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RESEARCH ON THE BIOLOGY AND ECOLOGY OF *PANAGROLAIMUS RIGIDUS* (SCHNEIDER) THORNE VI. THE INFLUENCE OF THE POPULATION'S ORIGIN AND BREEDING CONDITIONS ON MORPHOMETRIC FEATURES

ABSTRACT: Considerable differences were observed as regards morphometric features of individuals in populations of nematodes of the species *Panagrolaimus rigidus* (Schneider 1866) Thorne 1937 differing as to their origin (isolated from soil under various cultures). The breeding conditions (agar + yolk, or soil with cereal seedlings) and duration of the laboratory culture also influenced the size of nematodes. Temperature affected the size of *P. rigidus*, whereas moisture had no effect.

Contents

1. INTRODUCTION

As a result of research on the effect of various factors on the morphometric features of nematodes their usefulness as systematic criteria is questioned quite frequently by many scientists. According to Clark (1962) the indices of de Man, used so far in classification of nematodes: a – ratio of body length to body width, b – ratio of body length to oesophagus length, c – ratio of body length to tail length and V – percentage of the head-vulva distance to body length, should be very carefully used.

Hooper (1969) has reached similar conclusions, and on the basis of results of many authors on various species gives examples of considerable variability in body dimensions of

nematodes under different environmental conditions. Hooper doubts the usefulness of indices *a*, *b*, *c* and *V* in systematics because of the variability of features forming these indices.

According to Geraert (1968) there is no correlation between the body length and width and between the length of body and length of oesophagus, and indices *c* and *V* can be only applied in some cases.

Author's study (Kozłowska and Mianowska 1971) on the effect of medium in indices *a*, *b*, *c* and *V* in a nematode of the species *Panagrolaimus rigidus* (Schneider 1866) Thorne 1937 has also shown a lack of correlation between the measurements forming these indices. Index *V* has been the most constant.

Results of studies of Frederick and Tarjan (1975) point to a considerable variability of indices: *a* and *c* for nematodes of the species *Xiphinema krugi* Lordello 1955 belonging to different populations. More constant are indices *b* and *V*.

The reasons for morphological variability can be very different. Thorne and Allen (1959 after Tarjan 1967) consider the morphological variability as a cathegory of genetic variability. Nevertheless the effect of temperature, origin, source of food or species of host plant, in the case of nematodes – plant parasites, have to be taken into consideration (Hooper 1969).

Classic research of Stephenson (1942 after Tarjan 1967) on *Rhabditis terrestris* Stephenson 1942 show that the body length of nematodes greatly depends on the amount of food available.

Monoson (1971) has studied the effect of food on the morphometric features of *Aphelenchus avenae* Bastian 1965. The results prove the considerable variability of body length depending on the fungi species on which the nematodes are cultivated. Other species of mycophagous nematodes also varied in body length, in indices *a*, *b*, *c* and *V* of individuals bred on different fungi species, and also within the group of nematodes cultivated on one host species (Pillai and Taylor 1967).

The studies of Anderson and Hooper (1971) point to considerable differences of measurements between nematodes of the species *Eucephalobus striatus* (Bastian 1865) Thorne 1937, and those cultivated on agar and in soil.

Bird and Mai (1967) have found that the geographic origin (22 populations of *Trichodorus christiei* Allen 1957 from 8 geographical points were examined) affects the body length and width, the length of front and back section of ovary duct.

As regards nematodes – plant parasites there are frequently considerable differences of morphometric features in individuals cultivated on different host plants. And so Bird and Mai (1967) have observed great differences in the body length and width, in the length of spicules of representants of the species *Trichodorus christiei* Allen 1957 cultivated on different plant species.

Taylor and Jenkins (1957) have shown that in four species of the genus *Pratylenchus* the body length, oesophagus length, stylet length, head-vulva distance depend to a considerable extent upon the species of host plant.

Temperature may indirectly or directly affect the morphometric features of nematodes. Gysels (1964 after Tarjan 1967) has observed that female nematodes of the species *Panagrellus silusiae* (de Man 1913) Goodey 1945 were shorter when cultivated at higher temperatures. The direct effect of temperature upon the size of nematodes has been also observed by Hoeppli (1926 after Tarjan 1967) who has found smaller individuals of *Dorylaimus therme* Cobb in Hoeppli 1926 in hot springs of Yellowstone Park, Wyoming than in the cold lake Utah. Indirect effect of temperature upon the size of nematodes has been recorded by Evans and Fischer (1970) when examining nematodes of the following

species: *Ditylenchus myceliophagus* Goodey 1958 and *D. destructor* Thorne 1945. In their opinion the temperature affected the size of nematodes by the size of population: the lower temperature the smaller population number and greater size of nematodes.

