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Preliminary data on wintering habits of an intra-urban population of hybrid newts *Triturus cristatus* X *Triturus dobrogicus* (Caudata: Salamandridae)

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Abstract. The study focuses on an intra – urban population of *Triturus cristatus* x *Triturus dobrogicus* located in the Cismigiu Garden in Bucharest. The aim was to observe how microhabitat choice influences the success of overwintering, translated not only in survival, but also in changes in body condition, in a situation of anthropogenic pressure. We also searched for answers as for what motivates differences in wintering habits (regarding shelter features as well as gregariousness) between individuals of different age groups and different reproductive condition.
Key words: *Triturus cristatus* superspecies, newts, hibernation, microhabitat choice, anthropogenic impact.

Rezumat. Prezentul studiu urmăreşte o populaţie intra – urbană de *Triturus cristatus* x *Triturus dobrogicus* din Grădina Cişmigiu. Scopul avut în vedere a fost de observa modul în care alegerea unui tip de microhabitat sau a altuia influenţează succesul iernării, reflectat nu doar prin supravieţuire, ci şi prin modificări de condiţie corporală, într-o situaţie de presiune antropică. De asemenea, am încercat să oferim posibile răspunsuri în privinţa motivaţiilor care determină diferenţe în obiceiuri de iernare (atât în ceea ce priveşte caracteristicile adăpostului, cât şi tendinţa de agregare) între indivizi din grupe de vârstă diferite.
Cuvinte cheie: *Triturus cristatus* superspecies, tritoni, hibernare, alegerea microhabitatelor, impact antropic.

Resume. Cet etude est focalise sur une population intra urbane de *Triturus cristatus* x *Triturus dobrogicus* localise dans le Jardin Cişmigiu, Bucarest. Le but d'étude était d'observer comment la décision sur un type de micro habitat influence le succès de hibernation, traduit comme survivre en bonnes conditions corporelles, dans une situation de pression humaine. Nous ont aussi recherché des réponses concernant les motifs qui déterminent les différences des moeurs d'hibernation chez différentes groupes d'age concernant la tendance d'aggregation ou les caractéristiques des micro habitats choisis.
Mots clés: *Triturus cristatus* superspecies, triton, micro habitat chois, impacte humain.

Introduction. It is becoming increasingly clear that the terrestrial environment plays an important role in the amphibian life cycle (Denton & Beebee 1994; Dodd & Cade 1998; Semlitsch 1998; Jehle & Arntzen 2000). Still, little is known on what features make an individual (or group) prefer one type of microhabitat over another, when several options are available (Garner & Gregory 2006), on the influence of this

choice on successfully over-wintering, or on the way the animal's body condition is affected by hibernation, or, on the contrary, by the lack of it. The primary goal is to avoid death by freezing or hypoxia (Pinder et al 1992; Sas & Covaciu-Marcov 2007), but the success a clever choice brings can translate into a smaller loss of energy during the winter and a better body condition when the mating season begins, ensuring further a higher reproductive success (Sas & Covaciu-Marcov 2007; Hartel et al 2007), on one hand, and making slower – awakening juveniles less vulnerable to predation, on the other hand.

The chosen study group presented an additional interest, coming from an intra–urban population of *Triturus cristatus* x *Triturus dobrogicus* hybrids (Burlacu et al 2009), based on meristic – RBV – , and metric criteria, as well as chromatic analysis), as the two species hybridize frequently in the areas where their areals overlap (Mikulicek et al 2004). Jehle & Arntzen (2000) report a current expansion of the range of *Triturus cristatus* as a potential consequence of human – induced changes to natural landscapes. Negative effects of urbanization on amphibians have been discussed in previous years (Beebee 1996; Wilson & Dorcas 2003; Price et al 2006), because the consequences of urbanization are broad, no single taxon or ecological response can adequately characterize the effects of urbanization (Ryan et al 2008).

Materials and Method

Study area. The population is located in Cismigiu Garden, a park located in the center of Bucharest, as can be observed in Figure 1, in 3 artificial ponds (Site 1, Site 2, Site 3 (Figure 2) located at Lat. = 44°26'14,07" N, Long.= 26°05'27,85" E and elevation 76m for site 1, Lat.= 44°26'13,34" N, Long.=26°05'26,29" E and elevation 75m for site 2 and Lat. =44°26'14,15" N, Long. = 26°05'25,85"E and elevation of 75m for site 3. The three sites are sittuated at a relatively small distance one from the other, the average distance being of 25 m, and are separated by (highly circulated) park alleys. The average depth of the studied pools is 0.8 m, and the average distance to the main waterbody is of 50 m.

