The Occurrence of a Fungal Parasite on a *Tetraselmis* (*Prasinophyceae*) Species

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Abstract

The occurrence of a chytridial type of parasite on a *Tetraselmis* species during the dying-off stage of an algal bloom is examined. The main ultrastructural features in identifying the alga, such as the scales and hairs on the four flagella, as well as the situation of the four flagella are described. The infection of the algal host cells by the parasites, their reactions and final ultrastructural collapse are discussed.

Introduction

Algal blooms develop annually from August to early September in the lower parts of the Vaal River Barrage. These blooms usually last for two to three weeks after which they decline rapidly. The algae responsible for the blooms were found to be almost a monoculture of a *Tetraselmis* (Stein) (*Platymonas* (West)) species together with occasional *Pediastrum* species.

Microscopic investigations of the algae in the dying-off stage of the bloom show that most of them were parasitized by a chytridial type of parasite. Parasitic fungi of the subdivision Mastigomycotina have previously been recorded in fresh water algae (Schnepf, Hegewald and Soeder, 1971; Schnepf, Deichgräber, Hegewald and Soeder, 1971; Schnepf, 1972) and in marine algae (Kazama and Fuller, 1970; Kazama, 1972). The fungal parasites caused certain ultrastructural changes in the host algae.

Since no ultrastructural studies of a chytridial type of fungus on *Tetraselmis* have been done, the development of the parasite-host contact, as well as the infection of the host cell and its reaction were investigated. This study examines some of the cytological features of the chytrid parasite and *Tetraselmis* host as well as some of the host parasite interactions.

Materials and Methods

The material was fixed for 90 min at room temperature in 6% glutaraldehyde in 0,05 M sodium cacodylate buffer, pH 7,0. After rinsing in three changes of buffer the material was post-fixed for 90 min in 2% OsO_4 in 0,05 M sodium cacodylate buffer, pH 7,0. Dehydration was carried out in a graded ethyl alcohol series and the material embedded in a Maraglas/Cardolite resin mixture. Sections were stained with uranyl acetate and lead citrate (Reynolds, 1963), and observed with a Siemens 101 electron microscope.

Results

The alga was identified as *Tetraselmis* (*Platymonas*) using keys by Thompson (1959) and Butcher (1959). The identification was confirmed by the ultrastructure of the organism (Manton and Parke, 1965) and Manton (personal communication). Although the ultrastructure of *Tetraselmis* is described by Manton and Parke (1965), it is important to describe certain structural characteristics for identification purposes.

The four flagella (Figs. 1 and 2; F) are situated in a trough-like depression and are covered with scales and hairs (Fig. 3; S, HA). The single pyrenoid (Figs. 1 and 4; P) is situated in the cup shaped chloroplast (Figs. 1 and 4; C). Starch grains (Figs. 1 and 4; SP) surround the homogeneous protein core. Branched cytoplasmic invaginations (Fig. 4; CI) are present in the pyrenoid. The eyespot (Fig. 5; E) consists of two layers of globules separated by a membrane sac (Fig. 5; MS).

Preliminary identification of the parasitic fungus was done with the key of Sparrow (1960) and is probably a *Chytridium* species.

The fungus (Fig. 6; FU) is characterized by the presence of lipid globules (Fig. 6, 7; L). Typical cell structures like the nucleus (Fig. 7; N), ribosomes (Fig. 7; R), mitochondria (Fig. 7; M), endoplasmic reticulum (Fig. 7; ER), dictyosomes (Fig. 7; D) and plasmalemma (Fig. 7; PM) are present. The cell wall is 35 – 40 nm thick and a number of small vacuoles (Fig. 7; V) are present.

With penetration of the host cell wall by the parasite haustorium and further development of the haustorium, a part of the parasite cytoplasm moves into the haustorium. The actual way in which the parasite attaches to the host, as well as the way in which the haustorium penetrates, could not be ascertained in this investigation.

