

*Plant Parasitic Nematodes*

*VOLUME II*

**Cytogenetics, Host-Parasite Interactions,  
and Physiology**

# *Plant Parasitic Nematodes*

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## Preface

This two-volume treatise was written to provide an up-to-date reference source for students, teachers, and research and extension workers in plant nematology and related fields. Nematological advancements made since the publication of a similar book approximately ten years ago are discussed. A high proportion of the available knowledge obtained during this time has been in such important areas of nematology as ultrastructure, enzymology, chemistry of body composition, culturing, virus transmission, biological races, and nature of plant resistance. Thus, this is the first comprehensive reference work in nematology to include information from these new areas as well as from traditional ones.

An attempt has been made to coordinate and evaluate the phenomenal amount of research data of these years. In order to include the best possible coverage of the many diverse and specialized topics, a number of authors were invited to contribute to the text; many are actively engaged in the field about which they have written. Although each chapter was edited, the data and opinions expressed are those of the contributors.

Volume I includes a discussion of the history of plant nematology, the current status of research, and information pertaining to professional societies and publications. It also deals with nematode morphology, anatomy, taxonomy, and ecology, emphasizing plant parasitic forms and, where pertinent, drawing examples from free-living and animal parasitic nematodes.

Volume II deals with plant parasitic nematode genetics and cytology, host-parasite interactions, biochemistry, and physiology. As in Volume I, useful information relating to free-living and animal parasitic nematodes is included.

We wish to thank the authors for the considerable time spent in preparing their contributions. Such comprehensive treatises of important areas of plant nematology are invaluable to progress in this biological discipline. In fact, without them it would be difficult or impossible for students to become familiar with and research workers to keep abreast of the knowledge in specific areas.

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# Genetics and Cytology

## CHAPTER 13

### Genetics and Cytology

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#### I. INTRODUCTION—HISTORICAL REVIEW

Most of the early cytogenetic work in nematodes involves animal parasitic and free-living forms and was conducted in the late part of the nineteenth century and the early part of this century. Some of this work is still considered as classic because it elucidated certain basic cytological and biological phenomena that were incomprehensible until that time. Thus, Bütschli (1873) observed that two nuclei are present in fertilized eggs of *Caenorhabditis dolichura* which unite in the center of the egg to

produce the nucleus of the first cell of the future embryo. This observation was one of the first steps toward understanding the "role of fertilization in reproduction," which was completely elucidated 2 years later by Hertwig in eggs of the sea urchin. Probably, the most significant contribution of nematode research in the field of cytology and heredity came a few years later when van Beneden (1883) discovered the process of "meiosis" occurring during maturation of the eggs of *Parascaris equorum* (*Ascaris megalocephala*). He demonstrated that during the formation of the polar bodies the chromosome number of the egg is reduced to one-half (meiosis) and that this number is doubled again in the cleavage nucleus which is formed by the fusion of the egg and sperm pronuclei (fertilization). Van Beneden (1883) and Boveri (1888) also introduced in cytology for the first time the concept of individuality and physical continuity of the chromosomes by demonstrating that *Parascaris* chromosomes persist during interphase. They observed that blastomere nuclei of *Parascaris equorum* eggs show a number of finger-shaped lobes which are formed at telophase by the free ends of their V-shaped chromosomes. During prophase of the following division the chromosomes reappear with their free ends lying in these lobes, indicating that chromosomal basic organization does not change during interphase when the chromosomes are not visible.

The processes of chromosome fragmentation and chromatin diminution were first discovered in nematodes. Boveri (1887) observed that the early cleavage divisions in *Parascaris equorum* eggs were unusual in several respects. The zygote nucleus of this nematode has four long chromosomes, each with a thin (euchromatic) central part, and two thicker (heterochromatic) club-shaped ends. During the first cleavage the chromosomes divide normally, mitotically. During the second cleavage, one of the blastomeres— $P_1$  or propagation cell—divides mitotically, but the other— $S_1$  or somatic cell—undergoes a peculiar division. Each of its chromosomes fragments into the two club-shaped ends and a number of small chromosomes that are derived from the thin central part (chromosome fragmentation). The small chromosomes then divide mitotically and the sister chromosomes migrate toward the spindle poles. The club-shaped ends, however, remain in the middle of the spindle and eventually are left in the cytoplasm of the daughter blastomeres where they degenerate (chromatin diminution). The process of chromosome fragmentation and chromatin diminution is repeated in one of the progeny of the P-cell line till the fifth cleavage. Eventually, all the cells of the embryo are diminished, except the last two P cells that will be enclosed in the genital primordium of the larva and will later give rise to all the gonial cells of that individual.