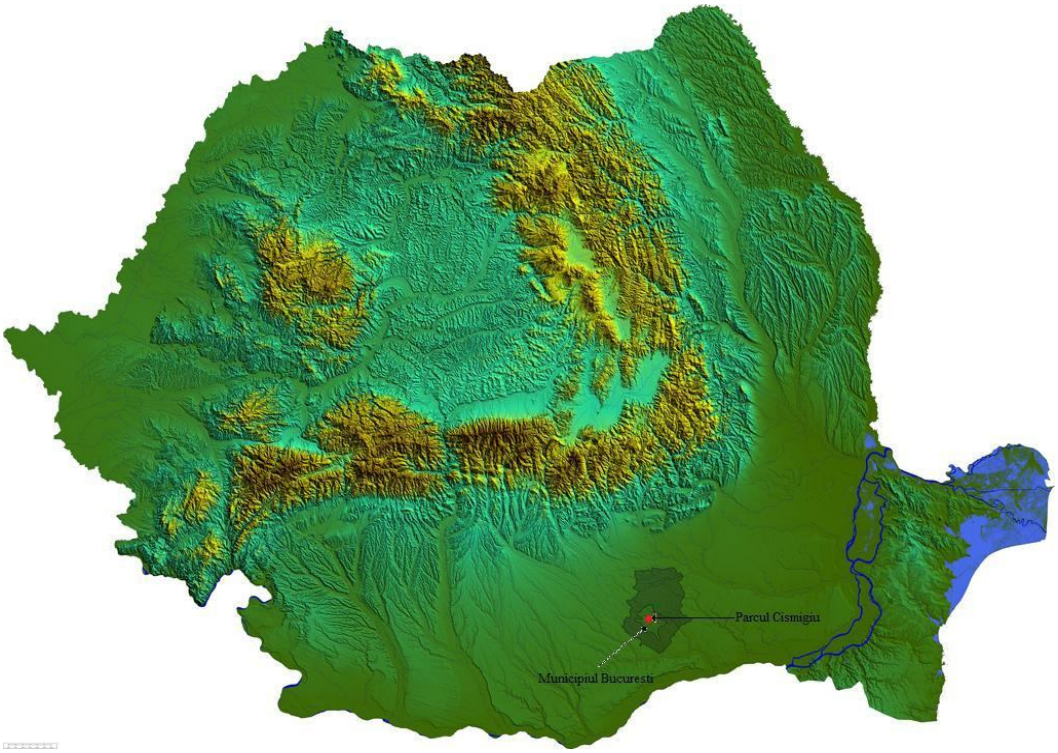

Figure 1. Location of the study.

Figure 2. Studied sites.

Regarding sun exposure, all ponds are shaded throughout the entire day. All but one pond had been drained and swept clean – there was no dead vegetation or mud available as an option for a wintering habitat; the pond with water held a small amount, probably gathered from precipitations, highly eutrophicated and with a large amount of inorganic waste.

Study period: from October 4th, before hibernation, to March 14th – as individuals collected on March 7th were observed in captivity after awakening from hibernation.

Study population

- 11 newts which were hibernating at March 7th, 9 adults and 3 subadults;

- 4 newts that didn't hibernate – they had been kept in semi-aquatic indoor tanks at a constant temperature, and had been active and feeding throughout the winter – all subadults;

- pre–hibernation data collected at October 4th: on the same location were found 20 specimens, out of which 13 were adults and were still in the water, and the 7 juveniles had gone under pavement rocks in the immediate vicinity of the ponds (2 to 5 meters away from the water), preparing for hibernation. Maximum air temperature at that date was 16 °C.

Method. We recorded temperature and pH; on March, 7th, 2009: air temperature was 12-12.5 °C, soil temperature was 10.5 °C, water temperature was 8.2 °C; soil pH was: 7.5, water pH: 6.5-7.

