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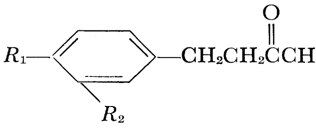
Anisylacetone, Synthetic Attractant for Male Melon Fly

Attractants have recently received increasing attention as an effective means for combating insect pests. By baiting traps with specific attractants, it is possible to discover an insect infestation at an early stage, and control measures may then be initiated immediately. Attractants in traps are also of use in delineating the areas that must be treated and in following the progress of the control program. Certain attractants in combination with a suitable insecticide may also be used to lure insects to their death.

The Agricultural Research Service has been conducting investigations on attractants for fruit flies at its Honolulu and Mexico City laboratories for many years (*1–3*). Since November 1955 chemists at Beltsville have been synthesizing and supplying candidate chemicals for testing in these laboratories. Recently, as a result of these studies, it was found that anisylacetone (formula Ib) is an effective attractant for the male melon fly (*Dacus cucurbitae* Coq.) (*4*).



(I)

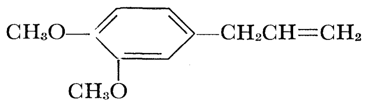
$R_1 = R_2 = \text{H}$ (Ia)

$R_1 = \text{CH}_3\text{O}; R_2 = \text{H}$ (anisylacetone) (Ib)

$R_1 = R_2 = \text{CH}_3\text{O}$ (Ic)

After more than 1000 compounds had been screened in Hawaii, it was found that the male melon fly was attracted by a number of aromatic ketones. Benzylacetone (formula Ia) and anisylacetone [formula Ib, 4-(*p*-methoxyphenyl)-2-butanone)] were the most attractive compounds, but field tests indicated that the latter was superior. These pleasant-smelling compounds are prepared by condensing an aromatic aldehyde such as anisaldehyde with acetone and hydrogenating the product. Anisylacetone has been described by Sosa (*5*), and by Chen and Barthel (*6*), who used it as an intermediate in the preparation of pyrethrinlike esters.

Analogous compounds are not as attractive as anisylacetone. Introduction of another methoxyl on the benzene ring gives a compound (formula Ic) which is no longer attractive to the melon fly but which is rather attractive to the oriental fruit fly (*Dacus dorsalis* Hendel). The similarity of this compound to the outstanding attractant for the oriental fruit fly, methyl eugenol (formula II) (*2*), is apparent.



(II)

The discovery and practical evaluation of the attractiveness of anisylacetone were timely. The compound was put to use almost immediately in California to determine the extent of a possible melon fly infestation. With the help of the attractant, the state and federal officials promptly undertook an extensive trapping program, and it became apparent that the single melon fly found was an isolated specimen of unknown origin.

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Diurnal Cycles and Learning in Earthworms

Recent studies of learning in earthworms (*1*) are important, not only for the data accumulated relative to a science of comparative psychology, but also because they appear to have relevance to a general theory of behavior. Because of these considerations, as well as the general lack of basic behavioral studies concerned with this species, an attempt was made (*2*) to follow up an incidental observation of a previous study (*3*) which had indicated that the diurnal cycle of the earthworm plays a significant role in the rate at which this organism acquires a particular turning tendency in a T-maze.

An early study by Baldwin (*4*), in which observations were made of *Lumbricus terrestris* placed in dirt between two parallel glass plates ⅜ in. apart, revealed that, in terms of crawling movements, feeding, and ejection of waste products, earthworms have definite activity cycles, with the active period occurring between 6 P.M. and 12 P.M. The present report attempts to extend these observations through study of the effect on learning of the earthworm activity cycle.

The apparatus used in this study was a T-maze constructed by fastening pieces of Lucite to a sheet of plywood. The stem and crosspiece of the maze were each 25.4 cm long, 2 cm wide, and 2 cm high; a Lucite cover was hinged in such a manner that each third of the runway could be opened independently. The left arm (negative goal) was 10.1 cm long and contained a 1-cm piece of 2/0 sandpaper 8 cm from the choice-point, followed immediately by two copper bell wires 0.25 in. apart, through which a shock of 1 v could be administered. The right arm (positive goal) was 15.3 cm long and ended in a beaker containing moist earth and moss, which was wrapped in paper in order to provide a dark interior.

One group of six earthworms (*L. terrestris*) was given five trials per day between 8 P.M. and midnight, with the negative and positive reinforcement, until a criterion of seven consecutive correct trials was achieved. The same procedure was utilized with a second group of six earthworms, except that the training was carried out between 8 A.M. and noon. A trial was considered correct if the worm entered the beaker without being shocked. Between trials on successive days, the worms were kept in a refrigerator. It should be noted that the conditions of the experimental room yielded no differential day-night cues to the two groups of worms.

Stimulation was applied by a camel's-hair paint brush if the worm remained motionless in the maze for 30 seconds. If, after 5 seconds of stimulation with the brush, the worm continued to remain motionless, a flashlight was turned on and held vertically slightly behind the anterior end of the worm. In all cases the worm began to move when the light was applied.