Because animal parasitic nematodes were recognized early as favorable material for cytological work, they were the second animal group after the insects to be studied extensively with regard to chromosomal mechanisms of sex determination. The X-O and multiple X chromosome situations were discovered in various nematodes as early as 1910 by Boveri, Gulick, Edwards, and others.

Nematodes provided also the first example of "gynogenesis" (pseudogamy or pseudofertilization) as a normal, nonpathological method of reproduction in animals. Krüger (1913) described this peculiar method of reproduction in the hermaphroditic *Rhabditis aberrans*. She observed that the sperm penetrates and thus activates the oocytes for further development but subsequently degenerates in the cytoplasm without fusing with the egg nucleus. Gynogenesis was confirmed later and demonstrated beyond doubt in a mutant line of *Rhabditis pellio* by Hertwig (1920) and by various workers in other nematodes and other animals.

Although these classic studies indicated that nematodes are favorable for karyological research, relatively little work followed. Most of this work involved studies on gametogenesis and the chromosomes of animal parasitic nematodes (Walton, 1924) and studies on gametogenesis, reproduction, and sexuality of free-living soil forms (Nigon, 1949).

The first attempt to study gametogenesis and the chromosomes of plant parasitic nematodes was made by Mulvey (1955). Although part of the early work is not very accurate, it indicated the potential usefulness of such studies in demonstrating cytogenetic differences among related nematodes. Cytogenetic work by various workers since 1960 has added much information regarding gametogenesis, chromosome numbers, mode of reproduction, and sexuality of plant parasitic nematodes. Purely genetic studies apparently have been hindered by difficulties involved in breeding experiments with plant parasitic nematodes and the almost complete absence of distinct morphological and physiological characters that could be used as genetic markers for genetic analysis. Therefore, this chapter is limited to an analysis of gametogenesis, reproduction, sexuality, and the cytogenetic aspects of evolution of plant parasitic nematodes. Some relevant information about common free-living soil nematodes is also included.

## II. GAMETOGENESIS AND CYTOLOGICAL FEATURES OF REPRODUCTION

Most species of soil and plant parasitic nematodes are bisexual, i.e., have males and females easily recognizable by primary and secondary



sex characters. Species in which males and females appear in approximately equal numbers usually reproduce by amphimixis (cross-fertilization). In some species males appear in relatively small numbers, are rare, or absent. Reproduction in such species is usually by parthenogenesis. A number of species are hermaphroditic and reproduce by automixis (self-fertilization). However, most hermaphroditic species, besides the hermaphrodites, also have a variable number of males (incomplete or unbalanced hermaphroditism), which may or may not be functional. When the males are functional, reproduction is partially by automixis and partially by amphimixis. Some hermaphroditic and some bisexual species are pseudogamous (see cytological features of pseudogamy, Section II, E). Detailed accounts of gametogenesis with emphasis on free-living soil nematodes can be found in Nigon's (1965) extensive treatise. Reproduction of plant and soil nematodes has been reviewed by Triantaphyllou and Hirschmann (1964). Also, reviews of gametogenesis, particularly of animal parasitic nematodes, have been presented by Walton (1940, 1959). In the following, a general account of gametogenesis in free-living and especially in plant parasitic nematodes will be given. *Anguina tritici* will be used as the main example since it is amphimictic and the only plant parasitic nematode in which both oogenesis and spermatogenesis have been studied in detail (Triantaphyllou and Hirschmann, 1966).

### A. Oogenesis

Oogenesis in soil and plant parasitic nematodes follows the same general pattern known in most animals, with slight deviations in regard to the behavior of the chromosomes at synapsis and during meiotic prophase.

Multiplication of oogonia occurs in the apical, "germinal" zone of the ovary (Fig. 1). Oogonial cells are straight line descendants of the "propagation," P cells of the embryo and appear to be set aside from the remaining somatic cells during the early cleavage divisions. Oogonial divisions usually start in third-stage larvae and continue up to the early adult stage. Most divisions, however, occur during the fourth larval stage and the fourth molt. They are normal mitotic divisions and result in the production of a large number of oogonia, all of which have the somatic ( $2n$ ) chromosome number. Oogonial divisions appear to be synchronized in some species (*Anguina tritici*, Fig. 1A), i.e., all oogonia of a region of the germinal zone may undergo division at the same time but occur with no particular order in others (*Meloidogyne* and *Heterodera*). The chromosomes in oogonial divisions of most soil and plant parasitic nematodes usually are not discrete (*Heterodera*, *Pratylenchus*, *Caenorhabditis*, and