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Size structure, relative growth and sanitary condition of a crayfish population (*Austropotamobius pallipes*) living in rocky pools

(Crustacea, Decapoda)

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Bio-ecological aspects of a crayfish population inhabiting two rocky pools in the Carpasina stream (Liguria Region, N.W. Italy) were investigated per three years. A different spatial distribution between young and adult individuals was observed: juveniles were found under stones in shallow water, while large individuals were commonly found in deep water and in burrows present at the bottom of the pools. Length-frequency analyses showed that males are bigger than females. The sex-ratio was estimated at 1:1. Morphological variations related to sexual maturity were analysed in both sexes. Size at sexual maturity, based on allometric growth of the abdomen and chela width, was estimated at approximately 19 mm CL for both sexes. The male chelae, however, showed a further step at about 28 mm CL, indicating the existence of one third kind of male. The adaptative value of the three different kinds of males was discussed in terms of physiological and morphological efficiency. Morphological analysis of the chelae indicated that both sexes begin their life as homochelids, but in the course of the life cycle they may lose their chelae. This seem to be related to the frequent intraspecific aggressions due to the limited space that the rocky pools offer to the species. A scarce number of crayfish was found affected both by pathologies than by branchiobdellids.

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Introduction

The biology of different European populations of *Austropotamobius pallipes* (Lereboullet) has been well studied (see for a general review Alderman & Polglase 1988, Laurent 1988, Lowery 1988). On the contrary, in Italy little has been done on the biology of *A. pallipes* (Ninni 1865, Mazzarelli 1903, Vinciguerra 1899, Supino 1925), nevertheless there has been a recent interest on this species (Mori et al. 1988, 1991, Specchi et al. 1991, De Luise 1991, Salvidio et al. 1993).

In Liguria (north-western Italian administrative Region) the freshwater crayfish populations are still widespread both in Alpine and Apennine water courses (Salvidio et al. 1993). In this region the

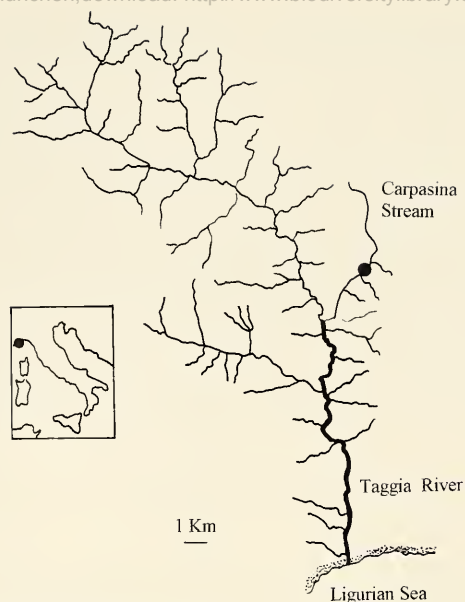


Fig. 1. Geographical location of the Carpasina stream.

crayfish are mainly distributed along the stream beds, but it is also possible to find them inside isolated rocky pools, mainly in those streams that present a steep gradient. The rocky pool environment could be characterised by fluctuations in a number of physio-chemical factors. No studies have been performed on the populations inhabiting these peculiar environments.

So we have conducted a study on a population of the crayfish *A. pallipes* inhabiting two rocky pools located in the Carpasina stream (Province of Imperia, N.W. Italy) with the aim to follow the temporal variation of its demographic structure and to estimate some of its biological parameters useful for a rationale utilisation and management of the species.

Materials and Methods

Fieldwork was conducted in the Carpasina stream (560 m above sea level), tributary of Taggia river, located in the Imperia Province (western Liguria, Italy) (Fig. 1). The study was performed along a transect constituted by two rocky pools, followed by terraces, of a total length of about 42 m, the width ranging from 10 to 2.8 m and a maximum depth of 3 m (Fig. 2).

