

# Gastrointestinal Helminths of the Oriental Tree Frog *Hyla orientalis* Bedriaga, 1890 (Amphibia: Hylidae) from İzmir Province, Western Turkey

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**Abstract:** Thirty *Hyla orientalis* (oriental tree frog) from Western Turkey (İzmir Province) were examined for helminth parasites. The frogs were collected in the spring of 2013. Four species of helminth parasites were recorded: three species of Nematoda (*Cosmocerca ornata*, *Oswaldocruzia filiformis*, *Oxysomatium brevicaudatum*), and one species of Acanthocephala (*Acanthocephalus ranae*). *Oxysomatium brevicaudatum* was recorded for the first time from *Hyla orientalis* in Turkey. A list of helminth parasites of *Hyla* spp. in Turkey is presented.

**Key words:** *Hyla orientalis*, Nematoda, Acanthocephala, Turkey

## Introduction

Previous studies based on morphological and bio-ecological data (KAYA, SIMMONS 1999, SCHNEIDER 2000, KAYA 2001, SCHNEIDER 2001) suggested the occurrence of two species of the genus *Hyla* Laurenti, 1768 in Turkey, i.e. *Hyla arborea* (Linnaeus, 1758) and *Hyla savignyi* Audouin, 1827. However, recent study based on molecular data showed that South-eastern European and Western Anatolian *Hyla arborea* should be considered a distinct species, *Hyla orientalis* Bedriaga, 1890 (STÖCK *et al.* 2008). *Hyla orientalis* was found in Northern, Western and South-western Turkey. Oriental tree frog lives in humid meadows, bushes, trees, shrubs, and prefers well-vegetated clear waters in breeding period (BARAN *et al.* 2012).

There are three published reports on parasites in *Hyla orientalis* from Turkey: DÜŞEN, ÖZ (2004) recorded six helminth parasites from South-western Turkey. YILDIRIMHAN *et al.* (2006) detected three helminths in *Hyla orientalis* (formerly *Hyla arborea*) from Edirne, Bursa and Sakarya Provinces of Turkey. DÜŞEN, YAKA (2014) reported six helminths from Denizli Province (the eastern part of the Aegean Region) in Turkey.

The aim of this study was to reveal the composition of helminth parasites of *Hyla orientalis* and to characterise their mean abundance in relation to different localities and sex. This is the first detailed helminthological study conducted on *H. orientalis* from the İzmir Province (Western Turkey).

## Materials and Methods

Thirty *Hyla orientalis* (23 male and seven female) were collected by hand in spring (March and April) of 2013 from two localities in İzmir Province: Ödemiş District (38°18'43"N, 28°01'37"E; n = 16) and Seferihisar District (38°16'53"N, 26°51'01"E; n = 14; Fig. 1). The mean ( $\pm$  SD) snout-vent length (SVL) of the specimens was  $41.96 \pm 4.42$  mm, with a range 37-54 mm.

The frogs were euthanised with sodium pentobarbital. The digestive tract was removed and placed in petri dishes containing a physiological saline and examined for helminth parasites under a dissecting microscope. Helminths were killed in a hot saline solution; nematodes were fixed in 70% ethanol and

mounted in glycerol; acanthocephalans were fixed in 70% ethanol, stained with iron acetocarmine (GEORGIEV *et al.* 1986), cleaned with clove oil and mounted in Entellan® for examination with a compound microscope. Prevalence, mean intensity and mean abundance were calculated according to BUSH *et al.* (1997). The species identification was done according to YORKE, MAPLESTONE (1926), YAMAGUTI (1961, 1963), BAKER (1987).

The Mann-Whitney U test was used to compare differences in parasite abundance between the localities. The effect of host sex on abundance of parasites was tested also using the Mann-Whitney U test. The significance level of  $\alpha \leq 0.05$  was used. All statistics analyses were performed using SPSS version 22.0.

## Results

Four species of helminth parasites were detected in oriental tree frogs. These were *Cosmocerca ornata* (Dujardin, 1845), *Oswaldocruzia filiformis* (Goeze, 1782) and *Oxysomatium brevicaudatum* (Zeder, 1800) (Nematoda) as well as *Acanthocephalus ranae* (Schrank, 1788) (Acanthocephala). *O. filiformis* was the most prevalent species (Table 1).

Out of 30 *Hyla orientalis* individuals, 15 (50%) were infected with one or more parasites. Four parasite species (61 individuals) were collected from 15 oriental tree frogs. Of the infected oriental tree frogs, 13 (86.66%) harboured one parasite species and two (13.33%) harboured two parasite species. There were  $1.13 \pm 0.09$  (mean  $\pm$  1 SE) helminth species per infected host and  $4.06 \pm 1.49$  helminth individuals per infected host.

Fourteen specimens were collected from Seferihisar District and nine (64.28%) of them were infected with *Oswaldocruzia filiformis*, *Cosmocerca ornata* and *Oxysomatium brevicaudatum*. Sixteen *H. orientalis* were collected from Ödemiş District. Six oriental tree frogs (37.50%) from Ödemiş were infected with *Oswaldocruzia filiformis*, *Oxysomatium brevicaudatum* and *Acanthocephalus ranae*.

