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PARTHENOGENESIS IN A CYST-FORMING NEMATODE, HETERODERA TRIFOLII (NEMATODA: HETERODERIDAE)[1]

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Abstract

The clover cyst nematode, *Heterodera trifolii* (Goffart, 1932) Raski and Hart, 1953, reproduced in the absence of males. Under greenhouse conditions nematodes reared from single larvae and from mass cyst culture were diploid-parthenogenetic. During maturation only one polar body was produced. The diploid number (24 ?) of chromosomes was not reduced and no male was found.

Introduction

The clover cyst nematode, *Heterodera trifolii* (Goffart, 1932) Raski and Hart, 1953, which attacks several cultivated legumes, is fairly widespread in Canada (7). Examination of a large volume of infected plant material from the greenhouse at Ottawa and from fields near Ottawa revealed no males on the roots. This agrees with Hirschmann (4), Gerdemann and Lindford (3), and Raski and Hart (10). However, Franklin (2) and McBeth (5) described males of this species. Males are abundant in populations of the soybean cyst nematode, *H. glycines* Ichinoe, 1952, a closely related form, and also in the sugar-beet nematode, *H. schachtii* Schmidt, 1871.

This is a report on oogenesis and reproduction in *H. trifolii*.

Materials and Methods

The material used was identified by the writer as of *H. trifolii* by host preference (white Dutch clover was heavily attacked, and extensive tests showed that the nematode would not attack red beet or rutabaga root), by the yellow phase during cyst formation, by fenestral length, and by underbridge size and structure.

An ample supply of gravid white females was made available for mass cultures of the nematode on the roots of white Dutch clover, *Trifolium repens* L., grown in the greenhouse at Ottawa. Cultures from single larvae were reared as follows: White Dutch clover seed was sown in flats containing sterilized soil, and after a 2-week growing period single plants were transferred to 250-ml. beakers containing sterilized sand. Each plant was then carefully inoculated with a single active second-stage larva by placing the larva close to the plant roots. After 10 days the plants were removed from the beakers, carefully washed under running water, and replanted singly in glass jars (250 ml.) containing vermiculite. A nutrient solution was added to each jar to maintain growth. The level of this solution was maintained by adding solution as required. The live female nematodes were prepared for study by the squash technique and in the manner described by Mulvey (6, 8).

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Oogonial Development

Oogonial development follows closely that described by Mulvey (6) for a species of *Heterodera* (probably *H. trifolii*) on hairy vetch (*Vicia villosa* Huds.), except that no prochromosomes were observed.

