

NOTE

Marine virioplankton produced by infected *Ectocarpus siliculosus* (Phaeophyceae)

Dieter G. Müller

Fakultät für Biologie der Universität, W-7750 Konstanz, Germany

ABSTRACT: The benthic marine brown alga *Ectocarpus siliculosus* can be infected by a virus which is expressed in reproductive organs of the host. In cultures, liberation of virus particles was stimulated by addition of fresh culture medium together with a rise in temperature, suggesting the incoming tide as a triggering factor in the natural habitat.

Ultra-small particles between 20 and 200 nm diameter suspended in aquatic habitats are referred to as femto-plankton (Sieburth et al. 1978). Direct counts in coastal and offshore waters yield high numbers of up to 10^8 particles ml^{-1} (Bergh et al. 1989). Proctor & Fuhrman (1990) found significant numbers of phage-infected bacteria and cyanophytes, which therefore can be a potential source of marine planktonic viruses. While Børsheim et al. (1990) considered viruses from eukaryotic hosts to be of minor significance, Suttle et al. (1990) found a strong impact of virus samples from natural habitats on the vitality of laboratory cultures of unicellular eukaryotic phytoplankton species such as diatoms, cryptophytes and prasinophytes.

The benthic brown alga *Ectocarpus siliculosus* has been shown to act as a host for a marine virus (Müller et al. 1990). Laboratory cultures were used to study virus release and to estimate the productivity of this system.

Material and methods. Virus-infected gametophytes of *Ectocarpus siliculosus* (Dillw.) Lyngb. isolated from the New Zealand coast (Müller 1991) were used to generate a sporophyte strain with rich and stable virus expression. Bacteria-free stock cultures were maintained in Petri dishes on Provasoli ES-medium solidified with 1% agar. Four to 6 wk after transfer to sterile liquid medium the *E. siliculosus* plants reached maturity. Release of virus clouds was observed in microscopic mounts under dark field illumination on a Leitz dissecting scope at $63\times$ magnification. Size of virus particles and their number per sporangium compartment were derived from electron micrographs (Figs. 1 & 3).

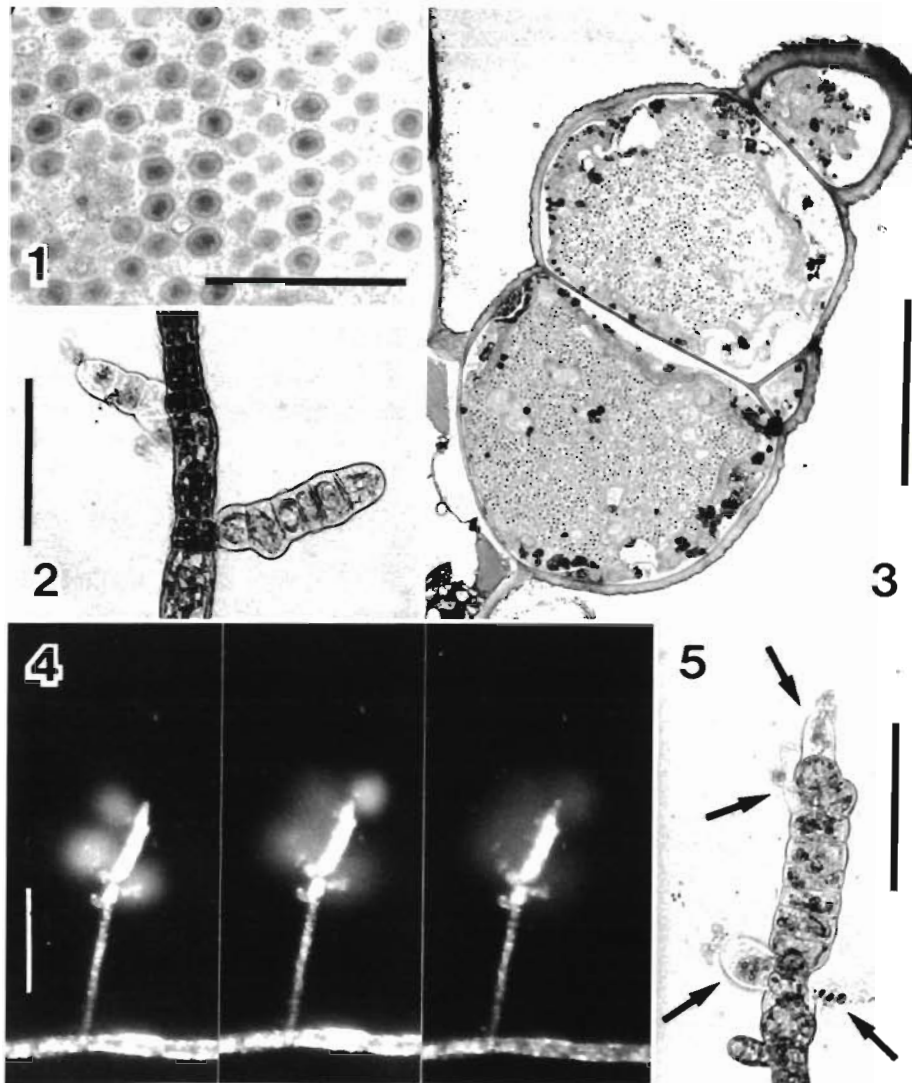
Observations. A typical infected *Ectocarpus siliculosus* sporophyte of a few cm in length forms several hundred non-functional sporangia (Fig. 2). Each compartment in such a sporangium contains an average of 2×10^6 virus particles. Quantitative measurements gave values between 1.7 and 3.2×10^{13} virus particles per g fresh weight of infected algae. Virus liberation was stimulated by transferring the host plants to fresh culture medium and raising temperature from 12 to 20 °C. Sporangial walls burst, and masses of virus particles were discharged into the surrounding seawater (Figs. 4 & 5). They showed intense Brownian movement and fanned out to plumes, which gradually dispersed and became invisible after 10 to 15 min.