The helminth communities of *H. orientalis* presented few differences in the two localities. *Oswaldocruzia filiformis* in Seferihisar exhibited higher mean abundance when compared to Ödemiş ( $p = 0.005$ ). *Cosmocerca ornata* was recorded only in Seferihisar. *Acanthocephalus ranae* was recorded only in Ödemiş. *Oxysomatium brevicaudatum* was recorded in both localities. There were no significant differences between mean abundances of these two localities ( $p = 0.885$ ). Mean abundance of each parasite species did not differ significantly between male and female frogs ( $p > 0.05$ ).



**Fig..** Collecting localities of *Hyla orientalis* from Western Part of Turkey: 1. Seferihisar; 2. Ödemiş

## Discussion

Four helminth parasites were recorded in *H. orientalis* in the present study: *Oswaldocruzia filiformis*, *Cosmocerca ornata*, *Oxysomatium brevicaudatum* and *Acanthocephalus ranae*. Among these parasites, *Oxysomatium brevicaudatum* was the new host record for *H. orientalis* in Turkey.

According to recent studies, *Hyla arborea* was subdivided into three species (STÖCK *et al.* 2008). In Turkey, there are two hylid species, *H. orientalis* and *H. savignyi*. There are several studies on helminth parasites of these hylids species from other countries. HRISTOVSKI, RIGGIO (1974) reported *Opisthioglyphe ranae* (Frölich, 1791), *Cosmocerca ornata*, *Oswaldocruzia filiformis*, *Nematotaenia dispar* (Goeze, 1782) and *Icosiella neglecta* (Diesing, 1851) in *H. arborea* from former Yugoslavia, Greece and Corsica. AL-BARWARI, NASSIR (1983) reported *Nematotaenia dispar* in *H. arborea* from Iraq. BUCHVAROV (1984) reported *Polystoma skrjabini* Buchvarov, 1984 in *H. arborea* from Bulgaria. MASHAII (2005) reported *Polystoma viridis* Euzet, Combes et Batchvarov, 1974, *Nematotaenia dispar* and *Aplectana* sp. in *H. arborea savignyi* from Iran.

There are several previous studies on the helminth parasites of *Hyla orientalis* in Turkey. DÜŞEN, ÖZ (2004) reported *Polystoma skrjabini*, *Pleurogenoides medians* (Olsson, 1876), *Encyclometra colubrimurorum* (Rudolphi, 1819), *Proteocephalus* sp., *Cosmocerca commutata* (Diesing, 1851) and *Acanthocephalus ranae* from the Antalya Province, SW Turkey. Among these parasites, only *Acanthocephalus ranae* was recorded in our study. YILDIRIMHAN *et al.* (2006) recorded three

**Table 1.** Prevalence, mean intensity and mean abundance of parasites in *Hyla orientalis*

Parasite species	Site of infection	Number of infected frogs (%)	Mean intensity	Min-Max	Mean abundance
<i>Oswaldocruzia filiformis</i>	Intestine	12 (40.00)	4.42	1-18	1.76
<i>Cosmocerca ornata</i>	Intestine	1 (3.33)	2.00	2	0.06
<i>Oxysomatium brevicaudatum</i>	Intestine	2 (6.66)	1.50	1-2	0.10
<i>Acanthocephalus ranae</i>	Intestine	2 (6.66)	1.50	1-2	0.10

**Table 2.** Helminth species reported for hylid members from Turkey

Helminth species	Site of infection	Host	Reference
<b>Monogenea</b> <i>Polystoma skrjabini</i>	Urinary bladder	<i>Hyla orientalis</i>	DÜŞEN, ÖZ (2004); YILDIRIMHAN <i>et al.</i> (2006); DÜŞEN, YAKA (2014)
<i>Polystoma integerrimum</i>	Urinary bladder	<i>Hyla savignyi</i>	YILDIRIMHAN <i>et al.</i> (2012)
<b>Digenea</b> <i>Pleurogenoides medians</i>	Intestine	<i>Hyla orientalis</i> , <i>Hyla savignyi</i>	DÜŞEN, ÖZ (2004); YILDIRIMHAN <i>et al.</i> (2012); DÜŞEN, YAKA (2014)
<i>Encyclometra colubrimurorum</i>	Kidney, liver, muscle, ovary, spinal cord, testis	<i>Hyla orientalis</i>	DÜŞEN, ÖZ (2004)
<i>Diplodiscus subclavatus</i>	Intestine	<i>Hyla savignyi</i>	YILDIRIMHAN <i>et al.</i> (2012)
<i>Halipegus kessleri</i>	Stomach	<i>Hyla savignyi</i>	YILDIRIMHAN <i>et al.</i> (2012)
<b>Cestoda</b> <i>Proteocephalus</i> sp.	Intestine	<i>Hyla orientalis</i>	DÜŞEN, ÖZ (2004)
<i>Nematotaenia dispar</i>	Intestine	<i>Hyla savignyi</i>	YILDIRIMHAN <i>et al.</i> (2012)
<b>Nematoda</b> <i>Cosmocerca commutata</i>	Intestine	<i>Hyla orientalis</i> , <i>Hyla savignyi</i>	DÜŞEN, ÖZ (2004), YILDIRIMHAN <i>et al.</i> (2012)
<i>Cosmocerca ornata</i>	Intestine	<i>Hyla orientalis</i>	YILDIRIMHAN <i>et al.</i> (2006); DÜŞEN, YAKA (2014); present study
<i>Aplectana brumpti</i>	Intestine	<i>Hyla savignyi</i>	YILDIRIMHAN <i>et al.</i> (2012)
<i>Oswaldocruzia filiformis</i>	Intestine	<i>Hyla orientalis</i>	YILDIRIMHAN <i>et al.</i> (2006); DÜŞEN, YAKA (2014); present study
<i>Oxysomatium brevicaudatum</i>	Intestine	<i>Hyla orientalis</i>	Present study
<i>Abbreviata</i> sp.	Submucosa of stomach and intestine	<i>Hyla orientalis</i>	DÜŞEN, YAKA (2014)
<b>Acanthocephala</b> <i>Acanthocephalus ranae</i>	Intestine	<i>Hyla orientalis</i>	DÜŞEN, ÖZ (2004); DÜŞEN, YAKA (2014); present study

