TEMPORARY PONDS AS BREEDING SITES OF AMPHIBIANS AT A LOCALITY IN SOUTHWESTERN SPAIN

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ABSTRACT

Breeding habitats of 10 amphibian species were investigated and classified by their physical characteristics. The relative larval abundance of the species was analysed in 15 temporary ponds, resulting in positive correlations with the area of the ponds, whereas negative correlations were found between species diversity and the persistence of the ponds. Regarding the use of their habitats, one species was not correlated with any one, and among the others, two groups of species were differentiated because of their negative associations.

INTRODUCTION

Breeding sites are of great importance for amphibian reproduction, and therefore play a decisive role in the conservation of the species (see e.g. Cooke and Ferguson, 1976, Beebee, 1983). Some general characteristics of the breeding habitats of several Iberian and European amphibians have been listed in fieldguides (e.g. Boulenger, 1897-98, Smith, 1954, Fretey, 1975, García Paris, 1985). More details have been given of the breeding habitats of newts by Cooke and Frazer (1976) and De Fonseca and Jocqué (1982) in Belgium, and in Norway by Dolmen (1980). The habitats of natterjack toads have been described in several papers (Beebee and Griffin, 1977, Andrén and Nilson, 1985a, b) and those of tree frogs were described by Stumpel and Hanekamp (1986) in Holland. Other works have been published on the characteristics of the breeding sites used by the amphibian communities from different localities. Beebee (1977, 1979, 1980, 1983, 1985) and Banks and Laverick (1986) have made an inventory of the breeding sites of amphibians in England, comparing the occurrence of different types of ponds and analysing their characteristics. Strijbosch (1980) related the breeding of several anuran species in the Netherlands to physicochemical variables and to the plant communities of the ponds. The amphibian community examined in this present paper was also the subject of a previous study made of the abundance of larval amphibian species in relation to pond features (Díaz-Paniagua, 1983).

In this study some physical characteristics of breeding sites were investigated, in an area where 10 species occurred. These were seven anurans: *Pelobates cultripes*, *Discoglossus galganoi*, *Bufo bufo*, *Bufo calamita*, *Rana perezi*, *Pelodytes punctatus*, *Hyla meridionalis*, and three urodeles: *Pleurodeles waltl*, *Triturus marmoratus* and *Triturus boscai*. The relative abundance of larvae and the coexistence of the different species breeding in the ponds are considered.

STUDY AREA

The Biological Reserve of Doñana is located in Southwestern Spain between the Atlantic Ocean and the marshes formed by the mouth of Guadalquivir River. This area has been described in detail by several authors (Allier, *et al.*, 1974, García Novo, *et al.*, 1978, Amat, *et al.*, 1979).

The climate is Mediterranean with an Atlantic influence, characterised by hot, dry summers, and rains that occur mainly in autumn. Winter temperatures do not commonly reach negative values (a description of the variation of temperature and rainfall during the study period is given in Díaz-Paniagua, 1986).

Temporary ponds in the area are commonly flooded by the autumn rains. A great number occur in an extensive sandy zone of the Reserve, where the predominant vegetation is the scrub of Halimium halimifolium and Erica scoparia; isolated specimens of Quercus suber and small groups of Pinus pinea constitute the arboreal vegetation. Numerous temporary ponds in an area bordering the marsh are also flooded. Slow-moving currents of water flow from these into the marsh during the autumn and winter months but later in the year they remain as still ponds, locally known as 'caños'. Some extensive lagoons constitute the permanent waters of the Reserve and these have not been included in this study, but the numerous temporary ponds in the meadows that surround them are included.

METHODS

This study was conducted during the amphibian breeding season 1984-85. In autumn 1984, 15 temporary ponds were selected for sampling larval amphibian populations. These samples were taken by means of a dipnet, during daytime, at different locations in each pond, where the numbers of larvae of



PLATE 1: Two temporary ponds included in this study: CHT (left) and PIN (right).

each species were recorded. This was carried out once every two weeks in the first two months. Later on, samples were taken monthly until no more larvae were found in ponds. A detailed description of some of the ponds has been given in a previous paper (Diaz-Paniagua, 1983). For this study, only physical characteristics were taken into account for the classification of the ponds (Table 1):

The persistence of water in the ponds was calculated in relation to the length of the amphibian breeding season (see description in Díaz-Paniagua, 1986), which is considered to be from the time the first species started breeding to the end of metamorphosis of the last larvae in the ponds. The following categories were considered: A) Ponds that persisted before and after the amphibian breeding season (flooded with autumnal rains and dried up in late summer). B) The breeding season was equivalent to the time of water persistence (flooded with autumnal rains and dried up in early summer). C) Ponds that were flooded once the breeding season had started but persisted up to the end (flooded in winter and dried up in summer. D) Ponds that were flooded once the breeding season had started and dried up before it had finished (flooded in winter and dried up in spring). E) Ponds that flooded and dried up intermittently during the breeding season.