All individuals were weighed and obtained data was divided by age categories and sexes; additionally, the data was also organized into 3 categories: pre–

hibernation, post–hibernation, and no hibernation (only females) and processed by means of Microsoft Excel with XLStat patch and Statistica 6 software.

The eleven newts found dormant were collected (by hand, as were the ones captured at Oct., 4th; the adults then found in the water were captured using an aquarium fish net) and placed in tanks covered with a thick (app. 10 cm) bryophyte layer, in order to simulate as closely as possible their natural hibernating environment and to keep an adequate level of humidity; once they were active, they were fed *Tubifex sp.,* earthworms (Oligochaeta) and various aquarium fish feeds (Tetra Pro, Tetra Diskus). They were collected for several reasons: as we continued to monitor the microhabitat, in order to observe the moment and conditions when the rest of the population would *naturally* become active, we wanted to avoid recapturing the individuals we had awaken; also, we were interested in subsequent ethological observation regarding reproduction and related differences between individuals that hibernated versus individuals that did not.

Results and Discussion

Wintering habitat selection

- *subadults:* one was found alone, beneath a vertical pavement rock, on a side of one of the dried ponds, app. 2 m away from the water; the other two were found under *separate* horizontal rocks on the side of the pond with the water; the ground beneath the rocks was very moist, and there were deep galleries burrowed beneath;

- *adults:* were found in groups of 2 or 3, as follows: three chose a crack filled with roots, between vertical paving rocks, 2 m from the water; five – beneath and between vertical paving rocks, 5 m from the water.

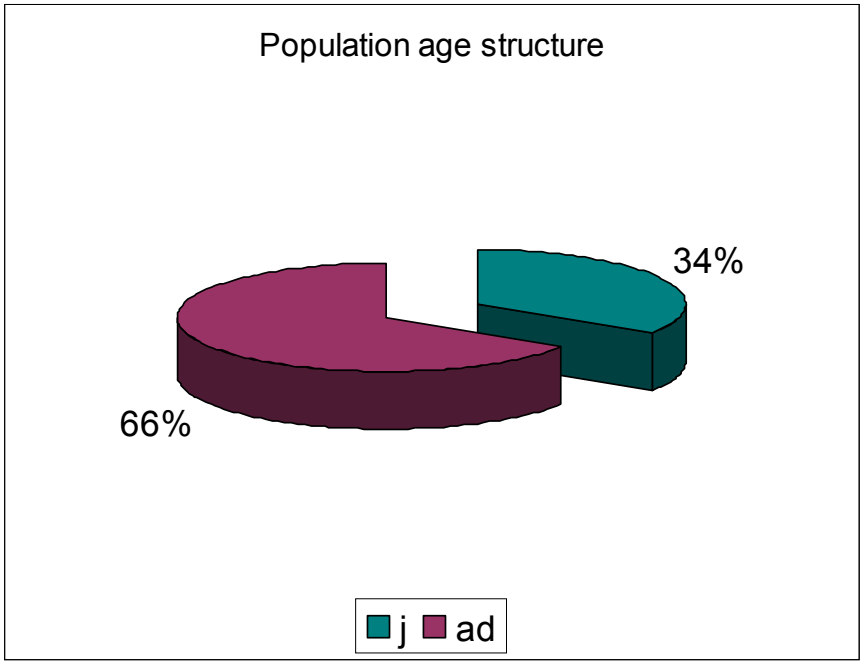

Figure 3. Population age structures. (j=subadults; ad=adults)

