76° ± 1° F. They were fed daily one medium-size, live goldfish. The eels always located the goldfish, stunned them with their electric discharge, and then swallowed them whole.

After about a month one of the bigger eels developed the skin disease. The first noticeable symptom was a loss of appetite. After 5–6 days a whitish patch, 1 cm in diameter, appeared on the skin (dorsal part) and also on the edge of the ventral fin. It had the appearance of a fungal disease of tropical fish. All the recommended and available tropical fish remedies were used in the dose recommended by the manufacturers of the drugs. 'Bacticide' (streptomycin sulphate and erythromycin thiocianate) was used without any good results. The disease advanced, and more patches appeared on the body and on the edge of the fin. We then tried tetracycline in a dose of 250 mg/gallon water after the first water was gradually replaced with fresh. We touched the patches on the skin with a solution of 1 part per thousand thymerosal after removing the eel from the tank. We also tried a salt bath (sea-salt : 1 spoon per gallon) with the same negative results. After seven days the eel died. A microscopic examination of a biopsy failed to show anything other than epithelial cells and mucus. After two months from death of the first eel, the other two eels also developed the same disease with the same symptoms.

This time we decided to use another treatment. We used 'Mysteclin-*F*' containing per capsule 250 mg tetracycline hydrochloride and 50 mg amphotericin *B*, in the proportion of 1 capsule/5 gallons water and water-soluble acromycin 50 mg/5 gallons of water. This stopped the disease from advancing further, and the fish were in a sort of equilibrium status with it. We then started painting the affected areas of the eels with a paste made of (per cent) salicylic acid 1 ; benzoic acid 1 ; thymol 1 ; zinc oxide 5 ; zinc bacitracin 400 units ; neomycin sulphate 0·5 ; borax 1 ; vitamins A (1,000 units), D (5,000 units) and E (100 units) ; hexachlorophene 0·5 ; benzocaine 0·5 ; phenol undecylene 0·5 ; thyrothricin 3 ; aerosporin polymixin sulphate 5,000 units ; and 30 mg of amphotericin *B* in a 30-g mixture of neutral 'Vaseline', lanolin and silicone fluid. After two weeks some of the patches began to disappear and a general change toward recovery was noticed. After another week the medicated water was replaced gradually over two days with aged water with green algae. One day after the water was changed the patches on the skin disappeared completely and the electric eels regained their normal appetite.

More than two months have passed since the eels were cured and they present no disease symptoms and they are healthy.

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ENTOMOLOGY

Breeding of the Rabbit Flea (*Spilopsyllus cuniculi* (Dale)) controlled by the Reproductive Hormones of the Host

THE rabbit flea (*S. cuniculi*) is a host-specific parasite of the rabbit. Before the outbreak of myxomatosis in Britain all populations of rabbits examined from the mainland were infested and the flea index was high[1,2]. The first hint that the breeding cycle of the flea was linked to the hormone cycle of the host came from Allan's[1] observation that shortly before the breeding season commenced, female rabbits carried an extra load of fleas. Further observation enhanced this view, and Rothschild[2,3] suggested that the breeding cycle, especially copulation, was under the influence of the sex hormones of the host. This suggestion was followed up by Mead-Briggs[4], who proved that: (*a*) maturation of the ovaries of female rabbit fleas could only take place on a pregnant female rabbit; (*b*) that immediately after parturition the fleas from the does were to be found in the nest containing the young rabbits, where they fed on the young and laid eggs.

The factor which stimulated and sustained ovarian development was not then known, although Mead-Briggs[4] postulated "a factor, perhaps nutritional, present only in the blood of pregnant rabbits". It can now be demonstrated that this factor is one of the hormones secreted by the anterior lobe of the pituitary gland. It has been possible to induce full maturation and egg production in laboratory-reared fleas feeding on: (*a*) doe rabbits not previously mated, undergoing pseudo-pregnancies after pairing with vasectomized bucks; (*b*) virgin doe rabbits injected with daily doses of 2 ml. 'Ambinon *B*' (Organon and Co., London, serial No. 12961, 10 U.S.P. (100 Heyl Laquer) units thyrotrophin and gonadotrophic factors); (*c*) castrated buck rabbits injected with daily doses of 2 ml. 'Ambinon *B*'. No development of the ovaries of female rabbit fleas occurred on virgin doe rabbits and castrated buck rabbits fed with (*a*) 1·2 mg 'Norlutin *A*' (norethisterone acetate, Parke Davis and Co., No. 894) daily by mouth for the period of normal pregnancy, that is, 30 days; (*b*) 0·2 mg of œstriol dissolved in 1 ml. olive oil by mouth for 30 consecutive days; (*c*) injected with 2 ml. 'Progestin' (B.D.H. progesterone Nos. 68900/1009/2204 and 94119/1010/2304) intradermally for 30 consecutive days. Fleas from the same source subsequently matured their ovaries on the same rabbits injected with 2 ml. daily of 'Ambinon *B*'.

