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VARIABILITY OF SPINE SHAPE AND OF COENOBIA FORMATION IN A STRAIN OF *SCENEDESMUS QUADRICAUDA* (TURPIN) BRÉBISSON

Mit deutscher und kroatischer Zusammenfassung
Sa sadržajem na njemačkom i hrvatskom jeziku

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A strain of *Scenedesmus* isolated from a small pond in the Botanical Garden of Faculty of Sciences, University Zagreb, has been identified as typical *Scenedesmus quadricauda* (Turpin) Brébisson (with single spines on the poles of terminal cells) when grown in an axenic culture. The alga has been cultivated in a simple inorganic medium (pH 6,7 by Sager and Granick 1954) for *Chlamydomonas*, which has been slightly modified by the author (= medium CHLA). The cultures were continuously air-bubbled and exposed to cycles of 14 hours light and 10 hours darkness. Detailed data about culture methods as well as preparations of *Euglena*[1]-culture filtrates will be published elsewhere.

During the experiments about the effects of algal culture filtrates on growth of *Scenedesmus* several morphological features have been changed so that cells resembling *Scenedesmus longus* (with additional spines restricted to the poles of any cells) (plate I, fig. 3, 7, 8), *S. abundans* (with spines on the medium surface of terminal cells) (plate I, fig. 5 and 9) and *S. bijugatus* (lacking spines with ovate cell shape) (plate I, fig. 1 and 6) as well as unicells with or without spines have been observed. Spines of varying length, occasionally very short, resembling those described by Trainor and Roskosky (1967a) for *S. brasiliensis*, often bent and asymmetrical, have also been observed (plate I, fig. 3 and 4). Similar variations have been found in different *Scenedesmus*-isolates in field and

[1] from *Euglena gracilis*, strain »Z« (kindly obtained from Dr. C. Birnboim)

laboratory cultures which could not be readily identified (Trainor (1964a and b), Trainor and Hilton (1963). According to Trainor (1964) a typical *Scenedesmus quadricauda* displays little variability regardless of medium used and growth conditions. Irregularities have been noted only in old cultures.

Acidifying of the medium with HCl to different pH has shown that lowering of the pH to 5,6 — 5 induces the formation of unicells in young cultures and prevents spine formation (plate I, fig. 1 and 6). With time 2 — 8 celled coenobia occur, but spines are lacking as long as the pH does not rise (90 per cent cells lacking spines on the 6th day of growth and 10 per cent with very short spines). Addition of dried *Euglena*-culture filtrate (in the case when *Euglena* has been grown at pH 4,8) to the medium also favours the production of a unicell population (65 per cent unicells on 5th day of growth) for the same reason — lowering the pH of the medium. A high percentage of unicells has been found also in a heavily contaminated culture with bacteria and in cultures grown in liquid medium after inoculation with *Scenedesmus* from old agar cultures. Unicell formations were noted also in other *Scenedesmus* isolates in media containing ammonium ion at high pH (Trainor and Roskosky 1967b), during contamination with bacteria (Trainor 1963) and in soil-water medium (Trainor and Hilton 1963). Overbeck and Stange-Bursche (1965) reported unicell formation in one strain of *S. quadricauda,* which was however not axenic.

Aberrant formations as bent, short, more abundant than normal or partly missing spines occured in normal young cultures from up to 7 per cent. A higher percentage of aberrant forms has been found in very acid (pH under 4) and in old cultures as well as in media with very high (up to 5—10 times higher than normal) salt concentration (see table I). Addition of dried *Euglena*-culture filtrate or growing *Scenedesmus* in *Euglena*-filtrates (autoclaved or not) produced similar irregularities in spine shape and distribution (table I). In nitrogen deficient *Euglena*-filtrates often 2-celled coenobia of *Scenedesmus* occured. The cells revealed very delicate bristles when prepared after the method of Trainor and Burg (1965) and Burg and Trainor (1967) (plate II, fig. 10). Very short spines seen on fig. 5 seem to be the pores in the pectic layer of the cell wall (Bisalputra and Weier 1963, Bisalputra et al. 1964) corresponding to the insertion place for bristles (Burg and Trainor 1967, Trainor and Roskosky 1968).

All observed changes in cell wall formations have been unstable, i. e. they were reversible as typical 4-spined—cells were produced again after washing and inoculation into fresh media.

The results indicate that most morphological changes in spine shape and distribution are dependent upon some unfavourable growth conditions (e. g. too high salt concentration, too low pH of the medium, nutritional deficiencies or presence of some inhibitory substances). Therefore in such cases the cell division is lowered or inhibited. Unicell formation, however, is in no way coupled with any inhibition of cell division the growth being quite normal.