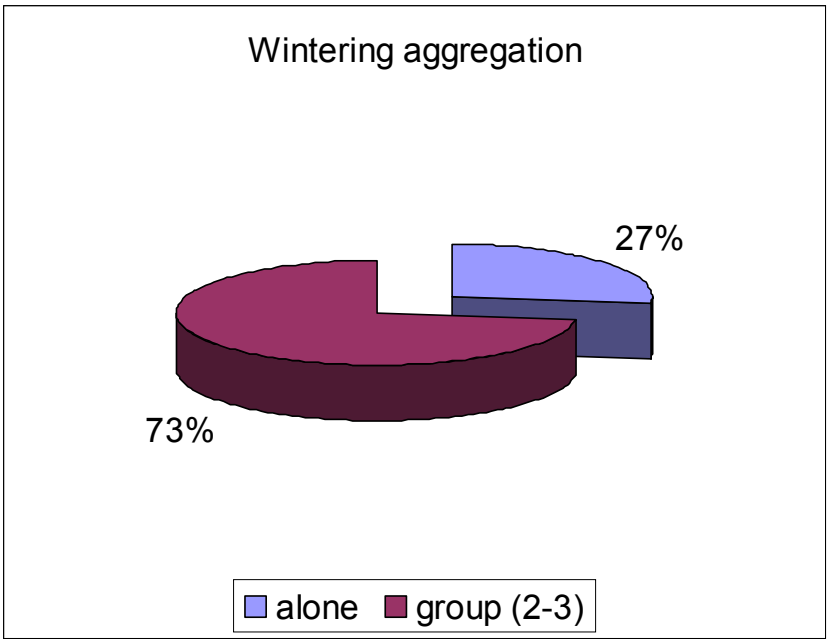

Figure 4. Wintering aggregation occurrence.

Table 1

Microhabitat features preferences on age categories

Age category / *Preffered microhabitat*	*Cracks*	*Vertical pavement stones*	*Horrizontal pavement stones*	*Roots*
Subadults	no	yes	yes	no
Adults	yes	yes	no	yes

Anthropogenic impact. A characteristic of the Cismigiu Garden microhabitats, is the high amount of wastes that visitors dump in the ponds, including beer cans, plastic packages and food and drinks soluble wastes, which can explain the population age structure of 34 % subadults to 66 % adults (Figure 3); Azous (1991) indicates a high correlation between conductivity, a general indicator of dissolved substance concentrations, and amphibian mortality. Rainfall pattern disturbances and prolonged periods of drought, as a consequence of climate changes, might also influence a lower amount of subadults, as these aspects have been reported to induce a decrease in fecundity and limit energy intake (Reading 2007). Also, lower energy intakes reduce the energy deposits needed to overwinter, affecting survivorship potential, particularly in the case of subadults or adults with lower body mass.

Another human–induced negative impact in Cismigiu Garden is sweeping off dried vegetation from the bottom of the basins once they are drained, for 'aesthetic reasons', thus depriving amphibians of a possible refuge.

Other observations

- The adult to subadult ratio might also be a consequence of limited hibernation sites, a reported cause of high juvenile mortality (Jehle et al 1995).

- One subadult was found in the same burrow (on the side of the pond with the rain water) with a *Bufo viridis* individual, which shows that different amphibian groups may share the same over-wintering place and possibly share criteria for choosing a specific type of microhabitat.

- The subadults showed an app. 10-minute 'lethargy – like' behavior when uncovered, afterwards slowly beginning to move, while the adults reacted immediately, trying to avoid being captured; this difference may be caused by the body mass difference between the age groups, a body with a larger fat deposit being able to quicker mobilize a larger amount of energy; it also constitutes a risk at the moment of the 'awakening', making subadults highly vulnerable to predation.

- In October, most subadults were observed in large groups, underneath man – made pavement stones, which make up small paths between the ponds; these locations were checked in March as well, yet no newt was found there. This indicates that newts do 'move around' after they get to a terrestrial shelter. This is in line with the observations made by Schabetsberger et al (2004), who recorded movements of up to 1 meter after the newts reached their shelter. We do not know wether this happened right before the hibernation, we do not exclude the possibility that they might have awaken earlier, moved to another shelter, and then remain burrowed again, due to temperature oscillations; however, on March, 7th, the subadults were found dormant, in a prolonged lethargic state; also, if they had been awakened due to a temporary increase in temperature and had been heading to the water, so would have the adults, to which 'the stake' of reproduction success is much higher and depends of the moment when they arrive in the water; yet, in March, the adults were found further from the water than the subadults. Therefore, we believe the latter changed their shelter before hibernation; as for the difference in distance from the water for the two age categories, our hypothesis is presented bellow, in the Conclusions section.

- One adult had the tip of the tail bit off and lower mass value that the others; this is in line with observed intraspecific predatorism (Burlacu et al 2009).

