

## Description of *Chordodes anthophorus* (Gordiida) for the first time in Iran with an emphasis on scanning electron microscopy characters

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### Article info

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### Summary

We report a female *Chordodes anthophorus* from a Giant Asian Mantis (*Hierodula membranacea*) for the first time from Iran. Scanning electron microscopy (SEM) was used to describe the characters and substructures precisely. We demonstrate characteristic cuticular patterns for *Chordodes anthophorus*. The presence of five types of areoles including simple, tubercle, crowned and circum-cluster areoles and also crowned areoles with long filaments which is a common feature in females, confirm our investigation.

**Keywords:** Nematomorpha; Gordiida; *Chordodes*; Scanning electron microscopy; Iran

### Introduction

Nematomorpha, commonly known as horsehair worms, parasitize terrestrial and aquatic arthropods. Approximately 360 species in the taxon Gordiida are parasites of terrestrial insects (Hanelt *et al.*, 2005; Schmidt-Rhaesa, 2013).

These worms mature in their host, manipulate its behavior and urge them towards water, where they emerge (Thomas *et al.*, 2000). After mating and laying eggs, the hatched larvae find aquatic invertebrates that serve as paratenic hosts. It is claimed that the terrestrial final hosts become infected by consuming the paratenic hosts. However, the direct pathways of infection are still unclear (Hanelt *et al.*, 2005).

Knowledge on biodiversity and distribution of Nematomorpha is still fragmentary. Many records were made more or less accidentally, which results in very scattered sampling and the fact that often only single specimens are available. Identifications are based on different methods and over the years scanning electron microscopy (SEM) has replaced the light microscopical investigation of

cuticular fragments. Molecular barcoding becomes more and more important (e.g. Chiu *et al.*, 2011, Hanelt *et al.*, 2015), but the database still includes few species. Therefore, new records, especially from new or scarcely sampled locations, should document specimens as good as possible.

About 100 species belong to the genus *Chordodes*, this is the largest genus in Gordiida (Nematomorpha) (Schmidt-Rhaesa, 2013; Schmidt-Rhaesa *et al.*, 2008). Most species occur in tropical and subtropical regions (Schmidt-Rhaesa, 2013; Schmidt-Rhaesa *et al.*, 2008). *Chordodes* species differ in their cuticular structure. The cuticular surface is shaped into different structures called areoles. Crown-shaped areoles with apical filaments are characteristic of the genus *Chordodes*. According to the species, crowned areoles may occur single, in pairs or clusters with another areolar type. Other types of areoles are those with a finger-like extension on top (= tubercle areoles), those resembling a thorn (= thorn areoles), those higher than surrounding areoles (= bulging areoles), those surrounding crowned areoles (= circumcluster areoles) and those without any of these characteristics (= simple areoles). This

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Fig. 1. Map of Iran, with the location of Shiyadeh, Mazandaran province.

terminology follows Schmidt-Rhaesa *et al.* (2008). Due to the complicated cuticle characters and substructures of this genus, scanning electronic microscopy (SEM) has become the standard technique for the investigation of this genus (Schmidt-Rhaesa, 2013; Schmidt-Rhaesa *et al.*, 2008). *Chordodes* species parasitize different hosts, but praying mantids are the dominant host (e.g. Schmidt-Rhaesa & Ehrmann, 2001).

We report here a new record from Iran. The nematomorph fauna of Iran is almost unknown, previously only one species, *Chordodes bipilus* Kirjanova, 1957 has been recorded (Kirjanova, 1957). This study describes *Chordodes anthophorus* Kirjanova, 1950 for the first time in Iran, including its host.

### Material and Methods

During ecoparasitological studies in Shiyadeh, Mazandaran province, Iran, a female Giant Asian Mantis (*Hierodula membranacea*) was found infected with a horsehair worm. The worm was placed in 70 % ethanol. Scanning electron microscopy (SEM) was done at the University of Hamburg. Entire pieces from the midbody region (1 mm long) were dehydrated in an increasing ethanol series, critically point dried and coated with gold in a sputter coater. The observation took place using a LEO SEM 1524 at 10 kV and digital images were taken. The horsehair worm was deposited together with its host in the Zoological Museum in Hamburg under the accession number ZMH V13462.

### Ethical Approval and Informed Consent

For this study formal consent is not required. The article does not contain any studies with human participants.