Additional cell wall material, such as found in *Scenedesmus* (Schnepf, Hegewald, *et al.*, 1971), was not formed by the host cell around the haustorium (Figs. 8,9 and 10; H). The haustorium is enclosed, just as in the case of *Scenedesmus* (Schnepf, Deichgräber, *et al.*, 1971), by a haustorium wall. The host plasmalemma surrounding the haustorium is usually not well defined. Where the haustorium penetrates the host chloroplast the plasmalemma can be clearly seen (Fig. 9; PM). This corresponds with the findings of Schnepf, Deichgräber, *et al.* (1971), in *Chytridium* on *Scenedesmus* where some host cytoplasm is always present between the haustorium and the host chloroplast. The haustorium contains a great number of ribosomes (Figs. 8 and 9; R). Endoplasmic reticulum may sometimes be present and the tips of the haustoria are often filled with lamellae and vesicles (Fig. 10; LM). It could not be ascertained whether these are typical wall vesicles, originating from the dictyosomes.

The host cell can be penetrated by more than one haustorium from one or more parasites. The haustoria can also branch, so that a cross section often shows more than one haustorium profile (Figs. 10, 11 and 12; H). Further development of the parasite and haustorium system leads to degeneration of the host protoplasm (Figs. 11 and 12). A marked decrease in the cytoplasmic content of the host cell follows and although

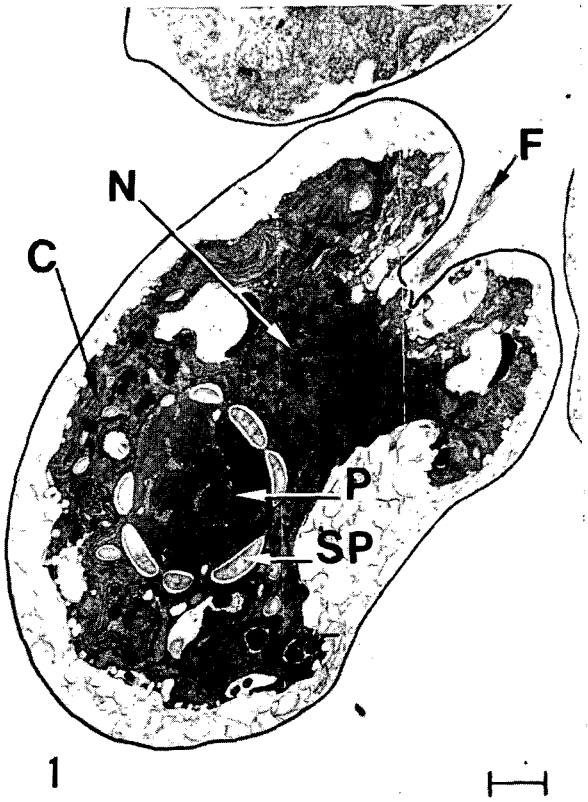

Figure 1
Median longitudinal section of ***Tetraselmis.***

Figure 2
Cross-section of ***Tetraselmis*** *to show the four flagella (F).*

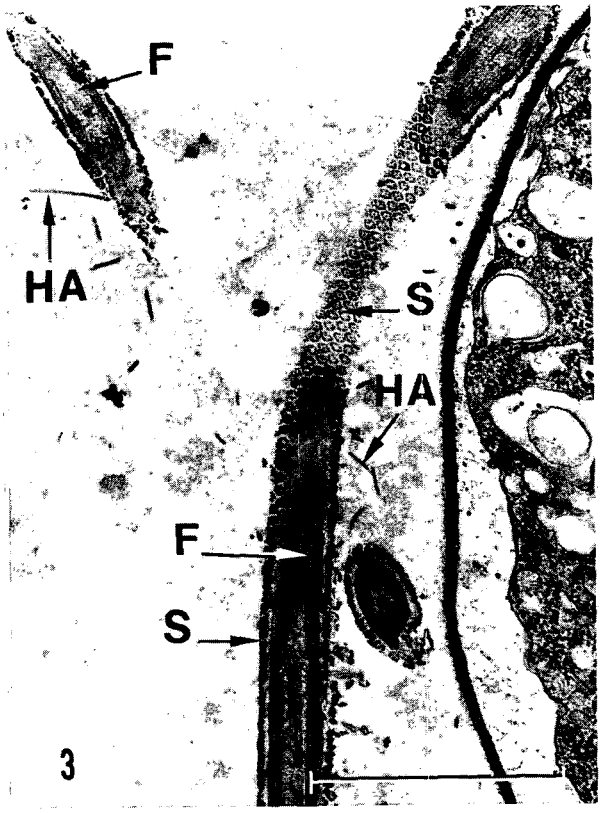

Figure 3
Longitudinal section of flagella (F).