The chemical and temperature stimuli inducing virus release also synchronize discharge of gametes and zoospores in healthy *Ectocarpus siliculosus* plants. These naked, free-swimming cells are attacked by the virus and used as entrance sites into a new host (Müller 1991). Since *E. siliculosus* grows in the intertidal zone, the stimulus of re-immersion with the rising tide is likely to trigger virus discharge, zoid release and infection processes in the field.

The laboratory observations described here demonstrate for the first time the existence of a potent eukaryotic virus production system in the marine coastal environment. Further studies in the natural habitat are needed to evaluate its ecological significance.

LITERATURE CITED

- Bergh, O., Børsheim, K. Y., Bratbak, G., Heldal, M. (1989). High abundance of viruses found in aquatic environments. *Nature*, Lond. 340: 467–468
- Børsheim, K. Y., Bratbak, G., Heldal, M. (1990). Enumeration and biomass estimation of planktonic bacteria and viruses by transmission electron microscopy. *Appl. envir. Microbiol.* 56: 352–356



Figs. 1 to 5. *Ectocarpus siliculosus*. **Fig. 1.** EM thin section showing virus particles in the sporangium of an infected plant. Scale bar = 1 μ m. **Fig. 2.** Sporangia of an infected host with intact and burst virus compartments. Scale bar = 100 μ m. **Fig. 3.** Longitudinal EM section through a sporangium with virus expression. Scale bar = 10 μ m. (From Müller 1991.) **Fig. 4.** Virus masses emerging from bursting compartments of a sporangium. Successive dark field exposures 6, 12 and 19 min after mounting. Scale bar = 200 μ m. **Fig. 5.** Same sporangium at 16 min in bright field. Arrows indicate remnants of cells burst in Fig. 4. Scale bar = 100 μ m

- Müller, D. G. (1991). Mendelian segregation of a virus genome during host meiosis in the marine brown alga *Ectocarpus siliculosus*. *J. Plant Physiol.* 137: 739–743
- Müller, D. G., Kawai, H., Stache, B., Lanka, S. (1990). A virus infection in the marine brown alga *Ectocarpus siliculosus*. *Botanica Acta* 103: 72–82
- Proctor, L. M., Fuhrman, J. A. (1990). Viral mortality of marine bacteria and cyanobacteria. *Nature, Lond.* 343: 60–62

This note was submitted to the editor

- Sieburth, J. M., Smetacek, V., Lenz, J. (1978). Pelagic ecosystem structure: heterotrophic compartments of the plankton and their relationship to plankton size fractions. *Limnol. Oceanogr.* 23: 1256–1263
- Suttle, C. A., Chan, A. M., Cottrell, T. (1990). Infection of phytoplankton by viruses and reduction of primary productivity. *Nature, Lond.* 347: 467–469

Manuscript first received: March 6, 1991
Revised version accepted June 25, 1991