Area was calculated by measuring the length and width of each pond and by its approximation to an elipsoid. The areas given in Table 1 correspond to this measurement recorded in December 1984.

Maximum depth: Although fluctuations during the whole study period were registered, only values in December were considered.

Relative depth was calculated by relating the two parameters described above (after Wetzel, 1975).

A classification of the ponds based on these physical characteristics was made by means of a cluster analysis of variables, calculating Euclidian distances as measures of similarity (BMD PIM, Dixon, 1985).

A description of the breeding sites of the species was also made from personal observations recorded in different locations of the study area during the last seven years.

Ponds	Persistence	Area (m²)	Relative Depth	Maximum Depth (cm)	Number of Species	Diversity	Relative Abundance (larvae/sample)
ORF	А	626	1.66	47.0	2 + 2	0.901	3.240
DS	А	1246	2.65	22.5	3 + 3	1.242	2.340
GAN	А	_	_	_	4 + 1	0.774	2.780
MAR	А	491	5.40	85.0	5 + 2	1.513	2.310
MP	В	48	6.01	47.0	1 + 2	0.856	2.500
BRE	В	1295	2.59	21.0	1 + 2	0.218	0.244
СМ	В	491	1.85	17.5	4 + 1	0.818	2.160
NSO	С	214	1.44	16.0	6 + 1	1.520	1.637
DOM	D	104	1.17	15.0	2 + 2	0.623	6.579
PIN	D	204	2.01	32.0	2 + 1	0.924	0.918
ACM	D	1200	2.48	20.0	4 + 0	1.136	1.970
CHT	В	1769	3.23	14.0	4 + 2	2.170	0.982
JH	E	406	1.10	24.0	3 + 2	0.760	3.000
ODU	E	304	1.57	16.0	3 + 2	0.761	5.730
NT	E	831	1.89	14.0	2 + 0	0.668	8.000

TABLE 1: Main physical characteristics of the ponds sampled besides number of species (anurans + urodeles), diversity and total relative abundance. (For explanation of codes of persistence: see the text).

Larval communities were described on the basis of relative abundance, considered as a ratio of the numbers of larvae of each species recorded to the number of samples taken. However, for calculating Shannon-Wiener diversity indexes, the frequency of individuals of each species over the total number recorded in each pond was considered. The same diversity index was considered as a measure of niche breadth for each species according to their presence in the different ponds. Multivariate factor (BMD P4M, Dixon, 1985) and cluster analysis (SPSS, 1986) were carried out on larval relative abundance matrix in order to analyse interspecific associations according to the use of their habitats. Non parametric correlation was employed to relate some physical variables characterising the ponds to other parameters of the larval community, such as diversity and relative abundance.

RESULTS

The physical characteristics of the ponds are described in Table 1. Flooding of the more persistent ponds occurred in early November. While the drying up of some was recorded from late March onwards, others persisted as smaller pools into the summer. Maximum depths of the ponds did not reach high values, all were under 100cm; relative depth gave some information on the morphology of the basin, the highest values corresponded to those ponds which have been modified by human activity (MP, MAR) while natural formations were usually shallower.

There was a wide variety in the area of the ponds studied, even though previous selection excluded very extensive stretches of water. The smallest pond measured 48m² (MP), with small fluctuations during most of its flooded period, whereas the others that reached larger dimensions exhibited broad variations between initial flooding and drying up.

The classification of the ponds based on the results from a cluster analysis is shown in Fig. 1 (because of the lack of some data, GAN was not included). Maximum and relative depths were the main variables contributing to the first grouping of the cases, thus one cluster included the ponds with the largest values and the other cluster included all the remaining ponds. In this second group, ponds were clustered mainly because of differences in their persistence.

The larval species in the ponds

Ten species bred in the study area, their presence and relative abundance in the ponds sampled are shown in Fig. 2, and commented on the following paragraphs:

Pelobates cultripes: This species commonly bred in very large water bodies such as the nearby marsh in the studyarea, but it also frequently used small temporary ponds and tadpoles were found in most of them. Its early breeding precluded it from using ponds that flood later in the season. This early occupation of temporary ponds, however, could produce mass mortality of the eggs or tadpoles, because of the great fluctuations in water level in the early phases of some ponds which may dry out and refill repeatedly (see Díaz-Paniagua, 1986).

