

Biology of banana population of *Heterodera oryzicola* (Nemata : Tylenchina)

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Summary – The biology of *Heterodera oryzicola* (banana population) studied on banana (*Musa* AAB cv. Nendran) had shown that the second stage juvenile invade the tertiary tender roots, feed and develop to maturity and reproduce. The infective juveniles were also observed to develop into adults after they had partially penetrated the root tissue. The period of one life cycle at 27 °C from egg to egg stage is 23 days while from the infective juvenile to the emergence of second stage larvae it is 25 days. The second stage juveniles after invasion develop into males and females in 12 days and in 17 days respectively. The average number of eggs produced per individual adult female is 197 in the cyst body and 88 in the egg sac.

Résumé – *Biologie de la « population bananier » de Heterodera oryzicola (Nemata : Tylenchina)* – L'étude de la biologie de la « population bananier » de *Heterodera oryzicola* – sur bananier (*Musa* AAB cv. Nendram) – montre que les juvéniles de deuxième stade (J2) envahissent les racines tertiaires tendres et que le nématode s'y nourrit jusqu'à la maturité et s'y reproduit. Il a été observé que les J2 se développent jusqu'aux stades adultes après n'avoir pénétré que partiellement dans les tissus racinaires. La durée du cycle, d'œuf à œuf, à 27 °C, est de 23 jours tandis que la durée entre la pénétration des J2 et la sortie des J2 de deuxième génération est de 25 jours. Les J2 produits conduisent à des mâles et des femelles 12 et 17 jours, respectivement, après qu'ils ont pénétré dans les racines. Le nombre moyen d'œufs est de 197 pour le kyste et 88 pour la masse d'œufs.

Key-words : Nematodes, *Heterodera*, life cycle, banana, *Musa*.

Charles and Venkitesan (1993) reported the pathogenicity of *Heterodera oryzicola* Rao & Jayaprakash, 1978 on banana (*Musa* AAB) cv. Nendran from the state of Kerala, India. Though the biology of *H. oryzicola* in rice was reported by Jayaprakash and Rao (1982), no such studies were made with the banana population. Considering the potential importance of *H. oryzicola* which attacks banana in Kerala, the present investigations were undertaken to study its life cycle and biology on banana cv. Nendran.

Material and methods

Cysts were hand picked from a pure culture of the nematode maintained on live banana plants and crushed using a cyst crusher and the eggs liberated into water at 25 °C. From this suspension single celled eggs were pipetted out using a micropipette. These eggs were kept in cavity blocks and incubated in tap water. Every 24 hours, a few eggs from the cavity blocks were picked out and placed in a cavity slide in distilled water and examined under a binocular microscope at 400 × for the development of the juvenile stage.

Cement tubs of size 75 cm × 120 cm (500 dm³ capacity) buried in soil at a spacing of 2 m × 2 m were filled with soil sand mixture of 2 : 3 proportion denematized

with 5.0 % formaldehyde and aerated for 10 days after 72 hours of exposure. Peeper banana suckers of *Musa* AAB, cv. Nendran collected from mother plants were pared and treated with hot water (55 °C for 10 min). One such treated sucker was planted in each cement tub. Healthy lateral roots from these suckers with secondary, tertiary and higher order feeder roots developed were exposed after one month of planting, and were placed in longitudinally split bamboo stakes, filled with the same soil and allowed to grow further (Fig. 1). These roots grown in bamboo splits were inoculated with 250 freshly hatched second stage juveniles near the feeder roots and properly marked with labels. Such inoculated tertiary lateral feeder roots were dissected out from the plant every 24 hours, washed free of soil particles and stained with 0.1 % boiling acid fuchsin in lactophenol. The different stages collected from roots were examined for the development and camera lucida drawings were made. The different stages were dissected out of the root pieces and slides were prepared for measurements. The duration taken for the development of each juvenile stage was also recorded. During the period of investigation the temperature of the soil at 15 cm depth in cement tubs around bamboo splits was maintained at 27 ± 2 °C by shading with dried coconut leaves.

