Seasonal Data on Morphology and Ecology of *Merismopoedia*-Like Marine Algae. Taxonomical Implications of the Observed Changes

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Abstract

Results of seasonal study on morphology and ecology of interstitial marine *Merismopoedia*-like algae are presented in this paper with the aim to compare classical taxonomy with that made by Drouet and Daily after their revision of coccoid Cyanophyceae:

— Shape and size of algal colonies, as well as number of cells per family, show strong variations along the year. Number of cells per colony ranges from 16 to 2560, single cells and 2—4 grouped ones, were often present.

— Cell size varies from 4 to 7 μ in diameter and from 6 to 19 μ in length.

— Colonies were found each month except February, showing an extense picture of related morphological forms.

— Growth takes place from March to August (r = 0.03). Mean values of duplication time were t = 25.8 days. The growth of a regeneration, to pass from families of 4 to 64 cells per colony, was 103.2 days.

— Morphological changes in the colonies and variation of cell size lead us to admit the validity of *Agmenellum thermale* (Kütz.) Drouet and Daily as taxon, because during a cycle of a year, the following taxa were related between them: *Merismopoedia convoluta* Breb., *Merismopoedia elegans* A. Br. var. *marina* Lagerh., *Merismopoedia glauca* (Ehrenb.) Kütz. (pro parte) and genus *Holopedium* Lagerh. and *Microcrocis* Ricker in Hauck and Ricker.

Individual cells can be related with the genera *Anacystis* and *Coccochloris*, both sensu Drouet and Daily.

Introduction

The taxonomic revision of coccoid Myxophyceae by Drouet and Daily (1956) acted among phycologists as a revulsive. Several authors as Bourrelly (1957), Padmaja and Desikachary (1967), Forest (1968) and Stanier *et al.* (1971) have criticised the criteria adopted by Drouet and Daily (1956) because of several different reasons.

Blue-green algae show an extreme high seasonal variability, reported to environmental conditions, and taxonomical descriptions were often made from few specimens or having enough specimens, from collections of one given season.

Following morphological changes in a given locality along the year it is possible to state the tolerance range of each species to different ecological factors (Komarek 1973) and the structural changes of taxonomical incidence.

High variability of taxonomical characteristics in some taxa causes confusion between themselves and suggests to reject the practice of basing species on habits. For this reason Drouet and Daily (1956) took their drastic taxonomical position.

Drouet and Daily (1956) include the colonial specimens belonging to the genus *Merismopoedia* Kütz and most of the genus *Holopedium* Lagerh., in the genus *Agmenellum* Brebisson, and suggest the existence of reports with other single-cell constituted taxa.

In this work we shall give evidence of the relation between several different "taxonomic entities" which are

different morphological manifestations of a single species. These observations support the validity of the taxa *Agmenellum thermale* (Kütz.) Drouet and Daily.

Methods

Samples to carry out the present work were collected in eight sandy-mud stations in the estuary of the Miñor River on the southern side of the Ria of Vigo in the NW of Spain (Fig. 1) Sites of collection are always protected from the direct wave action and placed between 0.74 and 1.90 m above the 0 level of tides

Collections were made monthly by means of a duplicate corer of 2.75 cm of section and 5 cm of depth into the sediment.

Salinity of interstitial water was measured with the Knudsen method (1902). Incident energy was calculated from Mosby's formula (1936) starting from data of the Meteorological Observatory of Vigo; mean values of different stations together with interstitial water temperature are shown in Figure 2.

In the area of study we found a *Cerastoderma edule — Scorbicularia plana* community in stations 1 to 6 and a degradated community of *Tellina tenuis — Tellina incrassata* in station 7 and 8.

Samples, fixed with a 5 % formaline solution, were washed in the laboratory according to Boisseau's (1957) modified method (Fig. 3) to remove animals and plants which were

further collected on plankton-net filters of 250, 125 and 62 μ and immediately stained with Bengala-red Water collected after filtration was carefully observed in order to localize individual cells, or small colonies composed by 2, 4 or 8 cells

Density of colonies (N_k) was counted in a modified Renaud-Mornant chamber (1966). Number of colonies is expressed per sample (two corers) to avoid extrapolation errors in the expression related to larger surfaces.

Cells per colony (C_k), morphology and dimension of colonies were determined microscopically. Length measures of colonial cells were made after squashing them between the slide and cover-slide.

Results

Morphology

a) *Shape and Size of Colonies:* The studied population was composed by gelatinous tabular colonies with regular or shapeless fringes, measuring between 30 $\times$ 40 μ and 400 $\times$ 200 μ. These colonies were divided as a grid in families of 4, 16, 32, 64 to 128 cells (Fig. 6 and Plate 1 g, h, i).

