prolonged the oestrus by 9 hours, while in the treated animals it was prolonged seven times as much. This suggests that not only the rise in

the dose of oestrogen but some other influence of the hormone had played a part.

Using 12 U of oestrone, oestrus in the group with liver lesions began 30 hours later, and lasted 194 hours. In the control group the oestrus also began 30 hours later, but lasted only 56 hours. Thus the oestrus cycle effected by a triple dose of oestrogen was nearly four times as long as the control group. The data are presented in Table II.

Other tests were made in 31 ovariectomised rats, of which 21 were given 1, 2, 4 and 8 U of oestrone acetate per 100 g. and 10 were used as controls. 1 U of oestrogen per 100 g. produced no measurable results, 2 U brought about a 24 hour reaction in the group with liver lesions, while in the control group this lasted 6 hours. With 4 U the duration of oestrus in the group with liver lesion was 48 hours, while in the control group it was 26·5 hours, and 8 U/100 g. produced a reaction lasting 94 hours in the loaded animals and 42 hours in the controls.

Discussion

From our results we may claim that the role of the liver in oestrogen metabolism and inactivation is established. This process is bound up in an intricate enzyme system and closely connected with bile excretion. The latter phenomenon has also been demonstrated by Heard's[8] tests with radioactive iodo-oestradiol. It should be remembered that the liver as well as deactivating substances also activates, in so far as it reduces the oestrone to the more effective oestradiol-17β. We found that the damaged liver quickly lost its ability to inactivate oestrogen. Presumably, therefore, any kind of interference preventing oestrogen inactivation may result also in the increased lesion of the liver. We observed that the capacity of the damaged liver to inactivate oestrogen decreased disproportionately with dosage of oestrogen. This was also shown by the difference between the oestrus cycles of control and loaded animals. The evaluation of our tests in ovariectomised animals supports the other experiments, and demonstrates the harmful effect of endogenous oestrogen in any case of liver lesion.

Ovariectomised animals react to an equal loading of oestrogen by a shorter oestrus—even after liver lesion—than do normal animals with loaded livers. Thus by ovariectomy the liver is protected.

In groups possessing active ovaries, the oestrus is not longer when compared with the groups of ovariectomised animals (67:48 hours, or 132:94 hours), because the amount of non-inactivated endogenous oestrogen is additive with the introduced oestrogen, as it is well known that on the introduction of oestrogen the production of endogenous oestrogen will cease. The difference between the two arises from the fact that endogenous oestrogen will further damage a liver poisoned with carbon tetrachloride at least in its function of inactivating oestrogen.

Our animal tests are in substantial agreement with what we know of human pathology, as in Long's[9] observation in a patient in whom *coma hepaticum* was induced by oestrogen administration. The patient had jaundice in childhood, and received treatment for primary amenorrhoea.

Autopsy revealed cirrhosis of the liver. In his paper this author reported on 29 patients suffering from cyclic disturbances that were relieved by liver therapy.

References

1. Talbot, *Endocrinol.*, 1939, **25**, 601.
2. Israel, Meranze and Johnston, *Amer. J. med. Sci.*, 1937, **194**, 835.
3. Riegel and Meyer, *Proc. Soc. exp. Biol., N.Y.*, 1952, **80**, 617.
4. Beretervide, Lazarri and Kravetez de Srulijes, *Prensa med. Argent.*, 1956, **24**. Ref. *Kongresszentralbl. f. ges. inn. Med.*, 1956, **172**.
5. Enzinger, *Wien. Zschr. inn. Med.*, 1955, **36**, 482.
6. Tagnon, Lieberman and Schulman, *J. clin. Invest.*, 1952, **31**, 346.
7. Kaufmann, *Beitr. Pathol. Anat.*, 1953, **113**, 253.
8. Heard, *Rec. progr. Hormone Res.*, 1949, **4**, 25.
9. Long and Simmons, *Arch. int. Med.*, 1951, **88**, 762.