

Growth and survival of several ascidian species from the northwestern Mediterranean

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ABSTRACT: Growth and survival parameters of 5 colonial ascidian species were studied in populations from the northwestern Mediterranean. Over a 2 yr monitoring period, the growth, mortality, reproduction and resistance periods of a number of colonies were recorded. A marked intraspecific variability in most parameters was evident, but a seasonal pattern emerged for most species, with more active growth in winter. Several types of resting and resistance periods, presumably of different biological significance, were observed in some species in the warm season. There were great differences in survival parameters among the species studied. These correlated with differences in total biomass/area and zooid/tunic proportions, thus indicating different biological strategies even in species of similar growth form and ecological distribution.

INTRODUCTION

The study of clonal organisms has attracted much attention in the last few decades, and theories and models have been developed to explain the consequences of clonal reproduction to the population biology of such organisms (see e.g. Larwood & Rosen 1979, Chapman 1981, Jackson et al. 1985, Harper et al. 1986). As a result of the indeterminate growth featured by most clonal organisms (Sebens 1987), the evolution of any clone is highly variable, depending on its particular history of phases of growth, degrowth, fission, fusion, regeneration and senescence. Due to such plastic, complex and heterogeneous life cycles, organisms of the same age diverge widely in their growth, survival and fecundity processes. Decoupling of size and age makes models based on age-classified populations unsuitable, and it has been pointed out that life-history theory and classical demographic methods have been developed for organisms without clonal reproduction (Caswell 1985, Hughes & Cancino 1985, Williams 1986). Life-history traits in clonal animals are likely to be tied more to size or life-cycle stage than to age (Hubbell & Werner 1979, Caswell 1982a, b, 1985, Jackson & Coates 1986). Moreover, in indeterminately growing organisms, accurate measurements of rele-

vant rates can be obtained only by following the fates of individual colonies over as much of their life-cycle as possible (Hughes & Cancino 1985, Stocker 1991). Underwood (1989) signaled the utility and limitations of monitoring programmes in estimating natural rates of change of populations.

An integrated theoretical life-history approach must be combined with data for particular species (Sebens 1987). Unfortunately, these data are very scarce, at least regarding clonal benthic animals on natural substrata, as has been long recognized (Ayling 1983, Connell & Keough 1985, Hughes & Cancino 1985, Jackson 1985). The aim of this work was to contribute to this issue by studying the main life-history traits (growth, reproduction, mortality) of some clonal ascidians in the Mediterranean Sea and examining the correlation of these traits with some parameters which have been considered relevant in the context of clonal organisms' development, such as habitat characteristics, size, shape, seasonality, and investment in reinforcement and defence (Buss 1979, 1980, Jackson 1979, Coates & Jackson 1985, Dyrinda 1986, Sebens 1986, 1987).

The data available for colonial ascidians are fragmentary (Fiala-Médioni 1987) and present a complex picture of phases of sexual and asexual reproduction,

mixed with phenomena such as recession, fission, fusion, senescence and rejuvenescence. A seasonal pattern is found in temperate waters (Millar 1971). Some studies in tropical species have shown a population biology dominated by a highly dynamic system of asexual propagation by fission and fusion (Bak et al. 1981, Ryland et al. 1984), which has also been reported in more temperate waters (Port Jackson, Australia; Stocker 1991).

MATERIAL AND METHODS

Study site, species and habitat. The study was carried out in Tossa de Mar (NE Spain) from May 1988 to May 1990. Data on the study site are given in Turon (1987). The following species were investigated: family Polycitoridae, *Cystodytes dellechiaiei* (Della Valle 1877); family Didemnidae, *Diplosoma spongiforme* (Giard 1872) and *Polysyncraton lacazei* (Giard 1872); family Polyclinidae, *Pseudodistoma crucigaster* Gaill 1972 and *Aplidium* aff. *conicum* (Olivi 1792) (for discussion of the taxonomic position of the latter species, see Turon 1987). All these species display an encrusting growth form (old colonies of *P. crucigaster* are somewhat massive in the shallow community studied here, but the young colonies monitored retained an encrusting morphology over the study period). Consequently, changes in the surface area of the colonies provided a good estimate of growth. These 5 species are abundant on rocky surfaces at the sublittoral level in this area. Their reproduction, ecological distribution and abundance are discussed in Turon (1988, 1990). There is a broad overlap among the habitats occupied by these species, although the relative abundances vary with depth.

In order to minimize the effect of habitat variables such as depth, orientation, currents, etc., all colonies of each species were selected from the same wall, within a narrow (2 m) depth range. The characteristics of the locations where the colonies were marked were as follows (see also Ros et al. 1985, Turon 1990): *Polysyncraton lacazei* and *Cystodytes dellechiaiei*, depth 10 to 12 m, vertical wall, orientation NE, sciaphilic assemblage; *Aplidium* aff. *conicum*, depth 15 to 17 m, subvertical wall facing SE, sciaphilic assemblage; *Diplosoma spongiforme*, depth 3 to 5 m, subvertical wall in calm waters, orientation SW, photophilic assemblage; *Pseudodistoma crucigaster*, depth 4 to 6 m, vertical wall with moderate wave action, orientation NE, photophilic assemblage. A total of 40 colonies of *Polysyncraton lacazei*, 14 of *Pseudodistoma crucigaster*, 39 of *Aplidium* aff. *conicum*, 8 of *Diplosoma spongiforme* and 11 of *Cystodytes dellechiaiei* were included in the survey.

Selection procedure and data collection. Colonies of all species except *Aplidium* aff. *conicum* were selected as follows: in April and May 1988, the adult colonies present in selected areas of the rocky walls were mapped and afterwards new recruits were located, starting in May and lasting until the end of the reproductive period of each species. In this way we could be sure that the colonies selected were new juveniles, not small or recessing colonies from previous years which had already been mapped in the April and May surveys. Some of the new colonies in the areas surveyed were haphazardly selected and marked using nails with plastic labels driven into the rock at a short distance from the specimens. The new colonies could be distinguished when still small, when they were made up of 1 to 3 systems (somewhat larger in *Diplosoma spongiforme*, whose transparency hindered the location of small colonies). These colonies were then monitored monthly until May 1990. Two additional surveys were carried out in September and November 1990 to observe the state of the colonies after the third summer. Additional colonies of the same generation of *Polysyncraton lacazei* were added to the survey during the second half of 1988 to compensate for the high mortality rate of this species.

Because of the resting period of *Aplidium* aff. *conicum* during the summer months, colonies of this species were not marked until September 1988, when they resumed activity. We assumed that very small colonies (under 4 cm²) had settled the previous winter, although confirmation of this was not possible.

