

development of the syncytium was not impeded as no degenerate syncytia were seen.

It is recognised that the cellular responses of partially or completely resistant cultivars can take different forms according to their complement of resistance genes (Rice & Stone, 1983). However, previous studies have concluded that all susceptible cultivars show similar cell responses (Hoopes, Anderson & Mai, 1978) viz. an insensitivity to tissue damage with necrosis confined to the cells adjacent to the nematode and with no necrosis around the syncytium, although Huijsman, Klinkenberg and Den Ouden (1969) noted greater or lesser amounts of necrosis around syncytia when they degenerated after the nematodes completed their development. In the present experiments Pentland Javelin exhibited the typical susceptible response. However, the susceptible cultivars Maris Peer and Maris Anchor both showed extensive necrosis in the tissues surrounding the nematode; both of these cultivars also have a lignified hypodermis and produce lignitubers in response to invading fungal hyphae. Pentland Javelin does not have a hypodermis and although it does produce lignitubers, it is not so prone to produce a hypersensitive response to the nematode.

A range of responses to nematode invasion is already recognised in resistant cultivars; apparently, a range of responses may also occur in susceptible cultivars.

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## INCREASE OF THE CHEMICAL OXYGEN DEMAND DURING THE GROWTH IN *HETERODERA SACCHARI*

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In nematodes species which remain vermiform at any stage of development, quantitative study of the growth may be approached by measuring increases of length and width (Bird, 1971; Ohba & Ishibashi, 1981). In the case of species which become swollen at later stages, such as *Meloidogyne javanica*, Bird (1959) showed that measurement of area with the camera lucida gave a better appreciation of the increase in size. This author however suggested that "measurements of volume or weighing would give better representation of the rate of growth. The size of these creatures predisposes against these techniques, however". In the present work, I

measured the growth of females of *Heterodera sacchari* by means of the chemical oxygen demand (COD), that is the oxygen required for the complete oxidation of the organic matter in the nematode with an oxidizing chemical (Reversat, 1981a)

### Material and methods

*Heterodera sacchari* was reared on two cultivars of rice (*Oryza sativa*): Morobérékan, traditional in Ivory Coast, and IR 1529, introduced from the International Rice

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Research Institute (IRRI) in Philippines to the Senegal. Two weeks old seedlings, grown in sterile soil, were transferred in hydroponic culture in 2 liter jars filled with a mineral nutritive solution (Hoagland & Arnon, 1950) diluted to 1/4 of its normal strength. Jars were maintained in a water bath at 28° and the solution aerated by air bubbling. After two weeks, when the root system was fully developed, the rice plant was taken out of the jars and the root system spread out on a thin layer of sterile soil in a plastic tray. The roots were immediately covered with another thin layer of sterile soil and watered. Each tray containing one rice plant was inoculated with 20 000 freshly hatched juveniles of *H. sacchari*. Juveniles were hatched from crushed cysts using potassium permanganate (Reversat, 1981b) and the inoculum was spread all over the area occupied by the root system. Trays were covered with an opaque plastic plate to prevent water loss and the effect of light. Three days later, roots were washed from soil with water and the plants were returned to hydroponic culture as described above. One, 2, 3, 4 and 5 weeks after the inoculation, the nematodes were extracted from the roots of two rice plants. Roots were placed on a 2 mm stainless steel sieve fitted on a 0.16 mm sieve. Females were dislodged from the roots using a fine and pressurized stream of water and recovered on the bottom sieve. Under the binocular, females were hand picked one by one and placed in samples tubes (Reversat, 1976) filled with distilled water. In order to improve the randomization of samples, five females were introduced in the first tube, five females in the second tube, etc., until six tubes of 25 females were obtained. At the first sampling date (one week), no females were available. When females settled at the bottom of sample tubes, the excess of water was removed and the tubes, were stored for 5 weeks at - 18° until the COD determination. COD of freshly hatched juveniles of *H. sacchari* was determined on six samples of 20 000 juveniles prepared from a suspension by the drop by drop method (Reversat, 1980). COD of the 54 tubes was determined as previously described (Reversat, 1981a). Results were expressed as  $\mu\text{l O}_2/\text{individual}$  (juvenile or female).

## Results

The COD of freshly hatched juveniles of *Heterodera sacchari* was 0.042  $\mu\text{l}/\text{indiv.}$  During the growth, the COD increased tremendously to 58.1  $\mu\text{l}/\text{indiv.}$  at 4 weeks on IR 1529 and 44.8  $\mu\text{l}/\text{indiv.}$  at 5 weeks on "Morobérékan" (Fig. 1). The ratio between the maximum value of the COD and the initial value of the COD (in freshly hatched juveniles) was 1 383 on "IR 1529" and 1 067 on "Morobérékan". With the area method, Bird (1959) observed a ratio of 32 between a fully developed female and a juvenile. This discrepancy