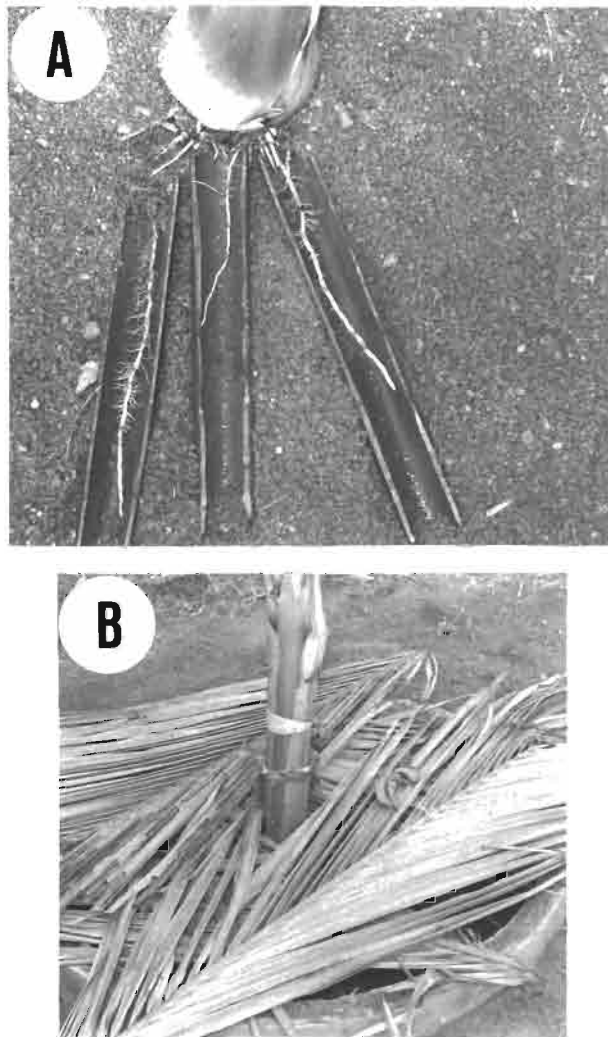


Fig. 1. Inoculation technique. *A* : Isolated roots with feeder rootlets placed inside longitudinally split bamboo and filled with denematized soil; *B* : After inoculation with nematode juveniles : root areas covered with sand, sprinkled water and shade provided with dried coconut leaves to avoid drying.

Results

INCUBATION PERIOD

Eggs of the unicellular stage were found to develop into the first juvenile stage with prominent spear 96 hours after placing them in tap water. The second stage juveniles emerged from the egg shell after 144 hours of development of first stage.

PENETRATION ALIGNMENT AND DEVELOPMENT

Details on the life cycle of *H. oryzaicola* on banana are presented in Fig. 2. The freshly hatched juveniles started penetration into feeder roots within 48 hours (L_1 ;

Fig. 2). Juveniles were found penetrating at the region of the growing tip and through the entire portion of the fine rootlets. The juveniles penetrated into the root tissues and oriented to a parallel position on the long axis of the roots with their heads attached perpendicular to the stele (L_2 ; Fig. 2). In several cases, the juveniles positioned their head after penetration towards the stelar tissue, without actually drawing their posterior body portion completely inside the roots. In such individuals the body thickness increased, with the posterior tail portion of juveniles protruding outside the roots (L_3 ; Fig. 2). On the fourth day after inoculation, the juveniles were found to increase in body width and the cuticle at the tail region loosened and started shedding towards the anterior exposing a small portion of blunt tail end posteriorly. The development of genital primordia was seen clearly on the sixth day after inoculation, indicating the early third juvenile stage of the nematode (L_3 ; Fig. 2). On the seventh day, sex difference was distinguishable. In certain juvenile stages the developing gonad was observed to be single, straight and increasing in length posteriorly and finally joined with the rectum, indicating their development into males (M_1 ; Fig. 2). The third moult of male juvenile began on the eighth day. The fourth stage juvenile with formative stage of vermiform body was observed on the tenth day. Development of the adult male was observed on the eleventh day. The moulting fourth stage male juvenile in coiled position was seen inside roots on the twelfth day after inoculation (M_2 ; Fig. 2). The fully developed adult males were recovered from the soil on the fourteenth day after inoculation (M_3 ; Fig. 2). In certain cases of developing juveniles the genital primordia were bifurcated and placed close to the posterior end of the body indicating them to be developing into females. The third-stage female juvenile with "V" shaped branched genital structures at the posterior end was observed on the ninth day after inoculation (F_1 ; Fig. 2). The moulting of the third female juvenile was observed on tenth day. These female juveniles were found to develop a slightly swollen body on eleventh day. After two days, the fourth-stage female juveniles transformed into a flask-shaped body, with well developed reproductive structures. On the fifteenth day, the developing females underwent the fourth moult. Two days later these became swollen white lemon-shaped females with a few eggs laid inside their body and their posterior part protruding by rupturing the roots (F_2 ; Fig. 2). On the nineteenth day, large gelatinous egg sacs were found formed and attached to each developed adult female. Eggs in the egg sac were observed on the twentieth day (F_3 ; Fig. 2). Adult females turned into light-brown cysts with eggs from the 23rd day changed into dark brown to black colour by 30th day (F_4 ; Fig. 2). Second-stage juveniles were recovered from the soil, on the 25th day. The average number of eggs in the posterior egg sac and cyst body was 88 and 197, respectively (average of ten replicates).

