The food composition of two brown frog populations
(Rana dalmatina and Rana temporaria) from Sălaj County, Romania

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Abstract. The studied individuals fed very intensely during the research. There are no important differences between the feeding of the two species or of the Rana dalmatina adults and sub-adults. The food competition between the two species was reduced, or even lacked, at least during the study. In the stomach contents of both species and size classes we identified vegetal matter, shed-skin fragments and animal preys belonging to 32 invertebrate groups. The Araneida was the most important prey taxa for R. dalmatina, while the Coleoptera was the most important one for R. temporaria. R. temporaria registered the highest food diversity, while the lowest one was recorded by the R. dalmatina sub-adults. Both species and R. dalmatina classes use the sit-and-wait strategy, consuming large preys. R. temporaria consumed the largest preys, while the R. dalmatina sub-adults captured the smallest ones. Both species seem to be opportunistic predators, without trophic selectivity.

Key words: Rana dalmatina, Rana temporaria, brown frogs, food composition, invertebrate preys, Romania.

Introduction

Amphibians that reproduce in puddles usually depend on a higher complexity of the habitats, needing both aquatic habitats for reproduction and feeding and hibernation ones (Hartel et al. 2008a). The last two are usually terrestrial habitats. Among the amphibians from Romania that are found in this situation, the most known ones are the brown frogs, among which two species are common in the country, Rana dalmatina and Rana temporaria. At least in the western part of Romania, they appear very often in the same localities, using both the same aquatic and terrestrial habitats (Hartel 2005, Covaciuc-Marcov et al. 2006a, 2007, 2008a). However, R. temporaria is usually connected to higher altitudes, being present from 200 m upwards (Cogălniceanu et al. 2000), while R. dalmatina also appears at the plain (Fuhn 1960, Cogălniceanu et al. 2000). Recently however, even R. temporaria was recorded in the western part of the country under the normal altitudinal distribution limit (Covaciuc-Marcov & Ferenși 2008a, Covaciuc-Marcov et al. 2008b). Generally, the two species inhabit forested or wet areas (Cogălniceanu et al. 2000, Covaciuc-Marcov et al. 2008c, 2009a). The situation is also valid in Sălaj County, where the two species usually appear in the same habitats (Covaciuc-Marcov et al. 2009b).

Despite the wide distribution of the two species in western Romania, studies regarding their ecology are relatively few (eg. Hartel 2005, 2008, Hartel et al. 2008b). Both species are considered to be vulnerable in the country (Iftime 2005). The rarity of the data regarding the ecology of the species can affect the future protection programs, knowing the environmental factors responsible for the distribution and the survival of the organisms being an important challenge of the actual conservationist biology (Hartel & Öllerer 2009). Within the context, feeding is an important parameter, the food diversity being an indicator of the quality of the environment in which the amphibians live (Kovács et al. 2007). The feeding information is important for understanding the life history, the population fluctuations or the impact of the habitat modifications upon the amphibians (Anderson et al. 1999). Knowing the feeding ecology of some syntopic species contributes to the understanding of the organization of the respective communities (Juncă & Eterovick 2007).

Consequently we have proposed to analyse the feeding of the two brown frog species from Almăș-Agrij Depression from Sălaj County. There are no comparative studies regarding the feeding of the two species in Romania, only data concerning R. dalmatina exist (Aszalós et al. 2005). The objectives of our study were: i.) establishing the food composition of the two species; ii.) analysing the differences between the food of the two species and, moreover, in the case of R. dalmatina between the adults and sub-adults.
Material and Methods

The study was realised at the beginning of October 2009. The investigated region lies in the northern part of Almăș–Agriș Depression, near Gâlgăul Almașului locality. The locality is situated at approximately 10 km from Jibou town, at 250 m altitude. The habitat is represented by a wet area of about 2 km long, formed on the flanks of a temporary stream. The swamp is mainly covered by thick grass, and also by burs, sporadically presenting fallen logs from the former forestry operation. The swamp has a variable length, ranging from 20 to 100 m, being limited by forests on both sides. Initially they were oak forests, but they were cut down in the lower sector, from the village, being partially replaced with pine plantations. The frogs were captured starting from 1 km upstream of the village, on a length of approximately 500 m during one hour. The time was not enlarged, in order to avoid the digesting of the preys, amphibians rapidly digesting the stomach contents (Caldwell 1996).