Discoglossus galganoi: Its common breeding sites in the study area were small muddy ponds in meadows close to extensive stretches of water. The largest larval abundance detected corresponded to a small, shallow, intermittently flooded pond (ODU). Its ability to spawn repeatedly during the breeding season (Knoeppfler, 1962) and to metamorphose at a very



Fig. 1 Dendrograms showing combine distances for 14 temporary ponds based on their physical characteristics. Similarity was measured by Euclidean distances.



Fig. 2 Relative abundance of each species in the 15 ponds, expressed as percentages over the value of total relative abundance in each pond (see Table 1). (Abbreviations of specific names correspond to the initials of their scientific names.)

small size (Díaz-Paniagua, 1988) enabled this species to breed successfully in such temporary ponds.

Bufo bufo: Breeding of this species in the study area was clearly restricted to a zone characterised by the occurrence of permanent and extensive lagoons (not included in this study). The proximity of the two ponds where tadpoles were detected to large areas of water might have been an important reason for their occupation rather than any other physical characteristic.

Bufo calamita: The short duration of the larval period of this species allowed it to breed successfully in ephemeral pools. Its typical breeding sites in Doñana were small, shallow ponds, but it also has been found to breed in the shallow margins of large ones. This species was abundant in those ponds showing the lower values of relative depth (DOM and JH, see Fig. 2). Even ephemeral puddles have sometimes been observed as breeding sites for this species.

Rana perezi: Adults of this species were common in most areas of water in the study area (temporary ponds, permanent ponds and marshes). High larval abundance was only recorded in GAN, a large pond which persisted much longer into the summer than the others. The characteristic late onset of reproduction in this species (Díaz-Paniagua, 1986, 1988) probably limited its breeding sites to those ponds still in existence in late spring and summer.

Pelodytes punctatus: Breeding of this species in the study area typically occurred in the nearby marsh, where it was found during the autumn. Occasionally it was observed in temporary ponds surrounding this habitat (MAR).

Hyla meridionalis: This species had the largest relative abundance of all the species observed (see also Díaz-

Paniagua, 1983) and its presence in most of the ponds sampled suggested that it may breed in a wide variety of temporary ponds. Their tadpoles were recorded in every pond, with the exception of NT where the breeding season of this species did not coincide with the period of flooding of the pond (see Díaz-Paniagua, 1986, for a description of the breeding season).

Triturus marmoratus: This species typically bred in most temporary ponds in the area, although it was not found in two of the ponds studied. In one of these the reason was the drying up of the pond before the onset of the breeding season of these newts.

Triturus boscai: These newts seemed to have the same requirements for breeding sites as the species above (see Díaz-Paniagua, 1983). The low numbers found may be due to the difficulty in finding their larvae in the ponds.

Pleurodeles waltl: This was an abundant species in the area, breeding mainly in the nearby marsh, although small numbers were detected in the temporary ponds.

The larval communities in the ponds

A general view of the composition of larval communities in each pond, expressed after the relative abundance of each larval species is given in Fig. 2. The maximum number of species occupying one pond was seven, while for anurans the maximum number of species in a pond was six, and for urodeles it was three (all). However it should be noted that in several cases larvae did not utilise the space coincidentally (Díaz-Paniagua, 1988). The highest number of species using the ponds.

10	Persistence	Relative Depth	Area	
Diversity	-0.585* (n = 15)	-0.267 (n = 14)	0.089 (n = 14)	
Total Relative Abundance	-0.213 (n = 15)	0.267 (n = 14)	-0.634* (n = 14)	

TABLE 2: Spearman correlation coefficients calculated between variables describing physical characteristics of the habitats and variables regarding to the occurrence of larval amphibians (*:P < 0.05).

Species	Number of Ponds	Niche Breadth	
P. cultripes	10	1.857	
D. galganoi	7	1.122	
B. bufo	2	0.549	
B. calamita	8	1.480	
H. meridionalis	14	2.081	
R. perezi	4	0.128	
P. punctatus	1	0	
T. marmoratus	13	1.646	
T. boscai	6	1.225	
P. waltl	4	1.332	

TABLE 3: Number of ponds in which each species was recorded and values of niche breath calculated after the Shannon-Wiener diversity index.

The relative abundance of all the species was negatively and significantly correlated to the area of the ponds (Table 2) which affected the measurement because the larvae tended to be more concentrated in smaller ponds. Also a significant negative correlation existed between species diversity in the ponds and their duration, although large diversity values were also obtained for some long-lasting ponds. No significant correlation was found for relative depth between either species diversity or total relative abundance in the ponds.

The presence of one species in the ponds and the diversity index calculated for each species according to its frequency of occurrence in the ponds (used as niche breadth index) were used to compare the spatial requirements of the species (Table 3). Hyla meridionalis was the most generalist species, being found in 14 of the 15 ponds, and presenting the largest value of diversity. At the other extreme, species such as Bufo bufo and Rana perezi were restricted to small numbers of ponds and presented low niche breadths whereas high abundance was only reached under particular conditions. Results obtained for Pelodytes punctatus and Pleurodeles waltl were not considered because of the small numbers found (see Fig. 2) thus suggesting that these are not their most characteristic breeding sites.