### Results

#### *Chordodes anthophorus* Kirjanova, 1950

**Material investigated.** Single female from Shiyadeh dam shore, Mazandaran province, Iran (36°19'43.5"N 52°30'27.7"E) (Fig. 1). Collected on September 9th, 2019 by Sina Mohtasebi, Mohammad Javad Abbaszadeh Afshar, Alireza Mohtasebi and Farbod Tabatabaie.

**Host.** Female Giant Asian Mantis (*Hierodula membranacea*)

**Description.** The worm was 37 cm in length and dark brown. The anterior end is tapering, it has a whitish tip (Fig. 2B). The posterior end is slightly swollen, the cloacal opening is terminal (Fig. 2C). Five areole types are present on the cuticle. Simple oval areoles range in size from 10 to 15  $\mu$ m and are of slightly different elevation (Fig. 3A, B). They vary in shape (Fig. 3B, C). The simple areoles can be smooth or covered with short bristles (2  $\mu$ m) arranged at the center of the apical surface (Fig. 3B, C). Among the simple areoles are tubercle areoles, which contain a finger-like projection called tubercle (Fig. 3B, C). Thorn areoles were not observed. Two other types of areoles occur in clusters. Crowned areoles are decorated with filaments on top and are surrounded by 8 – 12 elevated circumcluster areoles (Fig. 3D). The circumcluster areoles contain

