

# Reports

## Diurnal Cycles and Cannibalism in Planaria

**Abstract.** *Dugesia tigrina* attacks *Cura foremani* under certain circumstances. The length of fast prior to the attack and diurnal rhythms are involved in evoking such behavior.

The immature *Dugesia tigrina* sometimes attacks the planarian *Cura foremani* (1). Qualitative observations suggested a diurnal variation in likelihood of attack. Since neither this particular attacking behavior, nor its diurnal variation, has been previously reported, more systematic studies were undertaken.

When *C. foremani* and *D. tigrina* were removed from the dimly lighted conditions of maintenance in our laboratory, placed in a dish containing fresh spring water and illuminated by an overhead fluorescent light, they glided about in the dish. If a *D. tigrina* makes head contact with a *C. foremani* in the course of this gliding about, the former makes a slight withdrawal response. In contacts resulting in attack this withdrawal response is rapidly followed by a quick, snakelike lunge by *D. tigrina*. This results in an adhesive attachment of the *D. tigrina*'s head to the mid-dorsal surface of the *C. foremani* while the *D. tigrina*'s tail is anchored by adhesive attachment to the dish floor (Fig. 1b). The *D. tigrina* then contracts lengthwise and "hauls in" its prey (Fig. 1c) which, depending on its size relative to its attacker's, and the vigor of the attack, may escape.

On some occasions the attacks are "half-hearted"; on others, savage and persistent. If the *D. tigrina* manages to hang on, it works its head to a more rostral attachment (Fig. 1d), then loops

a tight coil about the *C. foremani* (Fig. 1e). From this position the *D. tigrina* uses its pharynx to tear out and ingest pieces (visible inside its digestive cavity) from its captive. *Cura foremani* was not observed to attack *D. tigrina* or another *C. foremani* although it readily attacks larger active prey such as mosquito wrigglers and smaller brine shrimp. *Dugesia tigrina* adheres to glass surfaces more tenaciously than *C. foremani*, indicating more powerful adhesive organs (2) and an advantage in combat, but not why *C. foremani* does not initiate attacks on *D. tigrina*.

For quantifying such attack behavior the following procedure was adopted. A *D. tigrina* (about 14 mm) was removed by random selection from a stock population of *D. tigrina* and placed in a bowl containing fresh water and approximately 25 specimens of *C. foremani*, ranging in size from 5 to 16 mm in length; the size distribution of the *C. foremani* population was maintained approximately constant in all tests. The bowl was observed on a white background under an overhead light. Each contact of *D. tigrina* with *C. foremani* was scored according to the three possibilities: (i) no attack, (ii) unsuccessful attack, (iii) successful attack. An attack was considered successful if, following it, ingested fragments of *C. foremani* were observed in the *D. tigrina*. When the digestive cavity of a *D. tigrina* was about half filled, that individual was removed and scored no more, in order to avoid satiation effects. If after 15 to 25 contacts the digestive cavity of the *D. tigrina* was still less than half filled, it was removed and scored no further. Once a *D. tigrina* had been used it was not used again.

When the number of contacts resulting in no attack is denoted as  $n_1$ , an unsuccessful attack as  $n_2$ , and a successful attack as  $n_3$ , the total number of contacts by the *D. tigrina* will be  $n = n_1 + n_2 + n_3$ . The "cannibalistic attack score,"  $s$ , for an individual *D. tigrina* at its time of testing is defined as

$$s = (0 \cdot n_1/n) + (1 \cdot n_2/n) + (3 \cdot n_3/n) \quad (1)$$

The score  $s$  is a measure of the attack behavior that is, to a first approximation, independent of the simple locomotor activity per se of either organism

at the time of assessment. Increased locomotor activity of the *D. tigrina* will increase the number of contacts per unit time but need not increase  $s$ . From Eq. 1 it is clear that  $0 \leq s \leq 3$  for any worm. Because a *D. tigrina* is not used again once it has been scored, each  $s$  is an independent measurement.

Since, in pilot studies, no attack behavior was ever exhibited by a *D. tigrina* recently fed to satiation, scoring was not conducted on a *D. tigrina* fasted less than 45 hours. At the outset of the fast period every *D. tigrina* was fed to satiation on raw beef liver. The laboratory temperature was maintained at 70°F during the period in question.

Table 1 gives  $s$  scores for the various *D. tigrina* specimens grouped according to length of fast and time of day of scoring. Measurements after 6 P.M. and before 6 A.M. are classified as "night," although most of them fall between 10 P.M. and 6 A.M. Most day measurements fall between noon and 6 P.M., but any after 6 A.M. and before 6 P.M. are classified as "day."