The monitoring process consisted of monthly drawings of the outlines of the colonies, carried out under water on acetate sheets. The accuracy of this method varied with the position of the colony and sea conditions. The error level was estimated at ca 2 to 5%. Whenever a colony was located where access was difficult (e.g. in a crevice), 3 drawings were made in each survey and the mean for the different parameters was used. Other parameters of interest, such as resting states, fissions or fusions, injuries, competing neighbouring species, etc., were also noted. One sample (from 5 to 10 colonies) of each species from the local populations was collected during each dive to ascertain the reproductive periods.

In addition, data were obtained on the biomass per area and the proportion of zooids relative to the tunic material for each species. Small pieces (ca 1 cm²) were cut from 3 colonies of each species (during the non-reproductive period). These pieces were then measured, the zooids were carefully taken out, and the dry weight of the zooids and the remaining tunic was noted. A further problem arose in *Aplidium* aff. *conicum*, the tunic of which is heavily encrusted with exogenous material (sand grains, shell debris, etc.). For this species,

after weighing the tunic a hot solution of sodium hypochlorite was used to dissolve all organic material, followed by several rinses in distilled water. The resulting weight of the sand material was then subtracted from the dry weight of the tunic with encrustations.

Data treatment. The outline drawings were digitized and analysed on an IBAS image treatment system (Kontron). The parameters recorded were the area and an index of circularity (area of the colon/area of a circle of equivalent perimeter) ranging from 1 (perfectly circular) to 0. A growth estimate was calculated for each colony and each month, from the expression: $r_i = (\text{surface in month } m_i - \text{surface in month } m_{i-1}) / \text{surface in month } m_{i-1}$.

Fission and fusion of colonies were rare in the species studied, except in *Polysyncraton lacazei*, for which these phenomena were relatively frequent (some instances were also recorded in *Diplosoma spongiforme*). For the purpose of the present study these phenomena were treated as follows: when 1 colony underwent fission, we used in subsequent months the sum of the areas of the resulting clonemates. When 2 colonies fused, their areas observed before the merging were added together to give one single value per survey. We are aware that this implies a simplification of the growth ecology of this species (*P. lacazei*), but due to the moderate number of fissions and fusions observed, we preferred to retain a simple presentation of the data in order to make them comparable with those for the other species.

Non-parametric statistical analyses were used whenever normality and homoscedasticity of the data could not be demonstrated by Kolmogorov-Smirnov and Bartlett tests, respectively (Zar 1984). All analyses were performed using a Systat 4.0 package, except the survival data, which were analysed using Programmes 11 and 21 of the BMDP package. The actuarial life table method (Cutler & Ederer 1958) was used to estimate the survival function, and rank statistics (analogous

to the Kruskal-Wallis test) were used to test the equality of the survival curves (Breslow 1970). Covariates were included in the survival analyses using Cox's (1972) proportional hazards regression model.

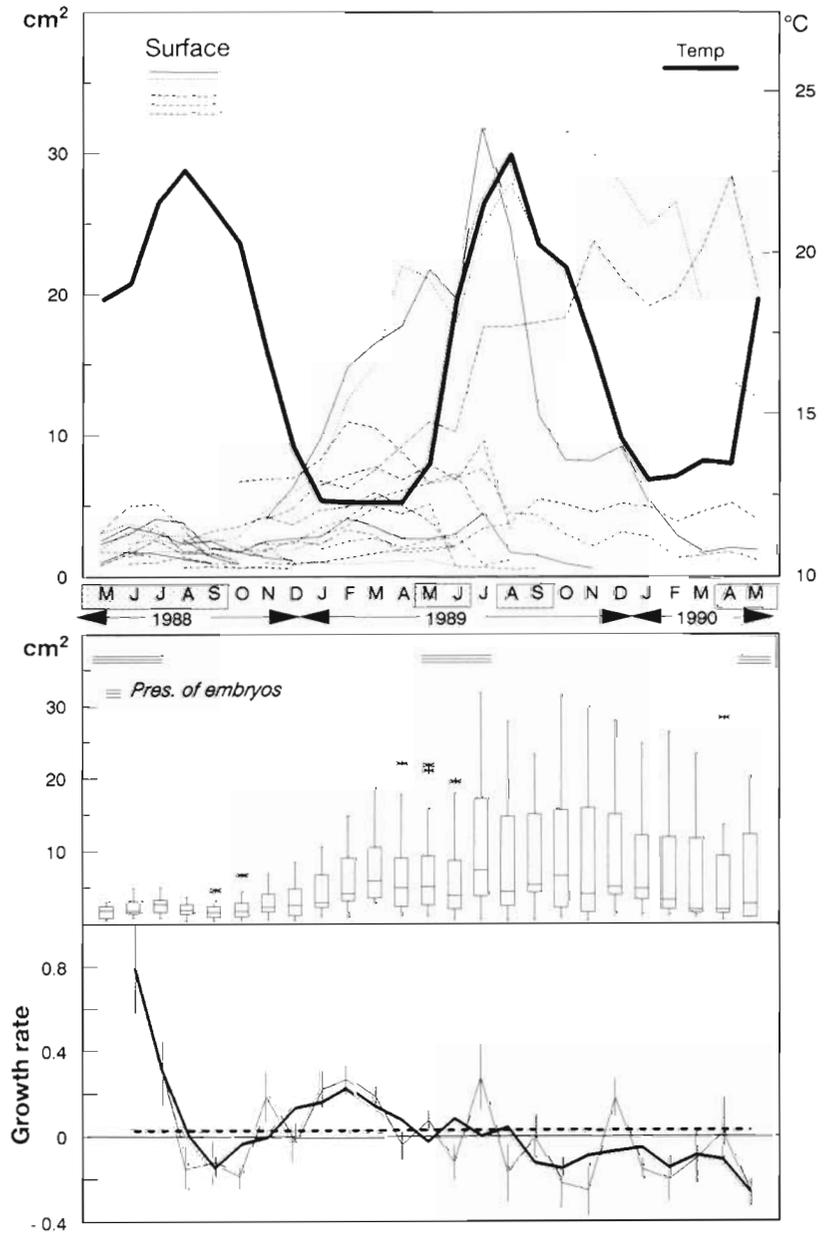


Fig. 1. *Polysyncraton lacazei*. Upper: Surface area of colonies studied during the survey. Water temperature graph at 10 m depth superimposed. Shaded months indicate periods in which resting forms were observed in the population. Middle: Box and whisker plot of surface area of the colonies. Extreme values (more than 1.5 times the interquartile distance) are marked with asterisks. Periods in which incubating embryos were present in the population are indicated by 3 parallel lines. Lower: Mean growth rates (thin line; error bars are standard deviations) and 'smoothed' values (thick line; see text)

RESULTS

Figs. 1 to 5 summarise the results on growth of the colonies monitored. Individual growth, the corresponding box and whisker graphs (McGill et al. 1978) and mean growth rates are represented. Line drawings of the latter were smoothed in order to make them less noisy and facilitate interpretation. A moving average 3 points wide was used as a simple filtering technique.