On a whole, we analysed the stomach contents from 66 frogs (Table 1). The amphibians were captured directly by hand, being afterwards held in different plastic buckets depending on their size. Regarding R. dalmatina, we captured adults and sub-adults, the analysis being differentiated depending on the size of the frog. In the case of R. temporaria, we only captured 5 juveniles, which were not taken into account because of their low number. We used the stomach flushing method in order to draw the stomach contents (Solé et al. 2005). Afterwards, the frogs were released in their habitat. The samples were stored in air tight test tubes, preserved in formaldehyde and determined in the laboratory. Food composition was evaluated by percentage abundance (%A) and frequency of occurrence (%f). Dietary diversity was estimated with the Shannon-Wiener (1949) diversity index (H) and the similarity with Sorensen index (1948). The signification index was calculated with the Mann-Whitney test (Mann & Whitney 1947).

Results

All of the individuals presented stomach contents. All of the identified preys were terrestrial animals, belonging to 32 invertebrate groups.

R. dalmatina consumed the most prey taxa, 29, while R. temporaria consumed 27. From the 32 prey taxa consumed on a whole, 24 were consumed by both species. The Araneida were the most important preys regarding R. dalmatina, concerning both adults and sub-adults (Table 2). The coleopterans occupied first place in the case of R. temporaria, concerning both the amount and the frequency of occurrence. In the case of R. dalmatina, 17 prey taxa were consumed by both adults and sub-adults. Generally, at this species, the adults consumed more prey taxa than the sub-adults did (25 compared to 21). Regarding both species, in the stomach contents were present prey taxa with low consumption values (under 1%), which were thus accidentally consumed. There are also differences between the amount and frequency of consumption regarding some prey taxa. Thus, at R. temporaria, the coleopterans occupy first place concerning the amount, but register only second place regarding the frequency. In the case of this species, the Lepidoptera larvae occupy first place regarding the consumption frequency, which, although represented only 18.3% from the total preys, were consumed by 95.2% of the studied individuals.

The feeding of both species was intensive during the study. Both the maximum and average number of preys per individual had high values, although most of the preys were relatively large in size (limax, myriapoda, orthoptera, coleoptera carabida). These va-

<table>
<thead>
<tr>
<th>Rana dalmatina</th>
<th>Rana temporaria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Juveniles</td>
<td>Adults</td>
</tr>
<tr>
<td>No. of studied individuals</td>
<td>22</td>
</tr>
<tr>
<td>Vegetal remains (%f)</td>
<td>68.2</td>
</tr>
<tr>
<td>Shed skin (%f)</td>
<td>22.7</td>
</tr>
<tr>
<td>No. of the consumed prey items</td>
<td>115</td>
</tr>
<tr>
<td>- maximum no.</td>
<td>9</td>
</tr>
<tr>
<td>- average no.</td>
<td>5.22</td>
</tr>
<tr>
<td>Feeding diversity (H)</td>
<td>2.38</td>
</tr>
<tr>
<td>Mean feeding similarity (S)</td>
<td>0.44</td>
</tr>
</tbody>
</table>
Table 2. The percentage abundance (%A) and the frequency of occurrence (%f) of the consumed prey items.