This brief literature review on the intrapopulational variability of nematodes points to the variety of factors responsible. Thus it would be worth-while to add here the results of studies on the effect of some ecological factors – population's origin and breeding conditions – on some morphometric features of nematode *Panagrolaimus rigidus*.

2. MATERIAL AND METHODS

The research was conducted in the years 1969–1975 on two populations of *Panagrolaimus rigidus* varying as to their origin. Individuals of population *A* were isolated from soil under potatoes (near Jadwisin, Warsaw district) in 1969. Individuals of population *B* were from the soil under rye (near Anin, Warsaw district) and were isolated in 1973. Nematodes belonging to both populations were extracted from soil and cultivated on agar with yolk as initial material for investigations. In order to examine the effect of culture conditions on the morphometric features of *P. rigidus*, nematodes cultivated on agar with yolk at 20°C and in soil with cereal seedlings were analysed. Soil cultivations were carried out in 25 ml beakers filled with sterile soil. In each beaker cereal seedlings grew. After 28 days nematodes were extracted from the soil and plants using Baerman's method. Soil cultures were conducted at three temperatures 10, 20 and 30°C and three moisture levels: 25, 50 and 100% of maximum soil water capacity. Thirty females and males of each experimental variant were measured. Nematodes cultivated on agar were measured for body length and width, stoma length, corpus length, isthmus length, bulbus length, oesophagus length, tail length, for females head-vulva distance, for males spicule length. For individuals cultivated in soil only the body length and width were measured. The significance of differences in measurements of nematodes was estimated by a statistical method of variance analysis.

3. RESULTS

3.1. The effect of population origin

In order to examine the effect of population origin on the variability of some morphometric features of nematodes of the species *P. rigidus* individuals both populations (*A* and *B*) bred under identical conditions were measured, namely: (1) on agar with yolk, (2) in soil with cereal seedlings.

Table 1 shows considerable differences between both populations cultivated on agar and yolk as regards the body length and width, oesophagus length, tail length, for females the head-vulva distance and for males the spicule length. The differences were so great that even the range of measurements frequently did not overlap or to a slight extent only. Much smaller differences were recorded in the stoma length and of the indices *a* and *V*. But the mean values and variability range of indices *b* and *c* for both sexes considerably varied in populations examined.

The differences in the body length and width of individuals from the populations examined were statistically analysed and were found to be significant[1].

[1] The significance of differences was estimated at the level of significance 0.05.

Table I. Morphometric features (in μm) and indices of females and males *Panagrolaimus rigidus* (Schneider 1866) Thorne 1937 differing as to origin, cultivated on agar with yolk
Population *A* is from soil under potatoes (isolated in 1969), population *B* from soil under rye (isolated in 1973)

Features and morphometric indices	Females		Males	
	population *A*	population *B*	population *A*	population *B*
Body length	1,126.9 (930–1,370)	767.7 (674–946)	988.7 (830–1,170)	661.1 (556–797)
Body width	62.4 (43–101)	41.0 (33–62)	50.6 (35–67)	26.3 (22–34)
Stoma length	10.9 (8.8–13.0)	11.9 (9.1–14.9)	10.8 (8.8–11.0)	11.4 (9.1–15.8)
Corpus length	94.7 (88–100)	89.3 (80–100)	88.5 (81–110)	78.4 (65–88)
Isthmus length	40.3 (29–54)	28.8 (18–36)	39.6 (29–50)	33.0 (26–39)
Bulbus length	31.2 (26–36)	26.7 (21–36)	26.6 (23–33)	21.8 (18–31)
Oesophagus length	179.0 (160–220)	144.0 (133–157)	169.0 (150–188)	133.3 (117–145)
Tail length	66.5 (54–74)	52.7 (42–69)	51.0 (44–65)	41.0 (34–47)
Length of the head--vulva region	661.3 (550–820)	464.5 (354–581)		
Length of spicules			28.3 (16–35)	16.6 (9–21)
a	18.0 (14–23)	15.8 (10–20)	19.5 (16–28)	20.9 (17–25)
b	6.3 (5.4–8.7)	4.4 (3.8–5.3)	5.8 (5.1–7.0)	4.6 (3.2–5.0)
c	17.0 (14–21)	12.4 (9–24)	19.4 (16–24)	13.4 (11–16)
V	58.7 (53–66)	50.0 (47–57)		

Table II. Body size (in μm) of females and males from two *P. rigidus* populations cultivated in soil with cereal seedlings
For explanations see Table I

	Females		Males	
	population *A*	population *B*	population *A*	population *B*
Body length	825.1 (560–1,140)	545.0 (462–643)	741.6 (580–960)	532.5 (449–641)
Body width	39.8 (24–62)	22.5 (19–27)	30.3 (21–44)	18.5 (17–23)

The differences in the measurements of nematodes of both populations cultivated in soil were also statistically significant (Table II).

