Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

ASSESSMENT OF THE POPULATION DENSITY OF *GALBA TRUNCATULA* SNAIL (MÜLLER, 1774) IN HABITATS OF TELEORMAN COUNTY, ROMANIA

Poliana Tudor [1,*], Dana Tăpăloagă [1]

[1]Faculty of Veterinary Medicine, University of Agronomic Sciences and Veterinary Medicine of Bucharest, Splaiul Independenței, no. 105, 050097, Bucharest, Romania

Current Trends in Natural Sciences

Abstract
Galba truncatula is a widespread gastropod, showing great adaptability and the ability to populate different types of habitats. The aim of this study to identify habitats populated by G. truncatula in two localities in the South of Teleorman County where ruminants are raised, as well as to establish the density of this gastropod in the identified habitats. Two areas were randomly established in each locality, of 10 m^2 each, from where specimens of G. truncatula were collected. The density estimation was performed using the method of counting the number of snails in an area of 1 m^2 in a unit of time. A number of 1761 specimens were collected from the 4 assessed habitats, and the density of snails varied from 20 to 71 specimens/m^2. Of all the examined quadrats, 60% recorded a density >40 specimens/m^2. No quadrat was recorded with a density <10 specimens/m^2, which proves that the evaluated habitats present optimal conditions for the development of the G. truncatula population.

Keywords: density, Galba truncatula, gastropod, habitats.

1. INTRODUCTION

The gastropod *Galba truncatula* (Müller, 1774), known until recently as *Lymnaea truncatula*, is a freshwater snail with a wide distribution throughout the world, having been identified in North and South America (Bolivia, Peru, Argentina, Chile, Venezuela) (Bargues et al, 2012), in parts of Africa (Morocco, Algeria, Tunisia, Egypt) (Mekroud et al., 2002) and Asia (Russia, Iran, Pakistan, India) (Glöer and Pešić, 2012; Yakhchali et al, 2015), as well as throughout Europe (Trouvé et al., 2005; Hörweg et al., 2011; Jones et al., 2015; Dreyfuss et al., 2016; Vignoles et al., 2017), including most Mediterranean islands, such as Corsica, Malta, Azores, Balearic, Canary, etc. In Romania, *G. truncatula* is widely spread (Glöer and Sîrbu, 2005), although an exact distribution of this species is not available.

G. truncatula is highly adaptable, inhabiting permanent aquatic habitats (streams, ponds, irrigation ditches) as well as temporary ones (ditches, puddles, swamps, wet or marshy meadows, and in "any land depression where water accumulates" such as animal hoofprints)(Truvé et al., 2003; Dreyfuss et al., 2016). They prefer areas with clayey, uneven, slightly calcareous soils. On larger areas of water, it prefers to be located at the edge, preferring muddy portions. The development of this gastropod is mainly dependent on climate, favorable conditions being represented by temperatures of 10-25°C and relatively high humidity, which is ensured by the presence of precipitation (Vignoles et al., 2017). *G. truncatula* feeds on chlorophilic and cyanophytic algae, as well as on

*Corresponding author, E-mail address: polianatudor@yahoo.com

Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

fresh or decaying plant parts, playing a crucial role in the functioning of freshwater habitats through their involvement in the local cycling of nutrients and plant matter in their environments (Mulero et al., 2021).

Similar to other freshwater gastropods (*Lymnaea palustris*, *Radix* spp.), *G. truncatula* has medical and veterinary importance as it is the main intermediate host for the trematode *Fasciola hepatica* (Jones et al., 2015; Vignoles et al., 2017), which causes significant losses in large and small ruminant livestock (Mehmood et al., 2017; Vignoles et al., 2017). Moreover, fasciolosis is a parasitic disease with important economic impacts, causing financial losses to farmers (through decreased productivity and increased animal mortality) and to traders (poor quality carcasses) (Mehmood et al., 2017). In addition, humans can be infested with this trematode, and human fasciolosis has alarming values worldwide (Mas-Coma et al., 2018).

The aim of this study was i) to identify the habitats populated by *G. truncatula* in two regions of Teleorman County, Southern Romania and ii) to evaluate the density of this gastropod in the selected habitats.

2. MATERIALS AND METHODS

Study area

During the period October-November 2019, open pastures located along the banks of the Teleorman stream, respectively Călmățui stream, where sheep and goats of the citizens of the two localities are raised, were visited (Figure 1).

