ALBERT CLAUDE

of the original extracts to produce tumors was due to the presence of inhibitory elements in the solution. The fact that agent and inhibitor can be separated by physical means would suggest either that the agent is not modified by its inhibitor or that the reaction between them is easily reversible. Whether or not this centrifugation method will separate potent agents from hitherto non-filterable tumors will be determined by further tests. So far, preliminary experiments with mammalian tumors have given negative results.

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THE FEEDING OF HOLLOW-SPEAR NEMA-TODES ON OTHER NEMATODES¹

THE habit of preying upon nematodes has been found by the writers to be well developed in two groups of nematodes in which it appears not to have been recorded: the genus Aphelenchoides Fischer, 1894, and the dorylaim genera Dorylaimus Dujardin, 1845, Discolaimus Cobb, 1913, and Actinolaimus Cobb, 1913. Equipped with hollow, protrustive oral spears or stylets, predators of these genera feed, not as do the types formerly recognized as feeding on nematodes, but rather by inserting their spears into their prey and holding them there while sucking out the body contents. The relatively large dorylaims, with their coarse spears and powerfully muscular esophaguses, disorganize their prey so quickly that there is little opportunity for struggle. The rather small Aphelenchoides, on the contrary, feed slowly, and here the evidence is definite for one species that the prey is paralyzed almost instantly when the very slender stylet is inserted, so that struggles are prevented. During the feeding of two species of Aphelenchoides, saliva has been seen flowing out from the large dorsal esophageal gland, through the esophageal tube and into the prev.

Soil-inhabiting nematodes formerly recognized to be predacious on other nematodes, such as species of *Mononchus* Bastian, 1865, and *Diplogaster vorax* Goodey, 1929, are equipped with fine grasping teeth, with cutting teeth or with both. Slender mural teeth which slash into prey but are withdrawn while feeding are possessed by the genera *Nygolaimus* Cobb, 1913, and *Sectonema* Thorne, 1930, which are known from the work of Thorne² to prey upon oligochaet worms. Cobb,³ in listing 16 genera of predacious free-living nematodes, most of which are marine forms, mentioned "Pharynx with acute clutching

¹ Published with the approval of the director as Technical Paper No. 97 of the Pineapple Experiment Station, University of Hawaii.

² G. Thorne, Jour. Agr. Research, 41: 445-466, 1930.

organs—onchia or denticles" as characteristic of such genera. Of these genera, only *Dorylaimus* feeds through a hollow oral spear.

Dorylaimus was included in Cobb's list of carnivorous forms, despite its lack of clutching organs, on the strength of his simultaneously published record of two nemas of this genus feeding upon mite eggs; of Steiner's⁴ earlier record of a seta of an oligochaet worm in the intestine of Dorylaimus regius de Man, 1884; and of Thorne's⁵ finding three instances of *Heterodera schachtii* Schmidt eggs impaled on the spears of young Dorylaimus obtusicaudatus.

We have repeatedly observed various species of this genus and allied genera, including larvae and adults of both sexes, feeding upon other nematodes including larvae of *Heterodera marioni* (Cornu) Goodey, 1932. All these observations, with two minor exceptions, have been in Petri dishes of agar. Unlike predators capable of grasping or slashing their prey, these forms which suck their food through hollow spears seem unable to capture prey suspended in water, for considerable pressure is required to thrust their relatively coarse, hollow spears into other nematodes. Even a very soft agar is unsatisfactory, the prey being pushed through the medium by the spear instead of being penetrated.

A predacious dorylaim finds its prey only by chance, but when its head makes contact with another nematode it responds immediately. It orients its head at right angles with the surface of the prey, so that its lips make firm contact, then protrudes its spear suddenly in an attempt to penetrate, and may do so repeatedly if not immediately successful. Once the spear enters the body of the prey it is held there while the heavily muscular esophagus of the predator begins a rhythmical sucking action which quickly disorganizes the body contents of the prey, sucking them out and leaving an empty, collapsed body wall.

A dorylaim holds its spear extended far into its prey, even during the periods of rest which alternate with periods of sucking. With a medium to large dorylaim, the spear tip reaches to the opposite body wall of such nematodes as the larvae of H. marioni. Such a position is advantageous in view of the long diagonally placed opening on the side of the spear.

Some of these predators have also been seen feeding on nematode eggs, both of H. marioni and of various free-living forms. When egg masses of the former are placed in agar dishes they prove most attractive to at least some of these species.