A cluster classification of the larval density of species in ponds is represented in Fig. 3, and the results obtained from a factor analysis (Table 4) contributed to its clarification. There did not seem to be a high association of species. Three main clusters appeared. A group with only one species, Rana perezi, which was not correlated with any of the others, high density only occurring in ponds where other species are not very abundant (see Fig. 2). The second cluster included the urodeles and two species of anurans: Bufo calamita which appeared closely associated with Triturus boscai and Hyla meridionalis which, being widely distributed, was negatively associated with the other four species of anurans. These last four species constituted the third cluster, including species negatively or not associated with those in the other groups.

DISCUSSION

The persistence of the temporary ponds may be considered as a limiting factor for the development of communities therein. For amphibian communities, the unpredictability of these habitats is compensated for by the flexibility of the breeding season (Díaz-Paniagua, 1986) and of the larval life duration with a



RESCALED DISTANCE CLUSTER COMBINE

Fig. 3 Dendrogram showing cluster combine distances for the larval species based on their relative abundance in the ponds. Cosine of vectors of variables is used as distance coefficient, rescaled in the figure in a range 1 to 25.

	Fl	F2	F3	F4	F5
P. waltl	0.868	-0.281	_		_
T. marmoratus	0.803	0.467	_	_	_
D. galganoi	-0.557	_	-0.340	·	_
H. meridionalis	_	0.916	_	_	_
T. boscai	_	_	0.848	_	_
B. calamita	_	-0.439	0.810	_	_
P. punctatus	_	_	_	0.909	_
P. cultripes	-0.347	-0.431	_	0.668	_
R. perezi	_	_	_	_	0.920
B. bufo	_	-0.466	-0.383		_
VP	1.907	1.780	1.751	1.394	1.148

TABLE 4: Rotated factor loadings resulting from a factor analysis based on the relative abundance of the species in each pond, and the proportion of variance (VP) accounted for by the common factors. (Loadings less than 0.250 are not shown.)

wide range of body sizes at metamorphosis (Wilbur and Collins, 1973).

Temporal reproductive patterns of the species studied have been described in a previous study, where a segregation in time was found to be of great importance (Díaz-Paniagua, 1988). In this study, differences in the occupancy of the available breeding habitats were also found, which may contribute to the segregation of the species. On the basis of larval abundance in the ponds, the amphibian community studied may be considered as composed of: a) species breeding mainly in temporary ponds, commonly persisting from winter to early summer (Hyla meridionalis, Triturus marmoratus. Triturus boscai; b) species which usually breed in very temporary ponds (Bufo calamita, Discoglossus galganoi); c) species breeding mainly in permanent or extensive waters, although they may also occur in temporary ponds (Pelobates cultripes, Bufo bufo, Rana perezi, Pelodytes punctatus, Pleurodeles waltl).

The high proportion of species in the group (c) was remarkable. Their larval life histories seem to be better suited to different habitat conditions from the temporary ponds although they can achieve an acceptable level of breeding success in them. The use of such ponds by these species provides another example of the flexibility of the breeding season of amphibians and probably this contributes to increasing the probabilities of success in reproduction under uncertain physical conditions.

The capability of inhabiting a great number of widely differing types of pond and of metamorphosing a large number of individuals may be considered as an indication of a species' ability to breed successfully in such kinds of ponds. In the same way, important variations in relative larval abundance are observed for some species depending on their coexistence with other larval species, resulting in negative associations, which seems to imply differences in competitive ability. Thus, some species appeared to occupy a great number of ponds and they were usually very abundant (e.g. *Hyla meridionalis, Triturus marmoratus, Pelobates cultripes*). Others appeared to be restricted to a particular kind of pond and only reached high

numbers when those more common species were not so abundant, as was the case with *Bufo calamita* and *Discoglossus galganoi* in intermittently flooded or ephemeral ponds.

No overriding relationships among the larval species were found in a previous work (Díaz-Paniagua, 1983), but a temporal segregation was found (Díaz-Paniagua, 1988). Habitat partitioning also contributed to a decrease in the overlap among the larval species, suggesting that interspecific relationships might have had an influence on the community organisation. This may also be confirmed by a decrease in larval density of some widely distributed species when they have to share their habitats with other species. As Heyer (1976) argued, it is possible that factors contributing to those segregations might have been important 'historically' and that interspecific competition is not an important factor in the organisation of the larval amphibian communities at present because of the occurrence of the temporal and habitat partitioning.

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