

COMPARATIVE STUDY OF THREE *TRITURUS CRISTATUS* (AMPHIBIA: SALAMANDRIDAE) POPULATIONS FROM SÃO MIGUEL ISLAND (AZORES)

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With 2 figures and 5 tables

ABSTRACT. This is the first study regarding *Triturus cristatus* life cycle and biometry in São Miguel island (Azores). Three populations from different habitats, at different altitudes, were compared. The results showed that the aquatic season of *T. cristatus* may begin as soon as November, lasting until July. The reproduction starts in December or January, reaching its peak in March or April. The first larvae may appear in February but most of them appear in April, completing the metamorphosis in July. Life cycle in the Azores differs from that in Europe, and no hibernation was found. Males are smaller, weight less and have a relatively bigger head and shorter tail, than females. Density estimates were higher in a tank with running water (86 ± 34 newts) and in a pond (76 ± 30), and lowest in a small tank (16 ± 3). In the last, the smallest and lightest newts were found. Evidence suggests that the cycle of newts varies between habitats. The long aquatic season of *T. cristatus* in the Azores further emphasises the high degree of specialisation to an aquatic life of this newt species. Two of the studied populations may be endangered, due to lack of resources, and human interference.

RESUMO. Este é o primeiro trabalho sobre o ciclo de vida e a biometria de *Triturus cristatus* na ilha de São Miguel (Açores). Compararam-se três populações de diferentes habitats, a diferentes altitudes. Os resultados demonstraram que a fase aquática de *T. cristatus* pode iniciar-se em Novembro, e prolongar-se até Julho. A reprodução inicia-se em Dezembro ou Janeiro, atingindo um máximo em Março ou Abril. As primeiras larvas aparecem em Fevereiro e são mais abundantes em Abril, completando a metamorfose em Julho. O ciclo de vida é diferente em relação ao das populações europeias, não se tendo encontrado evidências de hibernação. Os machos são menores e menos pesados do que as fêmeas, têm uma cabeça relativamente maior e uma cauda mais curta. As estimativas da densidade foram maiores para um tanque com água

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corrente (86 ± 34 tritões) e para um charco (76 ± 30), e menores para um pequeno tanque (16 ± 3). Neste último, encontraram-se tritões mais pequenos e mais leves. O ciclo de vida nos três habitats estava desfasado. A longa fase aquática de *T. cristatus* nos Açores realça a elevada especialização desta espécie de tritão para a vida aquática. Duas das populações estudadas poderão estar em declínio, devido à falta de recursos ou à interferência humana.

INTRODUCTION

Triturus cristatus (LAURENTI) (Amphibia: Salamandridae) a species not occurring in Mainland Portugal, was already present in São Miguel around 1922, the only azorean island where it is presently found (SVANBERG, 1975). Some work regarding crested newt distribution was initiated in the early 1990's (MACHADO, 1992), and concluded in 1996 (MACHADO *et al.*, in press). Azorean populations present those traits associated with subspecies *T. cristatus carnifex* (MACHADO *et al.*, in press).

The azorean population is submitted to the isolation associated with a oceanic island ecosystem, a situation not found in Europe. On the other hand, abiotic and biotic factors affecting crested newt life cycle in the Azores are probably different from those acting in Europe, where *T. cristatus* life cycle was extensively studied (DOLMEN & KOKSVIK, 1983; ARNOLD & BURTON, 1987; FASOLA & CANOVA, 1992).

In this work, research was conducted by comparing the life cycle and biometry of *T. cristatus* in three different habitats, at different altitudes, in São Miguel island.

MATERIALS AND METHODS

Study sites

In São Miguel many ponds and water tanks are found, associated with cattle farms, what allowed the establishment of *T. cristatus*. Mean annual temperature in São Miguel varies from 12°C at 550 m to 17°C at 70 m of altitude, and rain fall from 2309 to 1020 mm/year (INMG, 1991). At Macela Forest Park average annual temperature is 14,8°C, average maximum and minimum temperatures are 17,6 and 11,9°C, respectively (at 309 m of altitude). Relative humidity is generally above 80% and rain fall is about 1650 mm/year.