Individual growth, resistance periods and reproduction

The growth patterns of the 5 species studied reflected different growth strategies, although there was a clear trend towards higher growth in the winter season than in summer. A marked variability in the area attained by the colonies was also common.

Polysyncraton lacazei (Fig. 1) exhibited restricted, or even negative, growth during the first summer, only to give rise to a burgeoning development in winter. In the second summer, many colonies began to recede in area. Only 2 colonies were able to start growing again during the second winter, while the remainder failed to regain the area lost, surviving for longer or shorter periods in the form of small fragments (fragmentation into small units usually accompanies the recession toward death). Another decrease in size in the survivor specimens marked the arrival of the third summer at the end of the study. In the complementary survey in November 1990, only 3 colonies were still alive, but only one was in a 'healthy' (non-fragmented) state. The breeding period of this species lasts from spring to the first half of summer.

Another feature of *Polysyncraton lacazei* was the appearance of resting forms in spring and summer. This is a brief phenomenon, lasting from 1 to 4 wk, and it can easily be recognised by the sealing off of the siphonal apertures and the presence of a glassy pellicule on the surface of the colony. Fig. 1 also depicts (shaded months)

the surveys in which population members (but not necessarily the colonies monitored) of this species in the area were observed in resistance form. The percentage of colonies in resistance form was never high (the number never exceeded 30 % during this period).

Pseudodistoma crucigaster (Fig. 2) also showed active growth during the first winter, with a noticeable recession during the second summer, in June 1989; this recession was fairly synchronized and involved the switch to a resting form of nearly all the colonies. Growth was reestablished at the end of summer and during the second winter. The complementary obser-

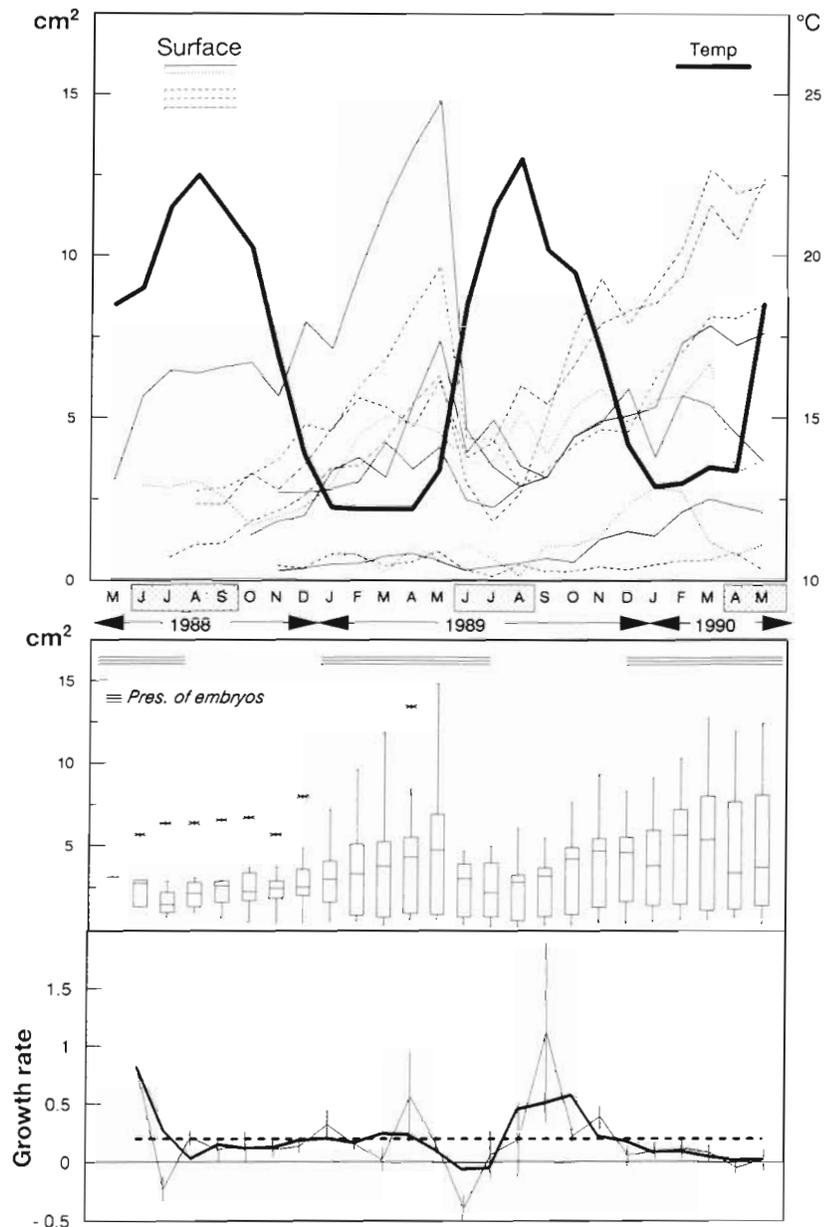


Fig. 2. *Pseudodistoma crucigaster*. Colony growth. Panels and symbols as in Fig. 1

variations of September and November 1990 seem to indicate that a recession also occurred in the third summer, with reactivation of growth as temperatures fell. Thus, a picture emerges of winter growth and summer recession. The breeding period of this species is remarkably long, and several larval release events are likely during this period. It may be noted that the colonies studied had settled at the beginning of the reproductive period. Some small colonies which had settled at the end of this period were added to the study in November (see Fig. 2). They did not exhibit winter growth in 1988. Instead, they remained small until they began moderate development in winter 1989.

A resting or non-feeding state also occurred in this species. As with *Polysyncraton lacazei*, this phenomenon was very brief (again, it was not observed for more than 4 wk on any given colony), and it was easily distinguishable by the presence of a glassy cuticle over the colonial surface. It occurred during summer, and also in spring 1990 (Fig. 2).

In *Aplidium* aff. *conicum* (Fig. 3) the developmental pattern was dominated by the occurrence of a resistance period during the months of highest water temperatures and affecting 100% of the population colonies; these characteristics differentiate this 'true' resistance phase from the non-feeding episodes of the above species. Growth was active in the first winter, but in the second winter mean growth rate was negative, resulting in a smaller mean cohort area at the end of the second winter than at the end of the first. A decrease in size preceded the onset of the resistance period in most colonies. It was impossible to measure the size of the colonies in 'aestivation', due to extensive fouling by other organisms, which made the colony outlines untraceable. Colony area receded during this resistance phase, although this might have been only apparent and due to the fact that, when reactivation occurs, it starts with the central system of zooids, while the basal part of the colony is still covered at the periphery by epibionts. The initial growth in winter might only represent

a recovery to the full size of the colony, with 'true' growth occurring afterwards. Similarly, in September 1990, the colonies began reactivation and most of them could be located in November 1990 in completely active form. The breeding period of this species is clearly restricted to the colder months.