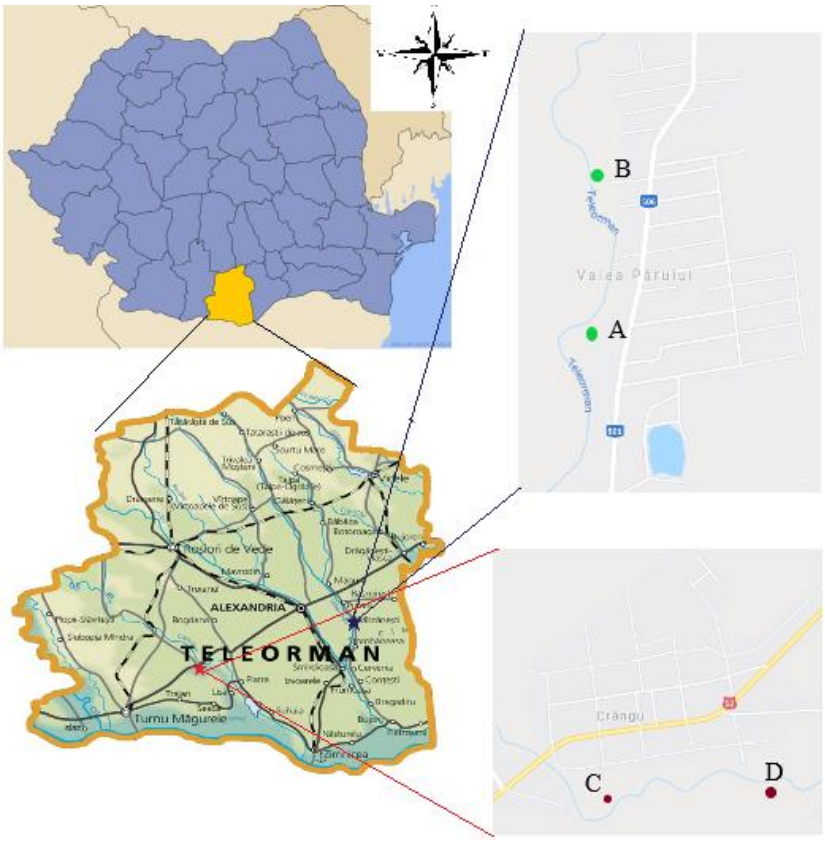

Figure 1. Geographical location of the visited sites for the identification of G. truncatula gastropod habitats ● in Valea Părului (Southern area - A; Northern area - B); ● in Crângu (Western area - C; Eastern area - D)

*Corresponding author, E-mail address: polianatudor@yahoo.com

Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

The first locality visited was the village of Valea Părului, commune of Mârzănești (43º55'12 "N 25º27'55 "E), situated at an altitude of 35 m above sea level. The second location was the village of Crângu, commune of Crângu (43º50'45 "N 25º04'24 "E), situated at an altitude of 38 m a.s.l. On the banks of the streams mentioned above (comprising pasture, stream bank and adjacent marshy areas) two plots of land (10/1 m each plot) were randomly demarcated in each locality (Figures 2 and 3) (Malone et al., 1984). In addition, each plot was divided into 10 quadrats of $1m^2$ each (numbered from 1 to 10) to determine the density of the snail population per m^2. The climate in both areas is temperate-continental, which is typical for the Muntenia region, and the mean annual precipitation ranges from 474 mm to 583.7 mm. The vegetation is predominantly that of lowland meadows, consisting of mesophilic and mesohygrophilous plant species, alternating with the characteristic vegetation of lake/stream banks (composed of reeds, canes and various species of the *Polygonaceae* family, which tolerate temporary water cover).

Figure 2. Pasture on the bank of the Teleorman stream (Valea Părului village)

Figure 3. Pasture on the bank of the Călmățui stream (Crângu village)

Sampling and identification of snails

From each plot, only specimens of *G. truncatula* were collected, mainly by hand, and in a few cases with the help of colanders. The specimens collected from each habitat were placed separately in plastic tubes with water from the respective habitat and identification data, i.e. perimeter and habitat of origin, were written on each container. Estimation of the population density of *G. truncatula* snails was performed by counting the number of snails in a marked area ($1m^2$) in a unit of time (Mekroud et al., 2002; Bargues et al., 2012) (Figure 4).

Statistical analyses

To assess significant differences between snail densities in the 4 plots, univariate one-way ANOVA analysis was used. To determine whether the observed differences are statistically significant, a value of significance of 0.05 was chosen. As a post hoc test, the Tukey-Kramer test was used to compare plots' means and identify significant differences among them. The mentioned statistical analysis were carried out by MedCalcTM v22.023 software, thus ensuring the correctness and precision of the results.