<table>
<thead>
<tr>
<th>Animal prey items</th>
<th>Juveniles</th>
<th>Adults</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% A</td>
<td>% f</td>
<td>% A</td>
</tr>
<tr>
<td>Anelida - Oligochaeta</td>
<td>0.87</td>
<td>4.55</td>
<td>-</td>
</tr>
<tr>
<td>Gastropoda - snails</td>
<td>4.35</td>
<td>18.2</td>
<td>2.83</td>
</tr>
<tr>
<td>- limax</td>
<td>1.74</td>
<td>9.09</td>
<td>2.83</td>
</tr>
<tr>
<td>Crustacea - Isopoda</td>
<td>5.22</td>
<td>18.2</td>
<td>0.94</td>
</tr>
<tr>
<td>Arachnida - Pseudoscorpionida</td>
<td>-</td>
<td>-</td>
<td>0.47</td>
</tr>
<tr>
<td>- Acaria</td>
<td>-</td>
<td>-</td>
<td>1.42</td>
</tr>
<tr>
<td>- Araneida</td>
<td>32.2</td>
<td>95.5</td>
<td>25</td>
</tr>
<tr>
<td>- Opilionida</td>
<td>-</td>
<td>-</td>
<td>0.94</td>
</tr>
<tr>
<td>Myriapoda - Chilopoda</td>
<td>-</td>
<td>-</td>
<td>4.72</td>
</tr>
<tr>
<td>- Diplopoda</td>
<td>1.74</td>
<td>9.09</td>
<td>1.89</td>
</tr>
<tr>
<td>Colembola</td>
<td>3.48</td>
<td>13.6</td>
<td>-</td>
</tr>
<tr>
<td>Orthoptera</td>
<td>0.87</td>
<td>4.55</td>
<td>4.72</td>
</tr>
<tr>
<td>Dermaptera</td>
<td>-</td>
<td>-</td>
<td>1.89</td>
</tr>
<tr>
<td>Heteroptera</td>
<td>1.74</td>
<td>4.55</td>
<td>3.77</td>
</tr>
<tr>
<td>Homoptera - Afidina</td>
<td>3.48</td>
<td>4.55</td>
<td>-</td>
</tr>
<tr>
<td>- Cicadina</td>
<td>7.83</td>
<td>36.4</td>
<td>10.8</td>
</tr>
<tr>
<td>Lepidoptera [L]</td>
<td>16.5</td>
<td>59.1</td>
<td>14.2</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td>-</td>
<td>-</td>
<td>0.47</td>
</tr>
<tr>
<td>Coleoptera [L]</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Coleoptera - undet.</td>
<td>6.96</td>
<td>36.4</td>
<td>7.08</td>
</tr>
<tr>
<td>- Carabidae</td>
<td>0.87</td>
<td>4.55</td>
<td>5.19</td>
</tr>
<tr>
<td>- Crysomelidae</td>
<td>-</td>
<td>-</td>
<td>0.47</td>
</tr>
<tr>
<td>- Coccinellidae</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>- Curculionidae</td>
<td>2.61</td>
<td>13.6</td>
<td>1.89</td>
</tr>
<tr>
<td>- Scarabeidae</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>- Staphilinidae</td>
<td>1.74</td>
<td>9.09</td>
<td>1.42</td>
</tr>
<tr>
<td>- Cantaridae</td>
<td>-</td>
<td>-</td>
<td>0.47</td>
</tr>
<tr>
<td>Diptera - Nematocera</td>
<td>0.87</td>
<td>4.55</td>
<td>-</td>
</tr>
<tr>
<td>- Brahicera</td>
<td>0.87</td>
<td>4.55</td>
<td>2.36</td>
</tr>
<tr>
<td>Neuroptera</td>
<td>0.87</td>
<td>4.55</td>
<td>0.47</td>
</tr>
<tr>
<td>Hymenoptera - undet.</td>
<td>2.61</td>
<td>13.6</td>
<td>1.42</td>
</tr>
<tr>
<td>- Formicidae</td>
<td>2.61</td>
<td>9.09</td>
<td>2.36</td>
</tr>
</tbody>
</table>

The food composition of two brown frog populations (Rana dalmatina & R. temporaria) were higher in the case of R. temporaria (Table 1).