Number of cells per colony (C_k) ranges from 16 to 2560. The lamina becomes thicker with growth because of increasing of cell length from 6 to 16 μ (Figs. 4 and 5).

We found sometimes irregular colonies of disordered cells (16). The presence of individual (or grouped by 2 or
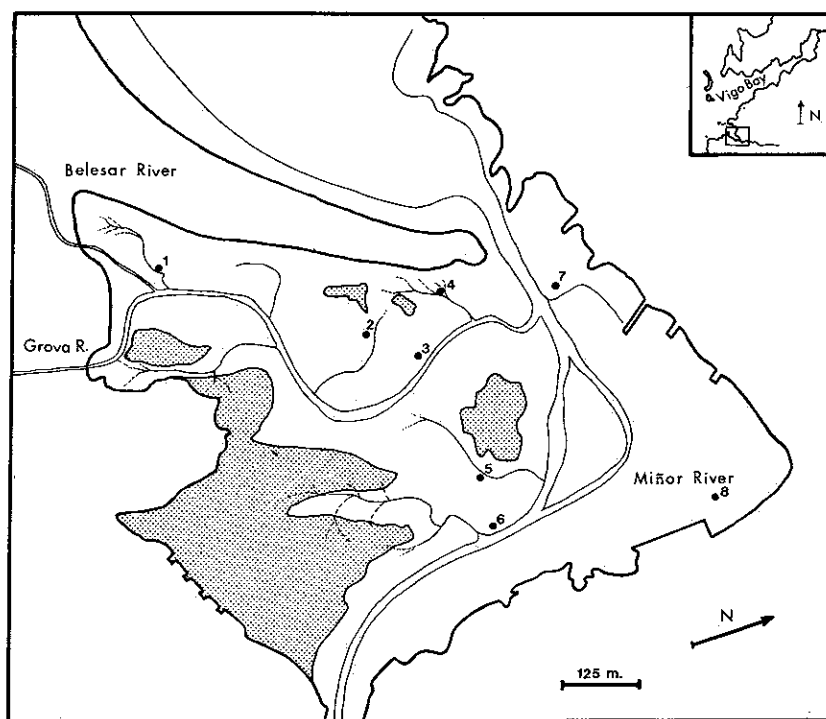

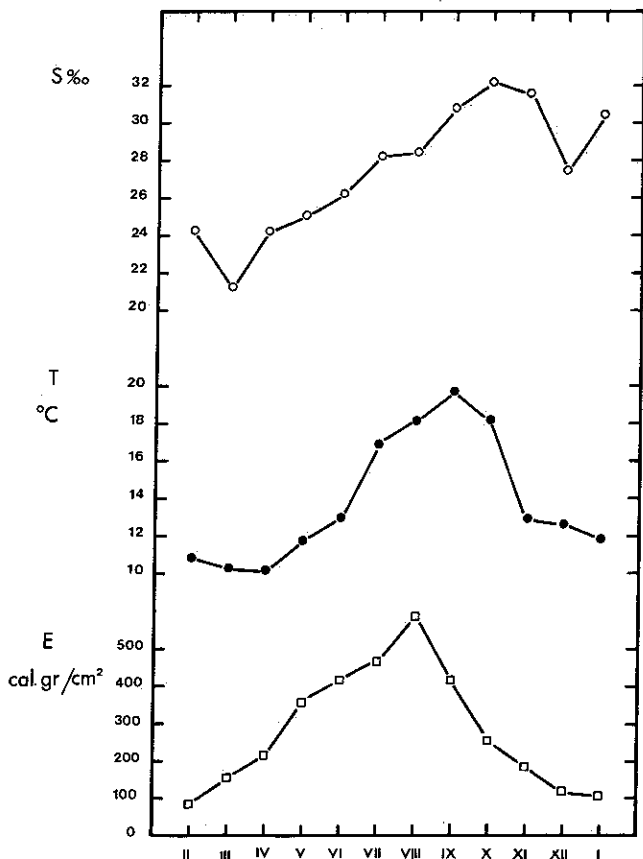Fig. 1 Situation of the localities where samples were collected. Shaded area show salt-marsh vegetation.