*Corresponding author, E-mail address: polianatudor@yahoo.com

Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

3. RESULTS AND DISCUSSIONS

A total of four grazing plots on the banks of the Teleorman and Călmățui streams were analysed. In the village of Valea Părului one plot was assessed in the Southern part of the village (marked A) and one in the Northern part (marked B), at a distance of 5 km from each other, and in the village of Crângu one plot was assessed in the Western part (marked C) and one in the Eastern part (marked D), at a distance of 7 km from each other (Figure 1). During the visit, the ambient temperature varied between 22 and 26°C and the sky was sunny. All assessed plots were classified as permanent semi-aquatic habitats, although this species can populate both permanent and temporary aquatic habitats (Trouvé et al., 2003).

A total of 1761 specimens of *G. truncatula* were collected from the four habitats, with the following taxonomic classification: Regn: Animalia; Order: Mollusca; Class: Gastropoda; Family: Lymnaeidae; Subfamily: Lymnaeinae; Genus: *Galba*. *G. truncatula* is a gastropod that has reverted to aquatic life, breathing through a "lung" that fills with water, allowing it to use air from the aquatic environment (Tudor, 2020). The shell is a hard, grey-brown calcareous structure with a slightly ridged surface. It has several coils (up to 5-6 coils), is twisted to the right (*dextral* shell) and its diameter decreases towards the apex. The length of the shell varies between 5 and 12 mm and the width between 2.5 and 6 mm (Tudor, 2020).

In the four visited plots, the density of snails varied from one perimeter to another (Table 1). In the village of Valea Părului, 917 specimens were identified and the density varied between 20 and 52 specimens/m^2 (with an average of 35.5 specimens/m^2) in the Northern area and between 32 and 71 specimens/m^2 (with an average of 56.2 specimens/m^2) in the Southern area. In the village of Crângu, a total of 844 specimens were identified and the density varied between 33 and 47 specimens/m^2 (with an average of 41.1 specimens/m^2) in the Western area and between 21 and 56 specimens/m^2 (with an average of 43.3 specimens/m^2) in the Eastern area.

The results show the presence of the *G. truncatula* in all four evaluated plots. Although the two localities are approximately 40 km apart, the obtained results indicate that these areas represent good habitats for the development of the gastropod population.

Table 1. Number of specimens sampled in each area (of 1m^2)

Marked plot	**The quadrat in each plot**									
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
A	71	62	32	58	59	58	62	61	49	50
B	52	20	28	37	48	27	30	33	48	32
C	43	38	47	33	39	40	47	45	37	42
D	56	49	51	52	44	47	45	39	29	21
Total	**222**	**169**	**158**	**180**	**190**	**172**	**184**	**178**	**163**	**145**

In the village of Valea Părului, there was a significant difference in snail population size between the two plots. On the plot in the Northern area, located upstream and close to the population's households and farmland, was recorded the lowest value of the number of gastropods sampled. Although previous studies report variations in snail population prevalence in correlation with the type of habitat (Dreyfuss et al., 2016), the assessed habitats in the current study do not show different characteristics, especially with regard to soil type or ambient temperature. The reduced population recorded in the Northern area of Valea Părului village may be a consequence of the fact that the stream bed is shallower and narrower in that sector, presenting also a lower flow rate, and

*Corresponding author, E-mail address: polianatudor@yahoo.com

Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

the vegetation is also more reduced. In fact, it is known that vegetation, microclimate conditions and pasture topography are factors that influence the development of *G. truncatula* populations, but also of larval forms of the trematode *F. hepatica* (Vignoles et al., 2017). In addition, in recent years, the area has been used increasingly less by shepherds as a grazing area, due to its location (in the vicinity of people's homes and farmlands). On the contrary, the area has been used as an access road to plots of land situated at the edge of the village, on which vegetables and cereals are cultivated. Previous studies have reported that the survival of gastropod populations in a habitat depends on many parameters, such as meteorological factors, soil type and habitat type (Truvé et al., 2003). It has also been shown that stress caused by lack of water in a habitat is an important factor for gastropod populations. However, adult snails can survive in drought conditions for several weeks or even months (Chapuis et al., 2007), having the ability to enter estivation ('summer sleep'). In the Southern part of the village of Valea Părului, the stream bed is much wider, the banks almost absent and the vegetation much more abundant. The adjacent land is more marshy, and the pasture opening much more generous, being used by a large number of animals as grazing land. The differences between the plots located in the village of Crângu were not significant, the habitat characteristics being similar in both plots.