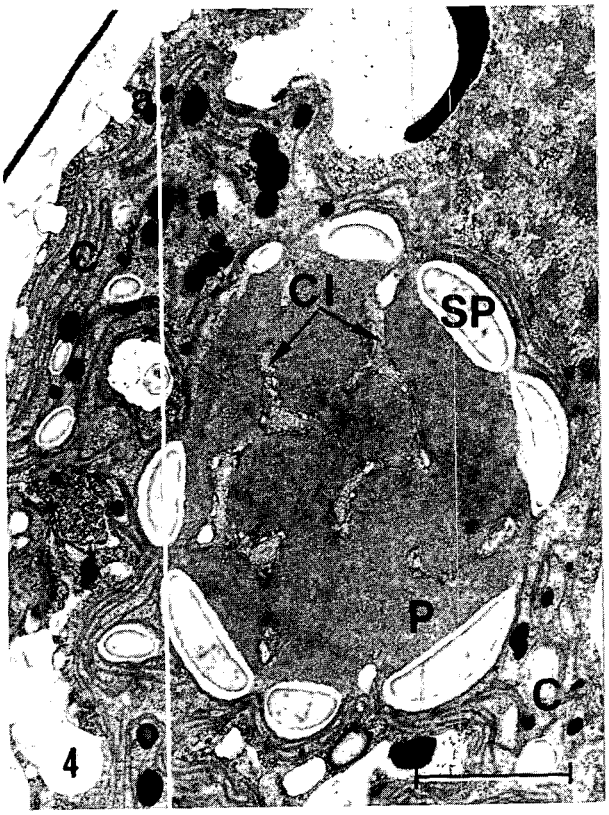

Figure 4
Cross-section of ***Tetraselmis.***

(C = chloroplast, CI = cytoplasmic invagination, F = flagellum, HA = hairs, N = nucleus, P = pyrenoid, S = scales, SP = starch. Marker = 1 μm.)