The predators in question have not yet been identified specifically, but it is our judgment that ten species of *Dorylaimus*, two of *Discolaimus* and one of Ac-

⁴ G. Steiner, Jour. Agr. Research, 28: 1062–1064, 1924. ⁵ G. Thorne, Jour. Agr. Research, 37: 575, 1928.

³ N. A. Cobb, Jour. Parasitology, 15: 284-285, 1929.

tinolaimus are now represented in our collection of individuals which have been preserved after having been seen feeding on other nematodes. These represent collections from pineapple fields, gardens, wet forest and arid waste land on the island of Oahu and from association with weed roots from Christmas Island, a small coral atoll lying at 21° 57′ N., 157° 27′ W. Further details with specific identifications will be reported later.

From observations on their occurrence in Hawaiian pineapple fields it appears that these dorylaims are vastly more important as predators than species of *Mononchus*. Cassidy⁶ reported four species of that genus from pineapple field soils, but these are very rare. The dorylaim genera *Dorylaimus* and *Dis*colaimus, on the contrary, are generally present and often numerous. For instance, a sample of 252 grams soil with 46 grams roots from under a single pineapple plant contained dorylaims of two species at the rate of 1.3 per gram, but no specimen of *Mononchus* was found.

The predacious species of *Aphelenchoides* are more remarkable in several ways, being morphologically very similar to several plant-parasitic species and being members of a group in which no predacious tendencies seem to have been suspected. The genus includes species parasitic on higher plants as well as at least one fungus-sucking species.⁷ Some other species are free-living forms of unknown food preference but commonly regarded as saprophytic.

We have found two species of this genus to be highly specialized predators which, given a sufficient population of prey, multiply rapidly in agar culture. Both are cultured readily by transferring them to cultures of *Aphelenchus avenae* Bastian, 1865, or *Aphelenchoides parietinus* (Bastian) Steiner, 1932, previously established in an agar culture of a suitable fungus. Neither predator feeds on fungi.

One of these species appears to be Aphelenchoides tenuicaudatus (de Man, 1895) Goodey, 1933, while the other may require description as a new species. Both have very slender hollow stylets, with which they penetrate their prey and through which their food is drawn. A. tenuicaudatus attacks nematode eggs as well as larvae and adults, while the other appears to feed only on nematodes.

These two predators are both small, well under one millimeter in length, but they attack successfully larvae of H. marioni and both adults and larvae of Anguillulina pratensis (de Man) Goffart, 1929, as well as of various free-living species including forms

larger than themselves. Both species appear to exert a paralyzing action on their prey, which helps to compensate for their inferior size, for nematodes penetrated by the stylet become inactive almost immediately and lie still during the relatively slow process of being sucked out.

Paralysis of prey was clearly indicated for the undescribed species of *Aphelenchoides* when a larva only one hour old was seen to pierce an *A. avenae* twice its own length. This prey was moving rapidly at the time, so that the predator lost its hold and its stylet was within the prey for only an instant; still this nematode stopped in half its own length and lay quiet for several hours before being fed upon by another predator.

Such paralysis probably results from an injection of saliva from the predator. No injection at the instant of penetration has been seen, but repeated salivary injection during feeding has been observed in many instances with both species of *Aphelenchoides*. The saliva itself has come from the dorsal esophageal gland, which is highly developed in this genus.

During periods of active sucking and continuing into the many alternating periods when the esophageal bulb is at rest, saliva may be seen flowing anteriorly from the long dorsal esophageal gland, through a slender noncuticular duct dorsad of the intestine, and into the posterior end of the esophageal bulb dorsad of its lumen. Here the duct widens, and the saliva is seen to continue its anterior flow within the confines of the bulb but dorsad of its cuticular crescentic plates, and to enter the anterior nonstriated part of the bulb. In both species this part is distinctly alveolate, and into its cavities the saliva flows. Accumulation here is apparent while the muscular part of the bulb is at rest. This alveolate region may be seen to twitch irregularly as it fills, and after it is filled some return flow from it posteriorly towards the dorsal gland often occurs.

As soon as the muscular bulb resumes activity, saliva again flows anteriorly from the dorsal gland. Presumably some of this saliva enters the lumen of the esophagus and mixes with the food rushing posteriorly towards the intestine. Very clearly, however, during momentary interruptions of the inflow of food, some of it rushes anteriorly from the bulb through the lumen of the esophagus and stylet and into the body of the prey where it appears to start the process of digestion. These details have been observed repeatedly in both predacious species of *Aphelenchoides*.

