

Figure 1. Competition of steroids with  ${}^3\text{H-estradiol-17}\beta$  for binding sites of SHBP from plasma of female dogfish. (T) testosterone, (DHT) dihydrotestosterone-5 , (P) progesterone, (E<sub>2</sub>) estradiol-17 $\beta$ , (E<sub>1</sub>) estrone, (E<sub>3</sub>) estriol, (DES) diethylstilbestrol, [S] concentration of unlabeled steroids in M. Specific binding of  ${}^3\text{H-estradiol}$  in the absence of unlabeled steroid was set at 100%, and the binding of  ${}^3\text{H-E}_2$  in the presence of other unlabeled steroids was expressed as percentage of maximum.

capacity (M) of embryonic plasma for <sup>3</sup>H-estradiol was only half that found in mature fish (Table 1), and a sexual difference was apparent; female plasma having a higher binding capacity.

In summary, a high affinity, limited capacity sex-hormone binding protein (SHBP) has been demonstrated in the plasma of mature and embryonic dogfish (Squalus acanthias). This SHBP clearly differs from amphibian and mammalian SHBPs in its steroid specificity as it binds C<sub>21</sub> steroid (progesterone and corticosterone) as well as C<sub>18</sub> and C<sub>19</sub> sex-steroids. The binding affinities for both testosterone and estradiol were ten times less than that of mammalian SHBPs. However, the binding capacities for both steroids were about 100 times higher than those of mammals.

Although the physiological role of high affinity plasma binders for steroid hormones is still speculative, it is likely that they regulate steroid concentration available for receptor binding at the target organs. Supported by NSF grant #PCM 78-08201 to IPC.

## INDUCTION OF VITELLOGENIN SYNTHESIS IN THE SPINY DOGFISH, SQUALUS ACANTHIAS

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In non-mammalian vertebrates, estrogens cause hepatic synthesis of a yolk-protein precursor, vitellogenic females. Injection of estrogens into males or non-vitellogenic females readily induces the production of this protein in teleosts, amphibians, reptiles and birds. In the oviparous elasmobranch Scyliorhinus canicula (a continuous breeder), although vitellogenin production continues throughout the year, it is more pronounced from September to February (Craik, Comp. Biochem. Physiol. 60B, 9–18, 1978). We have studied the spiny dogfish, Squalus acanthias, a yolk-sac dependent, aplacental, ovoviviparous species, with a gestation period of 20–22 months (Hisaw and Albert, Biol. Bull. 92, 187–199, 1947). The effect of steroid injections in females at either Stage A (early pregnancy, embryo < 17 mm) of Stage C (late pregnancy; embryos between 17–24 cm) and males were studied as follows:

TABLE 1

	Control (sesame oil)	Estradiol 17β*	Estradiol 17β* + progesterone
Stage A female	2 <sup>a</sup>	5	2
Stage B female	2	5	2
Males	2	2	2

<sup>\*</sup>Hormone dosage: estradiol- $17\beta = 3 \times 1.0$  mg, progesterone =  $3 \times 2.0$  mg. Hormone was administered at days 0, 2, and 4 and plasma samples were taken at day 0, day 3, day 5, day 7, day 14.

adenotes the number of animals in the group.

The appearance of a vitellogenin band was monitored by sodium-dodecyl-sulfate gel electrophorens (7-15%). Vitellogenin was not detectable in 25-50 ul samples of male and early pregnancy female plasma but low levels were detectable in late pregnancy female plasma. Treatment with estradiol- $17\beta$  alone or with estradiol  $17\beta$  + progesterone failed to induce detectable vitellogenin synthesis in males and early pregnancy females. However, injection of estradiol- $17\beta$  readily induced vitellogenin production in late pregnancy females (Fig. 1) and plasma levels continued

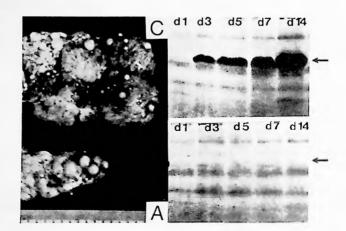


Figure 1. Portion of the ovary and gel electrophoretic pattern of plasma from different reproductive stages of the spiny dogfish Squalus acanthias. The left upper panel shows a portion of the ovary from a Stage C pregnancy fish with growing follicles and left lower from a Stage A pregnancy fish with small follicles. The right panel shows the electrophoretic pattern of the plasma from animals run on 0.1% sodium dodecyl sulphate, 7-15% polyacrylamide gels. Fish were injected with  $3 \times 1$  mg of estradiol- $17\beta$  in sesame oil and estrogen-induced vitellogenesis was observed only in Stage C animal (above) and not in Stage A animal (below).

to increase reaching maximum 15 days after the first injection. Concomittant injection of progesterone with estradiol-17ß, appeared to reduce the estrogen-induced response. Since early pregnant <u>S. acanthias</u> appear to possess active corpora lutea (Lance and Callard, Gen. Comp. Endocrinol. 13, 255-267, 1979) and progesterone has been demonstrated in elasmobranch plasma, the insensitivity of early pregnancy females to estradiol may be due to the presence of this steroid in the circulation. Further investigations of peripheral plasma progesterone concentration at various stages of pregnancy are required to support this suggestion. Supported by NSF grant #PCM 78-08201. \*Recipient of an NSF Undergraduate Research Participation Award.

## ESTROGEN SYNTHESIS IN DOGFISH TESTIS

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Although the ovary and placenta are generally considered to be the major sources of circulating estrogen, the testis can also synthesize large amounts of estrogen (Engel, Handbook of Physiology, Section 7, Vol. 2, Pt. 1, p. 467, 1974). Androgens, testosterone (T) and androstenedione (A), are the immediate precursors of estradiol-17 $\beta$  (E<sub>2</sub>) and estrone (E<sub>1</sub>), respectively, and, in all estrogen-synthesizing tissues, the reaction is governed by a P<sub>450</sub> enzyme complex termed aromatase that is found predominantly in microsomal subfractions. Both Leydig (interstitial) and Sertoli (tubular) cells have been implicated in testicular estrogen synthesis (Canick et al, Endocrinology 104, 285, 1979). The functional significance of aromatization in the testis has yet to be demonstrated, but it has been suggested that estrogen formed in situ governs testicular androgen production by a local feedback mechanism or regulates germ cell proliferation and maturation.