Fig. 2. Mean values of salinity (S), interstitial water temperature (T) for the eight stations; E are the incident energy values.
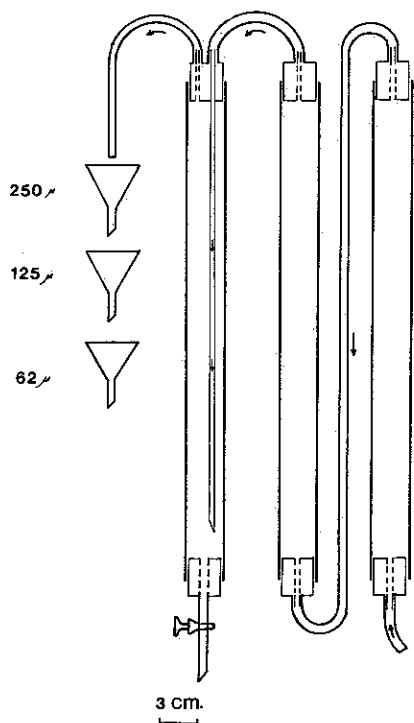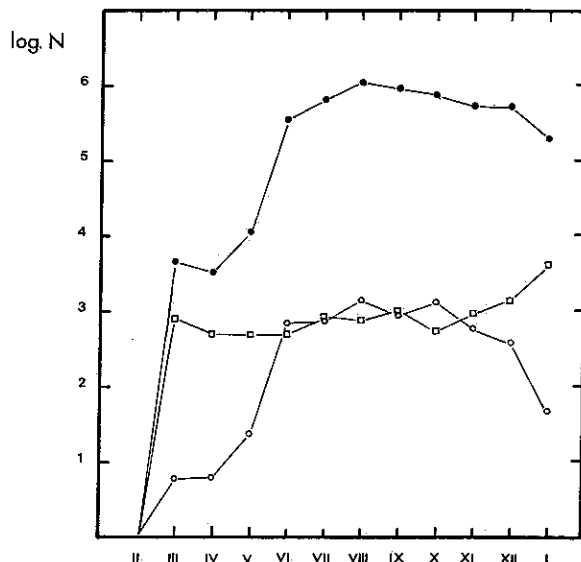

Fig. 4. Seasonal abundance of algae, ●: number of cells per sample, o: mean number of colony cells, ▫: density of colonies per sample.

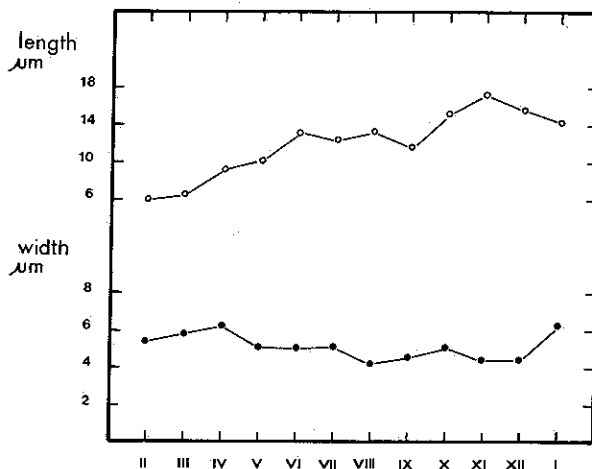Fig. 3. Schematic representation of the apparatus used to separate the sediment and living forms.

Fig. 5 Seasonal changes in mean values of length and width.

4) cells, was also frequent. Cells, which were found alone, had the same morphological aspect than those included in colonies (Pl. 1, b, e, f). Colonies frequently present (Pl. 1, h, j) empty places related to a next increase of free cells degaged from them.