A median test (3) comparison of night and day scores of those organisms fasted more than 98 hours gives  $P < .001$ , night being greater than day. Neither night nor day scores of animals fasted less than 90 hours differs significantly from zero. A comparison between day scores of the long- and short-fast groups gives  $P < .005$ , the longer fasted being greater. Night scores of the longer fasted are greater than those of the short fasted with  $P < .0015$ . Hence, at least two factors, length of fast and time of day, are determiners of the attack behavior of *D. tigrina*. The mechanisms of mediation in the present case are unknown. The effect of fasting and feeding on this behavior suggests involvement of some "hunger drive" mechanism.

The diurnal effect is not related to contemporary changes in temperature

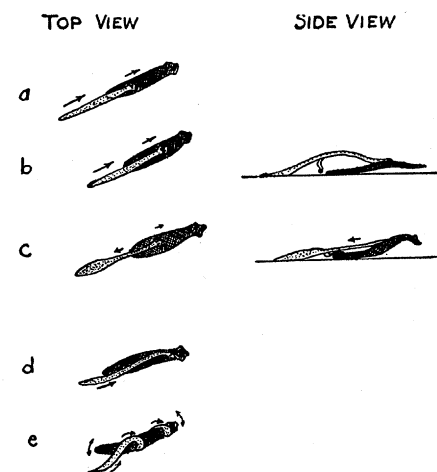


Fig. 1. *Dugesia tigrina* (stippled) attacking *Cura foremani* (dark).

**Instructions for preparing reports.** Begin the report with an abstract of from 45 to 55 words. The abstract should not repeat phrases employed in the title. It should work with the title to give the reader a summary of the results presented in the report proper.

Type manuscripts double-spaced and submit one ribbon copy and one carbon copy.

Limit the report proper to the equivalent of 1200 words. This space includes that occupied by illustrative material as well as by the references and notes.

Limit illustrative material to one 2-column figure (that is, a figure whose width equals two columns of text) or to one 2-column table or to two 1-column illustrations, which may consist of two figures or two tables or one of each.

For further details see "Suggestions to Contributors" [Science 125, 16 (1957)].

Table 1. Frequency distribution of attack scores for *D. tigrina* as a function of length of fast and time of day tested.

Score interval	Length of fast			
	45 to 90 hr		98 hr	
	Day	Night	Day	Night
$s = 0$	6	6	5	2
$0 < s \leq 0.1$	0	2	3	1
$0.1 < s \leq 0.4$	0	0	6	2
$0.4 < s \leq 0.6$	0	0	2	4
$0.6 < s \leq 0.8$	0	0	1	2
$0.8 < s \leq 1.0$	0	0	1	8
$1.0 < s \leq 1.5$	0	0	0	3
$1.5 < s \leq 3.0$	0	0	0	6

or illumination. Available facilities did not permit conclusion in regard to the "driven oscillator" model (4) as the means of synchronization to the solar day. Other species (5) exhibit similar diurnal rhythms in behavior (6).

JAY BOYD BEST

Department of Neurophysiology,  
Walter Reed Army Institute of  
Research, Washington, D.C.

#### References and Notes

- Species supplied as *D. dorotocephala* by Carolina Biological Supply Co., Elon College, N.C., but although black pigmented do not otherwise resemble these and have been identified as *C. foremani* by C. A. Miller and W. H. Johnson, *Ann. N.Y. Acad. Sci.* **77**, 87 (1959).
- L. H. Hyman, *The Invertebrates* (McGraw-Hill, New York, 1951), vol. 2, p. 80.
- A. M. Mood, *Introduction to the Theory of Statistics* (McGraw-Hill, New York, 1950), p. 395.
- C. S. Pittendrigh, et al., *Proc. Natl. Acad. Sci. U.S.A.* **44**, 965 (1958); V. G. Bruce and C. S. Pittendrigh, *Am. Naturalist* **92**, 295 (1958).
- G. P. Wells, *Symp. Soc. Exptl. Biol.* No. 4, 127 (1950); F. M. Baldwin, *J. Animal Behavior* **7**, 187 (1917); J. Arbit, *Science* **126**, 654 (1957); F. A. Brown, Jr., et al., *J. Exptl. Zool.* **123**, 29 (1953); *Sci. American* **190**, 34 (1954).
- I wish to acknowledge the assistance of Mr. David Schucker in conducting these observations.

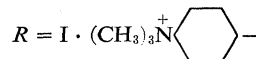
15 March 1960

### New Quaternary Ammonium Compounds with Adrenomimetic Action

**Abstract.** Quaternary salts of two triesters of *p*-dimethylaminothiophenol and phosphoric acid or thiophosphoric acid have effects similar to epinephrine on blood pressure, heart rate, and the nictitating membrane of the anesthetized cat. Two analogous compounds derived from phenol are depressor. None is a potent anticholinesterase. Further investigation is desirable because of the possibility of a relatively specific action on sympathetic ganglia.

