

**THE EFFECTS OF CULTURE FILTRATE  
ON SPORULATION OF THE GREEN ALGA  
*STIGEOCLONIUM PASCHERI*  
(VISCHER) COX AND BOLD**

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CULTURE filtrate from the blue-green alga *Cylindrospermum licheniforme* was shown to greatly stimulate sporulation in the same alga<sup>1</sup>. Differentiation of vegetative cells into sexual cells by treating the former with culture filtrate from cultures of male colonies of *Volvox carteri* was also demonstrated<sup>2,3</sup>. Dialyzable substances were detected in the culture filtrate of *Eudorina* sp., which could induce sperm packet formation<sup>4</sup>. In the present study effects of culture filtrate of the green alga *Stigeoclonium pascheri* on its sporulation are reported.

*Stigeoclonium pascheri* (Vischer) Cox and Bold, isolated from a fresh water pond at Sarnath, Varanasi, was grown in Bold's basal medium (BBM) (Cox and Bold)<sup>5</sup> at  $22 \pm 1^\circ\text{C}$ , illuminated with cool-white fluorescent tubes at an intensity of 2000 lux for 16 hr a day. The mode of reproduction of alga was observed to be through the formation of akinetes hereafter called as 'spores' which commence to appear in 30-day old cultures. The initiation of sporulation of cells is evident by the (i) changes in colour of filaments from deep green to light yellowish orange, (ii) constriction of cells at partition wall and consequently appearing barrel-shaped, and (iii) increase in the breadth of cells with their individual length more or less remaining constant. Another 30 days were required from the time of initiation to the maturation of a spore. Thus 60 days were necessary for its formation and maturation.

Culture filtrate was obtained as the centrifugal supernatant fluid from 35-day old cultures or 120-day old cultures, which is designated as conditioned

BBM. BBM and conditioned BBM were inoculated with the algal material from 7-day old or 25-day-old stock cultures, and were cultured in the same culture conditions as were the stock cultures. In some experiments, conditioned BBM was supplemented with all the constituents of BBM. Average percentage sporulation determined after 60 days of inoculation was the mean of three replicates from each of which about 70-100 sporulated filaments, each filament containing about 50-100 or occasionally more cells were handled.

In blue-green algae, *Cylindrospermum licheniforme*<sup>1</sup> and *Nodularia spumigena*<sup>6</sup>, culture filtrate from sporulating cultures induced vegetative cells to differentiate into spores, while, such an effect was not observed in blue-green algae *Aphanizomenon flos-aquae*<sup>7</sup> and *Nostoc* PCC 7524<sup>8</sup>. In the present study a gradual decrease of percentage sporulation of *S. pascheri* in 35-day and 120-day old conditioned BBM as compared to standard BBM was observed (Table I) indicating that the filaments of the alga release some toxic substance into the medium which is inhibitory to spore formation on reinoculation. A sporulation-inhibitory substance from vegetative thalli of the green alga *Ulva mutabilis* has also been reported by Nilsen and Nordby<sup>9</sup>. In the present study the toxicity of conditioned BBM increases with the age of filaments growing in the medium. Thus 120-day old conditioned BBM was more toxic to inhibit sporulation than 35-day old conditioned BBM. The vegetative cells from 25-day old stock culture seem to be more differentiated leading to sporulation than those in 7-day old stock culture and therefore, the former shows higher percentage sporulation in either of 35- or 120-day old conditioned BBM than the latter (Table I). Supplementation of conditioned BBM with all the constituents of BBM produced no significant change on the extent of percentage sporulation which results from conditioned BBM (Table I). Supplemented and conditioned BBM contains all the components of BBM in concentration equal to or greater than their concentration in BBM. The decrease in percentage sporulation as

TABLE I  
Effects of supplementing conditioned medium with components of fresh sporulation medium  
on the extent of sporulation of *S. pascheri*

% sporulation	Standard sporulating medium (BBM)	Conditioned BBM		Supplemented conditioned BBM	
		35 day	120 day	35 day	120 day
7-day old stock culture	63.5 $\pm$ 7.1	6.8 $\pm$ 1.2	1.5 $\pm$ 0.4	6.2 $\pm$ 1.8	2.8 $\pm$ 0.6
25-day old stock culture	65.7 $\pm$ 5.9	31.7 $\pm$ 4.9	4.6 $\pm$ 1.0	38.0 $\pm$ 5.3	4.9 $\pm$ 0.5

effected by conditioned BBM was not, therefore, the result of a depletion of the medium rather, it is reasonable to conclude that the filaments produced some toxic substances which were released into the medium which were responsible for decreased percentage sporulation of newly inoculated filaments.