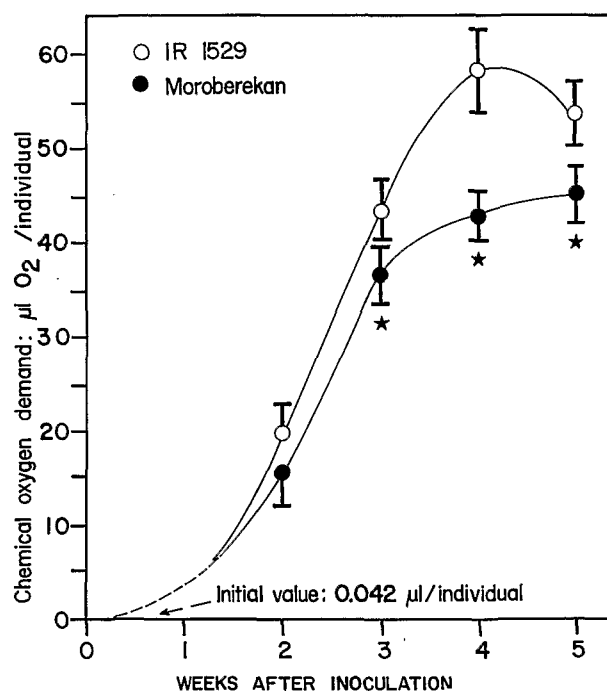


Fig. 1. Increase of the chemical oxygen demand during the growth in individual *Heterodera sacchari* on two rice varieties, IR 1529 and Morobérékan. (Each point represents the mean of six replicates of 25 individuals and the vertical bar equals the standard deviation of the mean; curves are fitted by eye; stars indicate the time where COD are significantly different between the two cultivars.)

can be explained by the following calculations. According to the mensurations of the *Heterodera sacchari* juveniles (Luc & Merny, 1963 : an average length of 480  $\mu\text{m}$  and an average width of 18.5  $\mu\text{m}$ ) and the formula of Andrassy (1956), the average volume of a *H. sacchari* juvenile was  $91.5 \times 10^3 \mu\text{m}^3$ . *H. sacchari* females were typically lemon shaped with an average length of 645  $\mu\text{m}$  and an average width of 445  $\mu\text{m}$ . Thus the volume of the average female can be estimated as the volume of the sphere with a diameter equal to the mean between the length and the width of the average female : 550  $\mu\text{m}$ . The ratio between this calculated volume ( $82.5 \times 10^6 \mu\text{m}^3$ ) and the volume of the juvenile calculated above ( $91.5 \times 10^3 \mu\text{m}^3$ ) equaled 952, which was of the same magnitude as the observed ratio for COD. Moreover, fully developed females of *H. sacchari* were filled with eggs for which, in some related species as *Meloidogyne arenaria*, lipid content was 40 % higher than in the juvenile (Krusberg, Hussey & Fletcher, 1973). Since the COD coefficient for lipids was two times as high as the COD coefficient for carbohydrates or proteins (Reversat, 1981a), the high value of the ratio, from 1 000 to 1 400, was justified. On

the other hand, the area method was applied to the average juvenile and the average female of *H. sacchari*. The area values were of  $237.6 \times 10^3 \mu\text{m}^3$  for the female and of  $8.9 \times 10^3 \mu\text{m}^2$  for the juvenile and the ratio equaled 26.7, which was very close to the value of 32 observed by Bird (1959).

Results were different on the two varieties. On "IR 1529", the development was more rapid than on "Morobérékan" and the maximum value of COD was higher. This effect of host varieties on the feeding of the parasite has been observed in *Heterodera avenae* by Cook (1977).

Between 4 and 5 weeks, the COD decreased on "IR 1529". This was related with the presence of some eggs in the nutritive solution of the jars. Thus, eggs formed by this species may be partly laid freely in soil. This decrease of COD could be also related with chemical changes occurring when females become cysts, especially the tanning of the cuticle, which involves a large oxygen uptake in *Globodera rostochiensis* (Hominick, 1983).

There was not enough material produced to evaluate the egg content of females at each time. This was done only at 5 weeks for "IR 1529". Five batches of 30 females were recovered and kept for 4 weeks in a 0.3 M NaCl solution, which inhibits hatching but allows the development (Dropkin, Martin & Johnson, 1958). The cysts were crushed and the juveniles hatched with potassium permanganate (Reversat, 1981b). The individual cyst content had an average of 327 juveniles (standard deviation : 41). In the COD value of the female, the part due to the eggs was  $13.8 \mu\text{l}$  ( $327 \times 0.042$ ), which represents 26 % of the total value.

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HOST RANGE OF *ANGUINA AMSINCKIAE* WITHIN THE GENUS *AMSINCKIA*

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*Anguina amsinckiae* (Steiner & Scott, 1934) Thorne, 1961 is a potential agent for biological weed control and is known to have three hosts under natural conditions : *Amsinckia intermedia* Fischer & Meyer, *A. lycopsoides*

Lehmann, and *A. gloriosa* Suksdorf (Pantone, Griesbach & Maggenti, 1986). The genus *Amsinckia* has four sections (Ray & Chisaki, 1957a,b,c), and all known hosts are either in the Muricatae or the Tessellatae

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