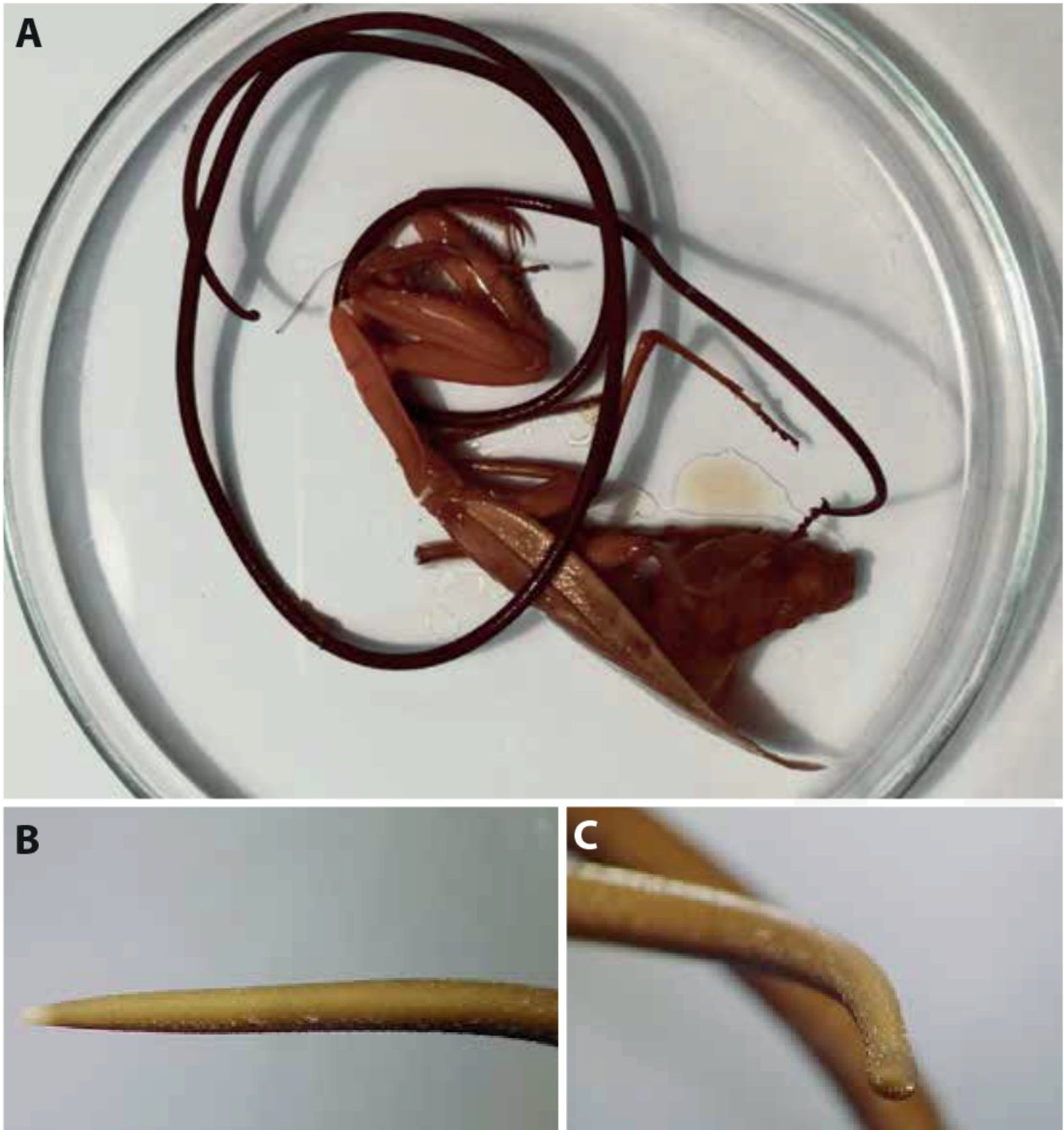


Fig. 2. *Chordodes anthophorus*. A. Specimen with its host, the Giant Asian Mantis (*Hierodula membranacea*). B. Anterior end. C. Posterior end.

