Maintenance of Pregnancy in Absence of Dietary Protein with Progesterone[1] (34730)

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Nelson and Evans (1) demonstrated that pregnant protein-deficient rats injected daily with 6 mg of progesterone from days 3 to 12 of gestation had live embryos on day 13 in 100% of cases; however, if injections were continued through day 20 and animals were sacrificed on day 21 only 60% had litters. Their explanation that reduction in maintenance at day 21 was "due to a progressive decrease in circulating estrin" is examined in light of the possibility that extension of hormone injections into the second half of pregnancy may have had deleterious effects. More recent work (2) has shown that administration of ovarian hormones may be discontinued as soon as the placenta has formed, *i.e.*, at midgestation.

Materials and Methods. Virgin Long-Evans rats, 77–87 days old and weighing 165–230 g, were bred with normal males and fed protein-free diet[2] *ad libitum* beginning the day of finding sperm in vaginal smear (day zero). Animals were kept in individual cages, on screens, until autopsy on day 21 of pregnancy. Injections of 6 mg of progesterone, or of 1 μg of estrone and 4 mg of progesterone, in 0.3 ml of sesame oil were given once daily from days 3 through 10 of pregnancy. Control rats received the vehicle only. Vaginal smears were examined daily. At autopsy uteri were removed and examined for implantation sites; living young and placentas, when present, were weighed.

Results. Table I summarizes the data on reproductive performance in protein-deficient animals injected from days 3 through 10 of gestation. More than 80% of rats injected with progesterone alone maintained pregnancy. There were no significant differences between animals injected daily with 6 mg of progesterone alone, and animals injected with both estrone and progesterone, with respect to weight loss during gestation, percentage of rats with living litters, average number of living young per litter, or average fetal or placental weight. Each of the above parameters of reproductive performance (except placental weight) was improved significantly in the progesterone-treated group compared with that of the sesame oil-injected (control) group.

Discussion. These data indicate that progesterone alone, when injected until day 10 of gestation, was almost as effective in the maintenance of pregnancy in protein-deficient rats as a combination of estrone and progesterone. Nelson and Evans (1) on the other hand found that at each level of progesterone injected, from 2 to 8 mg daily, incidence of reproductive failures was greater in animals injected through day 20 and autopsied on day 21 than in rats treated until day 12 and sacrificed the following day.

It is evident from the present data that pregnancy can be maintained to term in a high percentage of protein-deficient rats when injections of progesterone are terminated at

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[2] The protein-free diet was complete in all known essential dietary nutrients except protein. Fat-soluble vitamins were supplied weekly. Composition of the protein-free diet and of the fat-soluble vitamin supplement was reported previously (5).

TABLE I. Maintenance of Pregnancy with Ovarian Hormones Injected from Days 3 to 10 in Protein-Deficient Rats.

Daily dosage (mg)	Rats bred (no.)	Wt change during gestation (g)	Onset vaginal RBC (day)	Living litters (%)	Implantation sites/rat		Av wt	
					Total (no.)	Living (no.)	Fetus (g)	Placenta (mg)
4 P + .001 E[a]	19	−28 ± 2[b]	12.8 ± 0.2	100	9.0 ± 0.4	7.9 ± 0.5	3.4 ± 0.1	389 ± 12
6 P	16	−30 ± 4	13.4 ± 0.2	81	9.4 ± 0.5	6.5 ± 0.6	3.2 ± 0.1	348 ± 35
Sesame oil	6	−54 ± 6	8.5 ± 0.3	17	9.0[c]	1.0	2.1	323

[a] E = estrone; P = progesterone.
[b] Mean ± standard error.
[c] In five animals implantation sites were not visible at autopsy.

midgestation. Reduced reproductive performance in animals injected until day 20 of gestation thus was not due to a "decrease in circulating estrin" or to "an increase in the estrin requirement" during the latter half of pregnancy as Nelson and Evans suggested (1). It appears more likely that in their experiments exogenous progesterone upset hormonal balance from days 11 through 20. On the other hand, in animals of unspecified age, weight, and strain, Leathem (3) maintained pregnancy in 90% of rats injected with 4 mg of progesterone daily through day 20.

The effect of protein deficiency may be contrasted with that of ovariectomy. When rats were ovariectomized on day 7 the daily injection of 5 mg of progesterone resulted in only 44% maintenance of pregnancy (4); however, the injection of 5 mg of progesterone plus 1 μg of estrone resulted in 83% maintenance. This same degree of success was obtained in protein-deficient rats with progesterone alone; thus, there was a deficiency of circulating estrogen in ovariectomized rats but not in protein-deficient rats.

Summary. Pregnancy was maintained to term in protein-deficient rats when 6 mg of progesterone were injected daily from days 3 to 10. It is suggested that adequate amounts of circulating estrogen were present during the first half of pregnancy and that placental luteotropin stimulated adequate production of both estrogen and progestin during the latter half of gestation.

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