The highest food diversity (Table 1) is recorded at R. temporaria (H=2.68). In the case of the R. dalmatina adults, the value comes close to the one registered by R. temporaria (H=2.61). The R. dalmatina sub-adults record the lowest food diversity (H=2.38), fact that is expected due to the consumption of the lowest number of prey taxa. Regarding the food similarity, the values of this parameter were very close between the two species as well as between the adults and sub-adults of R. dalmatina (Table 1).

The differences between the adults and sub-adults of R. dalmatina and the ones between R. temporaria and R. dalmatina are not significant (Mann-Whitney p>0.05). Between the adults and juveniles of R. dalmatina, the absence of some significant differences is very obvious.
(p=0.81). However, the differences between the two species are more important, being almost significant (p=0.08).

Discussions

Food composition

On a whole, in the stomach contents of the 66 studied amphibians we identified 3 component categories: i.) vegetal parts, ii.) shed-skin fragments and iii.) animal prey, belonging to several prey taxa. All of the analysed stomachs contained animal content, the other two content categories always accompanying the animal preys. The fact indicates an intense feeding, and also a rich trophic basis that is available to the frogs. Individuals from different amphibian species have been recorded to consume exclusively vegetal or shed-skin fragments (Covaciu-Marcov et al. 2002a, 2003), or even not feed at all (Covaciu-Marcov et al. 2002b, 2004, Sas et al. 2003) in the case of unfavourable environmental conditions. Situations like these especially appear during spring, in the case of low temperatures, which decrease the feeding process, firstly through the lowering of the trophic offer (Guidali et al. 1999, Hirai & Matsui 2000). Cases in which a high number of individuals (18 from 54) did not present stomach content were previously signalled at R. temporaria (Stojanova & Mollov 2008). In this situation, the large number of empty stomachs is probably due to the method that was used, the authors examining the preserved animals from scientific collections. Thus, the time between the capturing and the conservation of the amphibians cannot be controlled. Probably, in the case of the animals without stomach contents, the time between the capturing and preservation was too long, which determined the digestion of the preys.

The cases when all of the individuals from the population fed indicate the existence of favourable trophic conditions (Sas et al. 2009). These are mainly expressed through a rich trophic offer, but also possibly through a high availability of the predators for feeding. Generally, the feeding is more intense at the amphibians during autumn (Sas et al. 2003, Kovács et al. 2007, Yu et al. 2009). The feeding intensity is also higher concerning our study, which was realised during autumn, in comparison to other studies performed in different periods (Aszalós et al. 2005, Stojanova & Mollov 2008). On the one hand, it can be explained through the existence of optimum conditions for a large diversity of preys, once with the passing of the summer drought. Meanwhile, the increase of the feeding intensity registered during autumn must be connected to the amphibians’ life cycle, the fact being explained by their need to build up the necessary energy for surviving the hibernation period (Holmweg & Reyer 2000).

Both of the studied species, including the adults and sub-adults of R. dalmatina, consumed vegetal parts. R. dalmatina adults register the highest consumption frequency of the vegetal fragments. The differences between the two sexes are low concerning the consumption of this element (Table 1). Amphibians, although are carnivores, generally consume vegetal parts (David et al. 2009, Hodişan et al. 2009). The fact is usually considered to be an accident (Solé & Pelz 2007, Stojanova & Mollov 2008), these parts being probably swallowed together with the aimed animal prey (Ferenţi & Covaciuc-Marcov 2009, Solé et al. 2009). Once with the increase of the feeding intensity, the accidental ingestion of vegetal fragments also rises (Kovács et al. 2007). The fact is also valid in the case of the studied brown frog populations, which consumed vegetal parts in large amounts, due to the intensive feeding.