Cells look close to one another in old colonies, and were slackly disposed in young ones (Plate 1); preceeding the cell separation some colonies present crevices between pairs of single cells offering the impression of an individual gelatinous cover (Fig. 6 and Pl. 1, specially h). All cells, in a given colony, are not equal in size, as pointed out by Fritsch (1952) and Drouet and Daily (1956); further, fringe cells can divide unequally giving two different cells: one sphaerical and one ellipsoidal (Fig. 7, Pl. 1, a, c, d).
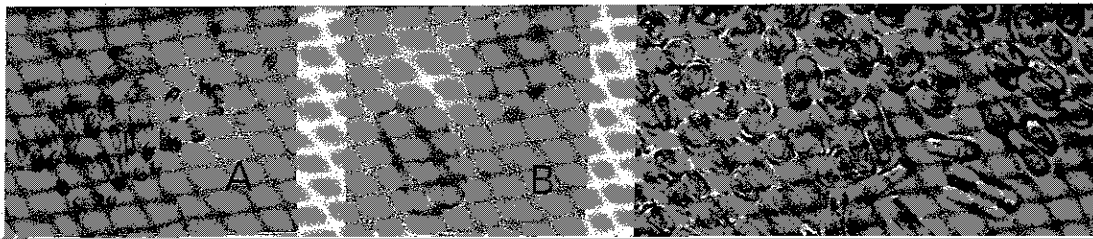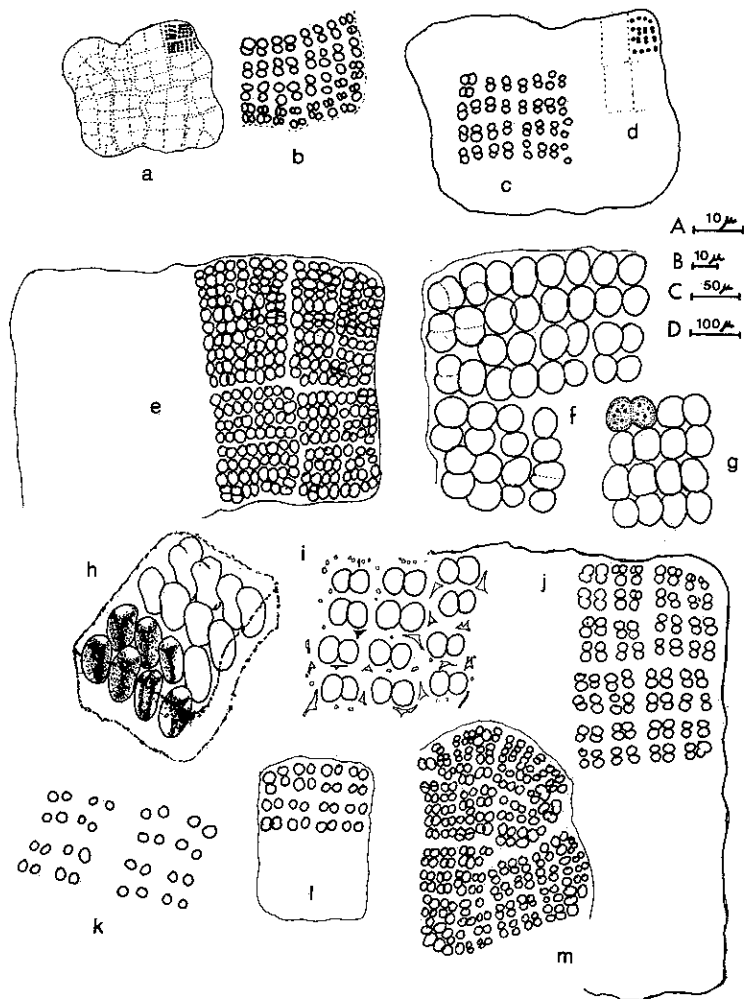
Fig. 6. Some morphological aspects of the algae studied: a) general aspect of an old colony; b) family of cells from a; c) and d) a young colony of families of 4 to 16 cells; f) fringe of a colony in an advanced state (16 to 64 cells per family); g) group of cells belonging to a colony as that represented in f showing a detail of plasmatic granulations h) general morphology of a 16 celled colony (January); i) aspect of crevices between cells in a young collected colony (April); j) corner of a colony with 64 celled families, initial phase (June); k) aspect of a very young colony with clearly 4 celled families (May); l) a 64 celled familys, degaged from an old-colony (September); m) a corner of a very old colony with compact and disordered cells (December).

b) *Cell Characteristics*: The plants studied have pale-green cells, homogenous or delicately granulous in content (Plate 1).

Dimensions very greatly along the year: 4–7 μ in diameter and 6 to 19 μ in length (Fig. 7).

c) *Spatial and Phenological Distribution*: Table 1 shows the density of colonies (N_k) per sample in each station during the year. Averaged values obtained monthly will be used in further calculations. In the same table, the results show total number of cells per sample (N_c) obtained multiplying (N_k) by (C_k) which expresses the mean number of cells per colony (Fig. 10). Temporal changes of these three parameters were compared above (Fig. 4).

(N_k) and (N_c) show a correlation of $r_{0.99} = 0.91$; C_k varies less along the year increasing considerably in winter before the total extinction of colonies in February.

Obviously N_c is a function of N_k and can be expressed by·

$$N_c = 83324.73 \pm 666.29 \, N_k \qquad (1)$$

with a goodness of 0.83.

Seasonal distribution in time shows a lack of *Merismopoedia* in winter and spring; maximal density of colonies was found in Station 2 at the end of summer. The cycle seems to be delayed in time high to low levels (Station 7 in Table 1).

d) *Cell and Colony Phenology*: Size of cells increases in time from February to November (Fig. 5); however, averaged diameters remain more or less constant along the year, with minima in May, August and November after cell division.