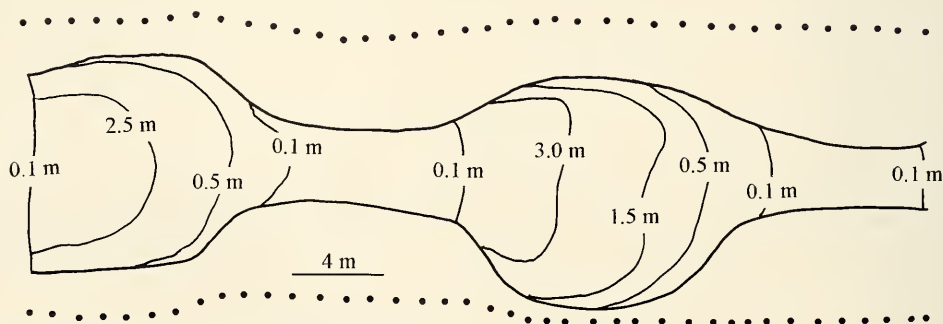


Fig. 2. Bathymetric map of the Carpasina stream.

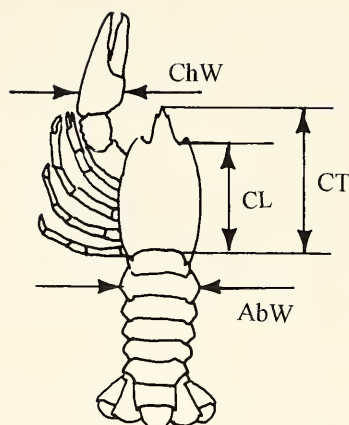


Fig. 3. *Austropotamobius pallipes*: measurements used to analyze relative growth. CL: carapace length; CT: cephalothorax length; AbW: abdomen width; ChW: chelar propodus width.

In this stream the postlarvae of *A. pallipes* are present from May to July. In order to sample all life stages, the crayfish population was inspected regularly once a year, during summer months (July–August) from 1993 to 1995. Crayfish were caught in the shallow water by manual sieving, while in deep water (>1 m) by diving. Once recorded the following parameters as sex, morphometric measurements, morphology of the chelae, external disease condition, scars and mutilation, the individuals were released.

Sex was readily identified from the morphology of the pleopod structures in individuals greater than 6 mm CL. Individuals smaller of this size were instead assigned as 50 % to the males and to the females, supposing that the primary sex ratio is unbiased.

In order to evaluate the external disease conditions and the morphological abnormalities of the population under examination, each individual was carefully inspected by use of a magnifying glass (3×).

The following morphometric measurements were also made on each specimen, using vernier callipers to the nearest 0.1 mm (Fig. 3): (CL) carapace length, from the eye socket to the mind-posterior margin of the carapace; (CT) cephalothorax length, from the tip of the rostrum to the mind-posterior margin of the carapace; (ChW) width of the chelar propodus; (AbW) width of the second abdominal segment. The body wet weight (WW) of each specimen was also recorded to the nearest 0.1 g by means of a Pesola dynamometer.

Relative growth was described using the traditional Model I regression equation for allometric growth: $\log Y = \log a + b \log X$ (Sokal & Rohlf 1981). Data were analysed after log-transformation with carapace length (CL) chosen as the main reference dimension. Allometry was determined by testing the slope of the regressions against the isometric slope of 1 with Student's t-test. Statistical equality of morphometric relationships between sexes, and among maturity instars were tested using ANCOVA, carried out by using BIOM-pc (Rohlf 1989).

Results

Population structure and Sex-ratio

Males were distributed between 3.5 and 40 mm CL and females between 3.5 and 32 mm CL (Fig. 4). The number of crayfish recorded showed a decrease from year to year, above all at the medium size. The distribution frequency of CL was significantly different in three years, above all considering males plus females (Tab. 1).

Annual sex-ratio was not found significantly different from 1:1, with the exception of July 1993, when, because of an insufficient sampling on the pool bottoms caused by a rupture of a aqualung, the proportion of males was 38.5 % (Fig. 4). The sex-ratio of crayfishes <22 mm CL was not significantly