Three populations of *T. cristatus* were followed monthly from February 1996 to February 1997 at Macela Forest Park (TABLE 1). The three sites were named Macela 1 (M1), Macela 2 (M2) and Macela 3 (M3). At site M1 a constant tap water flow was present, since it was used as a watering place for cattle. M2 was not in use, and M3 was also used as a watering place.

TABLE 1 - Characterisation of three sites in São Miguel island where *Triturus cristatus* populations were studied.

	Sites		
	M1	M2	M3
Altitude (m)	210	320	470
Surroundings	Road	Garden	Pasture
Habitat	Water tank	Water tank	Pond
Shape	Rectangular	Circular	Circular
Width (m)	1	-	-
Length (m)	46	-	-
Diameter (m)	-	3,8	10,2
Depth (m)	0,6	0,6	1
Capacity (m3)	28	14	54
Air temperature (°C)	17,6	15,9	13,9
Water temperature (°C)	15,3	15,2	13,8
pH	7,4	8,5	6,4

Temperatures and pH are means of values recorded during field work

Life cycle

At sites M1 to M3 monthly sampling consisted of 20 minutes hand-netting to capture adult newts and larvae, and direct observations of eggs. Number of males, females and pregnant females were recorded, as well as pH, and temperature.

Density estimates

The ventral surface of each captured adult was photographed, what allowed its recognition in subsequent recaptures, since adult newts have a orange ventral surface with a conspicuous black pattern, unique for each individual. Number of different newts, captured along the entire study at each of the three sites, was determined by analysis of the photos. At the peak of adult abundance the capture/recapture estimate of Petersen was used, with a modification by Chapman, to estimate population size (HEYER *et al.*, 1994).

Biometry

Adults were measured (head length, tail length, total length) with a ruler to the millimetre and weighted using a 100 g capacity spring scale *Pesola*. Frequency distributions for total length of males and females were determined. Comparisons between sites and sexes for weight and total length were performed using two factor ANOVA and Scheffé test when significant differences were found. The ratios head length/total length and tail length/total length found for males and females were compared with t-test.

RESULTS

Life Cycle

At M1 males are more abundant than females from November to April (Fig.1). From May to July males start to leave the water. Pregnant females were found from January to April, but some were caught as soon as December. Eggs and 1 cm larvae were found since February. Eggs were usually laid in a filamentous green algae. In April larvae were found with 2 to 3 cm and in July mean larval length was 6,3 cm, indicating the end of the larval development. In October only two larvae with an average length of 7,8 cm were found. In spite of the presence of pregnant females since December (1996), no larvae were found until the end of this study. This fact is probably the consequence of vegetation removal and adult capture by local people.

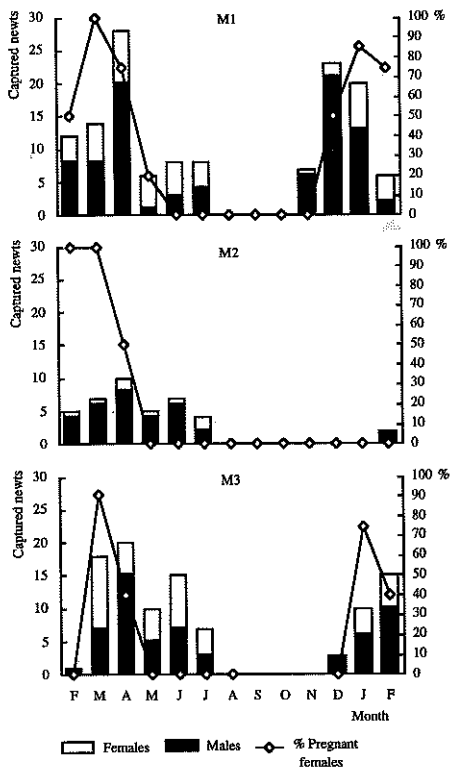


Fig. 1 - *Triturus cristatus* adults captured at three sites in Macela Forest Park, São Miguel island, from February 1996 to February 1997.

At M2 there were always more males than females (Fig.1). Adults appear only in February, leaving the water in July. Although some females were pregnant, no eggs or larvae were found at this site, and the population is apparently declining.