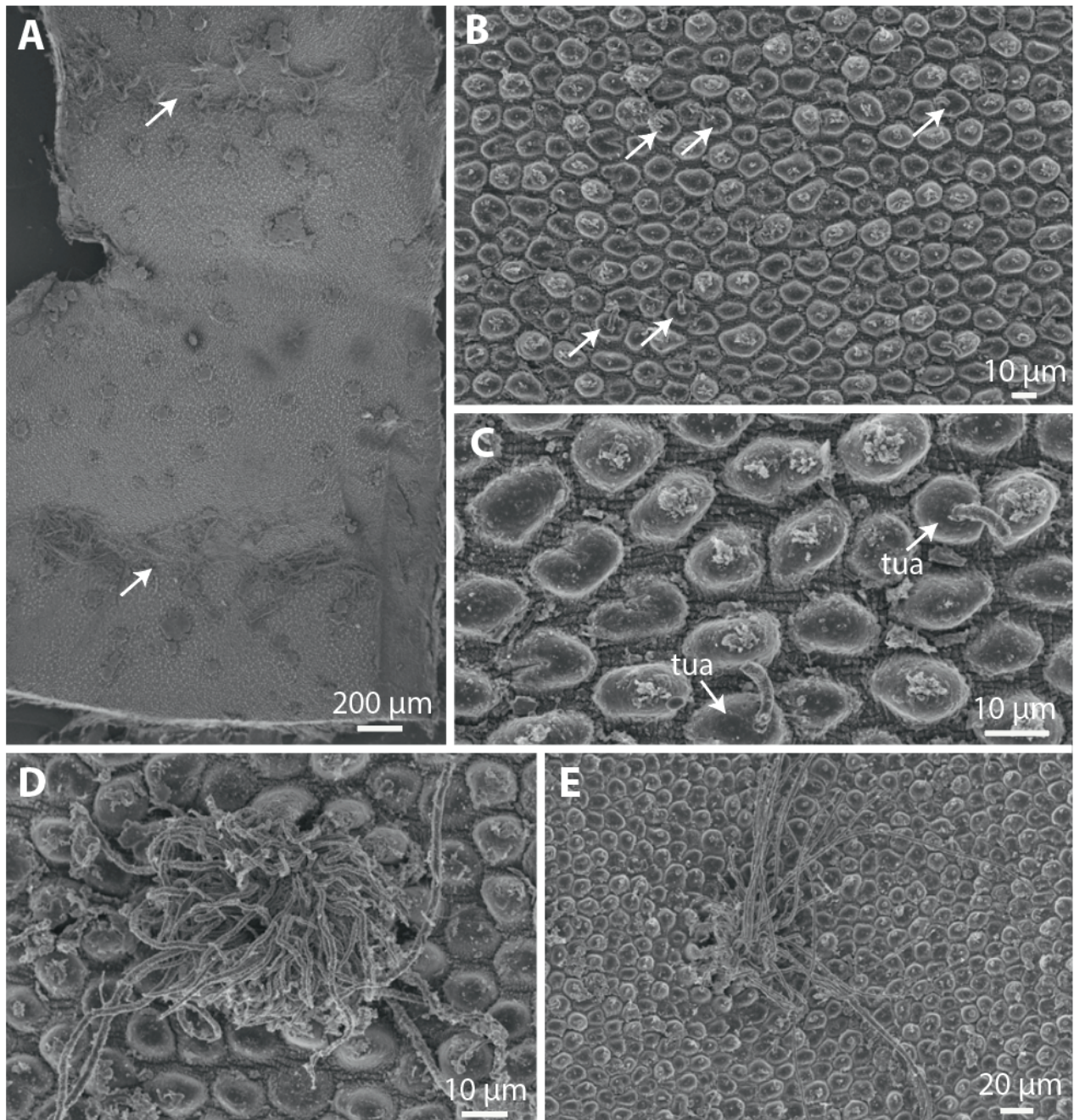


Fig. 3. *Chordodes anthophorus*, cuticular structure. A. Entire piece of cuticle from the entire circumference stretched flat. Crowned areoles with long apical filaments are present along the ventral and dorsal midline (arrows), clusters of crowned areoles with shorter filaments are visible on the remaining cuticle. B. Overview on the cuticle showing simple and tubercle (arrows) areoles. B. Magnification showing simple areoles with and without bristles as well as tubercle areoles (tua). C. Crowned areoles with short filaments surrounded by circumcluster areoles. D. Crowned areoles with long filaments.

short filaments at the top. Also, another type of crowned areoles with significantly longer filaments (80 µm) is present in the ventral and in the dorsal midline (Fig. 3A, E).

## Discussion

Global distribution patterns of nematomorphs are very patchy, because from several countries and regions, very few records are known. Initial random records can lead to a growing awareness in undersampled regions and lead to further records. For example, between 1853 and 2004, 13 nematomorph species were recorded from entire India (Schmidt-Rhaesa & Yadav, 2004). Until 2020, sampling activities just in the Eastern part of India raised the number to 22 species (Yadav *et al.*, 2020).

Little information is available about the phylum Nematomorpha in Iran. Previously there was only one single report of two females of *Chordodes bipilus*, one from Astrabade and the other from Beh-behan (province Farsistan) (Kirjanova, 1957). This species contains simple areoles with an irregular surface, tubercle and thorn areoles, crowned areole clusters composed of a pair of crowned areoles and 9 – 12 circumcluster areoles. The crowned areoles contain apical filaments of two sizes, longer and thicker ones (diameter 2 µm) as well as shorter and thinner ones (diameter 1 µm) (Kirjanova, 1957). The presence of thorn areoles, of two types of apical filaments in crowned areoles and the structure of simple areoles differ from the specimen described here.

Simple, bristle and tubercle areoles, clusters composed of crowned and circumcluster areoles and the presence of very long filaments in crowned areoles on the ventral (sometimes also on the dorsal side in females) are common hallmarks of this genus (Schmidt-Rhaesa *et al.*, 2008), but their fine structure and distribution patterns on the body cuticle are important characters for identification. The observed cuticular structures correspond well with the species *C. anthophorus*. This species was described by Kirjanova (1950) and was reinvestigated using SEM by De Villalobos *et al.* (2007). De Villalobos *et al.* describe that the simple areoles are present in two forms, with smooth surface or with a tuft of bristles, which corresponds to the newly reported specimen. De Villalobos *et al.* (2007) also report the absence of thorn areoles, which are present in many other *Chordodes* species. Finally, crowned areoles with long apical filaments were reported by De Villalobos *et al.* from both the ventral and the dorsal midline. In some species, this type of areoles appears to be present only along the ventral midline, but not all reports investigated the entire circumference of the worms, therefore it is not clear whether this character is absent or was just not observed. Therefore, the cuticular structure of the specimen from Iran very well corresponds with that of *C. anthophorus* as documented by De Villalobos *et al.* (2007).

De Villalobos *et al.* (2007) synonymized some other species with *Chordodes anthophorus*: *C. anthophorus*: *C. aqueductus*, *C. ferganensis* and *C. oscillates*. Including these synonyms, this

species is now recorded from Tajikistan, Uzbekistan, and Georgia (Schmidt-Rhaesa *et al.*, 2008). The new report from Iran fits well in this previous distribution pattern and makes it likely that *C. anthophorus* has a broad distribution east, west and south of the Caspian Sea.

This is the first report of a host of a nematomorph species from Iran. Mantids are a common host for the genus *Chordodes* (Schmidt-Rhaesa, 2013; Schmidt-Rhaesa & Ehrmann, 2001). The species *Hierodula membranacea* has been reported as host for *Gordius verrucosus* from Sri Lanka (Bell, 1885) and other or undetermined *Hierodula* species are reported for different *Chordodes* species (see summary in Schmidt-Rhaesa, 2013).

## Conflicts of Interest

The authors declare no conflict of interest.

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