graft versus host reaction. It remains to be shown conclusively for sarcomas and carcinomas, however, as has already been done for lymphomas, that the F_1 effect is really due to the difference at the H-2 locus between tumor and host. This can be achieved, for example, by using isoantigenic variant sublines isolated from tumors induced in F_1 hybrids between two isogenic resistant strains (*1*).

The absence of an F_1 effect detectable by skin transplantation may seem surprising in view of the present findings and this perhaps depends on differences in cell numbers inoculated (large in the case of a skin graft, small in the relevant experiments with tumor cells). Other differences are that hematopoietic and tumor cells are inoculated as suspensions of free cells, less protected than cells within a skin graft, and the fact that the tumor and hematopoietic cell populations studied grow logarithmically, while cell number is more stationary within a skin graft.

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Hormonal Activation of the Insect Brain

Abstract. *A new endocrine function has been shown to play a role in insect morphogenesis. The hormone named* proctodone *is secreted by specialized cells of the hindgut and appears to affect the brain, resulting in the activation of the brain hormone-producing system. Proctodone has been found in two lepidopterous species*—Ostrinia nubilalis *and* Galleria mellonella. *In the* O. nubilalis, *proctodone plays a part in both photoperiodism and diapause.*

Neurosecretory centers in the insect brain are thought to initiate the complex physiological processes leading to growth and molting. Diapause, as a state of arrested growth, is considered to be caused by an endocrine failure, usually that of neurosecretion; and the resumption of neurosecretory activity signals the termination of the diapause state (*1*). There is increasing evidence, however, that diapause is not invariably produced by a direct effect of genetic and environmental factors on the neurosecretory cells, but may be the result of changes in unidentified physiological processes to which neurosecretory processes are coupled (*2*).

Most of our experiments were with larvae of the European corn borer, *Ostrinia nubilalis*, in which larval diapause had been induced by short-day photoperiods (12 hr light/day). Borer larvae have been shown to be sensitive to photoperiod while in the diapause state, and diapause may be terminated by exposing the larvae to long-day photoperiods (16 hr light/day) for 10 to 12 days (*3*). According to the currently accepted concepts of insect growth, the larval brain is, in some unidentified way, reactivated, and the larvae resume morphogenesis leading to pupation.

When borer larvae in diapause were effectively bisected by the application of ligatures between the 6th and 7th abdominal segments, exposure to long-day photoperiods did not lead to the termination of diapause. Ligation at the 9th abdominal segment did not prevent diapause termination. From such experiments, it was concluded that the termination of diapause depended upon some event occurring in the 7th or 8th abdominal segments. All of the known endocrine organs are found in the head and thorax, and the photoperiodic response is also thought to originate in the cephalic parts of the insect (*4*). According to the known endocrine functions, abdominal ligations should not have prevented pupation of those portions of the larvae lying anterior to the ligatures.

A series of experiments, in which diapause larvae were exposed to long-day photoperiods for different intervals before the application of abdominal ligatures, demonstrated that the action of the abdominal system was required for a period of 10 to 12 days after the beginning of the long-day treatment. Ligations after this time did not prevent the termination of diapause, and the anterior portions of the ligated larvae pupated. Further ligation experiments demonstrated that the endocrine activity of the brain was required for several days after the abdominal system had completed its required function.

The experimental results strongly suggested that the function of the abdominal system was to activate the endocrine organs associated with the brain. This might be accomplished in either of two ways: (i) by nerve impulses arising in the posterior ventral ganglion in response to long-day photoperiods; or (ii) by hormones being produced in the 7th or 8th abdominal segments, with such production being regulated by photoperiod. The first of these alternatives was eliminated by the results of surgical experiments. Severing the ventral nerve cord at the 6th abdominal segment did not prevent the photoperiodic termination of diapause.

Table 1. Effect of tissue implants and abdominal ligations on morphogenesis of diapausing larvae of the European corn borer.

Material implanted or injected	Condition of larvae	Photoperiod	Larvae treated (No.)	Mortality (No.)	Pupae* (No.)
A. Diapause brain	Diapause and brainless	Long-day	12	1	11
B. Diapause brain	Diapause and abdominal ligation	Long-day	20	8	0
C. Nondiapause brain	Diapause	Short-day	18	0	18
D. Nondiapause brain	Diapause and abdominal ligation	Short-day	18	4	12
E. Proctodeal extract	Late diapause	Short-day	29	2	17
F. Saline control	Late diapause	Short-day	18	7	3

* Number pupating within 20 days after treatment.

The brains of diapausing borers implanted in brainless diapausing larvae did not prevent the photoperiodic response (Table 1, *A*).

