Plasma amino acids, though unchanged by fasting, were decreased by hormones, glucagon being more effective than epinephrine. Because liver glycogen levels were lower, it is not possible to ascertain whether this was a direct hormonal effect or the result of gluconeogenesis to replace liver glycogen stores. It seems unlikely that the latter is the case, in view of the fact that lowering of liver glycogen by fasting was not accompanied by a decrease in plasma amino acids.

It has been suggested by Foa and Galansino(1) that the main effect of glucagon is the transfer of glucose from the liver to the periphery, whereas the net effect of epinephrine is the reverse. Although this may be true in the untreated animal and appears to occur shortly after glucagon and epinephrine administration(11), it does not appear to be the principal effect of chronic administration. In our experiments both hormones depleted liver glycogen stores without significant increase in muscle glycogen or blood glucose. The hormones did not exhibit synergism or antagonism. Carbohydrate parameters were altered more by fasting than by hormone administration. Blood amino acid levels were, however, unchanged with fasting but lowered by glucagon and epinephrine.

Summary. Changes in body weight and in metabolic parameters were studied during chronic (twice daily) administration of glucagon 0.93 mg and/or epinephrine 0.1 mg to adult male rats. During the 5-day experimental period, fed control and epinephrine-treated animals lost no weight. Glucagon and glucagon-epinephrine animals lost 10-15% of their initial body weight. Fasted animals, control and hormone injected, lost approximately 20% of their initial body weight during the 5-day period. The weight loss by hormone-treated animals was not significantly greater than for controls. Liver glycogen was lowered by fasting and hormone administration. Muscle glycogen and blood glucose levels were lowered by fasting only. Plasma amino acid levels were not altered by fasting but were lower in hormone-treated animals. Changes in carbohydrate parameters usually associated with acute administration of glucagon and epinephrine, *i.e.*, hyperglycemia and glycogen shift from/to liver, to/from muscle, were not observed in adult rats during chronic administration of these hormones.

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Luteotropic Response in Pituitary-Ovarian Isografts in Male Mice.* (30420)

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Intraocular ovarian isografts in intact male mice develop mature follicles but show only sporadic luteinization. Administration of a single dose of bovine LH gives a quantitative luteal response(1,2). Similar grafts in ovariectomized females show cyclic formation of mature and luteinized follicles(3). The cor-

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TABLE I. Response in Terms of Formation of Corpora Lutea and of Development of Luteal Hyperemia in Ovarian Grafts After One or More Doses of LH.
(I: C3H; II, III, IV: Strong A)

Group	Grafts R/L	LH (5 μg)	No. grafts	% Grafts with: Corpora lutea	% Grafts with: Luteal hyperemia	Mean duration of luteal hyperemia
I	OP/O	1	26	84	84	6.6 ± 1.06
	" "	10	18	90	90	12.1 ± 2.00
II	OP/OP	1	21	71	71	6.1 ± .61
	" "	3	20	75	75	6.1 ± .44
	" "	10	18	86	86	8.7 ± .53
III	OP/O	3	19	89	79	5.1 ± .53
	OP/OP	"	18	83	83	7.5 ± .47
IV a	OP/OP	"	14	81	81	8.8 ± 1.07
b	O/O 4SC	"	11	91	91	10.0 ± .91
c	O/O 2SC	"	16	100	100	5.7 ± .36
d	OP/OP*	"	16	87	79	8.0 ± .98
e	O/O 4SC*	"	13	85	85	8.5 ± .49
f	O/O 2SC*	"	16	88	88	5.3 ± .45

* Pituitary grafts from male donors.

pora lutea become functional in the presence of pituitary grafts(4); duration of function depends upon the amount of pituitary grafted (5). Corpora are also functional during administration of ovine LTH; duration of function is influenced by concomitant administration of LH(6). The present report concerns the interaction of ovarian and pituitary grafts in male mice, including the effects of single or multiple doses of LH, contiguity of the two tissues, amount of pituitary grafts, and the sex of the donor.

Material and methods. Groups of 8 to 16 intact male mice received bilateral intraocular grafts of ⅛th of an ovary. In addition, they also received either ½ of an anterior pituitary in the right anterior chamber (OP/O), ½ in both anterior chambers (OP/OP), 2 placed subcutaneously (O/O 2SC), or 4 placed similarly (O/O 4SC). Host mice were 2 to 4 months of age and of the Strong A strain, unless otherwise stated. Donor mice were isologous and 3 to 6 months of age. Pituitary grafts, unless otherwise specified, were from female donors.