The mean number of trials to reach the criterion in the evening group was 32 (standard deviation, 4.01); the mean number of trials in the morning group was 45 (standard deviation, 5.27). A *t*-test shows that the evening group achieved the learning criterion in significantly ($p < 0.01$) fewer trials than did the group run in the morning hours. The use of the Mann-Whitney *U*-test, a nonparametric alternative to the *t*-test (*5*), confirms the significance of the difference in learning in the two groups. Analysis of brush and flashlight applications reveals that the evening group required significantly ($p < 0.05$) less stimulation per trial than did the morning group. Stimulation-per-trial measures were used because of the significant differences in the total number of trials that the two groups were run.

Since the morning group was run by one experimenter (J.P.M., *2*) and the evening group by me, an additional four animals were run in the evening by J.P.M. to insure that the procedure for running and stimulating animals was the same for both experimenters. No differences were noted between these four worms and the six others run in the evening group on any of the measures.

No specific control over temperature cues was exercised in this study; a record was kept, however, of the temperature of the room at the time of the first trial on each day for each worm. On the basis of the mean temperature for each worm, the 12 subjects were divided into a high-temperature group of six and a low-temperature group of six. No significant difference in the number of trials necessary to achieve the learning criterion was found between these two groups. Although this does not completely preclude the possibility that temperature may be an important variable (since there may have been temperature fluctuations during the five trials on any one day)—in particular, a temperature effect interacting with the time parameter—the experimental conditions of the present study did not lend themselves to greater control.

It may be concluded that the diurnal cycle plays an important role in maze learning in earthworms. This cycle appears to carry over to a learning situation where light-dark cues are eliminated from the environment. Wells (*6*) has noted the existence of diurnal activity cycles in the lugworm which continue for several days after the animal is placed in an environment free of diurnal cues, while other investigators report this persistence of cyclical activity in other species (*7*). It is possible that, like the feeding and irrigation cycles of the lugworm, the earthworm diurnal cycle is controlled by an "internal pacemaker."

At present there is relatively little evidence concerning the neurological structures which may act as pacemakers in regulating the biological activity of the worm. Scharrer and Scharrer (*8*) have noted the existence of neurosecretory cells in the brain of the earthworm; the function of these cells is as yet undetermined. Ziller-Perez (*9*) has indicated that the chromaffin cells in certain species of worms secrete an adrenalinlike substance. A tentative hypothesis might attribute the cyclical activity of the earthworm to the products of these cells. This hypothesis, however, sheds no light on the basic problem of the manner in which the cyclical pattern is imposed on the functioning of these cells.

Just as Schmidt (*1*) has shown that one cannot generalize from the avoidance behavior of one species of worm (*L. terrestris*) to the avoidance behavior of another species (*Eisenia foetida*, a manure worm), this study shows that time of learning relative to the worms' activity cycle is an important variable in research with Annelida.

JACK ARBIT*

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Extension of a Tracheole into Cytoplasm

In the course of an electron microscope study of spermatogenesis in the grasshopper (*Melanoplus differentialis*), a cell of the testicular theca was observed which is penetrated by a tracheole (*1*). The electron micrograph (Fig. 1) is presented because it illuminates two points: the structural details of the terminal portion of the tracheole and its position in relation to the cell.

The tracheole, which seemingly penetrates the cytoplasm of the thecal cell, clearly has taenidia (*TN*). This confirms the findings of Keister (*2*), Richards and Korda (*3*), and Beams and Anderson (*4*) that all respiratory tubes of the insect species examined possess taenidia. Furthermore, the taenidia are accompanied by cytoplasm. Buck (*5*), Keister (*2*), and Beams and Anderson (*4*) have observed tracheoles that emerge from their originating cells accompanied by protoplasmic processes. At the junction of the cytoplasm of the thecal cell and that of the tracheole is a double membrane, of which one part (*CL*) appears to be a limit of the cell cytoplasm and the other (*TCL*), the limit of the cytoplasm associated with the tracheole. At the region *D* each membrane appears to be double.

Fig. 1. (*A*) Thecal cell from grasshopper testis, penetrated by a tracheole (× 1850). (*B*) Enlargement of the tracheole-entered area revealing taenidia (*TN*), thecal cell membrane or limit (*CL*), and tracheolar cytoplasmic limit (*TCL*). At various regions (for example, *D*) each limit appears to be composed of a double membrane (× 15,000).

In this species, the terminal portion of the tracheole has an intima with recognizable taenidia, whose lumen measures 0.30 µ in diameter. In addition, beyond the limits of the taenidial border, this tracheole is surrounded by cytoplasm and has its own limiting membrane. The over-all diameter of the tracheole is about 0.67 µ. There is also evidence here that the tracheole does not end on the peripheral surface of the thecal cell but extends into the interior. However, the presence of the double membranes suggests that the tracheole has assumed its position, not by actual intracellular penetration, but by causing or following an invagination of the thecal cell membrane to the inner portion of the cell.

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5 August 1957