If the dosage of 'Ambinon *B*' was reduced or the injections stopped altogether, development of the fleas' ovaries was arrested and rapid regression followed.

One of the earliest visible manifestations of the onset of maturation in the rabbit flea is an accelerated rate of defæcation. In Nature, larvæ in the nest feed on the fæcal pellets of the adult flea, thereby obtaining a supply of blood which they require for satisfactory development[5].

In species like *Echinophaga gallinacea* (Westwood) which remain more or less permanently attached to the host, a very rapid rate of defæcation is frequently observed (one blood-red pellet may be passed out of the anus every minute when feeding has just commenced[6]). In the rabbit flea, which is only a semi-sedentary species, the defæcation rate is apparently greatly accelerated only at the onset of maturation; this is about a week before parturition of the rabbit at a period when the doe is making her nest, and it ensures that the fæcal pellets will be available in the nesting material when the larvæ hatch. Within 24–48 h of the injection of 2 ml. of 'Ambinon *B*' fleas can be observed with blood-red fæcal pellets adhering to the anus and accelerated defæcation commences. Sections of the abdomen of female fleas at this stage reveal an enormously enlarged rectal ampulla gorged with blood, whereas the mid-gut itself may be almost empty.

Throughout these experiments no pairing was observed, nor have fleas been seen to pair on a naturally pregnant host; furthermore, the spermatheca of female fleas before parturition of these hosts is empty, and it is assumed that a factor other than the hormone from the anterior lobe of the pituitary and the separation from the host is required to stimulate the male to copulate. Copulation of the rabbit flea was first observed by Mead-Briggs[7] on young rabbits in the nest.

The rabbit flea is extremely sensitive to changes of mood in the host, and the associated physiological changes which presumably occur. Thus, even the act of pairing between rabbits may agitate the fleas, which rise to the surface of the fur, and frequently an exchange of fleas takes place between the rabbits during copulation. The agitation of the doe at parturition could, in itself, be sufficient to account for fleas leaving the host and entering the nest. In the early stages of pregnancy the doe is particularly attractive to fleas and infested bucks lose their infestations—or, if not all, the majority of their fleas—to pregnant does if the two are confined together in hutches.

It is believed that the rabbit flea is the first recorded instance of a parasitic insect in which the reproductive cycle is under the control of the reproductive cycle of the host.

We thank Prof. G. W. Harris, who planned and directed the experiments which showed that secretions from the anterior lobe of the pituitary control maturation, and who performed the necessary surgical operations on the experimental rabbits. We also thank Dr. Deryk Frazer for supplying hormones for the preliminary experiments in 1959; Mr. F. G. A. M. Smit for dissecting the reproductive organs of male fleas at that time; Dr. John Parsons and Dr. John Godfrey for their assistance with the experiments with œstriol and 'Norlutin *A*', and Mr. Ronald Allan for his continued interest in this work and for cutting serial sections for us during the three-year period of these experiments. We thank Parke Davis and Co. for the gift of 'Norlutin *A*'. Work on this life-cycle has been greatly hampered and constantly interrupted by a shortage of material, and we would like to record our special gratitude to H.R.H. The Duke of Gloucester, the Earl of Fitzwilliam and Mr. J. Chudley for coming to our assistance on innumerable occasions with permission to collect rabbit fleas at Barnwell, Milton and Brigstock.

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Feeding of Dry Chemically Defined Diets, and Egg Production in the Adult House-fly

According to West[1], the adult house-fly, *Musca domestica* L., must ingest its food in liquid form. Probably because of this fact, and because it is easier to feed large numbers of insects with a single dietary preparation, most investigators have used liquid diets when studying the nutritional requirements of the house-fly and other adult Diptera[2–7]. A few investigators have presented dry diets and water separately, but simultaneously, to the adult house-fly and promoted egg production[8–10].

