Diversity of helminths in polish reptiles: a review

Natalia Kuśmierek1 · Ewa Pyrka1 · Marcin Popiolek1

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Abstract
The purpose of this review was to summarise the current species richness of helminths in the reptiles from Poland and to provide a host-parasite list along with studied localities. Between 1926 and 2018 the occurrence of at least 37 taxa of helminths were observed; of these, 19 belong to Trematoda, 14 to Nematoda, two to Cestoda, and two to Acanthocephala. In total, reports of 642 specimens of reptiles from 15 locations were included in this review, namely: 166 Natrix natrix (10 locations), 159 Vipera berus (6), 155 Lacerta agilis (6), 107 Zootoca vivipara (5), 52 Anguis fragilis sensu lato (4), and 3 Zamenis longissimus (1). The highest species richness of internal parasites was observed in N. natrix (at least 26 helminth taxa) and V. b er u s (20). Two out of nine species of reptiles, Coronella austriaca and Emys orbicularis, have not yet formed part of any helminthological surveys in Poland. To date, research conducted on helminths infecting reptiles has primarily been throughout eastern Poland. Over 60% of studied reptiles were caught in the Bieszczady Mts. or in Warsaw and its surroundings. Based on the current review, future research should focus on host species for which intestinal parasite data are poorly known i.e. E. orbicularis, Z. longissimus, C. austriaca. In general surveys should also expand into the western part of the country. Additionally, interspecies differences between two cryptic species of slow worms, Anguis fragilis and A. colchica, would provide interesting data on parasite host specificity.

Keywords  Endoparasites · Digenea · Nematoda · Lizards · Snakes · Poland

Introduction
Reptiles play an important role as final or intermediate hosts in the life cycles of a wide variety of metazoan parasites (Jacobson 2007; Zelmer and Platt 2008). Because these ectotherm organisms inhabit a wide spectrum of habitats, present a range of body sizes, life history traits, and modes of reproduction and feeding activity, they afford great opportunity for the study of patterns and processes influencing the organisation, distribution and abundance of the helminth community. Moreover, research of reptile parasites is important for understanding the relationship between the host, parasite, and the environment. Although several checklists concerning the helminths of reptiles, from different parts of the world, have recently been published (e.g. Lunaschi and Drago 2007; Werneck and Da Silva 2016; San-Martín-Ordenes et al. 2019), data from Central Europe are still lacking.

Currently, reptiles native to Poland comprise nine species, including one species of turtle: the European pond turtle Emys orbicularis (Linnaeus, 1758), four species of lizards: the sand lizard Lacerta agilis Linnaeus, 1758, the viviparous lizard Zootoca vivipara (Lichtenstein, 1823), the common European slow worm Anguis fragilis Linnaeus, 1758, and the Eastern slow worm A. colchica (Nordmann, 1840), and four species of snakes: the grass snake Natrix natrix (Linnaeus, 1758), the common European viper Vipera berus (Linnaeus, 1758), the smooth snake Coronella austriaca Laurenti, 1768, and the Aesculapian snake Zamenis longissimus (Laurenti, 1768). The green lizard Lacerta viridis (Laurenti, 1768) is considered extinct (Głowaciński 2001); the status of two other taxa, the dice snake Natrix tessellata (Laurenti, 1768) and the common wall lizard Podarcis muralis (Laurenti, 1768), is uncertain (Sura 2018a, 2018b). Only one juvenile N. tessellata was reported near the Czech border in 2009 (Vlček et al. 2010), but in the case of P. muralis it is not known whether the two populations inhabiting southwestern Poland were introduced or should be considered relicts (Wirga and Majtyka 2013; Kolenda et al. 2019).
Data of reptile-helminth relationships from Poland are scarce, with the majority of available reports of native reptile gut parasites from the last century. The first record of helminth parasites in a reptile from Poland was described in 1926 (Ruszkowski 1926). Subsequent reports generally referred to sporadic observations or were focused on specific helminth species (e.g. Grabda-Kazubska 1967; Okulewicz 1976; Bertman and Okulewicz 1984, 1987; Kondzior et al. 2018). Increased interest in native reptile helminths emerged in the 1990s (Lewin 1990, 1992a, 1992b, 1992c, 1993; Lewin and Grabda-Kazubska 1997). Although several efforts to summarise the knowledge of helminths in Polish reptiles have been made in the past, these reports failed to include all the available data (Grabda-Kazubska 1963, 1972; Pojmanska and Niewiadomska 2003; Pojmanska et al. 2007).