Number cells grouped in each family (C_k) varies in time starting from four to 128. In Figure 8, a graphic of bio-modal values of length for each month is presented, this picture is a basis to provide schema of the general picture

Fig. 8. Representation of the two modal classes in the length measures of cells along the year, this graphic provides a basis to support the hypothesis of a general cycle drawn in Figure 10.
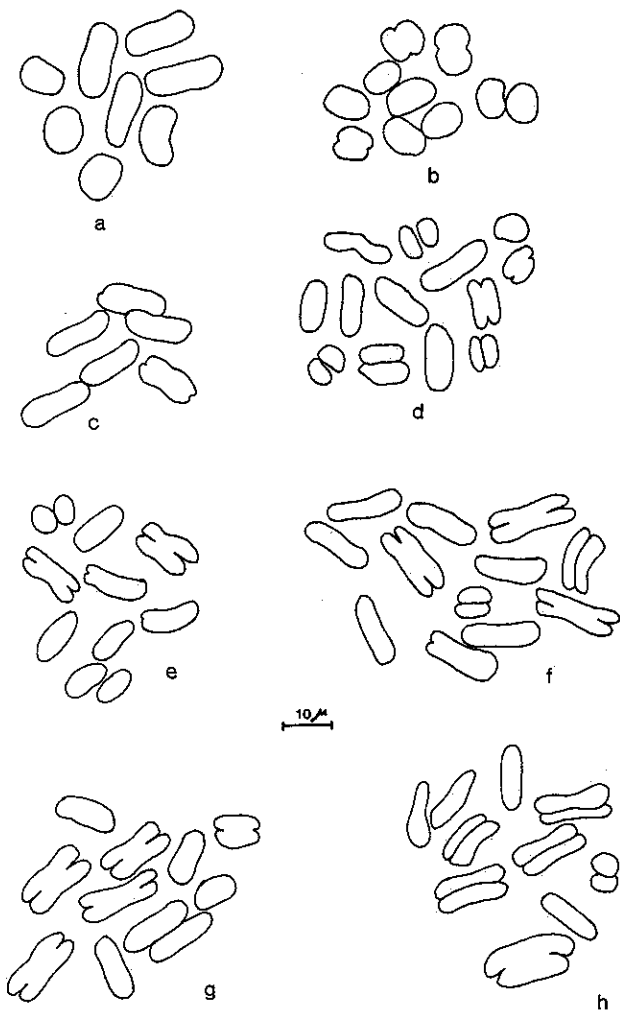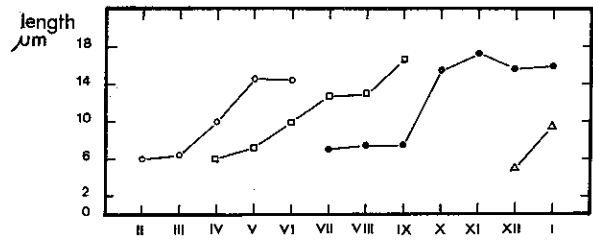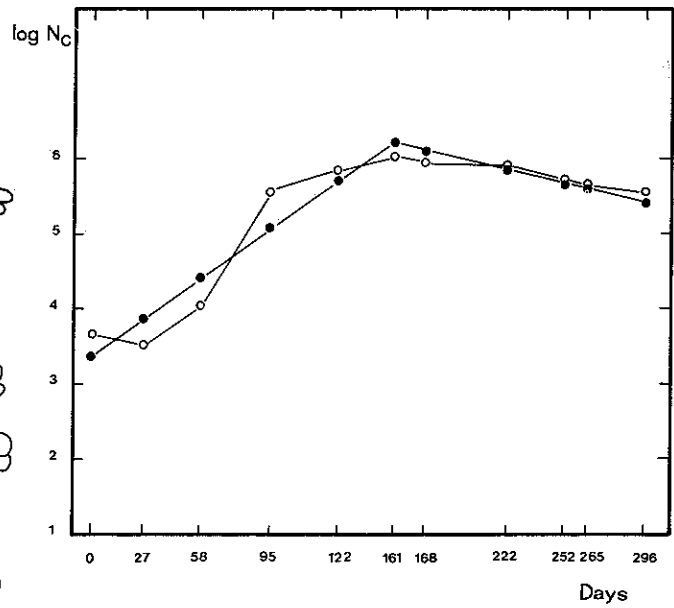

Fig. 7. Schematic figures of cells obtained squashing colonies along the year: a) January; b) April; c) May; d) July; e) August; f) September; g) November; h) December

Fig. 9. Growth values of the population studied in this work. o: obtained values. ●: calculated values.