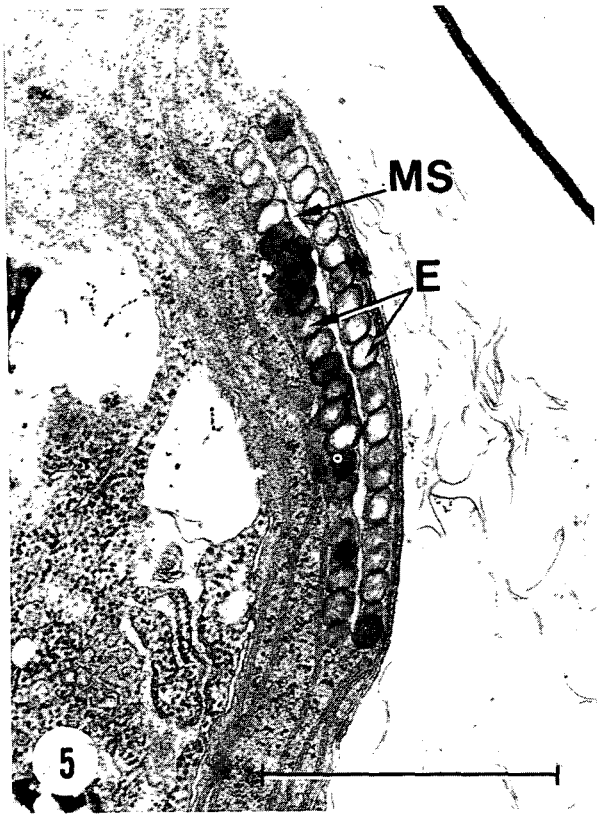

Figure 5
Longitudinal section of ***Tetraselmis.***

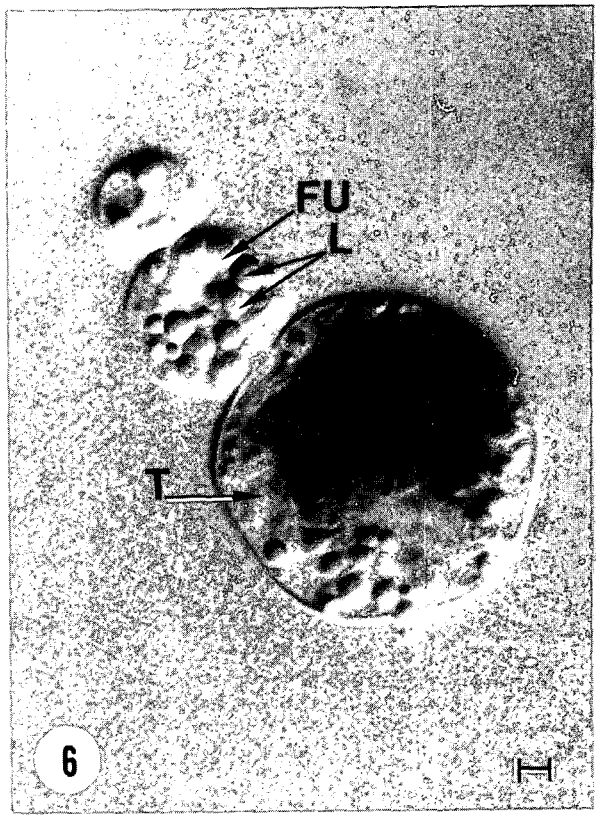

Figure 6
Tetraselmis *host (T) with fungal parasite (FU).*

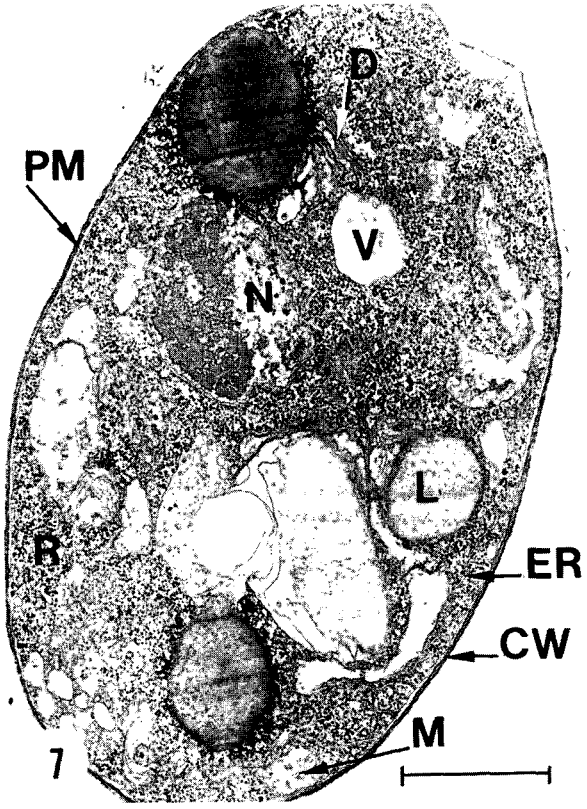

Figure 7
Cross-section of fungus.

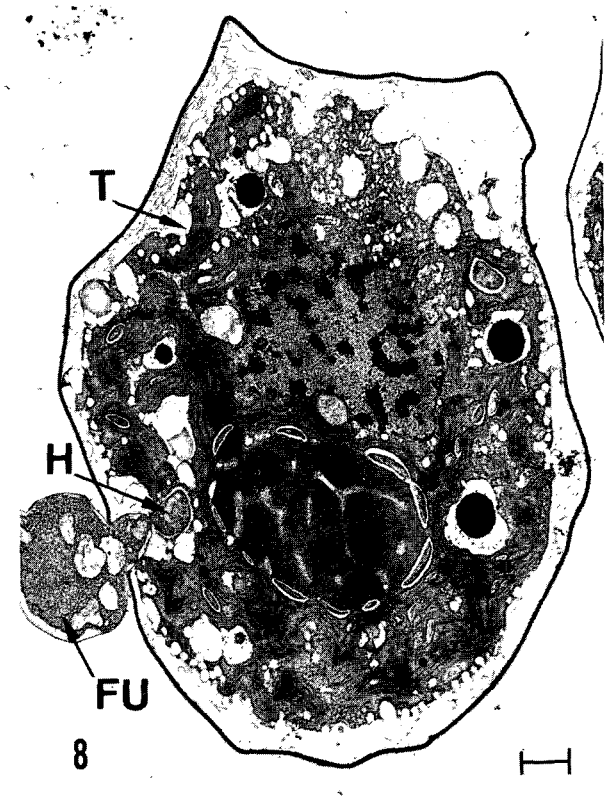

Figure 8
Section to show the host-parasite relationship.

