

## Southern Section

*Tulane University, May 17, 1935.*

8145 P

### Experimental Propagation of *Strongyloides* in Culture.\*

TED DE VINNE BEACH. (Introduced by Ernest Carroll Faust.)

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The nematode parasite *Strongyloides simiae* of New World monkeys has as a part of its life cycle a free-living phase outside the body of the host. The eggs laid by the parasitic females are passed in the feces, rhabditiform larvae hatched from them feed on bacteria and debris, and under suitable conditions, metamorphose into free-living, sexually mature forms. These adults mate and produce eggs and larvae supposedly identical with those of the first generation. Such second generation individuals have been considered capable only of developing into filariform larvae. Leuckart's<sup>1</sup> views on the fate of the second generation larvae probably have influenced the general belief that only one sexual generation of the free-living phase could develop. The only statement regarding the probable continued propagation of the free-living phase of this organism was that by Faust,<sup>2</sup> who predicted indefinite free-living development.

By planting a known number of *S. simiae* eggs from the stool of *Cebus capucinus imitator* which harbored the parasitic females, on a culture medium consisting of a nutrient agar, 2 gm., filtered, aqueous extract of monkey feces, 25 cc., and distilled water, 75 cc., it was possible to observe the entire developmental cycle of larvae and adults and their descendants.

In 4 experiments using the above mentioned medium, free-living sexually mature second generation males were produced on 11 cul-

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\* Supported by a grant from the Committee on Scientific Research, American Medical Association.

<sup>1</sup> Leuckart, R., 1882.

<sup>2</sup> Faust, E. C., *Human Helminthology*, Phila., 1929, p. 34.

tures. Five cultures produced second generation females as well. Both sexes of the second generation instead of metamorphosing into filariform larvae retained their rhabditiform characteristics. The males widened posteriorly and spicules appeared. At first no eggs were seen in the young females but with an enlargement of the vulva, eggs appeared in the uterus. In some instances the first generation adults were dead before those of the second generation matured. In most cases, however, the former were present with the latter but soon died while those of the second generation persisted and laid eggs which hatched and developed into third generation filariform larvae. These second generation adults conformed morphologically and physiologically to those of the parent generation.

Unfertilized free-living females, developed on cultures without males, failed to produce viable embryos. It is, therefore, believed that parthenogenesis in the free-living phase does not exist.

In a later experiment 6 cultures produced second generation males and females which, in turn, produced third generation filariform larvae. On 3 of these cultures males as well as filariform larvae of the third generation developed and on one of these, 2 third generation females were present in addition to a male and filariform larvae. Eggs of the fourth generation were found on the sixth and last day. A culture with first, second and third generation sexual forms and larvae of all 3 generations, was fixed with hot 2% formalin and all worms were counted and classified according to the stage of development. There had been planted 9 *S. simiae* eggs which hatched and developed into 4 males, 4 females and 1 first generation filariform larva. On the third day, second generation rhabditiform larvae were present in abundance. The first generation males were dead on the fourth day but the 4 females were still alive and in addition there were 10 males and 6 females of the second generation, the latter laying third generation eggs. These eggs developed into males and rhabditiform and filariform larvae of the third generation. The count demonstrated the presence of 4 eggs, 53 rhabditiform larvae, 22 males, 6 females and 553 filariform larvae, a total of 638. Deducting the parent generation represented by 4 males, 4 females and 1 filariform larva there remain 629 descendants. This does not take into consideration the eggs and larvae which died within the first few days and had disintegrated beyond recognition. As only 6 females, the number corresponding to those of the second generation, were found at the final count, it is believed that the females of the first generation had died

and disappeared entirely. This must have been true for the original males as well, since it was known from previous culture work and controls of the experiment described that they consistently failed to survive as long as the females.

The recovery of over 600 worms could not possibly be accounted for without the intercalation of a second generation of sexual forms, since it was known from previous *in vitro* culture work with this species that the maximum number of offspring per female was 80 and the average 60. Thus 4 first generation females could not conceivably produce more than 320 and probably would not produce over 240 offspring. Considering 60 offspring as an average per female then one would expect 600 from the 4 females of the first generation and the 6 females of the second generation, a figure closely corresponding to the number actually recovered.

It is believed that this continued propagation as demonstrated on culture media occurs under suitable natural conditions. As far as is known this is the first demonstration of continued propagation of the free-living phase of any species of *Strongyloides*.

## 8146 C

### Absorption of Drugs Through the Oral Mucosa. II.

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The relative effectiveness of certain drugs by the sublingual method of administration has been previously reported.<sup>1</sup> In this earlier communication, approximate ratios were determined for effective sublingual doses and the similarly effective subcutaneous doses. In the present report, a similar ratio for 4 other drugs is described. These latter figures were determined principally for the purpose of studying the correlation between oral absorbability and certain fat-water solubility relationships. The general method of experimentation was the same as previously described. All drugs were used in the form of the hydrochloride salt and the dosage expressed on this basis.

*Cocaine.* Using excitement effects as the criterion of absorption,

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<sup>1</sup> Walton and Lacey, *J. Pharm. and Exp. Therap.*, 1935, **54**, 61.