At M3 the water level is very unstable. In fact, in January 1996, and from September to November 1996, the pond had no water (Fig.1). Adults start to appear in December, or later, depending on the water availability. Newts winter in groups up to six animals in the soil, near the water, under logs or inside galleries dug by other animals (rabbits, rats). Males are the first to go into the water, the females appear in the following month, and males are not always more abundant than females (Fig. 1). A peak of pregnant females may occur in January, although, in 1996, they only appear in March, what was probably due to a low water level of the pond until January. Eggs, mainly laid on *Callitriche* sp., and 1 cm larvae were found since February.

Density estimates

The number of different newts captured was highest at M1 (112) and lowest at M2 (16), and were within the range of the population estimate (TABLE 2). The number of captured females was lower than the number of captured males for the three sites. Sex-ratio was more balanced at M3, than in the other studied populations (TABLE 2).

TABLE 2 - Number of different *Triturus cristatus* adults captured at three sites (M1-M3) in Macela Forest Park (São Miguel), from February 1996 to February 1997. Sex-ratio (Males: Females) and population estimate.

	M1	M2	M3	Total
Males	74	12	49	135
Females	38	4	33	75
Total	112	16	82	210
Sex-ratio	1:0,5	1:0,3	1:0,7	
Population estimate*	86±34	16±3	76±30	

* Petersen estimate modified by Chapman, average \pm standard

Biometry

The frequency distribution of female total length is concentrated at higher values than that for the males (Fig 2). In fact, the average length is higher for females at the three sites (TABLE 3).

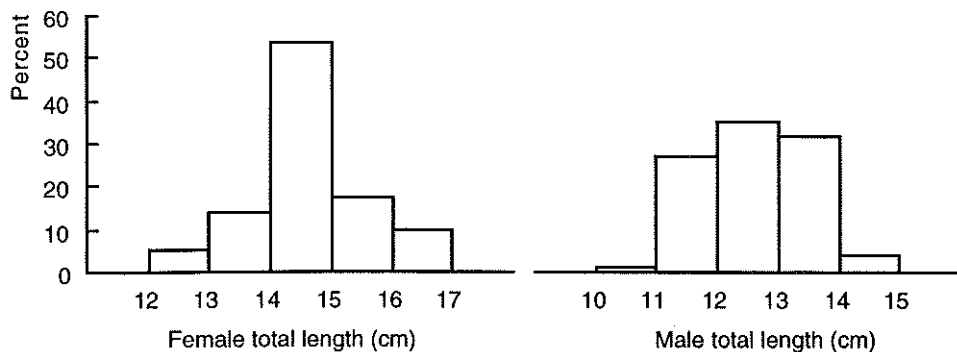


Fig. 2 - Frequency distribution of total length, from 135 males and 75 females of *Triturus cristatus*, captured at three sites in Macela Forest Park, São Miguel island.

TABLE 3 - Average total length (cm) of *Triturus cristatus* males (M) and females (F) from three sites (M1-M3) at Macela Forest Park, São Miguel island. From a total of 135 males and 75 females.

		Mean	Std. Dev.
M1	M	12,55	0,95
	F	14,78	0,73
M2	M	11,73	0,55
	F	13,52	0,41
M3	M	12,40	0,80
	F	14,08	0,95

A two factor ANOVA followed by a Scheffé test revealed significant differences between sexes ($p < 0,0001$) and between M2 and M1 ($p < 0,0001$), M2 and M3 ($p = 0,0002$). No significant differences were found between M1 and M3.

Comparison of the ratio head length/total length between males and females showed significant differences ($t = 5,17$; $p < 0,0001$), so that males have a relatively bigger head than females (TABLE 4). On the other hand a similar comparison for the ratio tail length/total length also showed a significant difference ($t = -3,16$; $p = 0,0018$), showing that females have a relatively bigger tail than males.

TABLE 4 - Comparison of the ratios head length/total length (A), and tail length/total length (B), between *Triturus cristatus* males and females. From a total of 135 males and 75 females of Macela Forest Park, São Miguel island.

A	Mean	Std. Dev.	B	Mean	Std. Dev.
Males	0,135	0,010	Males	0,410	0,030
Females	0,129	0,008	Females	0,431	0,067

Comparison of the weight, between the three populations, for males, females and pregnant females, revealed significant differences between sites and sexes (TABLE 5). Adults from M1 are the heaviest and those from M2 are the lightest. Females are heavier than males, and pregnant females are the heaviest.

