

ovaries, which latter were then stunted and the eggs were not developing. This situation is presumably the start of larval migration from the host. In the four infested males, however, the testes appeared normal and larval eelworms were not observed within these organs.

The larval eelworms showed the beginnings of differentiation into males and females. By analogy with other species of nematode parasites of insects, it appears possible that the larvæ come to maturity in a free-living state, outside the host. Possibly, also, it is the larva of *Drosophila* that is infested by the entry of spermatized but immature female eelworms, which gradually develop during the metamorphosis of the fly. Another nematode, *Tylenchinema oscinellae* Goodey, during the winter months parasitizes the larva of the fritfly (*Oscinella frit* L.) and passes over into the adult fly which emerges in May<sup>1</sup>. It is not known, however, whether larvæ of the above three species of *Drosophila* survive the winter, though Basden (unpublished) finds that *D. silvestris* passes this period in a puparium.

There appear to be two other records of parasitism of *Drosophila* adults by nematodes. Van Zwaluwenburg<sup>2</sup> cites Crawford as having found the larvæ of *Habronema megastoma* (Rud., 1819) (Spiruridae) in an undetermined species of Drosophilid in Ceylon in 1926, and Gershenson<sup>3</sup> found that most individuals of *D. melanogaster* Mg. and all those of *Parascaptomyza disticha* (Duda) (Drosophilidae) caught near Kiev were infested with a species of *Chondronema* (Allantonematidae).

The investigation is being continued in greater detail. We are grateful to A. A. Spence for many of the dissections.

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<sup>1</sup> Goodey, T., *J. Helminth.*, 8, (3), 123 (1930).

<sup>2</sup> Zwaluwenburg, R. H. van, *Bull. Hawaii. Sug. Assoc., Ent. Ser.*, 20, 1 (1923).

<sup>3</sup> Gershenson, S., *Drosophila Inform. Serv.*, 11, 44 (1939).

### A Method of isolating Vesicular Arbuscular Endophytes from Roots

THE exact systematic position of the vesicular arbuscular endophytes has been a topic of some controversy. In 1900 Dangeard<sup>1</sup> proposed the genus *Rhizophagus* for these fungi, and considered that they were closely related to the Endogonaceæ, but that they had lost the ability to form compound fructifications. Peyronel<sup>2</sup> also considered that these endophytes closely resembled *Endogone*.

Nicholls<sup>3</sup>, working in this Department, made a preliminary study of the endophyte present in the roots of *Allium ursinum* and pointed out the morphological similarity of this organism to certain Oomycetous fungi. From portions of root, surface sterilized with a solution of sodium hypochlorite, washed and placed on plain agar, she frequently isolated a strain of *Pythium ultimum*. Owing to the sensitivity of this fungus to the hypochlorite solution, this isolation technique often failed, while less severe surface sterilization of the roots allowed common soil fungi, notably species of *Fusarium*, which were probably

present as surface contaminants, to develop on the isolation plates to the exclusion of *Pythium*.

A more direct method of isolation of the endophyte has now been devised. Roots are thoroughly washed under the tap and then transferred to a flask of sterile water in which they are vigorously shaken for a few minutes. This process is repeated three more times, after which longitudinal sections of the roots are cut under aseptic conditions. The sections are then transferred to a sterile coverslip and covered with a few drops of plain agar which contains 0.2 ml. aureomycin per 15 ml. of medium. The coverslip is then sealed on to a glass ring as in the preparation of a hanging-drop chamber. The sections are examined daily until one of the intercellular hyphæ grows out from the cut edge. This hypha is then picked up and transferred to a plate of plain or malt agar.

The washing treatment is successful in removing most of the contaminating rhizosphere fungi, as roots treated in this way show only occasional growth of common soil organisms, and the position of these on the surface of the section can easily be seen. The addition of aureomycin to the agar inhibits the growth of any bacteria still adhering to the root.

By the use of this method, *P. ultimum* has again been isolated from infected roots of *Allium ursinum*.

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<sup>1</sup> Dangeard, P. A., *Botaniste*, 7 (1900).

<sup>2</sup> Peyronel, B., *Bull. Soc. Mycol. France*, 39, 119 (1923).

<sup>3</sup> Nicholls, V. O. (Mrs. J. Sankey), thesis for the degree of Ph.D., University of Bristol (1952).

### Embryo Development in Areca-nut

EMBRYO development in Palmae is unknown. There is only one description relating to the mature embryo of coconut, by Selvaratnam<sup>1</sup>, who compared it with the embryo of Gramineae. The present communication describes the embryo development in *Areca catechu* L., commonly known as the areca-nut or betel-nut palm.

The first division in the fertilized egg (Fig. 1) is transverse and results in two cells *ca* and *cb* (Fig. 2). The latter divides first by a vertical wall and then in all planes. Its derivatives contribute to the formation of the suspensor, which becomes detached from the embryo proper in advanced stages. The terminal cell *ca* divides by an obliquely vertical wall resulting in two rather unequal cells (Fig. 3), the bigger of which undergoes a further division by a slanting wall. As a result, a triangular cell, the epiphyseal initial (Fig. 4 e), distinguishable from the rest by its shape and position, is formed. The derivatives of this cell give rise to the stem tip, while those of the sub-epiphyseal cells contribute to the formation of the cotyledon and the rest of the embryo proper. During the development of the embryo, there is a marked growth of the cotyledon on one side, resulting in the shifting of the position of the stem tip as shown in Figs. 5-10. As the embryo proper is exclusively derived from the products of *ca*, the embryo development in *Areca catechu* conforms to the onagrad type of Johansen<sup>2</sup>. So far the formation of an epiphysis in any monocotyledon showing the onagrad type is not known.