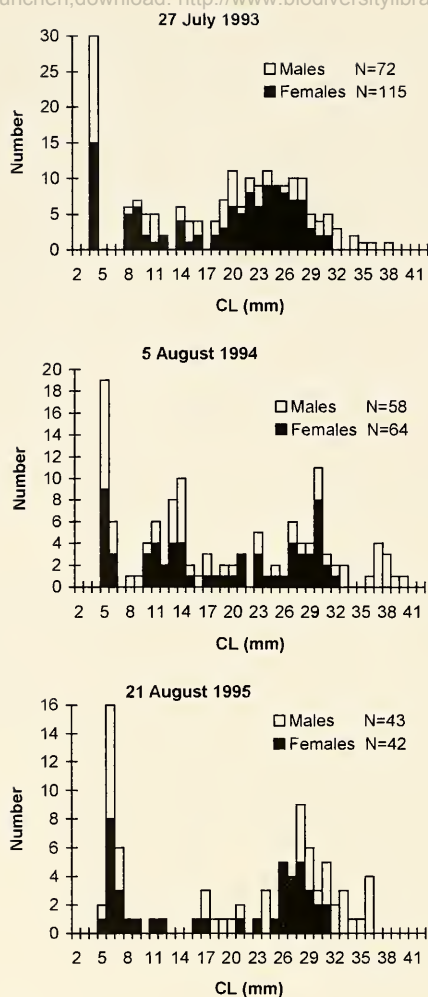


Fig. 4. Size (CL) frequency distributions of males and females of *Austropotamobius pallipes* collected and measured during the whole study performed in Carpasina stream (Liguria, N. W. Italy).

different from unity; females were significantly more numerous than males among 22-28 mm, and males dominated size classes >32 mm (Fig. 5).

Crayfish smaller than 13 mm CL were only found in shallow waters among in the riparian vegetation and under stones.

Tab. 1. Results of the Kolmogorov-Smirnov analysis (Siegel 1956) to test the null hypothesis that there is no difference in the size distribution of crayfish with year. Level of statistical significance: *: $p < 0.05$; **: $p < 0.01$; ns: no significance.

variables	Males + Females		Males		Females	
	D	P-level	D	P-level	D	P-level
1993 vs. 1994	0.178	*	0.182	ns	0.184	ns
1993 vs. 1995	0.265	**	0.261	ns	0.268	*
1994 vs. 1995	0.211	*	0.199	ns	0.262	ns

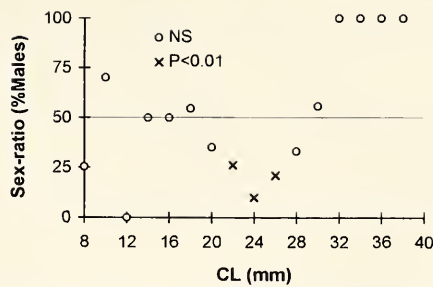


Fig. 5. Sex-ratio in relation to carapace length, recorded in 1993 (data grouped upon 2 mm intervals of CL).

Relative growth

The values of relative growth parameters of each morphometric character against carapace length estimated for overall males and females are given in Tab. 2. All correlation coefficients are high and significant ($P<0.01$). Negative allometric growth of cephalothorax length and of body wet weight both versus CL were revealed (Tab. 2) with no difference between males and females (Tab. 3). On the contrary a positive allometry either of the abdomen or of the chelae versus CL was revealed in both sexes (Tab. 2), but the former is larger in females than in males, while the chelae are larger in males than in females (Tab. 3).

As males and females increase in size certain of the body proportions change relative to CL. The width of the abdomen and chelar propodus plotted on a log-log scale against carapace length showed a change in the level of allometry, in both sexes, at about 19 mm CL (Fig. 6 and Tab. 2). The covariance analysis yielded significant differences between the different phases (Tab. 3). In the males, however, the growth of chelar propodus is more complex being it is triphasic, i.e., the regression line “broke” also at about 28 mm CL (Fig. 6 and Tab. 3). The covariance analysis yielded significative differences among

Tab. 2. Values of relative growth parameters of each morphometric characters against carapace length, estimated either for overall males and females or for different maturity instars of each sex. AbW: Abdomen width; ChW: Chela width; CT: Cephalothorax length; WW: Body wet weight; N: Number of individuals; Se: Standard error of the slope; r: Correlation coefficient; t : H_0 for t -test, $b=1$ ($b=3$ per WW); As: Allometric status; +: Positive allometry (+: $p<0.05$; ++: $p<0.01$); 0: Isometry; -: Negative allometry.