Tab. 1. Seasonal and spatial density of *Agmenellum*

| Month	1	2	3	4	5	6	7	8	$\overline{X}$	$\sigma\overline{X}$	$s\overline{X}$
						Station					
I	—	—	18	31	—	—	14	332	49,38	107,36	37,96
II	—	—	—	—	—	—	—	—	—	—	—
III	—	—	—	—	—	—	—	48	6,00	15,87	5,61
IV	—	50	—	—	—	—	—	—	6,25	16,54	5,85
V	30	66	16	—	18	—	18	45	23,88	20,76	7,34
VI	123	4671	627	136	38	18	17	—	703,75	1511,83	534,51
VII	617	3001	647	33	150	87	16	1332	735,38	954,25	337,38
VIII	450	7883	882	554	236	210	725	436	1422,00	2451,22	866,64
IX	438	2994	1280	575	248	196	662	575	871,00	860,56	304,26
X	1162	3538	1364	969	240	114	2794	655	1354,00	1134,45	401,09
XI	1808	349	514	44	121	72	1536	732	584,50	533,93	188,77
XII	640	1577	77	135	160	52	330	99	383,75	486,00	171,83
$\overline{X}$	397,33	2010,75	452,08	206,42	100,92	62,42	509,33	354,33	—	—	—
σ	444,16	2390,01	395,53	492,16	303,58	98,78	73,34	822,93	—	—	—
$s\overline{X}$	128,22	689,94	114,18	142,08	87,64	28,52	21,17	237,56	—	—	—
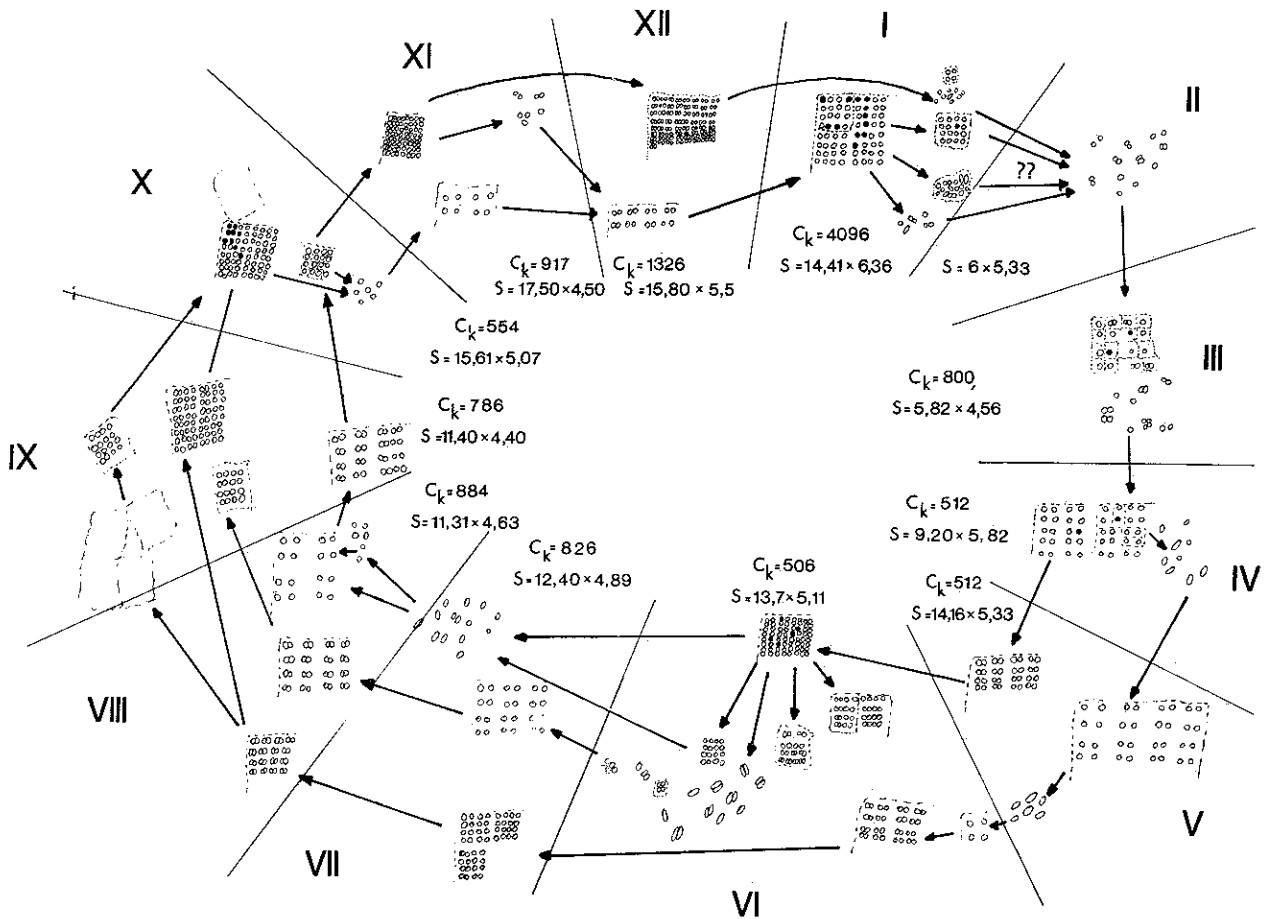
Fig. 10: Hypothetic and schematic cycle of *Agmenellum thermale* along the year C_k = number of cells per colony. S = cell size in μm (length x width).