In *Cystodytes dellechiaiei* (Fig. 4) the development pattern was quite unusual. Nearly all the colonies exhibited restricted growth or none whatsoever over the entire 2 yr period. Only one colony was able to grow

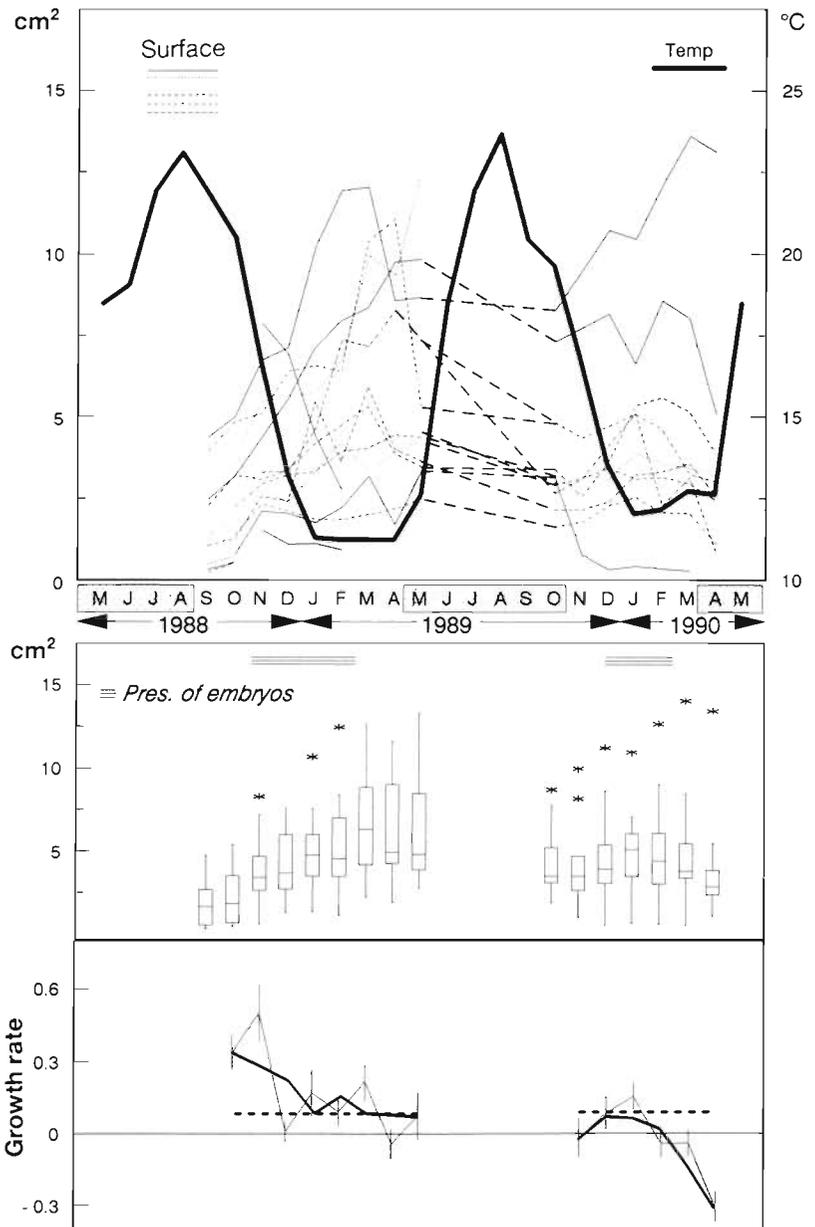


Fig. 3. *Aplidium* aff. *conicum*. Colony growth. Panels and symbols as in Fig. 1. During resistance periods no measurements of colonial surface were possible (see text). Dashed lines connect the surface values of colonies before and after the resistance period

actively, attaining a large size. This colony was located in more shaded conditions than the remaining ones, due to a slight change in the inclination of the wall. The breeding season occurs in spring, with larval release in May and June.

Diplosoma spongiforme (Fig. 5) behaves like an annual species. The colonies studied featured restricted growth during the first summer, with most of the colonies (6 out of 8) dying during this period. Only 2 colonies reached the cold season, at which point a noticeable development began, ending abruptly with the onset of high water temperatures in the following year, when the colonies underwent fragmentation and disappeared. Although the specimens monitored did not survive beyond spring, large colonies of the population were observed to survive a little longer, until August, when all the members of this generation apparently disappeared. Embryos were present in the population during spring and the first half of summer.

Growth rates

Relative growth rates showed a noticeable variability among colonies, as indicated by the large standard deviations. The general course of the growth rate means followed the periods of active growth and recession of the species considered.

The overall means (dashed horizontal line in Figs. 1 to 5) of the growth rates were in general >0 (i.e. effectively gaining surface area), although the monthly means clearly could fall below 0. In *Aplidium* aff. *conicum*, *Polysyncraton lacazei* and *Cystodytes dellechiajei* growth rates were clearly lower (and even negative in many months) in the second part of the study.

The possible relationship of relative growth rates with certain parameters was tested: there was no significant correlation between growth rates (monthly means) and temperatures (Table 1). It is also conceivable that during the period of sexual activity the growth rates were lower in order to allow energy to be allocated to reproduction. Table 1 also lists the mean growth rates during the active

(i.e. when gonads and/or larvae were present) and inactive periods for these species. Mean growth rates were slightly higher during the non-active periods, but the differences were not significant (*t*-test). This analysis was not performed for *Cystodytes dellechiajei*, since in this species gonads are present in the population for the greater part of the year. Finally, a correlation between colony size and growth rate was also to be expected. Table 1 displays the correlation between individual colony sizes and growth rates. The correlation was weak, but significant, in all species except for *Polysyncraton lacazei*. The correlations were all nega-