Measures/CL	Sex	N	CL range	intercept (Log a)	slope (b)	Se	r	t	As
LogAbW	Females	160	3-31	-0.4020	1.1778	0.011	0.993	16.14	++
	Immature	45	3-20	-0.2931	1.0568	0.026	0.986	2.15	+
	Mature	115	19-31	-0.4814	1.2377	0.035	0.956	6.66	++
	Males	84	3-40	-0.2722	1.0315	0.011	0.995	2.84	++
	Immature	36	3-20	-0.3217	1.0805	0.022	0.993	3.57	++
	Mature	48	19-40	0.0155	0.8351	0.028	0.974	5.73	-
LogChW	Females	117	3-31	-0.7874	1.2568	0.019	0.985	12.91	++
	Immature	35	3-20	-0.6556	1.1182	0.049	0.972	2.40	+
	Mature	82	19-31	-0.6895	1.1900	0.060	0.906	3.12	++
	Males	112	3-40	-0.7543	1.2793	0.016	0.990	16.47	++
	Immature	38	3-20	-0.6463	1.1678	0.015	0.996	10.56	++
	SubMature	38	19-32	-0.0434	1.0343	0.084	0.899	0.40	0
	Mature	36	28-40	-0.9683	1.4467	0.121	0.899	3.68	++
LogCT	Females	132	3-31	0.1803	0.9525	0.004	0.998	10.82	-
	Males	85	3-40	0.1813	0.9523	0.003	0.999	12.62	-
LogWW	Females	207	3-31	-2.9235	2.8150	0.023	0.992	7.86	-
	Males	157	3-40	-2.9769	2.8686	0.029	0.991	4.39	-

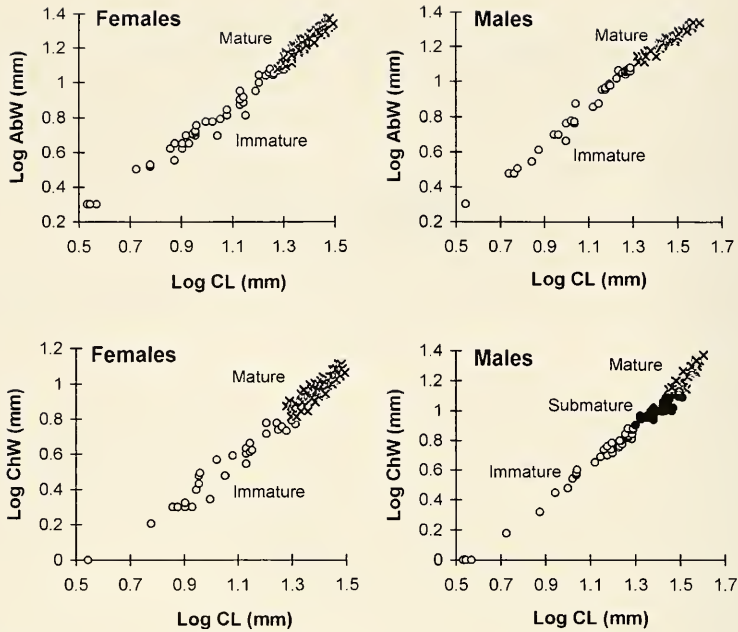


Fig. 6. Relationships among log morphometric dimensions and log carapace length, separated per maturity instars, for both sexes of *Austropotamobius pallipes*.

three male maturity instars (Tab. 3). No significant change in growth of the cephalotorax length and the body weight were detected in both sexes (Tab. 3).

Chelae morphology and pathologies

The percentage of males with homochelous, regenerated or missing chelae did not differ significantly in the three years of our study ($\chi^2 = 7.32$, $df = 4$, $P > 0.01$), as well as the females ($\chi^2 = 9.52$, $df = 4$,

Tab. 3. Comparison between male and female, and maturity instar log-log regression equations of each morphometric variable over CL, using ANCOVA. M: Males; F: Females; IF: Immature females; MF: Mature females; IM: Immature males; SM: Submature males; MM: Mature males.