Search for an endocrine organ in the 7th and 8th abdominal segments resulted in the discovery of secretory activity in the epithelial cells of the portion of the hindgut lying in the 7th and 8th abdominal segments. The secretory activity was detected in borer larvae undergoing diapause development, at which time the lumen of the intestinal tract was empty. No evidence of secretion into the gut lumen was detected; it was considered likely that the secretory products were being released into the hemolymph. This hypothesis was circumstantially supported by the observation that the intestinal muscularis was found to be very scant in this part of the digestive tract, and blood and hemocytes could be seen to be separated from the epithelial cells by only a very thin membrane. Cytological signs of secretory activity observed were: (i) multilobate nuclei, (ii) numerous cytoplasmic granular inclusions that stained dark purple with paraldehyde-fuchsin, and (iii) numerous small cytoplasmic vacuoles. This apparent secretory activity increased in intensity in response to biochemical treatments that have been shown to accelerate the termination of diapause (*5*). Evidence of a photoperiodically regulated daily secretory cycle was also found. Aqueous extracts of the proctodeal epithelium in question caused the termination of the diapause state when injected into diapause larvae that had undergone partial diapause development (Table 1, *E* and *F*). From this experimental evidence, it was concluded that the proctodeal epithelium constitutes a site of hormone production. Because of its source, the name PROCTODONE is proposed for this hormone.

A series of surgical experiments demonstrated the role of proctodone in the activation of the larval brain. Inactive (from diapause) or fully active (from nondiapause) larval brains were implanted into diapausing larvae, some of which had been ligated between the 6th and 7th abdominal segments (Table 1, *A*, *B*, *C*, *D*). The operated larvae were held under an appropriate photoperiod and observed for pupation occurring within 20 days after treatment. The results showed that a brain of diapausing borer implanted into a diapause larva can be activated, unless the recipient larva had been subjected to an abdominal ligation. A brain from a nondiapausing insect, however, induced pupation whether or not the recipient larva had been ligated.

Like the ligation experiments already discussed, the surgical results indicated that the neuroendocrine processes of the fully activated brain were independent of the hormonal output of the proctodeal system. We were, therefore, led to the conclusion that the physiological function of proctodone is activation of the endocrine centers of the brain. The details of these experiments and of the probable relationships between endocrine functions and photoperiodism are being published elsewhere (*5*).

Using larvae of *Galleria mellonella*, which is a nonphotoperiodic, nondiapause species, we have found that a proctodone function also occurs. Larvae that had completed feeding and had left the rearing medium preparatory to pupation were subjected to abdominal ligation. Those ligated before cocoon spinning had begun were unable to complete their development, although they survived for several weeks. Ligatures applied after cocoon spinning had begun did not prevent pupation. As with the corn borer, *Galleria* larvae showed a proctodone requirement over a period of time that was somewhat shorter than the period of dependence on the brain hormone system. In both of these species, proctodone is apparently required for the activation of the endocrine functions associated with the brain. No other species have been investigated (*6*).

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Inhibition of Antibody Synthesis by L-Phenylalanine

Abstract. *Antibody synthesis in response to the injection of diphtheria toxoid into rabbits and rats was used to study the effects of an excess of the amino acid L-phenylalanine on protein synthesis. Injections of phenylalanine produced a marked inhibition of antibody synthesis in both the rat and the rabbit. The dosage of phenylalanine used caused an increase over the normal concentration of phenylalanine in the plasma and spleen of rats, but did not cause a loss of weight or serum protein changes in the treated animals.*

Sufficient evidence is available to indicate that antibodies are formed from the pool of free amino acids in the cell and that such synthesis requires a full complement of amino acids (*1*). It would appear that interference with this pool may have serious consequences on the antibody-forming system.

The administration of an excess of an amino acid to an animal on a nutritionally adequate diet might be expected to produce an alteration in some of the pools of free amino acids of the body. Such alterations could occur because the excess amino acid interfered with the transport of other amino acids into the cell. For example, Neame (*2*) has reported that phenylalanine interferes with the uptake in vitro of histidine, arginine, ornithine, and tyrosine by the brain. Other studies (*3*) have indicated that this type of inhibition may result from many amino acids entering the cell by way of a common carrier. An excess of one amino acid could saturate the carrier, thereby interfering with normal cellular transport.

Table 1. Hemagglutination titers of rats immunized with diphtheria toxoid. Group A received 0.85 percent saline. All groups also received an intraperitoneal injection of 0.2 ml of diphtheria toxoid adsorbed on aluminum phosphate which was followed by a second injection 3 weeks later. Serums were obtained 10 days after the second injection of toxoid. Initial injections of L-phenylalanine and diphtheria toxoid were given on the same day. The L-phenylalanine was injected intraperitoneally.

Group	Animals (No.)	Phenylalanine dosage (mg/kg)	Average titer*
A	7	0	3600
B	6	125	1200
C	6	180	700
D	6	240	180
E	5	300	140

* Reciprocal of dilution.