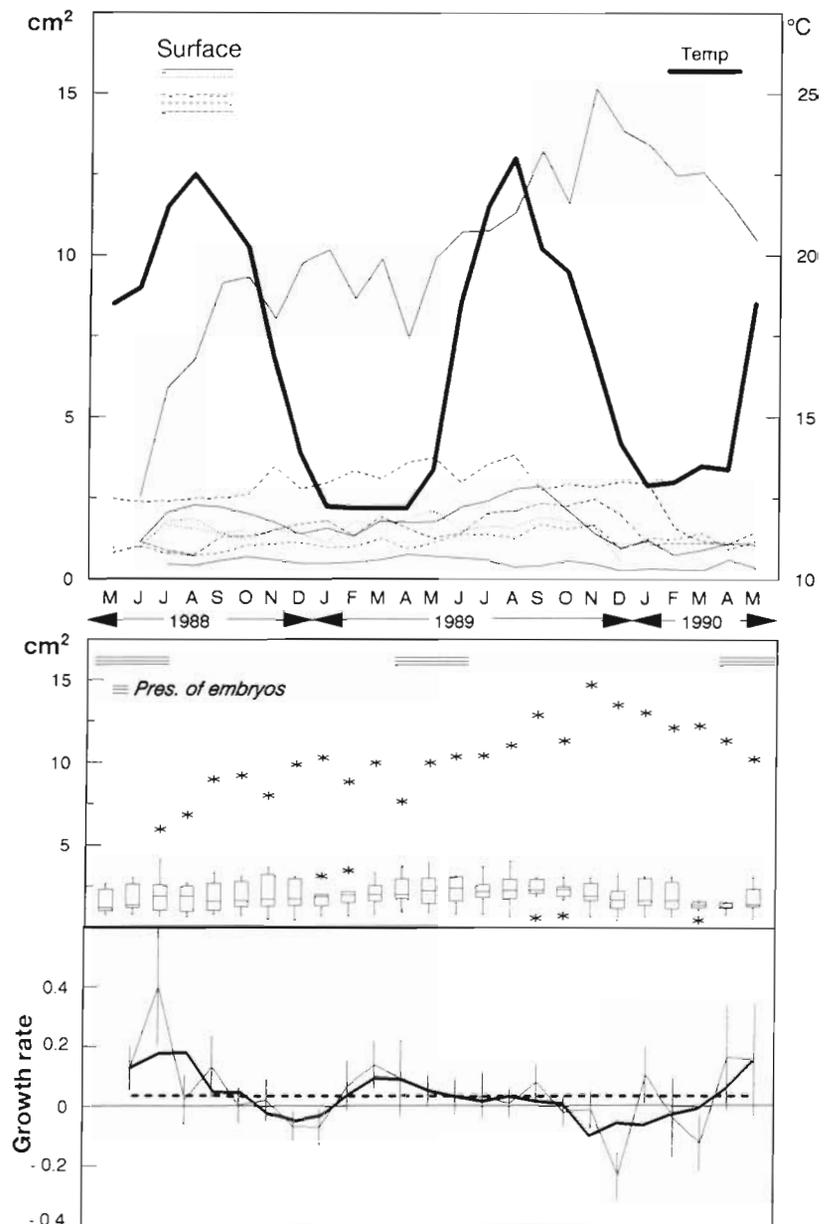


Fig. 4. *Cystodytes dellechiajei*. Colony growth. Panels and symbols as in Fig. 1

tive, indicating that small colonies tended to grow at a faster rate than larger ones. In fact, in most species the highest growth rate values were recorded during the first months.

Circularity index

In Fig. 6 circularity index is plotted against colonial surface area (logarithmic scale) for the 5 species studied. The regression between circularity index and log-transformed areas fits a linear function. The Pearson correlation coefficients are all negative and significant at $p < 0.001$.

A decrease in circularity with increasing colony area is to be expected, due to the impossibility of maintaining a circular shape while growing over a physically and biologically heterogeneous surface. However, it is of interest to examine whether the relationship is the same in all species or if some of these tend to adopt a more markedly irregular shape when growing. The regressions were compared using covariance analysis, and the assumption of homogeneity of slopes does not hold at $p < 0.001$. Multiple pairwise comparisons among slopes (Zar 1984) revealed that the regression coefficient of *Pseudodistoma crucigaster* was significantly smaller than for the other species (in other words, it retained a more circular form when growing). There were no significant differences in slope among the other 4 species. The covariance analysis was repeated with the latter 4 species, and significant differences in elevations were found. Multiple comparisons among elevations (Zar 1984) demonstrated that *Diplosoma spongiforme* had a significantly lower elevation (in other words, featured a more irregular outline for a given area) than the remaining 3 species.

Survival

The cumulative survival functions computed using the actuarial life table method (Fig. 7) were quite variable in the species studied. There was a phase of high mortality during the second half of the first summer and the beginning of the first winter. After this

period the survival curves became stabilised in *Aplidium* aff. *conicum*, *Pseudodistoma crucigaster* and *Cystodytes dellechiaiei* (cumulative survival was more than 0.7 by the end of the study in the latter 2 species). Conversely, by the second summer the marked colonies of *Diplosoma spongiforme* had died and most of the specimens of *Polysyncraton lacazei* had disappeared before the third summer.

Plots of the death density function (probability of death per unit time; Gross & Clark 1975) reflected this pattern of increased hazard levels for *Polysyncraton lacazei* and *Diplosoma spongiforme* and lower

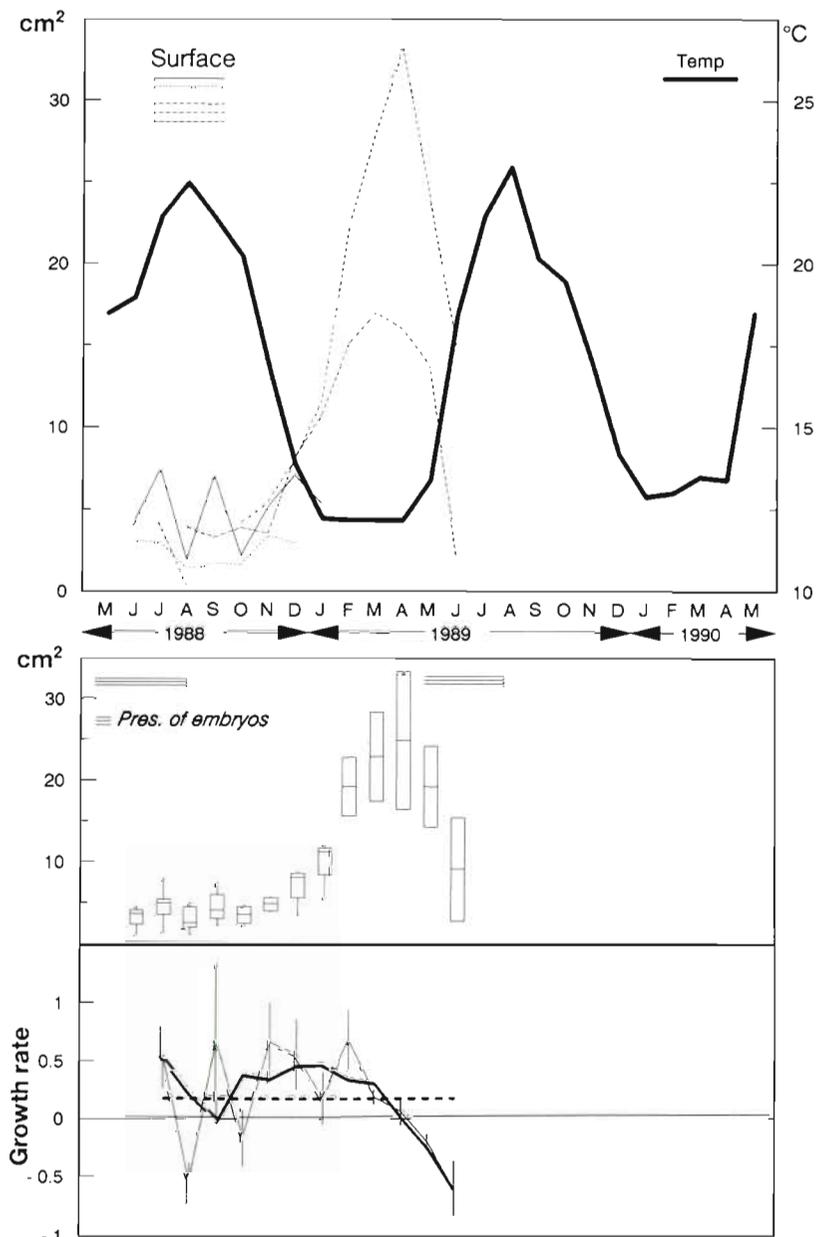


Fig. 5. *Diplosoma spongiforme*. Colony growth. Panels and symbols as in Fig. 1

Table 1. Correlation of growth rates with temperature, sexual activity and size of ascidian colonies. r_s : Spearman correlation coefficient; ns: not significant