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#### A NEW SPECIES OF SCENEDESMUS— *S. SERRATO-PERFORATUS* SP. NOV.

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DURING the study of Chlorococcales of Gujarat, the authors collected a new species of *Scenedesmus*, *S. serrato-perforatus* sp. nov. from a pond at Harni, near Baroda in 1970<sup>1-2</sup>. It has been described below.

*Scenedesmus serrato-perforatus* Patel et Isabella sp. nov. (Figs. 1-2)

Colonia 8-cellularis. Cellulae subquadratae, extremitatibus capitatis. Cellulae in serie signulati, raro, autem, irregulari ordinate. Latus exterius cellularum terminalium aliquantulum convexum rectumve, latus interius concavum. Cellulae interiores perforationes lineares lenticulares inter cellulas contiguas praebentes. Omnis cellula 1-3 series medias longitudinales denticulationum habens. Policellularum 1-4 dentibus parvis praediti. Cellulae 3.2-5.9  $\mu\text{m}$  lat., 10.6-

14.7  $\mu\text{m}$  long. Perforationes 0.3-1.2  $\mu\text{m}$  lat. Dentibus 0.3-1.3  $\mu\text{m}$  long. Colonia 8-cellularis 30  $\mu\text{m}$  long.

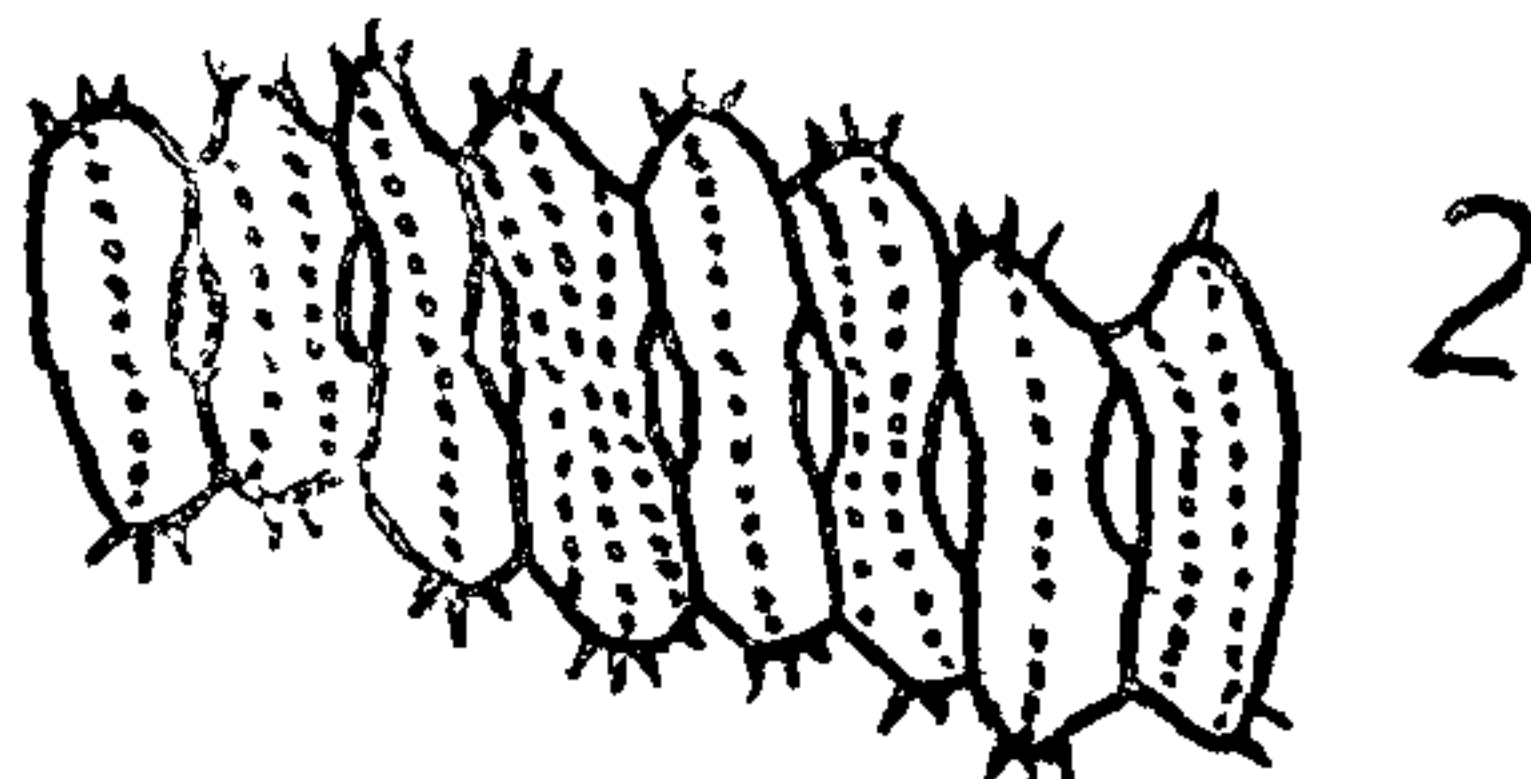
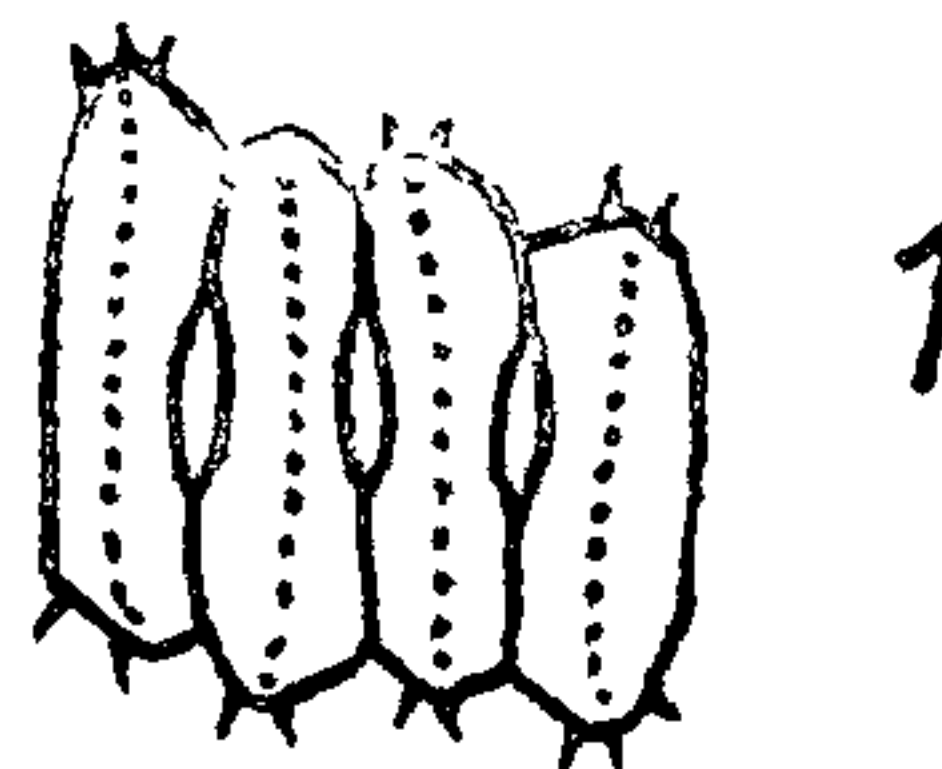