(CW = cell wall, D = dictyosome, E = eyespot, ER = endoplasmic reticulum, FU = fungal parasite, H = haustorium, L = lipid globule, R = ribosomes, T = ***Tetraselmis,*** *V = vacuole. Marker = 1 μm.)*

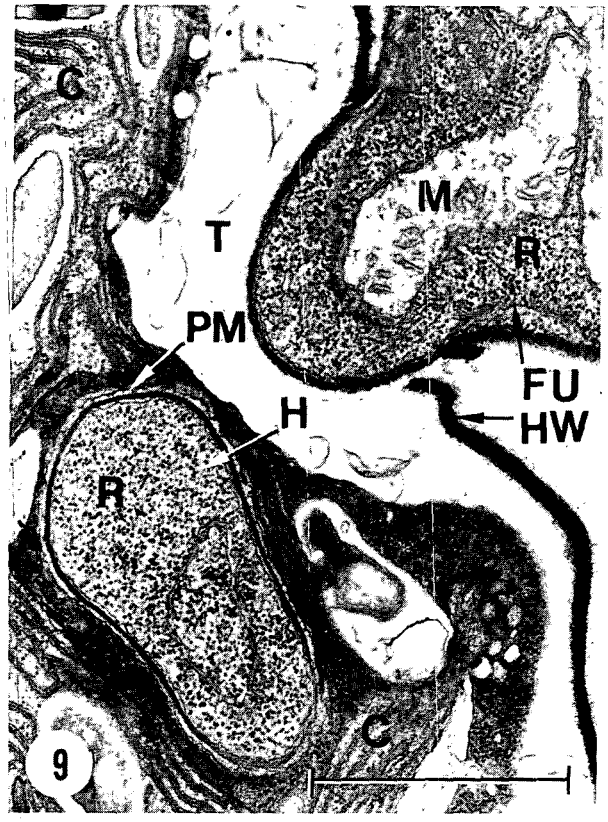

Figure 9
Section of host (T) and parasite (FU).

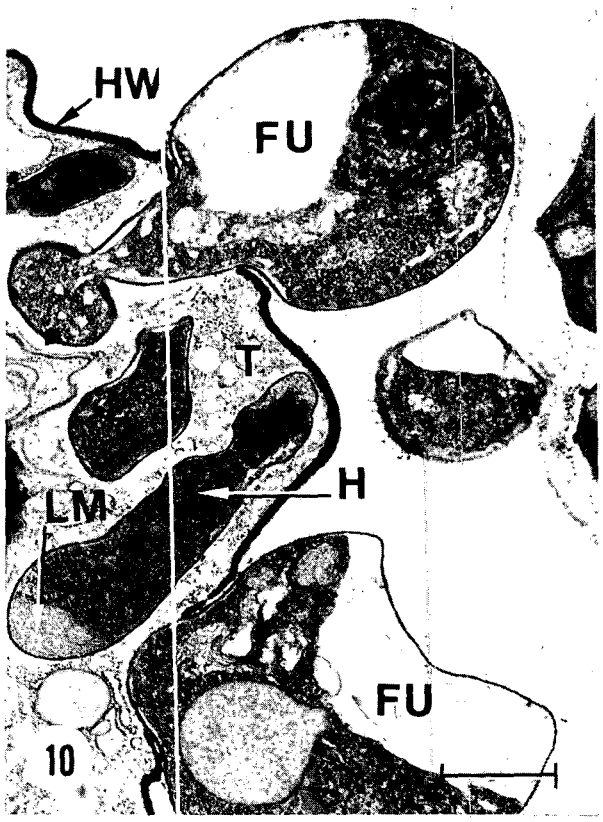

Figure 10
Section of host (T) and parasite (FU).