Most helminths recorded in native reptiles belong to obligatory, mostly autogenic, endoparasites, localized in the gastrointestinal tract or, more rarely, in the accompanying internal organs. There are two groups among them: with complex (all digenean and acanthocephalan), as well as simple (e.g. some nematodes) life cycles, for which reptiles can play the role of definitive, intermediate or paratenic hosts. Most helminths are characterized by relatively wide host specificity, occurring in several host species.

The aims of this study were to summarise the current species richness of helminths in native reptiles of Poland, to provide a host-parasite list with localities and numbers of reptiles examined, and, finally, to indicate directions for further research on intestinal parasites of native Polish reptiles.

Materials and methods

Information of helminths infecting native reptiles was found using search engines and databases: Scopus, Web of Science, Google Scholar, and Google Books. Additionally, data on reptile helminths (not available on the Internet) was found in local scientific journals and books. Articles written in different languages, namely Polish, English, German and French, were included. The data presented below was prepared strictly on the basis of scientific papers on endohelmints identified in Polish reptiles published between 1926 and 2018. Dissertations, theses, and abstracts from conferences were not considered.

In order to enhance the transparency of the presented data, all sampling sites originally described by authors in source articles were grouped into 15 locations: 1) Hel, 2) Sejny, 3) Gaudy Lake, 4) Iława, 5) Biebrza (Biebrza Valley, Biebrza National Park), 6) Białowieża Primeval Forest, 7) Gostynin-Włoclawek Landscape Park, 8) Warsaw (Okęcie district, environs of Warsaw, Kampinos National Park, Zielonka, Zabieniec), 9) Rawka near Skirniewicze, 10) environs of Dęblin, 11) Piotrówka near Kępno, 12) Wrocław (Swojczyce district), 13) Kielce (Białogon district, environs of Kielce, Małogoszcz, Skarżysko-Kamienna), 14) Roźnów, 15) Bieszczady Mts. (Bieszczady National Park, the Czarny Potok valley near Polana, the San valley between Sękowiec and Rajske, the Solinka valley near the village of Buk, environs of Ustrzyki Górne) (Fig. 1).

Results and discussions

In Poland, over a period of nearly 100 years, a total of 642 individuals representing seven species of reptiles were examined. At least 37 taxa of helminths were found: 19 (51.4%) trematodes (Trematoda), 14 (37.8%) nematodes (Nematoda), two (5.4%) tapeworms (Cestoda), and two (5.4%) acanthocephalans (Acanthocephala) (Table 1). However, species richness may be higher, as some parasites of digestive system were only identified to family or genus level.

Across the lizards (Lacertilia) screened, L. agilis represented the highest number of examined individuals and confirmed intestinal parasites: of the 155 specimens previously examined, 15 taxa of helminths were reported from six locations (Table 1, Fig. 1). Data on Z. vivipara was collected from five locations, in which six species of helminths were detected in a total of 107 individuals. Data on helminths in A. fragilis sensu lato was reported from four sites. In total, 52 studied individuals hosted seven gut parasite taxa. In Poland, there are currently two recognised species of Anguis, A. fragilis sensu stricto (until recently considered the only species in the country), and A. colchica (Gwoźdik et al. 2010). Regarding the distribution of both species of slow worms (Jablonski et al. 2017; Skórzewski 2017), it is possible that the individuals referred to in previous parasitological surveys, which came from the locations Bieszczady, Biebrza, and Kielce (n = 48), belonged to A. colchica. Whereas the slow worms (n = 4) from Hel were most likely correctly classified as A. fragilis s. s. The parapatric distribution of slow worms in Poland with contact zones across the country (Skórzewski 2017; Jablonski et al. 2017) may suggest interspecies differences in parasite communities; however, other reptile species are capable of hosting helminths specific to slow worms (see Table 1) and spreading them throughout the country.