	Medium	Coenobial type and spine formation
1.	CHLA	typical
2.	CHLA acidified with HCl pH 6	typical
3.	CHLA acidified with HCl pH 5 — 5,6	in young cultures unicells, in older cultures spineless colonies
4.	CHLA acidified with HCl pH 3,4	no growth, abnormal colonies
5.	CHLA + *Euglena* grown in dialysis sacks	various: typical, with spines (short, bent or more abundant than normally) as well as spineless
6.	CHLA + *Euglena* filtrate (dialysed)	typical
7.	CHLA + dried *Euglena* filtrate (*Euglena* previously grown at pH 4,8)	in young cultures unicells, in older cultures spineless colonies
8.	CHLA + dried *Euglena* filtrate (*Euglena* previously grown at pH 6,7)	various: typical, with spines (short, bent or more abundant than normally) as well as spineless
9.	*Euglena* filtrate autoclaved 10 min. at 120° C	various: typical, with spines (short, bent or more abundant than normally) as well as spineless
10.	*Euglena* filtrate not heated	various: typical, with spines (short, bent or more abundant than normally) as well as spineless
11.	Old culture	various: typical, with spines (short, bent or more abundant than normally) as well as spineless
12.	CHLA salts concentrated 5× higher than normally	various: typical (rarely), often with spines (short, bent, more abundant than normally) as well as spineless
13.	CHLA salts concentrated 10× higher than normally	abnormal colonies, growth much inhibited

Table I. Changes in spine formation and coenobial type of *Scenedesmus quadricauda* under different growth conditions.

SUMMARY

Scenedesmus quadricauda, isolated from a pool (Botanical Garden, Zagreb) has been grown in axenic culture. Changes in spine distribution and formation of unicells have been studied. Transitory irregularities in spine shape and distribution have been found in old cultures, in media with high salt concentration and after cultivation in *Euglena*-culture filtrates. Unicell formation as well as spineless forms could be induced by lowering the pH of the medium to about 5 — 5,6.

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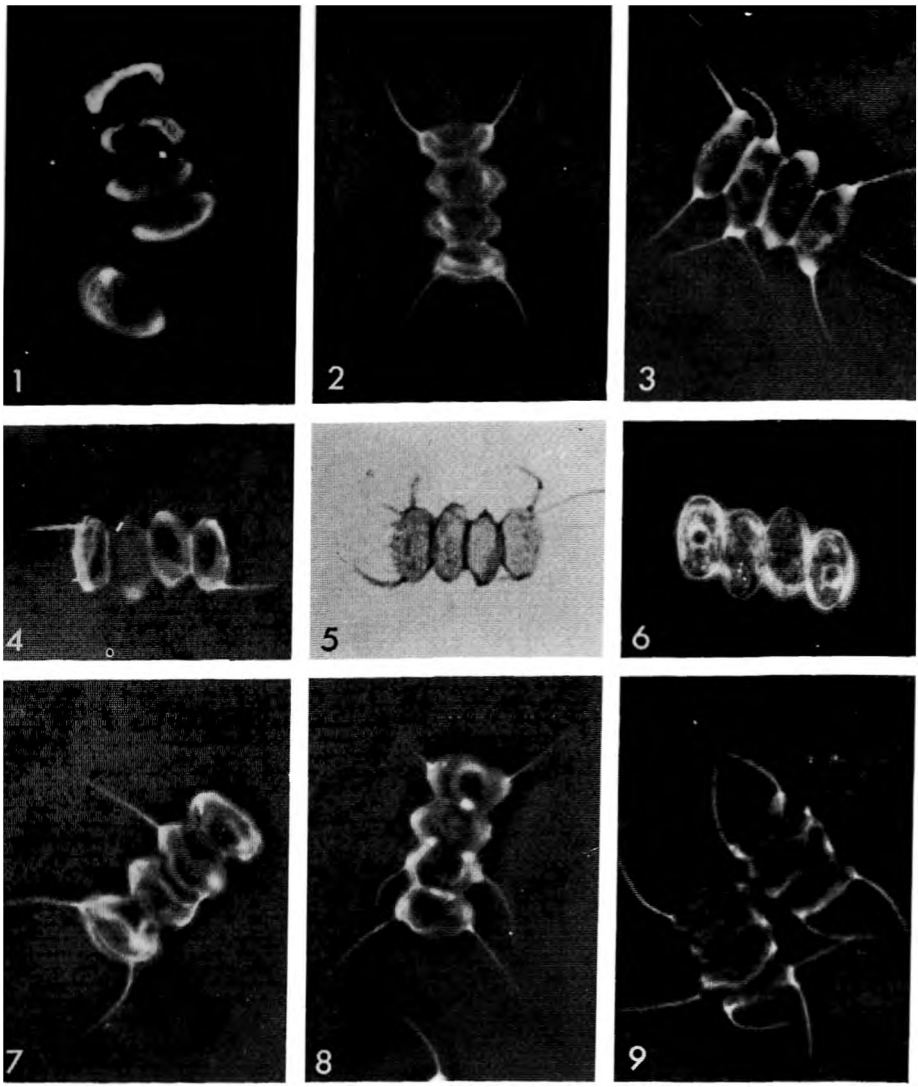