Post-hibernation behavior. After capture, the newts that were hibernating buried themselves beneath the bryophytes layer, remaining dormant for four subsequent days; afterwards they gradually became active and began feeding, but relatively restricted their activity beneath the briophyte layer; this fact would support the hypothesis that they prefer to over-winter in microhabitats supplied with their potential food. In the Cismigiu Garden, in all microhabitat types, in the same burrows and holes with the newts, we found earthworms, spiders and small crustaceans which are frequently included in the newts' diet. Observed intraspecific predatorism acts amongst faster–awakening adults, in conditions of scarceness of food, when it is impossible to search for food outside the burrow because of low temperature (also, the concrete basins hadn't been filled with water) supports this idea. It would seem that as soon as an individual awakens, it needs to feed immediately, especially when the hibernation conditions have led to a considerable energy consumption and it is vital for survival and reproduction success to become active as fast as possible; therefore, it would be profitable to hibernate in spots where food is available immediately. However, we have no evidence on whether or not newts feed occasionally during hibernation.

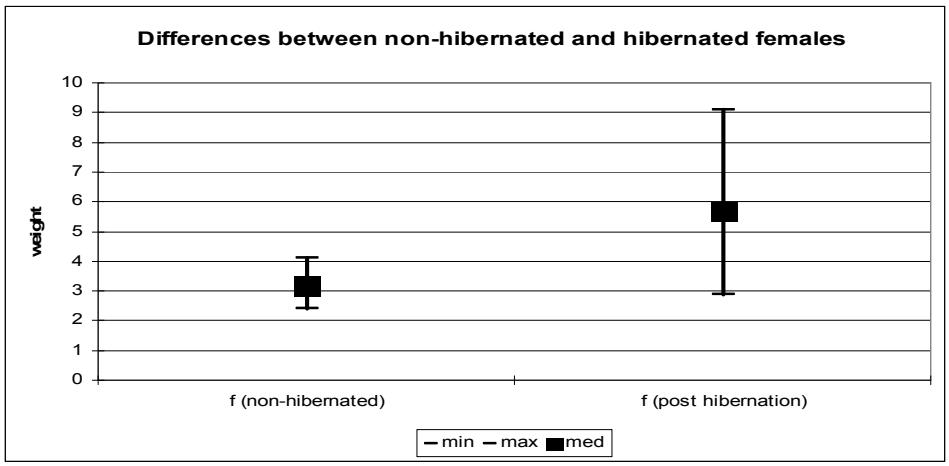

Figure 5. Weight differences between hibernated and non-hibernated females.

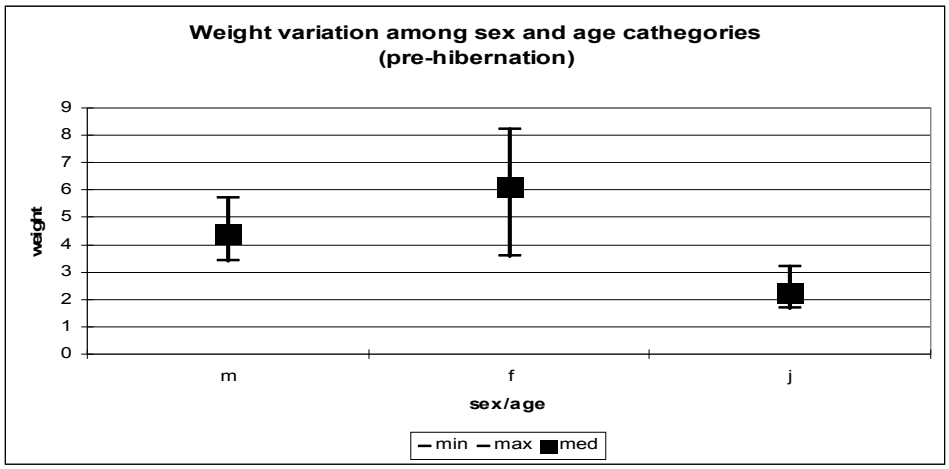

Figure 6. Weight variation in sexes and age categories (pre-hibernation).