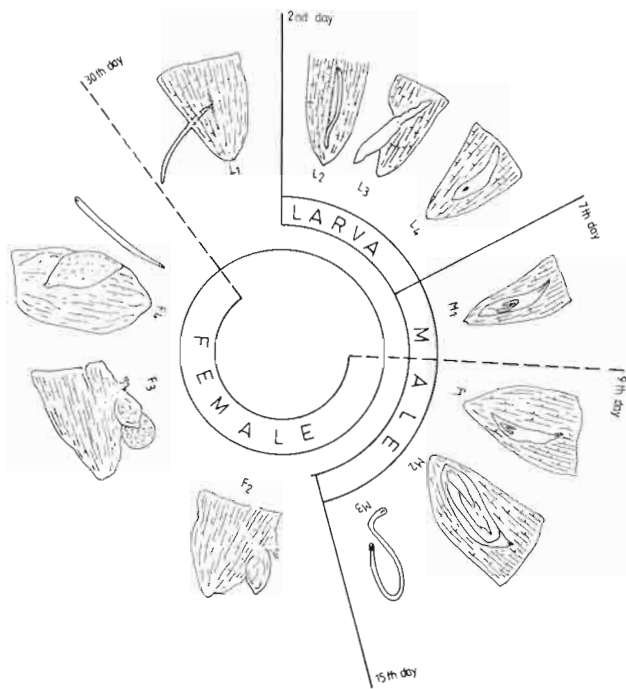


Fig. 2. Life cycle of *Heterodera oryzicola* on banana.

The development of male was completed on the twelfth day and the adult females in seventeenth day after inoculation of second stage juveniles. The total development period from egg to egg was 23 days and second stage juveniles to emergence of second stage juveniles was 25 days. For first the time, *H. oryzicola* was observed to successfully develop and reproduce as a semi-endoparasite on banana roots.

Discussion

Reciprocal matings between the banana isolate and *Heterodera oryzae* has been studied by Taylor (1978). He confirmed that the isolate from banana is conspecific with *H. oryzae*. Jayaprakash and Rao (1982), have reported the embryonic development of *H. oryzicola* isolate from rice and its emergence from egg masses was completed within 8 to 9 days at $28 \pm 1^\circ\text{C}$. Further, seeds of rice were germinated individually in plastic pots (6×6 cm) containing 100 g of soil with 40 ml water and when sprouts were 10 days-old, each was inoculated with 100 second stage juveniles. Adult males developed in 14 days and females in 22 days. Thus one life cycle of *H. oryzicola* on rice from egg to egg was completed in 30 days. In the present study, it was found that the emergence of larvae from egg occurred in six days and the development of larvae to adult females with eggs in seventeen days, with a total of 23 days to complete one life cycle from egg to egg stage. Generally, the duration

of one life cycle of *Heterodera* species varies from 16-32 days. *H. lespedezae* completed one life cycle in 26 days in roots of *Lespedeza striata* at $22-26^\circ\text{C}$ (Bhatti, 1975). *H. graminophila* in roots of *Echinochloa colonus* seedlings took 18 days for female development and lemon-shaped cysts emerged through the epidermis after 24 days (Birchfield, 1973). *H. zae* on maize at $27-38^\circ\text{C}$ (Srivastava & Sethi, 1985) took 16 days for one life cycle, while *H. sorghi* on maize took 24 days at $28-36^\circ\text{C}$ (Srivastava & Chawla, 1991). The life cycle duration can also vary within 10 or more days when situations under which it has been studied differ considerably. Thus, also, when different hosts were used for biology studies, the life cycle duration was found to fluctuate (Kalha & Edward, 1979). The shorter period taken for completion of one cycle in banana, indicated in the present study, as against the longer period reported in the case of the same nematode in paddy by Jayaprakash and Rao (1982) is explainable by the influence of different hosts. During the development of the second-stage juveniles into subsequent stage, it was found to moult from the posterior part to anterior region contrary to the normal behaviour of nematodes moulting from anterior region. This is similar to the report of Mulvey (1959) who observed the same phenomenon in the case of *H. trifolii*. Normally, *Heterodera* species are considered to be endoparasites which complete their development inside the root tissue after the invasion and penetration of the infective second stage juvenile. However, in the present case, the nematode was found to develop into adults after partial penetration of the infective juveniles, thus exhibiting the characteristics of a semi-endoparasite. This sort of phenomenon has also been reported by Koshy and Swarup (1971), in *H. cajani* infesting the pigeon pea. "As a part of the moulting process took place outside the root of banana instead of inside the root of rice, the time required was lessened because the mechanical strain of the root was exerted only on a restricted length." The egg production has been found to differ slightly from that reported by Jayaprakash and Rao (1982) in *H. oryzicola* on rice. They reported an average of 120 and 198 eggs in the cysts and egg sac, respectively, compared to 197 in the cysts and 88 in egg sacs in the present study. This variation may be ascribed to the effect of different hosts.

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