The results prove a considerable variability of some morphometric features of individuals belonging to two nematode populations belonging to the same species but differing as to their origin.

3.2. The effect of conditions and duration of laboratory culture

The earlier research (Kozłowska and Mianowska 1971) has shown that several morphometric features of nematodes of the species *P. rigidus* depend on a considerable extent on the medium on which nematodes are cultivated. There have been statistically significant differences in the measurements of nematodes cultivated on agar and yolk and those cultivated in soil with rye seedlings.

Table III. Body size (in μm) of females and males of *P. rigidus* population *B* cultivated on agar and in soil
For explanations see Table I

	Females		Males	
	agar with yolk	soil	agar with yolk	soil
Body length	767.7 (674–946)	545.0 (462–643)	661.1 (556–797)	532.5 (449–640)
Body width	41.0 (33–62)	22.5 (19–27)	26.3 (22–34)	18.5 (17–23)

The present research was conducted in order to examine the effect of: the kind of culture on morphometric features of nematodes from population *B*, duration of laboratory culture on size of population *A* individuals, temperature and moisture on size of nematodes belonging to both populations.

Comparison of the size of nematodes from population *B* cultivated on agar and yolk with those from the same population but cultivated in soil has shown that the latter were much shorter and thinner than those from the agar culture (Table III). These differences were statistically significant. The yolk provided very good conditions for the growth of bacteria on which *P. rigidus* mainly fed. This is probably why the nematodes from this culture were much bigger.

Table IV. Body size (in μm) of females and males *P. rigidus* population *A* from annual and five years' agar culture
For explanations see Table I

	Females		Males	
	annual culture	five years' culture	annual culture	five years' culture
Body length	825.1 (560–1,140)	542.8 (458–686)	741.6 (580–960)	528.9 (435–645)
Body width	39.8 (24–62)	24.5 (19–33)	30.3 (21–44)	22.2 (15–29)

Anderson and Hooper (1971) have observed also considerable differences in the size of nematodes of the species *Eucephalobus stratus* (Bastian 1865) Thorne 1937 cultivated in soil and those of the same species but cultivated on agar (the latter were longer and broader). The increase in body size of *P. rigidus* cultivated on agar and yolk seemed to be not constant. Already after 5 years of such laboratory culture the nematodes "degenerated" to some extent. Nematodes from population *A* after being cultivated for 5 years on agar and yolk were much shorter and thinner than nematodes of the same population from an annual agar culture (Table IV). These data are confirmed by earlier observations (Mianowska 1976) showing that population *A* (cultivated 5 years on agar) was less dynamic and more stenobiotic than population *B* (nematodes freshly isolated from soil).

Cultivation of *P. rigidus* at different temperatures and moisture allowed to estimate the effect of temperature and moisture on the size of nematodes of populations *A* and *B*. In both populations moisture did not affect the length and width of nematodes examined. There are no statistically significant differences in the size of nematodes cultivated at the same temperature and at various moisture levels. Also there are no statistically significant differences in the size of individuals cultivated at the same moisture level in variants at 20 and 30°C. Still statistically significant differences were observed between the size of nematodes cultivated at 10°C and those cultivated at 20 and 30°C. Individuals cultivated at the lowest temperature examined were much longer and broader (Table V).

Differences in measurements of nematodes varying in origin but cultivated under the same moisture and temperature conditions were not significant statistically. This was probably due to the fact that nematodes of population *A* in this series of experiments were from the five years' agar culture. After five years of culture under artificial conditions the size of nematodes from population *A* decreased to such an extent that the differences in the size of individuals belonging to both populations disappeared.

Table V. Body size (in μm) of females and males of two *P. rigidus* populations varying as to their origin, cultivated under different conditions of temperature and moisture
For explanations see Table I

Parameters		Females				Males			
		body length		body width		body length		body width	
Temperature (°C)	Moisture (%)	population *A*	population *B*	population *A*	population *B*	population *A*	population *B*	population *A*	population *B*
10	25	742.6 (633–884)	740.5 (468–1,017)	36.6 (31–42)	31.4 (27–50)		642.7 (516–842)		30.6 (19–39)
20	25	569.9 (443–803)	532.5 (445–778)	28.1 (19–39)	24.1 (17–33)	523.3 (385–666)	492.9 (393–566)	22.8 (19–33)	18.1 (12–22)
	50	542.8 (458–686)	545.0 (462–463)	24.5 (19–33)	22.5 (19–27)	528.9 (435–645)	532.5 (449–641)	22.2 (15–29)	18.5 (17–23)
	100	601.1 (426–721)		28.5 (21–60)		536.6 (420–686)		23.3 (19–31)	
30	25	572.0 (469–743)	569.9 (443–686)	26.8 (19–37)	21.1 (19–31)	539.1 (485–641)	515.8 (383–611)	22.7 (19–35)	19.1 (17–25)
	50	567.8 (464–665)	528.3 (426–630)	24.3 (21–39)	21.1 (17–31)	542.9 (412–624)	479.1 (422–603)	22.9 (19–33)	20.4 (12–27)

These results prove the considerable influence of various ecological factors on the size of nematodes of the species *P. rigidus*. Quite striking are the great differences in the size of some morphometric features of individuals of both *P. rigidus* populations differing only in their origin (prior to experiment the nematodes of both populations were for one year cultivated under identical conditions – on agar and yolk at 20°C).