	<i>Polysyncraton lacazei</i>	<i>Pseudodistoma crucigaster</i>	<i>Aplidium aff. conicum</i>	<i>Cystodytes dellechiajei</i>	<i>Diplosoma spongiforme</i>
Growth rate vs temperature					
r_s	-0.165 ns	0.026 ns	0.114 ns	0.027 ns	-0.211 ns
Growth rate vs sexual activity					
Active					
Mean	0.018	0.177	-0.021	-	0.115
SD	0.187	0.294	0.190	-	0.437
Inactive					
Mean	0.033	0.214	0.141	-	0.218
SD	0.347	0.383	0.176	-	0.561
t-test	ns	ns	ns	-	ns
Growth rate vs size					
r_s	-0.070 ns	-0.178 p < 0.02	-0.288 p < 0.005	-0.131 p < 0.05	-0.264 p < 0.05

death densities for the other 3 species (Fig. 7). There was no significant relationship (Spearman rank correlation) between temperature and the death density function.

Rank statistics were used to compare the survival curves (Table 2). The overall test was significant, and pairwise comparisons (the overall alpha level was 0.05) showed that *Polysyncraton lacazei* and *Diplosoma spongiforme* significantly differed from the other 3 species in having lower survival values. The low numbers of deaths recorded for *Cystodytes dellechiajei*, *Pseudodistoma crucigaster* and *Aplidium aff. conicum* hindered the search for any trends in these species; these should be examined over a larger time interval.

The possibility that the mortality observed was size-dependent was tested using the size attained by the colonies as a covariate in the hazard function (Cox's proportional hazards regression model). A regression coefficient could then be estimated and tested for significance (using the likelihood ratio test, which is compared with the chi-square distribution; Rao 1973). The results are shown in Table 2. All regression coefficients were negative, but only in the case of *Polysyncraton lacazei* and *Aplidium aff. conicum* were they significant, indicating that the larger the colony, the less the hazard of death, i.e. a positive relationship with survival.

Biomass

The biomass (dry weight) of zooids and tunic tissue per cm^2 and the relationship between zooid and tunic weights are presented in Fig. 8.

ANOVA analyses (Table 3) show that the biomass values were significantly different among the

species, as was the zooid/tunic weight ratio. It is noteworthy, however, that the dry weight of zooidal material per unit area was not significantly different among the 5 species.

Tukey's HSD multiple comparisons procedure was used to test for significant differences among pairs of means of the total biomass and of the zooid/tunic weight ratio. *Pseudodistoma crucigaster* had significantly higher biomass cm^{-2} than the remaining species. All other comparisons were not significant. Regarding the zooid/tunic ratio, *Diplosoma spongiforme* clearly displayed the highest value, and comparisons with all other species were significant. *Polysyncraton lacazei* also had significantly higher values than *Pseudodistoma crucigaster* and *Cystodytes dellechiajei*. The remaining comparisons were not significant.

DISCUSSION

The growth of the colonial ascidians studied featured a pattern in which periods of growth and recession alternated, and there was wide variation among specimens of the same species.

The Mediterranean displays marked seasonal fluctuations in environmental parameters (Zabala & Ballesteros 1989), which is reflected in some of the seasonal patterns in life cycles of ascidian populations. This seasonality is clear with respect to reproductive and resting periods, as well as to phases of active growth. In the species studied, summer appears to be the unfavourable season, with ascidians undergoing surface recession and resting periods. Only in *Cystodytes dellechiajei* is no seasonal pattern of growth apparent.