Concerning the snail density in the present study, it was assessed according to those previously published by Dreyfuss et al. (2016). Of the total examined quadrats (n = 40), the highest population density of *G. truncatula* (>40 specimens/m^2) was recorded in 60% of the marked quadrats. In 35% of the examined quadrats, the snail density was between 25.1 and 40 specimens/m^2 , and in 5% of the marked quadrats, the density was between 10.1 and 25 specimens/m^2. None of the examined quadrats recorded a density <10 specimens/m^2, which demonstrates the presence of favourable conditions for the development of the *G. truncatula* population in the evaluated habitats in this study. Previous studies have shown a higher prevalence of habitats containing 1 to 10 snails/m^2 (42.7%), while habitats containing 25.1 - 40 snails/m^2 or >40 snails/m^2 showed a lower prevalence (11.5% and 0.8%, respectively) (Dreyfuss et al., 2016). The differences may be a consequence of the different number of assessed habitats as well as different characteristics of those habitats.

Following the ANOVA statistical analysis of the recorded data regarding the density of snails in the two assessed habitats, respectively the 4 plots, assuming the two variances to be equal, a significance level of less than 0.05 was obtained, resulting in significant differences between the recorded values ($p < 0.001$), with a normal distribution (Shapiro-Wilk Test, P=0.1862). Applying the Tukey-Kramer post hoc test to compare the means of the plots and identify significant differences among them, it was found that the snail density values recorded in plots B, C and D differed significantly from the snail density value recorded in the A plot. Recorded differences can be associated to the characteristics of the habitat located in plot A, where the river stream is wider, muddy, and the banks are practically absent, favouring the frequent flooding of the pastures, thus providing much more favourable conditions for the development of gastropods.

G. truncatula is predominantly found around water sources and due to their role as intermediate host contributes to infestation of water plants and surrounding vegetation with larval forms of the trematode *F. hepatica*, subsequently leading to contamination of animals and humans. The World Health Organization lists fasciolosis as a Neglected Tropical Disease (NTD) in the group of food-borne trematodes (WHO, 2013), and WHO reports over 2.4 million people infected with *F. hepatica* worldwide (Mehmood et al., 2017). Consequently, identifying and monitoring the habitats of the gastropod *G. truncatula* is essential to prevent the transmission of fasciolosis to animals and humans, considering the known role of this snail as an intermediate host. Recent studies showed a

*Corresponding author, E-mail address: polianatudor@yahoo.com

Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

51.3% prevalence of *F. hepatica* infestation in snails collected from habitats located in the Eestern part of the country (Sîrbu et al., 2019).

4. CONCLUSIONS

The results of the present study show the presence of the gastropod *G. truncatula* in the evaluated habitats in Teleorman County, with a predominance of quadrats with a density >40 specimens/m^2. These results require further investigations to detect the infestation of this gastropod with the trematode *F. hepatica* and to assess the prevalence of infestation in the evaluated habitats.

5. ACKNOWLEDGEMENTS

This study was supported by the project no. 1488/2019 - MONGALFAS.

6. REFERENCES

Bargues, D.M., Artigas, P., Khoubbane, M., Ortiz, P., Naquira, C., Mas-Coma, S. (2012). Molecular characterisation of *Galba truncatula*, *Lymnaea neotropica* and *L. schirazensis* from Cajamarca, Peru and their potential role in transmission of human and animal fascioliasis. *Parasit Vectors*, *5*, 174.

Chapuis, E., Trouve, S., Facon, B., Degen, L., Goudet, J. (2007). High quantitative and no molecular differentiation of a frechwater snail (*Galba truncatula*) between temporary and permanent water habitats. *Mol Ecol*, *16*, 3484-3496.

Dreyfuss, G., Vignoles, P., Rondelaud, D. (2016). Current decline in the number and size of *Galba truncatula* and *Omphiscola glabra* populations, intermediate hosts of *fasciola hepatica*, on the acidic soils of Central France. *Parasite*, *23*, 46.

Glöer, P., Sîrbu, I. (2005). New freshwater molluscs species found in the Romanian fauna. *Haldia*, *6* (5/6), 229-238.

Glöer, P., Pešić, V. (2012). The freshwater snails (Gastropoda) of Iran, with description of two genera and eight new species. *ZooKeys*, *219*, 11-61.