The differences between the two species are very low regarding the shed-skin consumption (Table 1). But, concerning the adults and sub-adults of R. dalmatina, these variations are high (8.7% in comparison to 22.7%). There are also other species in which the shed-skin consumption records higher frequencies at the juveniles (Ferenţi et al. 2009). Generally, the shed-skin consumption at the amphibians is considered to be a method of recycling the epidermal proteins (Weldon et al. 1997), which has high frequencies in the cases in which the feeding is difficult due to certain unfavourable environmental conditions (Dimanca et al. 2008). The phenomenon is more frequent at the aquatic species, where the contact with the skin fragments is more easily obtained (Cicort-Lucaciuc et al. 2007, Ferenţi et al. 2008a).

In the present situation, the consumption of skin parts cannot be explained through the unfavourable trophic conditions and lack of other trophic elements. These data suggest that the shed-skin consumption is not necessarily a way of compensating the lack of food, but an act that follows moulting, regardless of the presence of other preys. In the case of the sub-adults, the high consumption frequency of shed-skin can be explained by their increased energetic needs, associated with the growing process (Ferenţi et al. 2009) and the survival over winter. The shed-skin consumption in the terrestrial environment has also been signalled at other amphibians (Kovács et al. 2007), shed-skin being basically a consistent and easily consumed trophic resource.

The animal preys are the most important stomach contents consumed by the two species, the amphibian adults being predators (Cogălniceanu et al. 2000). The R. dalmatina population from Gălgăului Almașului consumed almost the same prey taxa as the population from Livada, which was previously studied (Aszalós et al. 2009).
More prey taxa were consumed by the population from Livada, but the presence of the additional preys is a consequence of the fact that this study took place during a whole year. Thus, the Plecoptera and Trichoptera that were consumed at Livada are present only in certain periods of the year, while our study was conducted only in autumn. Comparing the amounts and frequencies of consumption of the most important prey taxa consumed by the two *R. dalmatina* populations, it can be stated that the spiders and coleopterans occupied the first places regarding both cases. Our results are similar to the ones obtained in Bulgaria in a study realised upon the two species (Angelov & Batschwarov 1972). In both studies, the same prey taxa were consumed in high amounts, being the most important for the two species.

Concerning *R. temporaria*, the food composition and the amount of the prey taxa are different from the ones registered at a population from Ukraine (Kuzmin 1990). At Gâlgăul Almașului, more prey taxa were consumed. The differences can be caused by the different period in which the two studies took place (in Ukraine, the research being conducted during summer), but most likely are determined by the very low number of frogs analysed in the study from Ukraine (Kuzmin 1990). Regarding the resemblance between the trophic spectrum of *R. temporaria* and *Salamandra salamandra* (Kuzmin 1990), this fact seems to be confirmed. Thus, comparing our results with the ones of the trophic spectrum of some *Salamandra salamandra* populations (Ferenți et al. 2008b, Cicort-Lucaciu 2009), the prey taxa and the amount of the most important preys are similar.

The two species mostly fed with terrestrial invertebrate preys. Flying forms were also included, such as butterflies, neuropterans or dipterans, although these prey record low amounts and consumption frequencies. At least *R. temporaria* is known as an efficient predator of terrestrial forms, in many cases even predators (Blackith & Speight 1974). Regarding the studied population, many of the consumed preys were also carnivorous (chilopoda, araneida), the situation also being similar for *R. dalmatina*. Although having a larger size, *R. temporaria* consumed slower preys (snails, diplopoda or Lepidoptera larvae) than *R. dalmatina*. The fact can be explained by the higher agility of *R. dalmatina*, species which can jump the longest from the brown Ranids from Romania (Fuhn 1960).

**Comparing the feeding between the species and the size class**

Both of the studied brown frog species, as well as both age classes hunted in the same territory. It was not observed the usage of different hunting micro-habitats, the existence of a certain trophic selectivity or the preferential orientation for a specific prey type. This fact is proved by the absence of some important differences between the feeding of the two species or between that of the adults and sub-adults of *R. dalmatina*. The situation can appear to be somehow surprising, differences between the hunting territory being signalled in the case of some amphibian species present in a smaller habitat (Covaciu-Marcov et al. 2010). Moreover, the usage of different microhabitats has been observed between the adults and juveniles of other species (Solé et al. 2009). The differences between the food of some syntopic amphibian species is due to their size, the used microhabitat and the different activity period (Van Sluys & Rocha 1998). The usage of different hunting microhabitats results in avoiding food competition (Lima & Magnusson 1998, Vignoli et al. 2009).