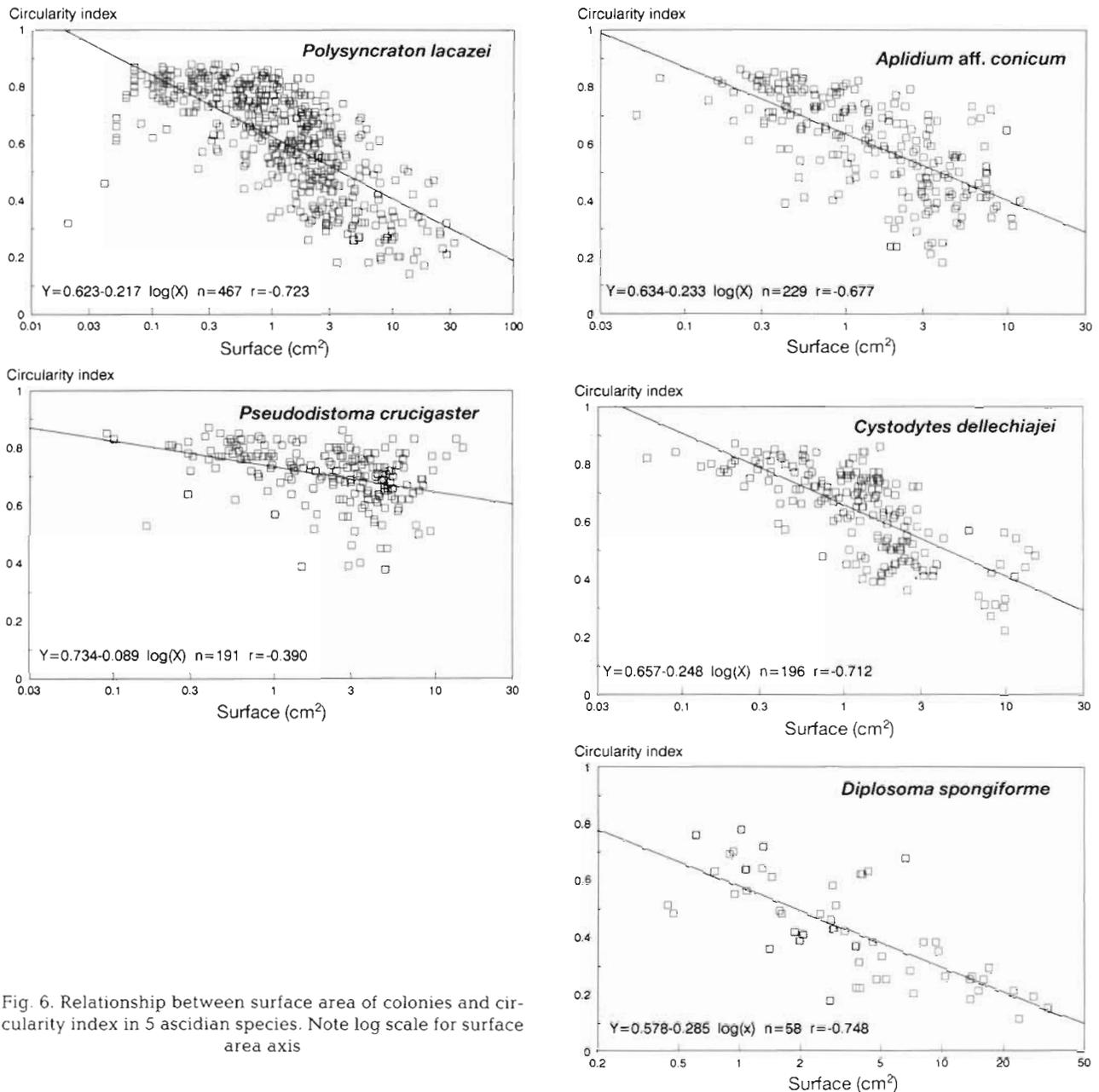


Fig. 6. Relationship between surface area of colonies and circularity index in 5 ascidian species. Note log scale for surface area axis

In this species, most of the colonies featured practically no growth during the study. Only one specimen attained a large size. This was probably linked to microhabitat conditions. In fact, colonies as large as, or even larger than, the one that grew actively in our study are not uncommon in shaded environments in the study site.

The high variability featured by the colonies of each species for nearly all the parameters studied (except for biomass and zooid/tunic values, which were quite constant within species) is worth noting. It cannot be ascribed with certainty to genetic differences or habitat conditions. Ambient parameters were kept as uni-

form as possible by sampling in a restricted and ecologically uniform zone, but small-scale variations and different biological interactions are unavoidable in field studies and may account for the variability observed. Studies on the growth of modular animals usually report such wide variability (see e.g. Brunetti & Copello 1978, Ayling 1983, Todd & Turner 1988, Stocker 1991, Wulff 1991). Indeed, plasticity is a characteristic of the indeterminate growth featured by these organisms (Sebens 1987).

Colony growth rates significantly decreased with increased colony area in most of the species studied. It has been suggested that relative growth rates of

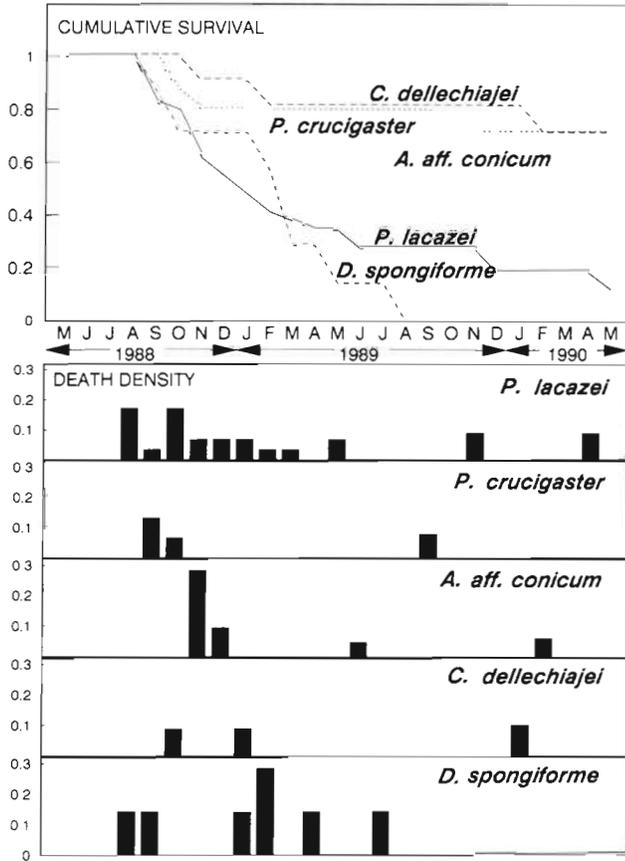


Fig 7. Upper: Cumulative survival function of 5 ascidian species. Lower: Histograms of death density function values (probability of death per unit time) of these species

colonial invertebrates should remain constant with increasing size or age (Jackson 1979, 1985). In fact, exponential growth has been recorded for some colonial tunicates, at least for part of the life span (Oka & Usui 1944, Yamaguchi 1975), but these measurements

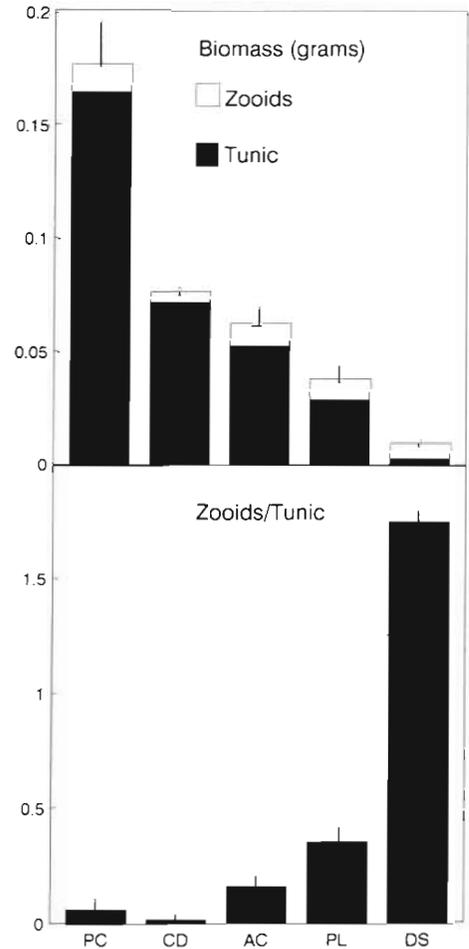


Fig. 8. Upper: Biomass (dry weight) cm^{-2} of zooids and tunic of *Pseudodistoma crucigaster* (PC), *Cystodytes dellechiajei* (CD), *Applidium aff. conicum* (AC), *Polysyncraton lacazei* (PL), *Diplosoma spongiforme* (DS). Vertical lines are standard deviations of the total biomass. Lower: Zooid/tunic ratio. Vertical lines are standard deviations

Table 2. Comparison of survival functions of ascidian colonies; and significance of regression of area on hazard values. ns: not significant