Variables		Differences among slopes		Differences among adjusted means (intercepts)		
		F	P-level	Common slope	F	P-level
AbW/CL	M vs. F	81.87	<0.001	1.116	173.53	<0.001
	IF vs. MF	14.18	<0.001	1.085	36.63	<0.001
	IM vs. MM	26.73	<0.001	1.049	1.59	ns
ChW/CL	M vs. F	0.72	ns	1.271	125.57	<0.001
	IF vs. MF	0.80	ns	1.134	20.02	<0.001
	IM vs. SM vs. MM	0.28	ns	1.167	75.81	<0.001
CT/CL	M vs. F	1.14	ns	0.951	0.10	ns
	IF vs. MF	2.55	ns	0.951	0.01	ns
	IM vs. MM	10.7	ns	0.948	0.65	ns
WW/CL	M vs. F	1.47	ns	2.836	2.05	ns

Tab. 4. Percentage of individuals with homochelous, regenerated or missing chelae according to five different carapace length classes. M: Males; F: Females.

Size-range CL (mm)	Chelae										No examined	
	Homochelous		Regenerated				Missing					
			Left		Right		One		Two			
	M	F	M	F	M	F	M	F	M	F	M	F
4-13	89.4	90.6	2.1	1.9	2.1	3.8	2.1	3.8	4.3	0.0	47	53
14-19	90.0	77.8	5.0	0.0	5.0	1.1	0.0	0.0	0.0	11.1	20	18
20-25	66.7	63.0	11.1	22.2	16.7	7.4	5.6	3.7	0.0	3.7	18	54
26-32	68.6	63.3	20.0	18.3	8.6	8.3	2.9	10.0	0.0	0.0	35	60
33-40	68.2	–	13.6	–	4.5	–	13.6	–	0.0	–	22	–
Total	74.8	69.7	13.3	15.4	6.7	7.4	3.7	5.1	1.5	2.3	142	185

$P>0.01$). So, data from different years were pooled and separated according to five different carapace length classes (Tab. 4). Most crayfish of both sexes were homochelous in the smaller size classes. Their largest number with regenerated or missing chelae was found in larger classes. No difference in chelae morphology was detected between sexes ($P>0.05$). A scarce number of crayfish was found affected by scars and mutilations (fingers, antennae and pereopods lacking) (Tab. 5).

A relatively small number of crayfish appeared affected by external pathologies (Tab. 6). *Branchiobdella* spec. was only recorded during the first year of investigation. These annelids have been always found on the ventral margin of the carapace and coxa of the legs. They range among 2 to 7 individuals per crayfish.

Discussion

Data obtained from catches performed at different depths in the stream indicate differences in the occupation of the habitat between large and small specimens of *A. pallipes*. Crayfish <13 mm CL were found only in the riparian vegetation or under stones in shallow water, whereas larger crayfish occurred at different depth or on the bottom of the pools. This distribution suggests that the first development of new-borns occurs in shallow waters and, after the first phase of their life cycle, they move to the main stream bed. Shallow waters are usually sheltered places, rich in organic deposition, where the young specimens are protected from predators and have a more suitable substratum in terms of feeding opportunities. Microhabitat changes related to predator pressure as well as feeding requirement seem also to be very common among fishes (Mazzoni & Caraschi 1995).

The annual decrease of the crayfish number observed during this study in the Carpasina stream is probably due to the poaching, because any natural predator of crayfish (brown trout, eel, bulhead, rat, and crow) was never observed in the area of study (S. Salvidio, data unpublished). Poaching is very frequent in Italy (Laurent 1988). In the rocky pools the illegal catching of crayfish seem to be principally addressed to the specimens of medium size because the crayfish of larger sizes, living on the bottom, can be only collected by diving.

Tab. 5. Frequency (%) of pathologies, scars and mutilations over body surface.

