

## Short Note

# Morphological and molecular characterisation of *Criconemoides informis* (Micoletzky, 1922) from Southern Russia

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Data on the species composition and distribution of plant-parasitic nematodes in the rhizosphere of grapevines in the southern regions of Russia are very limited. During routine nematological surveys conducted in 2016-2019 in the Crimean peninsula, *Criconemoides informis* was the most frequently encountered ring nematode in the rhizosphere of grapevines. One of the populations found in an abandoned vineyard in the Bakhchisarai region (44°52'34.619" N; 33°39'37.159" E) was morphometrically and molecularly characterised. The soil in the sampling site was a calcareous chernozem of heavy loam texture. Nematodes were extracted from the soil samples using a modification of the decanting and sieving method (Flegg, 1967). For morphological studies, the nematodes were killed with hot water, fixed in 5% formalin solution, and mounted in glycerin on slides using the Seinhorst technique (Seinhorst, 1959). For molecular studies, nematodes were frozen in distilled water. Their total DNA was extracted using the K-Sorb kit (Syntol LLC, Russia) according to the manufacturer's instructions. The forward 18S900F (5'-AAG ACG GAC TAC AGC GAA AG-3') and the reverse 18S1713R (5'-TCA CCT ACA GCT ACC TTG TTA CG-3') primers were used for the amplification of the 18S rRNA gene. The forward D2A (5'-ACA AGT ACC GTG AGG GAA AGT TG-3') and the reverse D3B (5'-TCG

GAA GGA ACC AGC TAC TA-3') primers were used for amplification of the D2-D3 expansion segments of the 28S rRNA gene. The partial cytochrome c oxidase subunit 1 (*COI*) gene was amplified with the forward primer COI-F5 (5'-AAT WTW GGT GTT GGA ACT TCT TGA AC-3') and the reverse primer COI-R9 (5'-CTT AAA ACA TAA TGR AAA TGW GCW ACW ACA TAA TAA GTA TC-3').

Sequencing of PCR products was carried out with the same primers using genetic analyser ABI 3130xl (Applied Biosystems, USA). Low-quality segments of sequences at the 5' and 3' ends were removed. Then, the newly obtained sequences were submitted to the GenBank database under accession numbers MT328749 (18S rRNA), MT332825 (28S rRNA) and MT328867 (*cox1*).

**Morphological characterisation.** Body slightly arcuate after fixation. Cuticular annuli retrorse with rounded edges, no lateral differentiation, margins smooth or slightly irregular. Anastomoses rare. First annulus directed sideways. Submedian lobes distinct, relatively large. Labial disc slightly elevated above lobes. Stylet very strong, knobs thick, anteriorly concave, with marginal processes directed anteriorly. Spermatheca empty. Vulval lips bulging but not projecting above body contour. Tail plump with rounded tip. Terminal annulus usually bilobed. Males were not found.

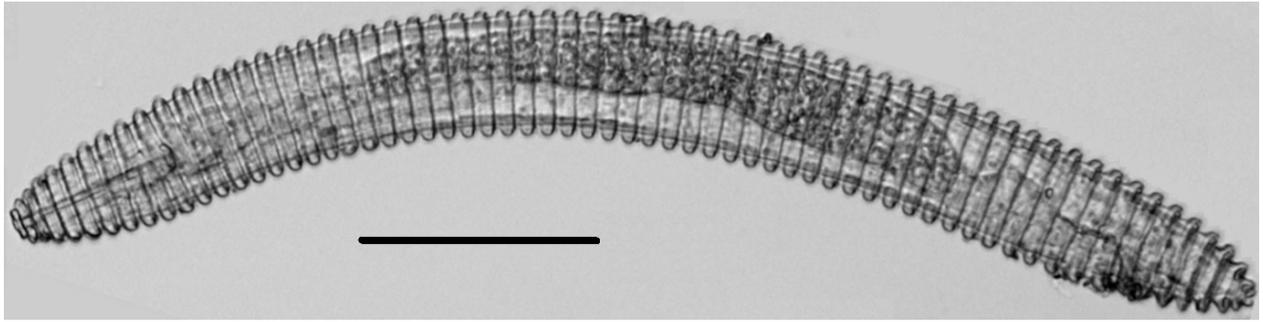


Fig. 1. *Criconemoides informis*, female. Scale bar: 100  $\mu$ m.

**Female (n=12):** L =  $454.6 \pm 53.9$  (390-540)  $\mu$ m; stylet =  $77.3 \pm 3.5$  (69-87.5)  $\mu$ m; oesophagus =  $132.5 \pm 11.3$  (122.5-152.5)  $\mu$ m; tail =  $25 \pm 2.1$  (20-27.5)  $\mu$ m; a =  $8.7 \pm 0.8$  (8.0-10.4); b =  $3.5 \pm 0.3$  (3.1-3.8); c =  $18.4 \pm 2.1$  (15.6-21.4); R =  $63.6 \pm 0.9$  (62-65); Rst =  $11.3 \pm 0.8$  (10-13); Roes =  $19.2 \pm 0.9$  (17-20); Rex =  $20.9 \pm 0.6$  (20-21); RV =  $6.5 \pm 0.7$  (6-8); Ran =  $3.5 \pm 0.5$  (3-4); V =  $90.9 \pm 1.4$  (88.4-91.2)%; VL/VB =  $1.2 \pm 0.1$  (1.1-1.3).

Thus, the specimens examined in this study agree well with the description of *C. informis* in Geraert (2010).

**Molecular characterisation.** The sequences of the 18S rRNA gene of the studied specimens were most similar to the *Criconemoides informis* sequences from Custer Gallatin National Forest, Park County, MT, USA (MF094902), with 99.07% similarity, and clustered with other criconematid nematodes: *Discocriconemella sinensis* (MK253543), *Neolobocriconema serratum* (MH668971) and *Ogma* sp. (KJ934175).

The sequences of the D2-D3 expansion segments of the 28S rRNA gene were most similar to the *Criconemoides informis* sequence from Greece (AY780970), with 99.44% similarity, share less similarity (98.89%) with *Criconemoides informis* from Iran (KU722386) and also clustered with *Discocriconemella sinensis* (MK253537).

The sequence of the *COI* gene shared 82.6% similarity with the *Criconemoides informis* sequence from Custer Gallatin National Forest, Park County, MT, USA (KJ787839) and 81% similarity with the *Criconemoides informis* sequence from Uncompahgre National Forest, San Miguel, CO, USA (MF770962). Thus, the sequences of *COI* gene of this species collected from different localities can be variable.

## ACKNOWLEDGEMENTS

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