Among both snakes (Serpentes) and reptiles in general, N. natrix is the most studied species in Poland to date. In total, 166 specimens from 10 locations were subjected to helminthological research; 26 taxa of gut parasites were found (Table 1, Fig. 1). The second most-studied reptile is V. berus. In six locations (n = 159), vipers were infected by a total of 20 taxa (Table 1, Fig. 1). The only data regarding Z. longissimus comes from three road-killed snakes from Bieszczady that hosted four taxa of intestinal parasites (Lewin 1993). However, it should be emphasised that the distribution of the Polish population of Z. longissimus, which amounts to only...
150–200 individuals (Głowaciński 2018), is limited to the Bieszczady Mountains, which represent the limit of its range. Therefore, further studies of helminths infesting this species may be difficult. The latter snake species, C. austriaca, has not been the subject of helminthological research. Only the presence of unidentified nematodes was reported in one individual from the Gostynin-Włocławek Landscape Park during a survey on the detection of Salmonella spp. in native snakes (Zajęczała et al. 2016). This snake is a secretive and notoriously difficult species to detect in the field (Profus et al. 2018), thus data on the helminths infecting this species throughout Poland and other Central European countries are scarce. In Belarus, 10 individuals hosted four species of helminths, Alaria alata (Goeze, 1782), Ascarops strongylina (Rudolphi, 1819), Physocephalus sexalatus (Molin, 1860), and Serpentirhabdias fuscovenosa (Railliet, 1899) (formerly Rhabdias fuscovenosa) (Shimalov and Shimalov 2010), whereas in the south-eastern parts of the Czech Republic only nematodes of the genus Abbreviata Travassos, 1920 were detected across three C. austriaca (Borkovcová and Kopřiva 2005). Two of the parasites observed in Belarus, A. alata and S. fuscovenosa, have also been detected in several species of reptiles from Poland (Table 1).