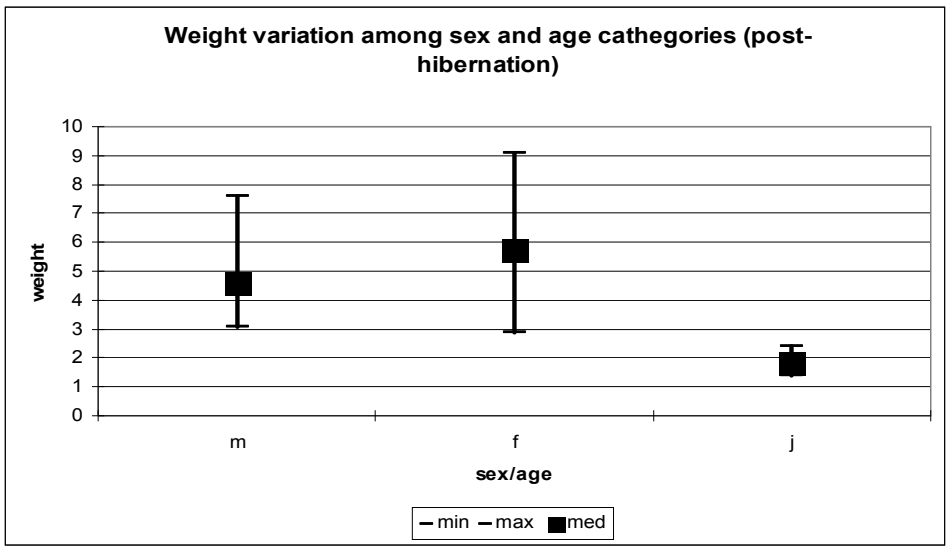

Figure 7. Weight variation in sexes and age categories (post-hibernation).

Changes in body condition

- Females that hibernated registered a wider weight variation than those that didn't hibernate (Figure 5), as the latter were kept under constant conditions, while the hibernated ones had opted for different microhabitat types, which would lead to differences in energy consumption during hibernation, according to the efficiency of the preferred shelter.

- Comparing Figure 6 and Figure 7, it is obvious that the average weight value had shifted closer to the minimum registered value for each category respectively, all groups lost weight during hibernation (probably due to the relative high temperatures for the winter months), but the subadults were most affected, indicating lower body mass as a cause; for the same reason, among adults, males were more affected than females. Post- and pre – hibernation, female adults were overall heavier than males (but their weight varied in a wider span).

Conclusions. Subadults preferred to hibernate in *individual* holes, while adults gathered in common burrows in small groups (Figure 4, Figure 10, Figure 11). This might be a means of protection against (or, perhaps, consequence of) intraspecific predatorism and cannibalism, as larger adults tend to bite either subadults or smaller adults when food is scarce; also, attacks with intention to feed inside the same group age is more frequent among subadults than adults (Burlacu et al 2009).

Subadults stay very close to the water, burrowing themselves in moist soil covered by flat rocks, while adults wandered further from the water (Figure 8 and Figure 9), choosing less humid cracks, sometimes filled with vegetation that may provide better thermal isolation (Table 1). This might be linked to the fact that subadults, due to their smaller body mass, tend to 'awaken' slower (up to 10 minutes of inactivity), which renders them very vulnerable to predation when hibernation is over; being closest to the water might spare them important time and possibly save their lives.

Figure 8. Crack without vegetation - shelter type preferred by adults.

Figure 9. Crack with vegetation – shelter type preferred by adults.

Figure 10. Adults found in group.

Figure 11. Subadult found alone.

Our observations did not support the hypothesis exposed by Schabetsberger et a. (2004) who found individuals of different age groups and even different *Triturus* species together in the same shelter, that subadults use shelters that have proven to be frost safe by older individuals.

Most females were below the average for their sex, which is probably due to their previous higher energy consumption involved in parental care (Orizaola & Brana 2003); for every category respectively, weight after hibernation was considerably lower, indicating that despite very low metabolic rates during hibernation, their body still needed a considerable amount of energy provided by fat deposits to maintain its vital functions, probably due to relatively high temperatures during this past winter, as warmer winters accelerate the consumption of energy reserves (Reading 2007).

In comparison, another intra-urban population of *Triturus cristatus* superspecies, from a pond in the Botanical Garden, Bucharest, chose microhabitats with vegetation, although they also had the option of the cracks beneath pavement rocks, which supports the idea of the first being more efficiently isolated against the cold, as it is preferred, when available. Also, an observed frozen anuran under a rock on the concrete bottom of the basin in the Botanical Garden, without any dry vegetation to act as 'insulation', might support this.

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