TABLE 5 - Average weight (g) of *Triturus cristatus* adults from three sites at São Miguel island (A), and comparison of the weights of males (M), females (F) and pregnant females (P), from three sites (M1-M3) at Macela Forest Park, São Miguel island. Results of a Scheffé test (B), performed following a two factor ANOVA.

A	Mean	Std. Dev.	B	Mean Diff.	P-Value	
M1	M	12,385	1,408	M1/M2	2,880	<0,0001
	F	14,382	1,024	M1/M3	0,865	<0,0001
	P	18,386	2,209	M2/M3	-2,015	<0,0001
M2	M	9,769	0,927	M/F	-1,966	<0,0001
	F	12,250	0,957	M/P	-5,940	<0,0001
	P	16,250	2,475	F/P	-3,975	<0,0001
M3	M	11,491	1,141			
	F	13,528	1,440			
	P	17,088	1,079			

DISCUSSION

In the Azores the aquatic activity of *T. cristatus* may begin as soon as November, lasting until July, beginning earlier than in central Norway where adults stay in the water

from the second half of May to July and sometimes August (DOLMEN & COKSVIK, 1983). Male adults start to leave the water in May, followed by the females. Between August and October (or as late as January, of the following year), adults are out of the water. Males are the first to return, again followed by the females.

The reproduction starts in December or January, reaching its peak in March or April. This contrasts with what was observed in Northern Italy, where reproduction occurs from May to July (FASOLA & CANOVA, 1992). The first larvae may appear as soon as February but most of them appear in April, finishing the metamorphosis in July. In Northern Italy, larvae are found between July and September (FASOLA & CANOVA, 1992) while in central Norway larvae metamorphose from August to October (DOLMEN & COKSVIK, 1983).

The observed differences in life cycle between the azorean populations and those from Europe, are due to climatic differences, since in the Azores winter is not so cold, with an average temperature of 12°C. There is no evidence of hibernation in the Azorean populations, what can also be justified by the amenity of the winter.

On the other hand, the long aquatic season of *T. cristatus* in the Azores (six months or more) further emphasises the high degree of specialisation to an aquatic life of this newt species (DOLMEN, 1988).

Total length of adults is in accordance with the range given by ARNOLD & BURTON (1987). As expected, there is a significant difference between the length and weight of males and females. Males are smaller, weight less and have a relatively bigger head and shorter tail, than females. Adult weights found in this study were higher than those recorded by FASOLA & CANOVA (1992) for newts captured in Northern Italy. These differences may be due to a longer residence in the water for the azorean populations, what may favour feeding and growth. Large size is advantageous because fecundity is related to body size (cf. FASOLA & CANOVA, 1992) and, in fact, a significant difference in weight was found between reproductive and non-reproductive females.

Comparing the three populations it was possible to detect a displacement, in the beginning of the aquatic activity and reproductive season, of about 1 to 3 months. This may be due (in the case of M1 and M3) to differences in altitude. On the other hand there is a clear difference in the total length, weight and sex-ratio, between M1, M3 and M2. Adults from M1 do not differ from M3 in length but are heavier. This may be due to a longer aquatic season, since at M3 periods of drought are common, limiting the length of the aquatic season.

M2 seems to be a declining population, with smaller and lighter newts, possibly

due to a combination of limited food and space, lack of vegetation and relatively high pH. Population M1 is also threatened due to the removal of vegetation and to the capture of adult newts. The population of M3 seems to be in better conditions, possibly because it uses a less humanised habitat, with less human interference, abundance of food, vegetation, and available space.

The technique utilised for identification of adult newts in capture-recapture sampling, namely to photograph the ventral surface of the newt, can be considered as practical, and efficient method, allowing to easily identify the individuals in subsequent recaptures, without having to clip their toes.

This work indicates that there are considerable differences within the Azorean populations and between them and the European ones. On the other hand it was perceived that *T. cristatus* populations in São Miguel face many threats. It is urgent that a wider study is undertaken, that allows a complete knowledge of the status of the Azorean populations, to better preserve this endangered species.

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