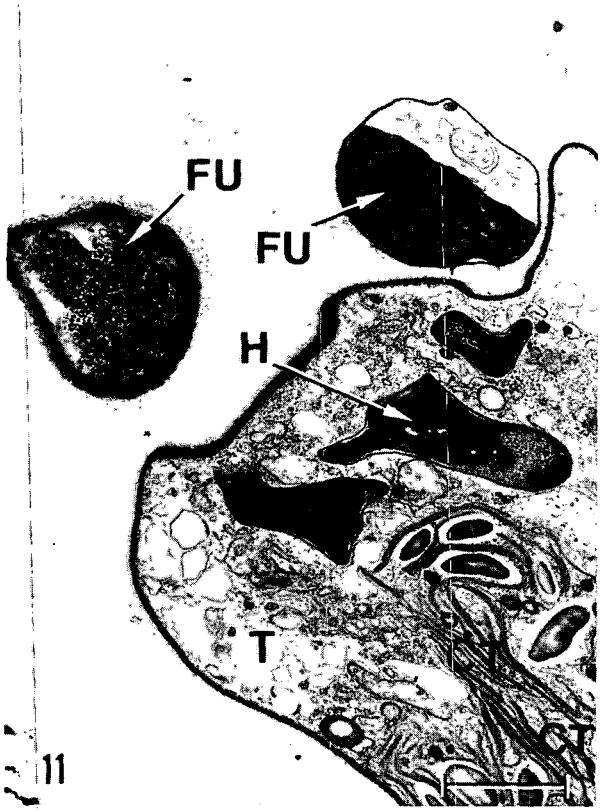

Figure 11
Section of host (H) in advanced stage of cytoplasmic degeneration.

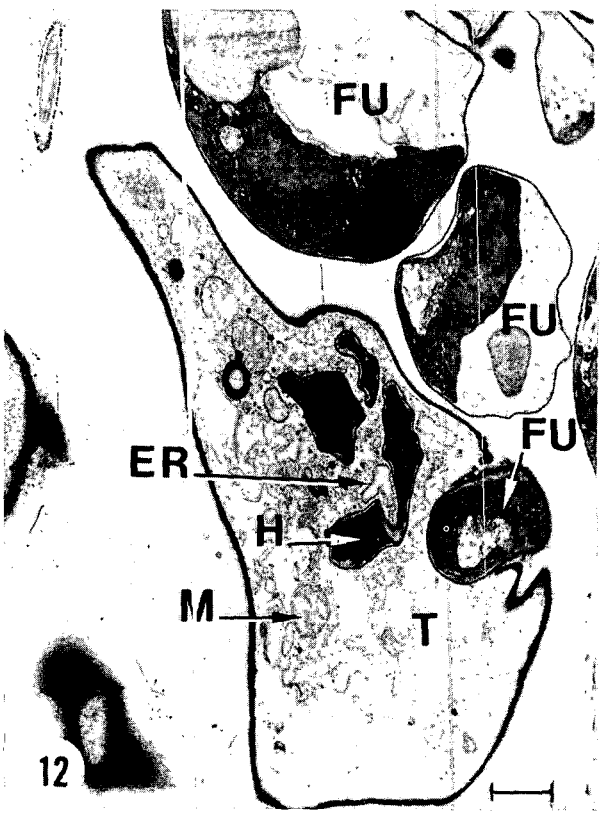

Figure 12
Section of host (T) in final stages of cytoplasmic degeneration.

*(C = chloroplast, CT = chloroplast thylakoids, ER = endoplasmic reticulum, FU = fungal parasite, H = haustorium, HW = host cell wall, LM = lamellae, M = mitochondrion, PM = host plasmalemma, R = ribosomes, T = **Tetraselmis** host. Marker = 1 μm.)*

chloroplast thylakoids can still be observed for quite a while (Fig. 11; CT), they also disappear until finally only a few mitochondria (Fig. 12; M), rough endoplasmic reticulum (Fig. 12; ER), and other membrane elements can be seen.

No zoosponrangial development of the fungal parasite could be observed.

Discussion

The structure of the fungal parasite of *Tetraselmis* corresponds with that of the *Chytridium* sp. described by Schnepf, Deichgräber, *et al.*, (1971). This applies especially to the structure of the encysted zoospore and the penetration and structure of the haustorium. The absence of the forming of additional wall material by the *Tetraselmis* host around the haustorium is the only real difference between this and the *Chytridium* parasite of *Scenedesmus* as described by Schnepf, Deichgräber, *et al.*, (1971), and may further support the idea that the parasite on *Tetraselmis* may be a chytrid.

The role of the parasite in controlling the bloom of *Tetraselmis* could not be assessed. However, if one takes into account that the parasitizing of the host cells invariably leads to destruction of the host protoplasm and the dying-off of the cells it can be accepted that the parasite does play a role in limiting the bloom.

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