of the relationships between the different morphological forms (Fig. 10).

e) *Growth and Production*: It is assumed that cellular bipartition and dispersion by breaking were the only systems of effective increasing of the studied *Merismopoedia* population.

The growth of the population can be expressed by

$$N_t = N_o \, e^{\alpha t} \tag{2}$$

were $\alpha = r - m$; r and m are increase and decrease rates of the total cell number. N_o is the initial amount of cells and N_t is the number of cells after a given time; seasonal changes of total cells fits with the equation

$$N_t = 4800 \, e^{0.03t} \tag{3}$$

in growth period, and

$$N_t = 1210.479 \, e^{-0.01t} \tag{4}$$

in the period of losses.

The empirical values show a goodness with the calculated ones of 0.91 for equation (3) and 0.93 for equation (4).

The number of individual cells considered as initial population, could be calculated with $N_k = 0$ in expression (1), then $N_c = 83324.73$.

The growth curve (Fig. 9) presents first a "production time" in which the population increases followed by a plateau until January when colonies dissapear gradually.

Growth curve fits, in this case, with a logistic function in which (k) is the "maximum supportable population density" in given environmental conditions, K can be obtained from the equation

$$\frac{d \, N_t}{d \, t} = (r - cN_t) \, N_t \tag{5}$$

where c is the reduction of growth rate per unit of N.

Doing $c = r/K$ the logistic equation can be rewritten as

$$\frac{d \, N_t}{d \, t} = r \, N \frac{K - N}{K} \tag{6}$$

Best procedure to obtain K is by iterative calculation till the obtention of the best fit between obtained values and a straight line. The equation is

$$\log c \, \frac{K - N}{N} = a - rt \tag{6}$$

In the studied case best K values are of 1.205.000 cells with a goodness of 0.9407.

The mean values of duplication time estimated from expression (2) for r = 0.03 constant were 25.8 days.

Number of divisions necessary to pass from the smallest (C_k = 4) families to the compact ones (C_k = 64) is 4, therefore each generation develops totally in 103.2 days giving rise to 3 generations along the year taking only the period into account in which colonies of *Merismopoedia* were conspicuous.

These generations are easily disjointable (Fig. 8) in function of monthly bimodal distributions of cell lengths.

Values of standing-crop were obtained by cell volume calculations, following Findenegg (1969).

Total cell volume (V_c) can be obtained approaching the cell shape to an ellipsoid.

$$V_c = 4/3 \; \Pi \; ab^2 \; N_c \tag{7}$$

being *a* and *b* the diameter and length of the cell.

Values of total surface (S_c) are, as Paasche (1960) pointed out, a measure of production capacity of the cells. For a supposed ellipsoidal body, total external surface of the population can be calculated by means of the following expression

$$S_c = 2 \; \frac{ab}{2} \; \sin^{-1} \epsilon \tag{8}$$

being ϵ eccentricity of the ellipse that generates the ellipsoid.

The ratio S_c/V_c is equivalent to the P/B quotient. Values concerning external volume surface and their relations are expressed in Table 2.

Total volume (V_c) increased between March and August as N_c did, and decreased between this month and February.

Cell volume changes seasonally being minimal after division time (May, August and November) and at the beginning of the cycle.

Changes of V_c values can be expressed by

$$V_c = 0.324 \; e^{0.039t} \tag{9}$$

Tab. 2. Mean values of volume (V), surface (S), S/V ratio and total standing-crop per sample (V_c).

Month	V(μ^3)	S(μ^2)	S/V	V_c mm^3 10^{-3}
I	2420.26	41435.70	16.98	493.56
II	714.20	12136.48	17.00	–
III	507.07	10432.77	20.58	2.433
IV	1305.71	22312.01	17.10	4.178
V	1685.51	35051.84	20.80	20.607
VI	1505.57	32514.84	21.60	535.558
VII	1291.47	27853.47	21.57	784.064
VIII	1015.89	23806.76	23.45	1135.452
IX	924.75	23032.45	24.93	951.153
X	1681.25	37773.14	22.47	1252.563
XI	1481.29	37951.67	25.63	793.272
XII	1997.70	40860.02	20.46	1052.846

after a given time t, with a goodness of r^2 = 0.914 in growth time, and

$$V_c = 188.929 \; e^{-0.008t} \tag{10}$$

after a given t, during the death time between August and January with a goodnes of r^2 = 0.719.