Habitatio: Stagnum Harni dictum in leco Harni near Baroda dicto, m. Aug. d. 5, 1970 (Nr. 1845).

*Scenedesmus serrato-perforatus* Patel et Isabella sp. nov. (Figs. 1-2)

Colony 8-celled. Cells subquadratae with capitate ends. Cells arranged in single series or rarely in an irregular series. Outer side of the terminal cells slightly convex or straight, inner side concave. Internal cells with linear lenticular perforations between the adjacent cells. All the cells with 1-3 median longitudinal row of denticulations. Poles of the cells with 1-4 small teeth. Cells 3.2-5.9  $\mu\text{m}$  broad, 10.6-14.7  $\mu\text{m}$  long. Perforations 0.3-1.2  $\mu\text{m}$  broad. Teeth 0.3-1.3  $\mu\text{m}$  long. 8-celled colony 30  $\mu\text{m}$  long.

Habitat: Harni pond, Harni near Baroda, 5-8-1970 (No. 1845).

As far as the authors are aware, no perforate form of *S. serratus* is known to which the present form is agreeable in general characters. This new alga bears the same relation to *S. serratus* as *S. perforatus* bears to *S. quadricauda* and *S. balatonicus* Hortobagyi to *S. bijugatus*<sup>1</sup>.



20  $\mu\text{m}$

FIGS. 1-2. *Scenedesmus serrato-perforatus* Patel et Isabella sp. nov.