Generally in all these tests, both the liquid and the dry diets contained one of many basic, refined natural products supplemented with various chemicals[2,3,8–11]. Recently more specific interest has been directed towards the significance of nutrition in insect reproduction[2,3,11]. Some investigators have fed anti-vitamins with natural products[3]; others have added varying levels of chemical supplements to semi-synthetic diets and determined their effect on fecundity, survival or fertility[11]. Until now, only one adult dipteran (*Aedes aegypti* L.), in addition to the house-fly, has been successfully fed a chemically defined liquid diet that resulted in egg production[6,7]. There have been no reports on ovarian maturation or fecundity when flies were fed on a chemically defined dry diet.

In all the investigations in which liquid diets have been used techniques have been devised to minimize the chance of dietary changes brought about by contaminating micro-organisms[6,8]. Although in such investigations sepsis can be kept to a minimum by adding antibacterial agents[12], or by presenting a fresh diet daily, it has long been recognized that dry diets are less subject to contamination over long periods of feeding[10,13]. Consequently, if at all possible it is important to develop and feed dry chemically defined diets in the study of insect nutrition and reproduction.

In our work on nutrition and reproduction in house-flies, a liquid chemically defined diet was developed which, when fed to adult females, resulted in a fecundity comparable with that of adults fed liquid milk[14,15]. The question arose—Would this diet when presented to the adult as a dry powder support egg production?

It was evident from the literature that a source of liquid should be provided in addition to the dry diet[16,17]. Our preliminary tests established that, for adults to survive for long periods, water must be supplied as sugar-water, although some investigators have fed only water, confining the sugar to the dry diet[8]. In the tests discussed here, sucrose was presented to the adult flies in two ways simultaneously, as a 0·1 M solution on an absorbent cotton wick, and in the dry diet to encourage aggregation and feeding.

The house-fly larvæ used in these experiments were reared by standard techniques on *CSMA* (Ralston Purina Co., St. Louis, Mo.)[14]. Dried brewers' yeast was the only supplement added to the medium. Twelve females and three males were separated immediately after pupation into a clean glass cylinder (180 mm high by 150 mm outside diameter). All cylinders were topped with nylon screening and placed on aluminium trays covered with filter paper. Dry test diets and sugar-water were renewed daily; flies were allowed to feed *ad lib*. Fresh milk, fed as a 'control', was also renewed daily. Although fecundity was recorded daily, dissections were also made of the developing ovaries.

Our principal interest in these tests was two-fold, to determine whether the adult house-fly would feed on a completely synthetic dry diet, and, if so, whether this diet formulation was nutritionally adequate for ovarian maturation. The results are a summary of five replicates for each test diet.

From the observation recorded in Table 1, it is evident that both diets *A* and *B* were acceptable and supported ovarian maturation. This was substantiated by dissections which revealed many mature eggs in the ovaries. However, with both synthetic diets, the fecundities, though low, were not significantly different over the period of the test. It is unlikely that the low fecunditics arose from

Table 1. Fecundity and Survival of Adult House-flies fed a Dry Synthetic Diet (plus 0·1 M Sugar-water separately) compared with that of flies fed Liquid Milk

Experimental diet	After 7 days: Mean No. eggs/cylinder	After 7 days: Mean No. eggs/exp.♀	After 7 days: Survival (%)	After 17 days: Mean No. eggs/cylinder	After 17 days: Mean No. eggs/exp.♀	After 17 days: Survival (%)
Diet *A*	176	15	83	235	20	20
Diet *B*	130	11	87	188	16	30
Fresh milk	725	60	83	1208	101	28

The chemically defined diet *A* contained the following: *l*-arginine 0·38 g, *l*-histidine 0·15 g, *l*-isoleucine 0·25 g, *l*-leucine 0·75 g, *l*-lysine 0·75 g, *l*-methionine 0·15 g, *l*-phenylalanine 1·20 g, *l*-threonine 0·15 g, *l*-tryptophan 0·30 g, *l*-valine 0·50 g, *l*-glutamic acid 1·00 g, salt mixture '*W*' (Nutritional Biochemical Corporation) 0·15 g, sucrose (Fisher reagent) 3·42 g. Diet *B* contained the same amount of the above basic components of diet *A*, but was supplemented with ribonucleic acid (yeast) 0·4 g, and the following B vitamins: folic acid 0·34 mg, niacin 0·30 mg, calcium pantothenate 0·44 mg, pyridoxal 0·90 mg, riboflavin 0·16 mg, thiamine 0·12 mg, biotin 0·05 mg, inositol 5·95 mg, choline chloride 2·00 mg. Both diet *A* and diet *B* were ground to a fine powder and stored at 5° C until fed.