These observations of saliva flow lend great weight to the hypotheses of various investigators to the effect that pathological states arising from nematode infestations result, at least in part, from an outpouring of salivary secretions.

⁶G. Cassidy, Hawaiian Planters' Record, 35: 305-339, 1931.

⁷ J. R. Christie and C. H. Arndt, *Phytopathology*, 26: 698-701, 1936.

SCIENCE

After this note was submitted for publication, Mr. Gerald Thorne, of the United States Bureau of Plant Industry, who was sent a manuscript copy, wrote as follows (personal communication): "There are many species of *Discolaimus* and *Actinolaimus* in my collections . . . which I know to be predacious. But it

r. was only about a month ago that Mr. McBeth . . . saw t our first *Aphelenchoides* feeding on another nema."

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SCIENTIFIC APPARATUS AND LABORATORY METHODS A CIRCULATION PUMP¹ air-valve is opened and the top-light is out. Air pres

In a series of experiments on adrenaline secretion² need arose for a pump with which to pump the blood from the adrenals of a cat into its carotid artery. A description of the main features of this pump, which has been found to function satisfactorily and which can be used for the perfusion of organs with blood or other fluids or for the maintenance of the circulation of a whole animal, follows.



The blood, leaving the vein, enters a vertical glasstube. Its top is connected with the outlet of a solenoid air-valve, the inlet of which leads to an air-cylinder. When this valve is opened, the pressure, delivered by the tank, acts on the blood in the glass-tube and pumps it out. Two glass-valves direct the blood from the tube into the artery during the systole and prevent back flow from the latter during the diastole of the pump. When the glass-tube is emptied the air-valve is closed, and new blood is thus allowed to enter.

The air-valve is opened and closed by a photoelectric cell activated by two beams from two light sources, which pass through the glass-tube and can be interrupted by the blood column. These light-beams are focussed by lenses in such a manner that one passes through the bottom of the tube, while the other one is focussed on it at any desired height above. The wiring of the relay-contacts of the photo-electric outfit, the solenoid valve and the light sources result in the following operations of the pump. During systole the

²Gerhard Katz and Gertrud Katz, Jour. Pharm. and Exp. Therap. (in press). air-valve is opened and the top-light is out. Air pressure pumps the blood out of the tube into the artery until the blood column reaches the beam of the bottomlight. This beam is thus allowed to pass through the glass-tube and activates the photo-cell, which then closes the air-valve and switches the top-light on. During the ensuing diastole the top-light keeps the solenoid-valve closed until the rising blood level cuts the light off the photo-cell, which, by opening the pressure valve and switching the top-light off, starts the next systole. A narrow permanent opening at the top of the glass-tube allows the blood to replace the air enclosed in the tube during the diastole.

The output per stroke is determined by the height at which the top-light-beam passes the glass-tube. With each stroke, the tube is emptied, since the lower beam passes through the lowest part of the glass-tube. Increase or decrease in venous return are answered by a corresponding change in rate. The desired systolic pressure is adjusted by the reduction valve at the pressure tank. A large inside diameter of the glasstube prevents significant changes in venous pressure. as small changes in level correspond to large changes in volume. An air-cushion chamber as used in other circulation pumps between arterial valve and artery maintains blood flow and pressure during the diastole. By placing the lower end of the glass-tube below the level of the vein, a slight suction may be obtained, which facilitates venous return. The parts of the pump which are placed close to the animal in order to avoid dead space in the connections between pump and vessels are: the two flow-valves, glass-tube, which by means of a metal fitting is screwed into the bottom of the air-valve, the two light-bulbs with lenses and the photo-electric cell (see Fig. 1). These parts are mounted on a metal holder and connected with the rest of the set-up by cables. All parts conducting blood can be heated electrically. They may also be kept sterile. We have used the pump with a maximum speed of about 100 strokes per minute, although usually a lower rate was preferred.

This pump resembles the artificial heart of O. S. Gibbs,⁸ as with a constant, but adjustable output per stroke its output per minute depends on the venous return. Unlike the Gibbs-pump it avoids the contact of rubber with the blood. All the major parts used ⁸ O. S. Gibbs, *Jour. Pharm. and Exp. Therap.*, 38: 197.

¹ Aided by a grant from the David Trautman Schwartz Research Fund.