helminths from Bursa, Edirne and Sakarya Provinces of Turkey: *Polystoma skrjabini*, *Cosmocerca ornata* and *Oswaldocruzia filiformis*. We did not record *Polystoma skrjabini* in our study. DÜŞEN, YAKA (2014) found *Polystoma skrjabini*, *Pleurogenoides medians*, *Cosmocerca ornata*, *Oswaldocruzia filiformis*, *Abbreviata* sp. and *Acanthocephalus ranae*.

Fourteen helminth species have been reported from *Hyla* spp. from Turkey (Table 2). Nematodes were dominant (six species), followed by digeneans. In the present study, nematodes also were the most abundant group. However, we did not record any digeneans.

The helminth species found in this study are common parasites of amphibians. *Cosmocerca or-*

*nata* was recorded for the first time in *Bufootes variabilis* (Laurenti, 1768) (formerly known as *Bufo viridis*), *Rana macrocnemis* Boulenger, 1885 and *Pelophylax ridibundus* (L., 1758) (formerly known as *Rana ridibunda*) in Turkey by SCHAD *et al.* (1960). In the present study, *C. ornata* had a lower prevalence (3.33%) when compared to the studies by YILDIRIMHAN *et al.* (2006) and DÜŞEN, YAKA (2014), who reported *C. ornata* prevalence of 7.41% and 11.76% for *H. orientalis*, respectively.

*Oswaldocruzia filiformis* was recorded for the first time in *Bufo regularis* and *Rana macrocnemis* in Turkey by SCHAD *et al.* (1960). The prevalence of *O. filiformis* in *H. orientalis* was 11.11% and 5.88% as reported by YILDIRIMHAN *et al.* (2006) and DÜŞEN,

YAKA (2014), respectively. In our study, the prevalence (40%) of *O. filiformis* was higher than in previous studies.

*Acanthocephalus ranae* was recorded for the first time in *Pelophylax ridibundus* in Turkey by OĞUZ *et al.* (1994). The prevalence (5.5%) of *Acanthocephalus ranae* reported by DÜŞEN, ÖZ (2004) was lower than that observed in the present study (6.66%). In contrast, the prevalence (11.76%) of this parasite reported by DÜŞEN, YAKA (2014) was higher with comparison to our study.

*Oxysomatium brevicaudatum* was recorded from *Bufo regularis* (SCHAD *et al.* 1960), *Bufoes variabilis* (SCHAD *et al.* 1960, TOPÇU, BAYRAK 2000, DÜŞEN *et al.* 2010, DÜŞEN, OĞUZ 2010, DÜŞEN 2011), *Rana macrocnemis* (SCHAD *et al.* 1960), *Pelophylax ridibundus* (SCHAD *et al.* 1960, DÜŞEN *et al.* 2010, DÜŞEN, OĞUZ 2010), *Bufo bufo* (YILDIRIMHAN *et*

*al.* 1997, DÜŞEN *et al.* 2010, DÜŞEN, OĞUZ 2010, DÜŞEN 2011), *Pelobates syriacus* (YILDIRIMHAN *et al.* 1997, YILDIRIMHAN, BURSEY 2010), *Ommatotriton ophryticus* (formerly known as *Triturus vittatus*) (YILDIRIMHAN 2008), *Triturus karelinii* (YILDIRIMHAN 2008), *Rana dalmatina* (DÜŞEN *et al.* 2009) in Turkey. Although *O. brevicaudatum* was reported from several amphibians, there was no record from *Hyla orientalis* in Turkey. So this parasite was new record for *H. orientalis* in Turkey.

There are several studies about parasite species of anurans in different countries and Turkey. These studies are important for the study of the geographical distribution of parasite species and relationship with their hosts. Our study contributed to determining the amphibian parasite fauna of Turkey. Additional studies on different populations of *H. orientalis* are needed for this purpose in the future.

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