Comparison of survival functions					
Overall:	Breslow test = 9.99		df = 11	p < 0.05	
Multiple comparisons:					
<i>Diplosoma spongiforme</i>	<i>Polysyncraton lacazei</i>	<i>Applidium aff. conicum</i>	<i>Cystodytes dellechiajei</i>	<i>Pseudodistoma crucigaster</i>	
Regression of area of hazard values					
	<i>Polysyncraton lacazei</i>	<i>Pseudodistoma crucigaster</i>	<i>Applidium aff. conicum</i>	<i>Cystodytes dellechiajei</i>	<i>Diplosoma spongiforme</i>
Regression coefficient	-0.244	-0.054	-0.244	-0.806	-0.005
Standard error of coefficient	0.097	0.123	0.097	0.651	0.037
Significance (L-ratio statistic)	p < 0.001	ns	p < 0.01	ns	ns

Table 3. ANOVA for total biomass, zooid weight and zooid/tunic ratio in 5 colonial ascidian species

Source		SS	df	MS	F-ratio	p
Total biomass cm ⁻²	Species	0.049	4	0.012	6.528	<0.01
	Error	0.019	10	0.002		
Zooid biomass cm ⁻²	Species	0.17 × 10 ⁻³	4	0.42 × 10 ⁻⁴	1.650	0.237
	Error	0.25 × 10 ⁻³	10	0.25 × 10 ⁻⁴		
Zooid/tunic biomass ratio	Species	6.477	4	1.619	328.207	<0.001
	Error	0.044	10	0.004		

were made under experimental conditions. Growth in natural conditions is constrained by environmental parameters (habitat boundaries, competition, partial predation), imposing a decline in growth rate with size even in animals with indeterminate growth (Sebens 1987). Other measurements on colonial ascidians also show a decrease in relative growth rates with size (Brunetti & Copello 1978, Brunetti et al. 1988, Stocker 1991).

A seasonal pattern in the mortality data was not apparent. The study period was too short to detect any trend in these parameters, at least in the species featuring the highest survival (*Aplidium* aff. *conicum*, *Cystodytes dellechiaiei*, *Pseudodistoma crucigaster*). A relation between size and mortality was not demonstrated, except for *Polysyncraton lacazei* and *Aplidium* aff. *conicum*, in which mortality was higher in small colonies. We did not see any direct evidence of predation or other sources of mortality. However, the abrupt disappearance of certain colonies (without prior shrinkage or fragmentation) may be interpreted as a predation or dislodgement event. Instances of these phenomena have been observed in particular for small colonies of *P. lacazei*.

A panoply of non-feeding and resistance forms was also encountered in 3 of the species studied, all of them occurring in the warm season. The resistance form of *Aplidium* aff. *conicum* is an example of the well-documented 'survival budding' (Nakauchi 1982) featured by many colonial ascidians. The non-feeding periods observed in *Polysyncraton lacazei* and *Pseudodistoma crucigaster* are clearly different; non-feeding in the former species is studied in more detail in Turon (in press). This kind of phenomenon may prove to be an important feature of the biology of colonial ascidians and warrants further investigation. A shrinkage and an apparent loss of antifouling abilities occur in all of the resting forms observed, which may be relevant in the context of space competition.

Except for *Diplosoma spongiforme*, which features an apparently annual life cycle (this finding should be interpreted cautiously due to the low number of

colonies studied), the ascidian species studied are perennial and iteroparous, with one breeding season per year. It is difficult to provide data on the longevity of these species, as their lifespans are longer than the period analysed. Only in the case of *Polysyncraton lacazei*, of which only one colony remained healthy 2½ yr after settlement, can we conclude that the average lifespan (for colonies surviving the high initial mortality rate) is 2 yr or less. Wahl & Lafargue (1990) estimate a lifespan of 3 yr for this species in Banyuls-Sur-Mer (France), a locality with colder waters than our study site.

Although all the species studied displayed an encrusting morphology, significant differences in shape were encountered. The degree of perimeter convolution (as indicated by the circularity index) with increasing size was significantly lower in *Pseudodistoma crucigaster* than in the other species. The circularity index has important implications regarding the competition for space between the species, since the perimeter of the colonies marks the zone of interaction with neighbouring species. The loss of circularity during colonial growth, resulting in an irregular shape of larger colonies, also reflects the colonies' ability to adapt to their spatial microenvironments and display some directionality in their growth (Buss 1979). *Diplosoma spongiforme* featured the most irregular growth form of all species studied. Moreover, a relationship between shape and mortality has been recently demonstrated in one colonial ascidian (Stocker 1991).

Fragmentation of colonies has been observed in *Polysyncraton lacazei* and, to a lesser degree, in *Diplosoma spongiforme*; it is less important as a dispersion phenomenon in ascidians than in other groups (e.g. sponges; Wulff 1991). Some ascidians undergo frequent fission, but the resulting colonies remain close to the parents (Bak et al. 1981, Ryland et al. 1984, Stocker 1991). However, fragmentation in *Polysyncraton lacazei* was much less extensive than in the above studies and we did not address this phenomenon specifically in this work.

The data on biomass are highly revealing in terms of the growth strategy of the species studied. The biomass of zooidal or 'active' tissue was not significantly different among the species studied, perhaps representing the limits imposed by food availability. Thus, the differences observed in biomass were attributable to differential investment in 'structural' (tunic) material, one of the defence mechanisms of modular, immotile animals (Dyrynda 1986). Svane & Lundalv (1982) also demonstrated the importance of the tunic/mantle ratio for some life-history parameters of solitary ascidians. In our species, *Diplosoma spongiforme* displayed the least biomass per unit area, and featured the highest proportion of zooid vs tunic material. This reflects a somewhat opportunistic strategy, allowing rapid growth under favourable conditions. This point is consistent with other parameters studied, such as the short lifespan, low survival, high growth rates and large areas of convoluted perimeter attained by some colonies. On the other hand, *Aplidium* aff. *conicum*, *Cystodytes dellechiaiei* and *Pseudodistoma crucigaster*, which have high relative biomasses per unit area (invest much more in the production of protective and structural material), featured the highest survival values and attained more moderate sizes; their growth rates can be very low (*C. dellechiaiei*). All these parameters seem to indicate a more conservative strategy. *Polysyncraton lacazei* has intermediate characteristics for nearly all the parameters studied. This is not, of course, an absolute or rigid classification. Rather, it is only an ordination involving the relative positions of the 5 species in accordance with the growth characteristics studied. A further datum is the presence of chemical defences in all species except *D. spongiforme* (they were tested during a study on bioactivity in Mediterranean benthic species; Uriz et al. 1991).

In conclusion, the life-history traits featured by the 5 species studied showed an important seasonal component, and appeared variously correlated with other parameters. Thus, different adaptations do occur in the same or very close habitats. Growth form (encrusting in all the species studied) is not as informative in describing the strategy followed as are other morphological parameters. Size appears related to some life-cycle parameters but not to others. The data most promising as indicators of ecological strategy concern the proportion of structural (tunic) material. In future studies this may prove useful for testing evolutionary divergence in this group.

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