Hörweg, C., Prosl, H., Wille-Piazzai, W., Joachim, A., Sattmann, H. (2011). Prevalence of *Fascioloides magna* in *Galba truncatula* in the Danube backwater area east of Vienna, Austria. *Wien Tierärztl Monatsschr - Vet. Med. Austria*, *98*, 261-267.

Jones, A.R., Williams, H.W., Dalesman, S., Brophy, M.P. (2015). Confirmation of *Galba truncatula* as an intermediate host snail for *Calicophoron daubneyi* in Great Britain, with evidence of alternative snail species hosting *Fasciola hepatica*. *Parasit Vectors*, *8*, 656.

Malone, J.B., Loyacano, A.F., Hugh-Jones, M.E., Corkum, K.C. (1984). A three-year study on sesonal transmission and control of *Fasciola hepatica* of cattle in Louisiana. *Prev Vet Med*, *3*, 131-141.

Mas-Coma, S., Bargues, M.D., Valero, M.A. (2018). Human fascioliasis infection sources, their diversity, incidence factors, analytical methods and prevention measures. *Parasitology*, *145*, 1665-1699.

MedCalc® Statistical Software version 22.023 (MedCalc Software Ltd, Ostend, Belgium. https//www.medcalc.org; 2024.

Mehmood, K., Zhang, H., Sabir, A.J., Abbas, R.Z., Ijaz, M., Durrani, A.Z., Saleem, M.H., Rehman, M.U., Iqbal, M.K., Wnag, Y., Ahmad, H.I., Abbas, T., Hussain, R., Ghori, M.T., Ali, S., Khan, A.U., Li, J. (2017). A review on epidemiology, global prevalence and economical losses of fasciolosis in ruminants. *Microb Pathog*, *109*, 253-262.

Mekroud, A., Benakha, A., Benlatreche, C., Rondelaud, D., Dreyfuss, G. 2002 - First studies on the habitats of *Galba truncatula* (Mollusca Gastropoda: Lymnaeidae), the snail host of *Fasciola hepatica*, and the dynamics of snail populations in Northeastern Algeria. *Revue Méd Vét, 153* (3), 181-188.

Mulero, S., Toulza, E., Loisier, A., Zimmerman, M., Allienne, J.F., Foata, J., Quilichini, Y., Pointier, J.P., Rey, O., Boissier, J. (2021). Malacological survey in a bottle of water: A comparative study between manual sampling and environmental DNA metabarcoding approaches. *Glob Ecol Conserv*, *25*, e01428.

Sîrbu, C., Oprescu, I., Dărăbuș, Gh., Imre, M., Suici, T., Jitea, B., Ilie, M.S., Morariu, S. (2019). PCR diagnosis of Fasciola hepatica in intermediary hosts-snails collected from the environment. *Scientifical Papers: Veterinary Medicine, Timisoara, vol LII* (1), 93-102.

Truvé, S., Degen, L., Renaud, F., Goudet, J. (2003). Evolutionary implications of a high selfing rate in the freswater snail *Lymnaea truncatula*. *Evolution*, *57* (10), 2303-2314.

*Corresponding author, E-mail address: polianatudor@yahoo.com

Current Trends in Natural Sciences
Vol. 13, Issue 25, pp. 200-206, 2024
https://doi.org/10.47068/ctns.2024.v13i25.024
Current Trends in Natural Sciences (on-line)
ISSN: 2284-953X
ISSN-L: 2284-9521
Current Trends in Natural Sciences (CD-Rom)
ISSN: 2284-9521
ISSN-L: 2284-9521

Trouvé, S., Degen, L., Goudet, J. (2005). Ecological components and evolution of selfing in the freshwater snail *Galba truncatula*. *J Evol Biol*, *18*, 358-370.

Tudor, P. (2020). Biologie Animale [Animal biology]. Printech Publishing House, Bucharest.

World Health Organization. (2013). Sustaining the drive to overcome the global impact of neglected tropical diseases. Second who report on neglected tropical diseases. 85- 90.

Yakhchali, M., Imani, Baran A., Malekzadeh-Viayeh, R. (2015). Molecular detection of the infection with *Fasciola hepatica* field-collected snails of *Galba truncatula* and *Lymnaea stagnalis* from West Azarbaijan, Iran. *Arch Razi Inst*, *70* (3), 195-202.

*Corresponding author, E-mail address: polianatudor@yahoo.com