There is also a lack of helminth data for the only native turtle, E. orbicularis (Testudines). So far, only one specimen of E. orbicularis, from an unknown locality (purchased in a pet shop in Warsaw), has been examined and found parasitized by the nematode, Spiroxys contortus (Rudolphi, 1819) (Łukasiak...
| Table 1 Helminth taxa found in Polish reptiles |
|---------------------------------------------|
| **Af/ Ac** | **La** | **Nn** | **Vb** | **Zl** | **Zv** | **Location** | **References** |
| **Digenea** | | | | | | | |
| Alaria alata (Goeze, 1782) | + | + | + | + | 2–5, 7–10, 14–15 | Grabda-Kazubska 1961; Sulgostowska 1971; Lewin 1992a, 1992b, 1992c; Lewin and Grabda-Kazubska 1997; Zając et al. 2018 |
| Brachylaima spp. Dujardin, 1845 | + | + | | | 15 | Lewin 1990, 1992a |
| Diplodiscus subclavatus (Pallas, 1760) | + | + | + | | 8, 13 | Lewin 1992a; Bertman 1993; Lewin and Grabda-Kazubska 1997 |
| Leptophallus nigrovenosus (Bellingham, 1844) | + | | | | | 3, 4, 8–10, 13, 15 | Grabda-Kazubska 1961, 1972; Lewin 1992c; Bertman 1998 |
| Macrodera longicollis (Abildgaard, 1788) | + | | | 3–5, 8–10 | | | Grabda-Kazubska 1961; Sulgostowska 1971; Lewin 1992c |
| Metalepophallus gracilimus (Lühe, 1909) | + | | | 3–5, 8–10, 15 | | | Grabda-Kazubska 1961; Lewin 1992c |
| Neodiplostomum minor (Dubois, 1936) | + | | | 3, 9, 10 | | | Grabda-Kazubska 1961 |
| Neodiplostomum spathoides Dubois, 1937 | + | + | | | 4, 8, 15 | | Lewin 1992c; Lewin and Grabda-Kazubska 1997 |
| Opisthioglyphe ranae (Fröhlich, 1791) | + | | | | 8 | Sulgostowska 1971; Lewin 1992c |
| Paralepoderma cloacicola (Lühe, 1909) | + | + | | | 4, 5, 8 | | Lewin 1992c; Lewin and Grabda-Kazubska 1997 |
| Plagiorchis mentulatus (Rudolphi, 1819) | + | + | + | | 8 | Grabda-Kazubska 1972; Lewin 1992a, 1992b; Lewin and Grabda-Kazubska 1997 |
| Plagiorchis molini Lent & Freitas, 1940 | + | + | + | | 8, 15 | Lewin 1992a, 1992b; Lewin and Grabda-Kazubska 1997 |
| Pleurogenoides medians (Olsson, 1876) | + | | | | 8 | Grabda-Kazubska 1972; Lewin 1992a |
| Prostocus confusus (Looss, 1894) | + | | | | 8 | | Lewin 1992a |
| Strigea falconis Szidat, 1928 | + | + | + | | 4, 5, 8, 15 | | Lewin 1992a, 1992b, 1992c; Lewin and Grabda-Kazubska 1997 |
| Strigea sphaerula (Rudolphi, 1803) | + | + | | | 4, 8, 15 | | Lewin 1992c; Bertman 1998; Lewin and Grabda-Kazubska 1997 |
| Strigea strigis (Schrank, 1788) | + | + | + | | 3–5, 8–10 15 | | Ruszkowski 1926; Grabda-Kazubska 1961; Lewin 1992a, 1992c; Lewin and Grabda-Kazubska 1997 |
| Telorchis assula (Dujardin, 1845) | + | | | | 3–5, 8–10, 15 | | Grabda-Kazubska 1961; Sulgostowska 1971; Lewin 1992c |
| Tetracotyle crystallina (Rudolphi, 1819) | + | | | | 3, 9, 10 | | Grabda-Kazubska 1961 |
| Cestoda | | | | | | | |
| Mesocestoides spp. Vaillant, 1863 | + | + | + | + | 2, 5, 8, 15 | | Lewin 1990, 1992a, 1992c, 1993; Lewin and Grabda-Kazubska 1997 |
| Spirometra erinaceieuropaei (Rudolphi, 1819) | + | | | | 6 | | Kondzior et al. 2018 |
| Nematoda | | | | | | | |
| Cosmocerca ornata (Dujardin, 1845) | + | + | | | 4, 8 | | Lewin 1992c; Lewin and Grabda-Kazubska 1997 |
| Entomelas dujardini (Maupas, 1916) | + | + | | | 8, 15 | | Bertman and Okulewicz 1984; Lewin and Grabda-Kazubska 1997 |
| Entomelas entomelas (Dujardin, 1845) | + | | | | 1, 15 | | Markowski 1933; Lewin 1990 |
| Hedrurus androphora Nitzsch, 1821 | + | | | | 8 | | Lewin 1992c |
| Kalicephalus viperae (Rudolphi, 1819) | + | | | | 15 | | Lewin 1993 |
| Mermis nigrescens Dujardin, 1842 | + | | | | 15 | | Lewin 1992b |
| Oswaldocruzia bialata (Molin, 1860) | + | | | | 8 | | Grabda-Kazubska 1972 |
| Oswaldocruzia dispar (Dujardin, 1845) | + | | | | 1 | | Markowski 1933 |
| Oswaldocruzia filiformis (Goeze, 1782) | + | + | + | + | 2, 4, 5, 8, 13–15 | | Bertman and Okulewicz 1987; Lewin 1990, 1992a, 1992b, 1992c; Lewin and Grabda-Kazubska 1997; Bertman 1998 |
1939). However, additional data is available from Polish Polesia, a region belonging to Poland during the interwar period. Between 1926 and 1938, over 130 specimens of *E. orbicularis* were collected, in which four species of flukes, *Astiotrema emydis* Ejsmont, 1930, *Cercorchis parvus* (Braun, 1901), *C. solivagus* (Odhner, 1902), and *Spirhapalum polesianum* Ejsmont, 1927, and two species of nematodes, *Spironoura armenica* Massino, 1924 and *S. contortus*, were detected (Ruszkowski 1926; Ejsmont 1927; Modrzejewska 1938). Due to the proximity of the Polesie Lubelskie, the area with the largest native population of *E. orbicularis* (Najbar and Rybacki 2018), it can be assumed that these turtles are hosts for the same species of helminths. In addition, in the same region of the country, the third stage of *S. contortus*, a nematode that utilizes *E. orbicularis* as the final host, was found in a lake minnow *Eupallasella percnurus* (Pallas, 1811) (Popiołek et al. 2005).