After 3 to 4 weeks, all animals in specific groups received one, 3, or 10 daily doses of 5 μg of LH-NIH-B2† (IP, in saline). Daily examinations of grafts commenced one to 5 days before hormonal administration and continued for as long as luteal function, indicated by hyperemia of corpora(4,6), continued in the grafts. Grafts without mature follicles at the time of LH administration were disregarded. Animals in other groups were untreated and their grafts examined daily for 10 to 30 days.

Results. Untreated. In C3H mice (OP/O), 6 of 48 grafts showed a single luteinization over a mean period of 16 days. Two grafts had non-hyperemic corpora; 4 showed hyperemic corpora for 2, 2, 3, and 4 days, respectively. In Strong A mice (OP/OP), 2 of 16 grafts exhibited a single luteinization over a 10-day period; one had non-hyperemic and the other hyperemic corpora for 4 days.

Single and multiple doses of LH. The percentage of grafts that formed corpora lutea was not materially affected by more than one dose of LH (Table I). Such corpora developed within 2 to 5 days after LH administration in 99% of responding grafts; all, or almost all, developed hyperemia immediately upon formation. In animals receiving one or 3 doses, only a single crop appeared but, in those on 10 doses, second, or even third, sets of such corpora appeared. However, the initial crop lost hyperemia one to 3 days after the appearance of the second (or the second after the third). Duration of hyperemia always refers to the initial crop.

In Group I (C3H mice), the mean duration of luteal hyperemia was approximately

† LH-NIH-B2, generously supplied by the Endocrinology Study Section, Nat. Inst. Health.

twice as long with 10 as with one dose of LH ($p<0.05$). In Group II, duration was not increased by 3 *vs* one dose but was longer by 2.6 days with 10 doses ($p<0.01$ for 10/1 doses; <0.001 for 10/3 doses).

Contiguity of ovarian-pituitary grafts. While the mean duration of luteal hyperemia in Group I, on one dose of LH, was 6.6 days for all grafts, it was 9.5 ± 2.21 in the right, and 4.5 ± 0.49 in the left grafts ($p<0.05$). On 10 doses, overall duration was 12.1 days but 19.3 ± 2.35 in the right, and 6.3 ± 1.41 in the left grafts ($p<0.001$). The difference in means for the right grafts on one or 10 doses was significant ($p<0.01$), but was not for the left grafts.

In Group III, the overall mean duration of hyperemia was longer in OP/OP than OP/O mice (7.5 and 5.1, respectively; $p<0.01$). However, in OP/O animals it was 6.1 ± 0.66 in the right, and 3.9 ± 0.63 in the left grafts, respectively ($p<0.05$). The means for right and left grafts did not differ in the OP/OP mice (nor in Group IV, a-f).

Amount and sex of pituitary graft. In Group IV, the means for animals with the same amount of pituitary (a/d; b/e/ c/f) from female or male donors did not differ statistically. On the other hand, regardless of pituitary sex, the means for O/O 2SC were shorter than those for OP/OP ($p< 0.05$) or O/O 4SC ($p<0.001$); those between OP/OP and O/O 4SC did not differ.

Discussion. Ovarian isografts in male mice (O/O) show from 14 to 83% luteinization in response to 0.2 to 0.8 μg of LH, given IV (2); the absence of any more than sporadic luteinization in untreated OP/O or OP/OP mice (as in untreated O/O mice) suggests that pituitary isografts secrete no LH. In O/O mice, hyperemic corpora appeared in less than a third of responsive grafts, and never lasted more than one day after LH administration. Consequently, the development of functional (hyperemic) corpora in OP/O or OP/OP mice, after LH dosage, and their persistence for similar periods to those found in untreated OP/O females(4), indicates that pituitary grafts secrete LTH as well in males as in females. Further, grafts from males or females were equally effective. Such grafts from either sex can induce mammary tumorigenesis in male mice(7) and those from males have been considered more effective than those from females in the same process in female hosts(8).