Plate I. Coenobia of *Scenedesmus quadricauda* in various media; dessicated cells (except in Fig. 6 which has been photographed living). Anoptral-contrast Fig. 1 — 4 and 7 — 9, Fig. 5 bright field illumination. Fig. 6 dark field illumination. 1200 ×

Fig. 1. Four-celled coenobium lacking spines and an unicell observed at at pH 5,5 (after 5 days of growth).

Fig. 2. Typical coenobium.

Fig. 3. 4. 5. 7, 8, 9. Four-celled coenobia with abnormal spine shape and distribution.

Fig. 6 As in Fig. 1 but living.

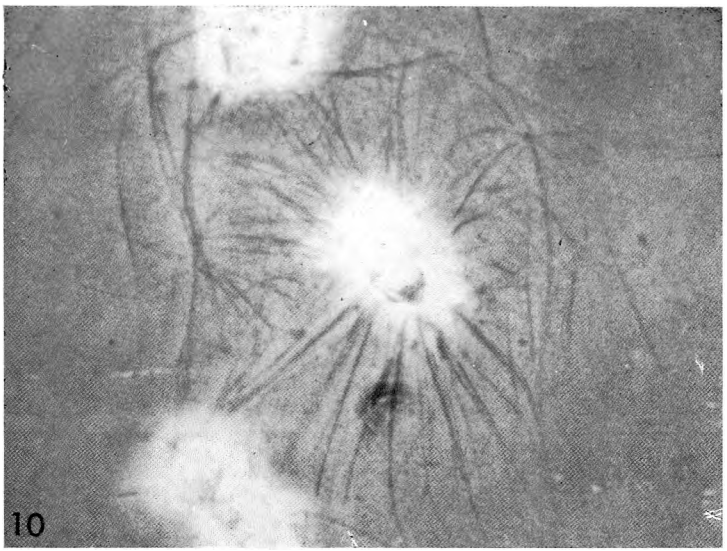

Plate II. Fig. 10. Dessicated preparation showing bristle formation. 900 x.

ZUSAMMENFASSUNG

VARIABILITÄT DER STACHELFORM UND DER COENOBIENBILDUNG EINES STAMMES VON *SCENEDESMUS QUADRICAUDA* (TURPIN) BRÉBISSON

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Scenedesmus quadricauda wurde aus einem Becken des Botanischen Gartens in Zagreb isoliert und bakterienfrei kultiviert. Coenobienformwechsel und Einzellen-Bildung wurden studiert. Vorübergehende Anomalien in Form und Verteilung der Stacheln konnten in alten Kulturen, in Nährlösungen mit hoher Salzkonzentration und nach Kultivieren in Eugenakultur-Filtraten beobachtet werden. Einzellige sowie stachelfreie Formen konnten besonders durch Herabsetzen des pH des Mediums auf 5 — 5,6 induziert werden.

SADRŽAJ

VARIJABILNOST OBLIKA BODLJI I STVARANJA CENOBIJA U SOJA VRSTE *SCENEDESMUS QUADRICAUDA* (TURPIN) BRÉBISSON

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Scenedesmus quadricauda izoliran iz bazena Botaničkog vrta u Zagrebu uzgojen je u čistoj kulturi. Proučavane su promjene oblika i položaja bodlji te stvaranje jednostaničnih populacija. Prolazne nepravilnosti u obliku i rasporedu bodlji primijećene su kod rasta u starim kulturama, u hranidbenim otopinama s visokom koncentracijom soli kao i kod rasta u filtratima kulture *Euglene*. Jednostanične populacije kao i oblici bez bodlji pojavile su se naročito kod sniženja pH medija na 5—5,6.