Previous reports on helminths in native reptile species suggest a tendency for research to be conducted in the eastern, and especially in the south-eastern part, of Poland. Among 642 reptile specimens studied to date, 210 (32.7%), belonging to six species, were collected in the Bieszczady Mts. (Fig. 1). Another popular location for helminthological surveys was Warsaw (including the city’s surroundings and Kampinos National Park). A total of 194 (30.2%) reptile specimens, representing four species, were collected there; 80 (12.5%; 3 species) were collected from the Biebrza locality, and the remainder (158, 24.6%) from other localities (Fig. 1). Selection of study sites was likely based on the location of the author’s affiliations or on the occurrence of the host species (e.g. the range of *Z. longissimus* does not cover the entire territory of Poland). Indeed, most authors collected their material close to the institution where they worked, and Bieszczady Mts., the location with the highest numbers of studied species and of individuals, is a Polish hotspot for native reptiles (Blązuk 2007).

Currently, parasitological surveys of wild reptiles in Poland is challenging due to the conservation status of these species (all of which are protected by Polish law) and the lethal procedures followed to collect internal parasites. These studies require permission from national authorities and ethics committees, which is difficult to obtain, especially for species that are rare and endangered (e.g. *E. orbicularis* or *Z. longissimus*). Considering that reptiles are globally threatened with extinction (Gibbons et al. 2000; Fitzgerald et al. 2018), and some species living in Poland are endangered on a national scale (Głowaciński 2001), the attention of researchers is being directed towards non-invasive methods, such as collecting road-killed carcasses (Lewin 1993). An alternative and non-invasive methods utilised are molecular analysis of fecal samples and coproscopy; however, in the case of the latter method, due to the morphological and biometric similarity of most reptile endoparasite dispersive forms (mainly eggs), the results are often inaccurate. In addition, improved collaboration between parasitologists and herpetologists, using specimens from museum collections, along with collection of reptile road-kills, may lead to the acquisition of a large number of hosts for the study of helminth and reptile relationships.

| Location | References |
|----------|------------|
| Warsaw | Lewin 1990, 1992a, 1992c; Lewin and Grabda-Kazubska 1997 |
| Bieszczady Mts. | Markowski 1933; Łukasiak 1939; Lewin 1990, 1992a, 1992c; Lewin and Grabda-Kazubska 1997 |
| Kampinos National Park | Lewin 1993 |
| *Eupallasella percnurus* | Lewin 1992a, 1992c, 1993; Lewin and Grabda-Kazubska 1997 |

**Table 1** (continued)

| Location | References |
|----------|------------|
| Warsaw | Lewin 1990, 1992a, 1992c; Lewin and Grabda-Kazubska 1997 |
| Bieszczady Mts. | Markowski 1933; Łukasiak 1939; Lewin 1990, 1992a, 1992c; Lewin and Grabda-Kazubska 1997 |
| Kampinos National Park | Lewin 1993 |
| *Eupallasella percnurus* | Lewin 1992a, 1992c, 1993; Lewin and Grabda-Kazubska 1997 |

Af/Ac – *Anguis fragilis/A. colchica*, La – *Lacerta agilis*, Nn – *Natrix natrix*, Zl – *Zamenis longissimus*, Zv – *Zootoca vivipara*, Vb – *Vipera berus*. Location numbers refer to those from materials and methods section.
Conclusions

Since reptiles are regarded as vulnerable and research focused on understanding the factors threatening them is a priority (Gibbons et al. 2000), the constant monitoring, which includes parasite surveys of native reptiles, is important. Thus, in order to determine the exact distribution of particular helminth species, further research should include new study areas, including western Poland, and should focus on host species, for which intestinal parasite communities are less known (i.e. E. orbicularis, Z. longissimus, C. austriaca, A. fragilis and A. colchica). Additionally, further research on the helminth fauna of native turtles should be a priority, especially regarding possible transfer of parasites between them and invasive Trachemys spp. (Demkowska-Kutrzepa et al. 2018).

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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