The limited span of luteal function, and its difference in pituitary-contiguous and non-contiguous ovarian grafts, resembled that found in females. For instance, in OP/O BALB/c females, the mean duration was 6.1 in the right, and 4.6 days in the left, grafts (5); here, in similar Strong A males, after 3 doses of LH, it was 6.1 and 3.9 days, respectively. Further, a half pituitary, placed intraocularly, was almost as effective as 2 placed subcutaneously, in females(5); here, in males 2 intraocular halves were more effective than 2 subcutaneous pituitaries. Finally, repeated doses of LH prolonged luteal function in OP/OP males here, indicating a synergism with graft LTH. Evidence for a similar synergism was found in O/O females receiving multiple doses of LTH and LH(6). All evidence suggests that the concept that differences in LH release in the male and female rodent are under hypothalamic control, rather than inherent in the pituitary(9), should be extended to include LTH.

Summary. Untreated intact male mice, bearing intraocular ovarian and pituitary isografts, showed only sporadic luteinization. Following administration of 5 μg of LH, over 70% of ovarian grafts developed functional (hyperemic) corpora lutea. Duration of this function was increased by 10 *vs* one or 3 doses. It was also increased by bilateral *vs* unilateral intraocular grafts of half pituitaries, and by 4 *vs* 2 subcutaneous grafts. In unilateral intraocular grafts, duration was longer in the ovarian tissue contiguous, than in that not contiguous, with the pituitary tissue. The sex of the pituitary donor did not influence luteal function.

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Tolerance to the Induction of Interferons by Endotoxin and Virus: Role of a Humoral Factor.* (30421)

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A single intravenous administration of bacterial endotoxin or virus induces circulating interferon or interferon-like viral inhibitors in experimental animals(1,2,3). We soon found that the response to a second inoculation of either endotoxin or virus in terms of interferon formation was markedly diminished. A preliminary report of this phenomenon of "tolerance" to the interferon-inducing effect of endotoxin and virus has been made (4). It has been found independently by Youngner and Stinebring(5).

It is well known that under certain circumstances, normal serum may inactivate or detoxify endotoxins(6,7). Whether tolerant serum has an enhanced capacity to do this is less well studied. In this report, in addition to describing tolerance to endotoxin as measured by its interferon-inducing activity, we wish to report on a factor in the serum of tolerant rabbits which inactivates this newly found biological activity of endotoxin.

Materials and methods. Albino rabbits weighing about 1 kg were inoculated intravenously in the marginal ear vein. Serum was obtained from blood collected by cardiac puncture or from the ear. Endotoxin extracted from *E. coli* (strain 0113) according to the Boivin method was provided by Dr. A. I. Braude. The use of endotoxin as well as Sindbis (AR339) virus pellet for induction of interferons has been previously described (2,8). To titer for interferon activity, serum collected 2 hours after injection of endotoxin, and 4 hours after injection of Sindbis virus was used. Tube cell cultures of rabbit kidney or rabbit embryo were incubated at 37°C for 18 hours with 2-fold serial dilutions of serum. These cultures were then challenged with about 1000 TCD_{50} vesicular stomatitis virus (Indiana, VSV) after the serum dilutions were decanted. Interferon effect was measured by the highest dilution in which 50% of the cultures showed inhibition of the cytopathology of VSV, as previously described(2, 8,9). The range of titers obtained was 1:16 to 1:512 for endotoxin induced interferon, and 1:40 to 1:10,000 for virus induced interferon.

The endotoxin inactivating factor (EIF) of rabbit serum was measured by the capacity of serum to eliminate the interferon-inducing capacity of endotoxin. A dose of endotoxin (5 or 10 μg) ordinarily sufficient to produce more than 1:16 interferon was diluted 1:5 in the serum to be tested, making a final volume of 0.5 to 1.0 ml. The mixture was incubated at 37°C for one to two hours, and given intravenously to a test rabbit. Serum was collected 2 hours later and titrated for interferon activity. If no interferon activity was detectable or if the titer was less than 1:8, it was assumed that the serum had eliminated or reduced the interferon inducing capacity of endotoxin. The serum obtained 24 hours after inoculation of endotoxin (*i.e.*, from tolerant animals) was called "tolerant serum."

The hemagglutination test against Type "O" human red blood cells coated with NaOH-treated endotoxin was performed in

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