The habitat from Gâlgăul Almașului has large dimensions, fact which contributes to the prevention of limiting the trophic resources. However, despite the size of the habitat, there are no differences in the used hunting territory between the two species and the adults and sub-adults of *R. dalmatina*. The frogs are accidentally distributed in the habitat, often appearing in the same place. The lack of differences connected to the intense feeding and to the high number of consumed prey taxa, indicate that at least during the study period, the available trophic offer is rich at the habitat from Gâlgăul Almașului. This allows the total satisfaction of the trophic necessities of the two species.

In the studied habitat, the competition between the two species does not exist, or in any case is much reduced. The competition is not avoided through different mechanisms or ecological accommodations, but is annulled from the start by the existence of a sufficient trophic offer in order to fully satisfy the needs of both species. *R. dalmatina* and *R. temporaria* exploit the same trophic offer, being important predators of the invertebrate communities from the studied habitat. The resemblance between the food consumed by the two species has been previously observed (Angelov & Batschwarov 1972), being also explained through the fact that the two species use the same habitats (Mollov et al. 2006). The absence or perhaps the existence of an insignificant trophic competition was also signalled between *R. temporaria* and another Ranide species, with which it appears in the same habitats, respectively *Rana arvalis* (Stojanova & Mollov 2008). These results seem to infirm the hypothesis according to which *R. dalmatina* and *R. temporaria* are found in a competition, excluding each other (Riss 1988).

Although the differences between the adults and sub-adults of *R. dalmatina* are not important, some variations can be observed determined by the size of the predator. This limits the capturing and swallowing of

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certain prey categories, which are too large to be swallowed. In the case of the amphibians, the size of the consumer limits the size of the prey which it can consume (Arroyo et al. 2008). These differences are harder to be observed between the two species, however *R. temporaria* consumed large caterpillars, even reaching 4 cm long, and myriapodes of similar size. The preference of the species for larger preys has been previously signalled (Kuzmin 1990). Although *R. dalmatina* consumed both prey types, sometimes even in a higher quantity, it did not capture preys with such a large size. The *R. dalmatina* sub-adults consumed fewer carabides and mieriapodes than the adults did, due to the larger size of the preys.

Regardless of the size and species, the studied amphibians also consumed in amounts and with variable frequencies smaller preys, such as pseudo-scorpionida, acarians or Collembola. For example, at *R. dalmatina*, the collembolanas were consumed only by the sub-adults, but these very small preys were however consumed by *R. temporaria*, which is a larger species. Meanwhile, the pseudoscorpionida were consumed only by the adults of both species, although they are small preys. It seems that, regardless of the size of the amphibians, the consumption of extremely reduced-sized preys in comparison to the size of the predator is possible (Santes et al. 2004, Covaciuc-Marcov et al. 2006b, Lima et al. 2010).

**Trophic strategies**

The lack of trophic preferences also indicates that the two species and both classes of *R. dalmatina* are opportunistic predators, which consume the most accessible and easily captured preys. This is generally the case of most of the amphibian species (Guidali et al. 2000, Dimancea & Covaciuc-Marcov 2009, Lima et al. 2010). Meanwhile, the consumption of a majority of large preys can indicate that the populations of the two studied species use the “sit-and-wait” feeding strategy (Duellman & Trueb 1986). Our results confirm that at least *R. temporaria* is a non-selective and sedentary predator (Blackith & Speight 1974). However, as in other cases, the feeding technique is not well highlighted and must not be generalised, because small preys were consumed even here, although in low amounts, and their consumption indicates the presence of the active foraging strategy. Feeding is usually adapted to the present situation (Bisa et al. 2007), amphibians not using a homogenous strategy.

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