Year	Pathology								Number	
	<i>Branchiobdella</i> sp.		Fungi		Scars		Mutilation			
	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀
1993	2.7	4.3	0	1.4	0	3.6	0	2.7	22	115
1994	0	0	0	2.3	2.3	0	2.3	0	43	42
1995	0	0	1.7	1.5	1.7	0	0	3.1	38	64
Total	0.9	0.9	1.0	1.8	1.9	1.8	0.9	2.3	103	221

In the study population both the maximum size and the sex-ratio were similar to those found in populations inhabiting hill streams or brooks (Brown & Bowler 1978, Arrignon & Roché 1983, Almaça 1990, Mori et al. 1996). Thus, the rocky pools seem to offer sufficient energy to crayfish development. Imbalanced sex-ratio among older specimens can be due to different reasons. A possible mechanism limiting female size in decapods could be a lengthening of the moult interval compared with males, caused by the onset of reproductive activity, which, at least in *Anomura* and *Brachyura*, is restricted to the intermoult period (Micheli et al. 1991). Furthermore, both moulting and reproduction entail considerable energy demands (Gherardi et al. 1989). A further hypothesis is that it may be due to their high mortality rate, caused by the high cost of egg production. Probably, both these hypotheses are valid because *A. pallipes* living in oligotrophic habitats mainly feeds on detritus (Reynolds 1979). According to Momot et al. (1978) the detritus, that consists of decomposing plant and animal fragments, and associated micro-organisms, i.e. bacteria, fungi, algae and protozoans, accounts for some 13.2-21.6 per cent by weight of total food consumed by natural crayfish population. However, whether the microbial organisms are or are not a sufficient source of energy for macrobenthic populations is still a subject open to discussion (Cammen 1989).

Employing CL as a reference dimension, this study has demonstrated that the cephalothorax length and the body weight are growing with a negative allometry and that no significant change in growth of these dimensions were detected in both sexes. The growth in width of the abdomen and of the chelar propodus instead demonstrates, in addition to the already demonstrated sexual dimorphism (Rhodes & Holdich 1979, Thomas 1981), marked allometric steps within each sex.. The first distinct alteration in growth of these dimensions at a CL of about 19 mm suggests a moult of puberty (sensu Pérez 1929), which demarcates the immature from mature phase. These results are in agreement with the smallest ovigerous females collected by Mori et al. (1988) in other streams of Liguria. The males presented a further step at about 28 mm CL, for that they may be separated in three different stages, i.e. in immature, submature and mature individuals. We have not performed any histological study to confirm this subdivision, however, three kinds of males has been already described in a different number of decapod species (Hartnoll 1982, Micheli et al., 1990, Mori et al. 1996). So, on the basis of these studies, it is possible to hypothesise that the immature males are physiologically immature individuals, the submature males are physiologically mature individuals (i.e. they have functional gonads) but morphometrically immature individuals to sustain agonistic interactions with the mature males that present bigger chelae than them. This pattern was not revealed by previous morphometric studies performed on *A. pallipes* (Rhodes & Holdich 1979, Lowery 1988, Mori et al. 1991). The morphometric changes seem to be related to hormonal changes (Cornier et al. 1992). However, whether physiological and morphological maturities are linked in *A. pallipes* is still unknown. For this reason, further studies to determine at which size *A. pallipes* reaches its physiological maturity are needed.

Various interpretations have been given because the males of the decapod crustaceans have larger and wider chelae than females (see Hartnoll 1982 and Vannini & Gherardi 1988). In some species the role of the chelae has been related to the feeding habits, but for *A. pallipes* it can be dismissed because this species is principally a detritus feeder. So, the chelae are mainly used by crayfish to capture and hold females during mating or in agonistic interactions for keeping or conquering a burrow. The high incidence of individuals with regenerated or lacking chelae (first pereopods) indicates that intraspecific aggressions are frequent. Probably this is related to the limited space that the rocky pool offer to the species. Such combats, however, did not involve the body or other pereopods because a small number of individuals with scars and mutilation was found.

Fungi were found in a small number of crayfish, but in this study only the external part of the crayfish body was inspected, while a much wider number of diseases may be recognised in the internal part of the crayfish body (Alderman & Polglase 1988).

Crayfish infested by branchiobdellids were only recorded during the first year of investigation, but their number has been probably underestimated because other branchiobdellid species can live inside the gill chambers (Alderman & Polglase 1988, Gelder et al. 1994) It is also possible that the branchiobdellids move inside and out of the gill chambers in relation to the light, because during an investigation performed in a streams of the Liguria we have recorded by day a lower number of crayfish infested by branchiobdellids than by night. Such a supposition seems to be confirmed by the position of the branchiobdellids, always recorded, during this study, on the ventral margin of the carapace and coxa of the legs. Also this subject poses question for future research.

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