Relation with Some Ecological Factors

N_c shows a correlation with salinity (S) r = 0.896, air temperature r = 0.797, measured in the collection month. N_c shows also a correlation with salinity (r = 0.668), air temperature (0.91) and incident energy (r = 0.576) when these variables were measured a month before collection. All values are significative at levels of 0.95.

Total volume shows correlation with temperature of water (r = 0.877), and salinity (r = 0.786) measured one month before collection of algae.

Discussion

The most important conclusions arising from the data exposed in this work are taxonomic.

Genus *Merismopoedia* Kütz. includes colonial Cyanophytes with sphaerical or narrow ellipsoidal cells, arranged to families in a gelatinous mass, divisible into two planes perpendicular to the colony. Colonies which are first square or rectangular-shaped become irregular at the edge with time

Fremy (1929–1933) mentioned six species of marine *Merismopoedia*, included also by Geitler in his classification (1932). In these classical works were the first typifying species after different cell-size.

Classification of the Specimens of the Miñor River Estuary

Specimens found in the Miñor River always have cells greater than 4 µm in diameter (Fig. 5) excluding for this reason their relationship to *M. affixa* P. Richt, *M. warmingiana* Lagert., *M. gardneri* (Collins) Setchell and *M. tenuissima* Lemm.

The cell-measures of our specimens makes it possible to classify them among the following species: *M. littoralis* (Oersted) Rab. and *M. convoluta* Breb. in Kütz.

With a wider criteria the genus *Merismopoedia* was included in the genus *Agmenellum* Breb. by Drouet and Daily (1956), dividing it in to two species: *A. quadruplicatum* Breb. and *A. thermale* (Kütz.) Dr. and Daily. Specimens collected in the Miñor estuary belong to *A. thermale* (Kütz.) Drouet and Daily because they never show cells smaller than 3.5 µ.

Most of the specimens collected can be classified as *Merismopoedia convoluta* Breb. but because of their dimensions (4–6 µ of diameter) belong to *M. elegans* A. Br. var. *marina* Lagerh. this latter species measures

(6 x 10–12 μ) and can be considered as a neighbour of *M. convoluta* Breb., the differences between both taxa being trivial

In the general cycle (Fig. 10) it is suggested that much of the 16 (32) celled colonies derive themselves from other ones by disgregarion. In short, *M. elegans* A. Br. var. *marina* Lagerh. in respect to the presented characteristic must be also considered as a part of *M. convoluta* Breb. in Kütz.

We have found some colonies formed of cells longer than 12 μ; this cell size leads to classify them as *M. elegans* A. Br. var. *mandalensis* (= *M. maior* (Smith) Gertter) or *M. littorale* (Oersted) Rabenhorst; but the cell-diameters of 7–10 μ and 7 μ given in the literature for these taxa are too broad to include our specimens.

Following the classical taxonomy the genus *Merismopoedia* Kütz. and *Holopedium* Lagerh. differ in the length width relation of cells. Figure 7 shows how these differences depend on the state of growth and division of cells. In the same colony, cells may differ in size (central and fringe ones for example) as shown in Plate 1 and Figure 7.

Drouet and Daily (1956) include *Holopedium* subgenus *Euholopedium* Forti in the genus *Agmenellum* Breb. and on the basis of a disordered disposion of cells in the colonies give a generic entity to the subgenus *Microcrocis* Forti. We have observed that disposition of cells in the colony also changes in time; so, some specimens collected in January (Fig. 10), had a structure presenting the features of *Microcrocis geminata* Geitler and Engler in Praute.

This relation between *Microcrocis* and *Agmenellum* is suggested by Drouet and Daily (1956) in page 92 of their work: "It is highly probable that plants of this genus (*Microcrocis*) are growth-forms of *Agmenellum thermale*",

these authors point out that Bulnheim in Rabenhorst (1856) and Rabenhorst (1878) has suggested the same relation

Individual cells, as represented schematically in Fig. 10, are often present in samples and they present the same morphological features as those forming colonies (Pl. 1).

The identity of *Coccochloris* Sprengel and *Anacystis* Mengh. with the single cells degaged from "merismopoedia" colonies suggested by Drouet and Dialy (1956) are well supported by data obtained in this work. Other evidences on the same way were given by Stanier *et al.* (1971), who obtained "merismopoedia" shaped plants from an *Aphanocapsa* (*sensu stricto*) culture.

Acknowledgements

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