In the course of a study of anticholinesterases, certain triaryl phosphate esters bearing quaternary ammonium groups were synthesized (1) which failed to show the expected activity. For four of these, the code number,

structure, and relative activity in vitro as cholinesterase inhibitors compared to neostigmine are: CP46, (RO)<sub>3</sub>P=S, 1.8; CP99, (RS)<sub>3</sub>P=O, 0.53; CP98, (RS)<sub>3</sub>P=S, 3.8; CP27, (R'O)<sub>3</sub>P=S, 0.25; neostigmine 100, where



and *R'* is the corresponding *meta* isomer of *R*. This is in marked contrast to related quaternary ammonium phosphate esters, such as the dialkoxyposphoryl esters of *m*-dimethylaminophenol methiodide (phosphostigmines) (2), which are potent inhibitors of cholinesterase. The low activity of compound 27 indicates that this is not a question of *meta* as against *para* orientation. All of these compounds were tested on the anesthetized cat for effects on blood pressure, respiration, heart rate, and the nictitating membrane.

Animals were anesthetized with Dial. Blood pressure was recorded with the Sanborn electromanometer; respiration, with a Krogh spirometer recycled four times per minute, from which rate and minute volume could be read; heart rate was recorded with a Thorp counter integrated over a 10-second interval; and contractions of the nictitating membrane, with an isotonic lever. All injections were given into the femoral vein. Test doses of acetylcholine (1 µg/kg) and of epinephrine (1 to 8 µg/kg) were given before and after injection of the experimental compound.

None of these substances induced muscle fasciculation or the typical profound muscarinic effects characteristic of systemically administered anticholinesterases. Compounds 27 and 46 were found to be moderately long-acting depressors. Compound 27 has been studied clinically in peripheral vascular disease as a vasodilator (3). Its mechanism of action is in doubt and does not fall into any obvious classification. Compounds 98 and 99, on the other hand, were pressor and their actions were as follows: compound 99 showed a minimal effect on the blood pressure at 0.1 mg per kg which consisted in a slight and transient rise or fall or biphasic response. Similar effects were seen with epinephrine at 1 µg/kg. At higher doses of compound 99 the blood pressure was consistently elevated, sometimes showing a bimodal curve as illustrated in Fig. 1. The onset and duration of the pressor action appeared to be the same for compound 99 and epinephrine at equally pressor doses. The heart rate did not change markedly and moved in the same direction as after test doses of epinephrine. In small to moderate doses, compound 99 caused a transient stimulation of respiratory minute volume without alteration in rate. At high doses, 6 to 12 mg/kg,

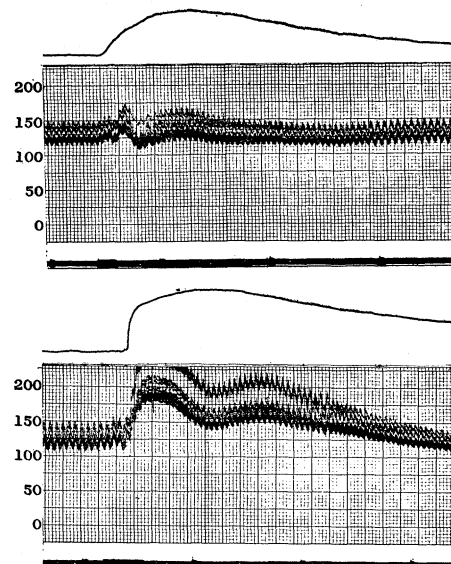


Fig. 1. Female cat, 2.7 kg, Dial anesthesia. From top to bottom: isotonic contraction of the nictitating membrane, blood pressure in millimeters of mercury, time in minutes. At the signal in the upper tracing epinephrine (0.004 mg/kg) was injected. At the signal in the lower tracing compound 99 (2 mg/kg) was injected. Drugs were given intravenously.

the animals died as a result of immediate respiratory arrest. Contraction of the nictitating membrane occurred at dosage levels showing effects on blood pressure, and the two effects had the same duration. A fall in blood pressure was observed with both compound 99 and epinephrine in animals previously given 15 mg of dibenamine per kilogram intravenously in three divided doses. Duration was comparable to that of the pressor effect before dibenamine.

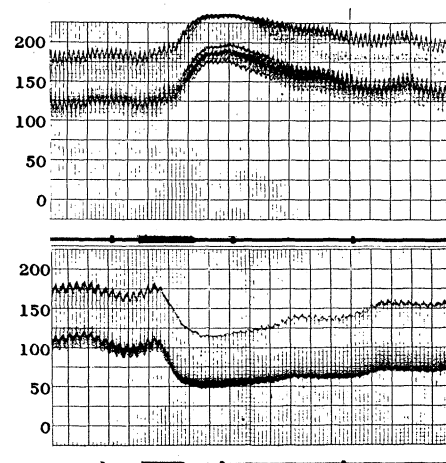


Fig. 2. Female cat, 3.2 kg, Dial anesthesia. From top to bottom: blood pressure in millimeters of mercury, time in minutes. At each signal compound 98 (5.3 mg/kg) was injected. Between the upper and lower tracings three doses of dibenamine (5 mg/kg) were given. Drugs were given intravenously.