Oocytes

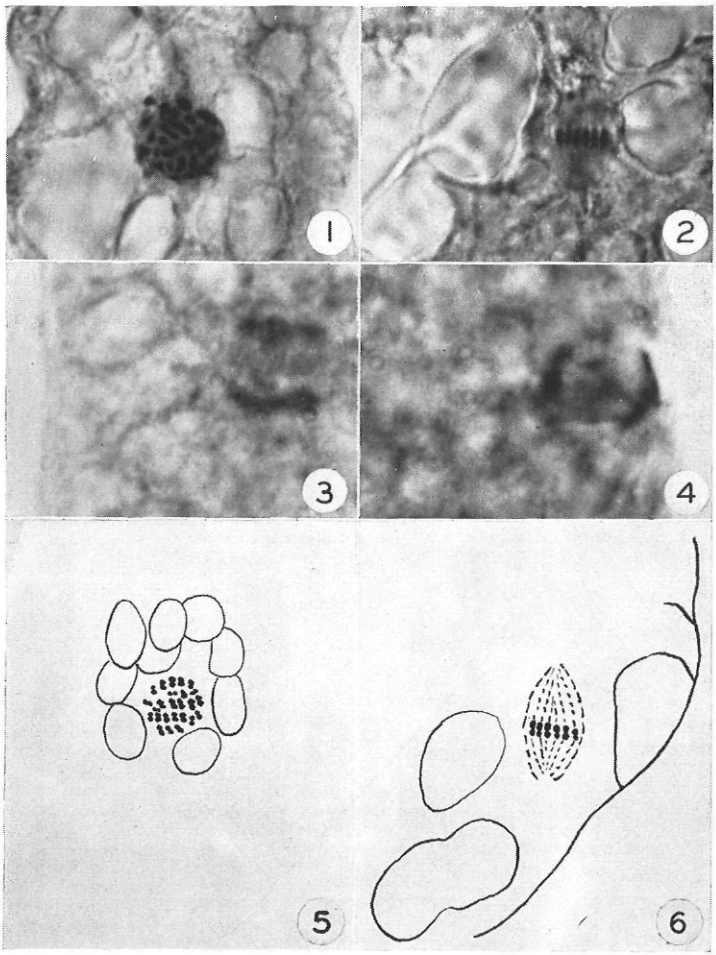
Oocytal development also follows closely that described by Mulvey (6) in the species of *Heterodera* on hairy vetch. After completion of the growth period the chromatin is organized into discernible chromosomes. Duplication of each chromosome becomes discernible with no pairing of the homologous chromosomes. Therefore, only the dyad condition is reached (Figs. 1, 2, 5, 6). Mulvey (8) reported that the homologous chromosomes pair in the females of the bisexual nematode *H. schachtii*. The achromatic material is well defined (Fig. 2) and the cytoplasm of the egg densely vacuolated. During early metaphase the spindle and its nuclear elements lie at the center of the egg. Eventually the chromosomes separate (Fig. 3) and the spindle, which is normally parallel to the long axis of the egg, rotates through an angle of 90° and comes to rest with one of its poles against the cell membrane (Fig. 4). Only one polar body is produced and the diploid number of chromosomes is not reduced. The chromosomes are very small and, therefore, accurate counting was not possible. However, the diploid number appears to be at least 24.

Although many females were examined no sperm was found. Cultures from single larvae, each of which developed into a gravid female in the absence of males, in turn produced gravid females. Second-generation females contained no sperms.

Discussion

These studies indicate that *H. trifolii* is capable of reproducing parthenogenetically, at least under greenhouse conditions. According to Walton (11), only in rare cases does the nematode egg develop without fertilization by the sperm. Belar (1) reported that two parthenogenetic species of *Rhabditis* show a single maturation division and no reduction in the chromosome number. Mulvey (6) described a species of *Heterodera* in which the chromosome number was not reduced during maturation division but he did not observe polar body formation. Hertwig (cited by Walton (11, p. 208)), investigating a dioecious culture of *Rhabditis pellio* (Schneider, 1866) Butschli, 1873, found a mutant that produced only one polar cell without reduction and, therefore, retained the diploid number of 14 chromosomes. None of the eggs developed without sperm being present in the egg although the sperm did not enter the cleavage nucleus. Nigon (9) reported that in the free-living nematode *Rhabditis bellari* Nigon, 1949 some eggs undergo two reductional divisions and their pronuclei fuse; these produce males and females. Other eggs, which produce only females, form a single polar body; at the same time the bivalents divide twice and in these pseudogamous eggs development proceeds from the diploid female nucleus, the sperm remaining inert.

PLATE I



FIGS. 1–4. Photomicrographs showing maturation stage in a cyst-forming nematode, *Heterodera trifolii*. 1500×

1. Polar view of early metaphase stage in primary oocyte showing dyads. 2. Side view of metaphase stage showing achromatic material and densely vacuolated cytoplasm of the primary oocyte. 3. Side view of early anaphase stage in primary oocyte. 4. Side view of late anaphase stage of primary oocyte. The polar body is produced at this stage.

FIGS. 5–6. Camera lucida drawings showing finer details in Figs. 1 and 2. 1500×

Acknowledgments

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