It is interesting that the nematodes cultivated under artificial conditions on agar were bigger than nematodes of the same population cultivated in soil and thus in a natural life environment for nematodes of the species *P. rigidus*. However, five years' cultivation of *P. rigidus* in unnatural conditions unfavourably affected the nematodes and as a result they became much smaller.

4. SUMMARY

A research was conducted on the effect of the origin of the population, conditions and the duration of laboratory culture on the morphometric features of nematodes of the species *Panagrolaimus rigidus* (Schneider 1866) Thorne 1937. Two populations varying in their origin were examined. Individuals of population *A* were isolated in 1969 from the soil under potatoes near Jadwisin in the Warsaw district, individuals of population *B* were isolated in 1973 from the soil under rye near Anin in the Warsaw district. Nematodes of both populations were cultivated under identical conditions before the measurements – on agar with yolk at 20°C. Considerable differences were observed in the body length and width, oesophagus length, tail length, in females of the head-vulva distance and in males of the spicule length between both populations cultivated on agar and yolk. Much smaller differences were recorded in the stoma length and in the indices *a* and *V*. Still the indices *b* and *c* of both sexes differed considerably in populations examined (Table I). There were also considerable differences in the size of nematodes belonging to both populations cultivated for one month in soil (Table II). The nematodes of population *B* cultivated on agar and yolk were much longer and broader than those from the soil culture (Table III). Five years' culture of nematodes on agar unfavourably affected their size. Nematodes from this culture were much shorter and thinner than those cultivated for one year (Table IV). Measurements of nematodes cultivated at different temperatures showed that individuals cultivated at 10°C were much longer and broader than those cultivated at 20 and 30°C. No essential differences were observed in the size of nematodes of both populations cultivated in soil at three moisture levels: 25, 50 and 100% of maximum soil water capacity (Table V).

5. POLISH SUMMARY (STRESZCZENIE)

Przeprowadzono badania nad wpływem pochodzenia populacji, warunków i czasu prowadzenia hodowli laboratoryjnej na cechy morfometryczne nicieni z gatunku *Panagrolaimus rigidus* (Schneider 1866) Thorne 1937. Badania prowadzono na dwóch populacjach różniących się pochodzeniem. Przedstawiciele populacji *A* zostali wyizolowani w 1969 roku z gleby pod uprawą ziemniaków z okolic Jadwisina w woj. warszawskim, przedstawiciele populacji *B* w 1973 roku z gleby pod uprawą żyta z okolic Anina k. Warszawy.

Nicienie należące do obu populacji przed wykonaniem pomiarów hodowano w jednakowych warunkach, na agarze z dodatkiem żółtka jaja kurzego w temp. 20°C. Stwierdzono znaczne różnice w długości i szerokości ciała, długości przełyku i długości ogona oraz u samic długości odcinka głowa-vulva, a u samców długości spikul między obiema populacjami hodowanymi na agarze z żółtkiem. Znacznie mniejsze różnice zaobserwowano w długości torebki gębowej oraz we wskaźnikach: *a* i *V*. Natomiast wskaźniki: *b* i *c* u obu płci w badanych populacjach różniły się wyraźnie (tab. I). Stwierdzono również znaczne różnice w wymiarach nicieni należących do obu populacji hodowanych przez okres jednego miesiąca w glebie (tab. II). Stwierdzono, że nicienie z populacji *B* hodowane na agarze z żółtkiem były znacznie dłuższe i szersze niż nicienie z hodowli glebowej (tab. III). Zaobserwowano, że pięcioletnia hodowla nicieni na agarze wpłynęła niekorzystnie na ich wymiary. Nicienie z tej hodowli były znacznie krótsze i węższe niż nicienie hodowane przez jeden rok (tab. IV). Pomiary nicieni hodowanych w różnych temperaturach wykazały, że osobniki hodowane w temperaturze 10°C były znacznie dłuższe i szersze niż nicienie hodowane w temperaturach 20 i 30°C. Nie stwierdzono natomiast istotnych różnic w wymiarach nicieni obu populacji hodowanych w glebie przy trzech poziomach wilgotności: 25, 50 i 100% maksymalnej pojemności wodnej gleby (tab. V).

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