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Bacteriophage Ecology Group (BEG) News

Dedicated to the *ecology* and *evolutionary biology* of the parasites of unicellular organisms (UOPs)

© Stephen T. Abedon (editor)

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October 1, 2004 issue (volume 22)

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Editorial

The Bacteriophage Literome - Part I

As many of you are aware, part of the function of the **Bacteriophage Ecology Group** is defining just what phage ecology is. To a large extent this is accomplished through collection of phage ecology literature. To date this "BEG bibliography" consists of nearly **6,000 references** and over **3,000 associated abstracts**. Currently only 722 of those references are from the pre-1967 literature, a literature consisting of over 10,000 phage reports, books, theses, and papers. The great majority of that pre-1967 literature was collected into two volumes by the late **Hansjürgen Raettig**. We are currently in the process of entering these Raettig references into an electronic bibliography and we hope to make them available to you in an **online searchable form**. For what we currently have online, including the entire BEG bibliography, see www.phage.org/beg_bibliography.htm.

As though the above were not enough, a grander plan is to collect the entire bacteriophage "Literome," a collection of all but the most recent phage references (ecology and otherwise). Presentation of that literature again is to be accomplished using an online, searchable format. By our admittedly rough estimation, there are 45,000 (plus or minus 10,000) phage references out there, a figure that does not include papers on such peripheral topics as phage display or the use of phage-derived molecules to engineer non-phage organisms. The yet grander plan—the ultimate goal of the phage literome—is collection of all of these materials into not just a searchable format, one that includes both abstracts and keywords, but also as full-text PDFs (Portable Document Format). And not just full-text PDFs, but also PDFs that are OCRed (Optical Character Recognized) so that they are electronically searchable and, if posted online, indexable by such search engines as **Google** (e.g., click [here](#)).

With sufficient funding, and a lot of cooperation by a lot of different publishers, all of the above is achievable. In the mean time, without funding and no time to contact publishers for copyright permission, we've been concentrating on references alone. As a demonstration project we have attempted to create a "Cyanophage Literome." This was inspired (and, indeed, enabled) by the 1979 effort by Safferman and Rohr to collect all of the cyanophage literature into a single report (Safferman, R. S. and M. E. Rohr. 1979. *The Practical Directory to the Phycovirus Literature*. U.S. Environmental Protection Agency, Cincinnati, Ohio). As this quarter's **submission** we present an update of Safferman and Rohr's report, one now consisting of 338 cyanophage references, 107 entered with abstracts. We hope that you find the product of these efforts useful.

- [BEG: What we are, Where we are, Where we're going](#) by Stephen T. Abedon
- [When Grown *In Vitro*, do Parasites of Multicellular Organisms \(MOPs\) become Unicellular Organism Parasites \(UOPs\)?](#) by Stephen T. Abedon
- [Bacteriophages as Model Systems](#) by Stephen T. Abedon
- [2000 and Sun: A Phage Odyssey](#) by Stephen T. Abedon
- [Lytic, Lysogenic, Temperate, Chronic, Virulent, Quoi?](#) by Stephen T. Abedon
- [Which Ecology are You?](#) by Stephen T. Abedon
- [Science NetWatch October 13, 2000](#)
- [The Best of Times, the Worst of Times](#) by Ry Young
- [Naming Bacteriophages](#) by Hans-Wolfgang Ackermann and Stephen T. Abedon
- [The Bacteriophage Rise](#) by Stephen T. Abedon
- [Mathematics for Microbiologists](#) by Stephen T. Abedon
- [Shipping Phages](#) by Hans-Wolfgang Ackermann
- [Calling a Phage a "Phage"](#) by Stephen T. Abedon
- [Phage or Phages](#) by Hans-Wolfgang Ackermann
- [The Phage Manifesto](#) by Ry Young
- [The Félix d'Hérelle Phage Center Changes Hands](#) by Hans-Wolfgang Ackermann
- [Phage T4 Meets Microbial Diversity](#) by Jim D. Karam
- [Phage T1: A lambdoid phage with attitude?](#) by Andrew Kropinski
- [ASM Conference on the New Phage Biology](#)
- [A Brief History of Phage Art](#)
- [Declining Electron Microscopy](#) by Hans-Wolfgang Ackermann
- [The Bacteriophage Literome - Part I](#)

Editorials should be written on subjects relevant to The Bacteriophage Ecology Group as an organization, to *BEG News* (either the concept or a given issue of *BEG News*), or the science of Bacteriophage Ecology. While my assumption is that I will be writing the bulk of these editorials, [I wish to encourage as many people as possible to seek to relieve me of this duty, as often as possible](#). Additionally, I welcome suggestions of topics that may be addressed. Please address all correspondences to microdude+@osu.edu or to "Editorials," *Bacteriophage Ecology Group News*, care of Stephen T. Abedon, Department of Microbiology, The Ohio State University, 1680 University Dr., Mansfield, Ohio 44906. Please send all submissions as Microsoft Word documents, if possible (I'll let you know if I have trouble converting other document formats), and in English.

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New BEG Members

Please welcome our newest members

name (home page links)	status	e-mail	address
Steven Ripp	PI	utk.edu @saripp	The University of Tennessee, 676 Dabney Hall, Knoxville TN 37996
	interests:	Bacteriophage for bioluminescent monitoring of pathogens. (contents BEG members top of page)	
Ruth-Anne Sandaa	---	ruth.sandaa @bio.uib.no	Department of Biology, Postboks 7800, University of Bergen, Norway
	interests:	Molecular diversity and ecology of viruses and bacteria in environmental samples. Especially the influence of phages on bacterial population dynamics in marine ecosystems. (contents BEG members top of page)	

The [BEG members page](#) can be found at www.phage.org/beg_members.htm. There are two ways of "joining" BEG. One, the "traditional" way, is to have your name listed on the web page and on the list server. The second, the "non-traditional" way, is to have your name only listed on the list server. The latter I refer to as "non-members" on that list. Members, e.g., individuals listed on the [BEG members list page](#), should be limited to individuals who are actively involved in science (research, instruction, outreach, industry) and who can serve as a phage ecology resource to interested individuals. If you have an interest in phage ecology but no real expertise in the area, then you should join as a non-member. To join as a member, please contact BEG using the following link: microdude+@osu.edu. Include:

- your name
- your e-mail address
- your snail-mail address
- the URL of your home page (if you have one)
- a statement of whether or not you are the principal investigator
- a statement of your research interests (or phage ecology interests)
- a list of your phage ecology references, if any

Note that it is preferable that you include the full reference, including the abstract, if the reference is not already present in the [BEG bibliography](#). Responsibility of members includes keeping the information listed on the [BEG members page](#) up to date including supplying on a reasonably timely basis the full references of your new phage ecology publications. Reprints

Meetings

Please send photos, etc. from meetings, etc. for inclusion in this section.

Follow up to ASM Conference on the New Phage Biology

Please send any photos and powerpoint presentations from the summer, 2004, phage meeting to microdude+@osu.edu. We hope to present this material in a subsequent issue of BEG News.

The BEG [Meetings](#) link will continue. Reminders of upcoming meetings will be placed in this section of *BEG News*. If you know of any meetings that might be of interest to BEG members, or would like to recap a meeting that you've attended, then please send this information for posting to microdude+@osu.edu or to "BEG Meetings," *Bacteriophage Ecology Group News*, care of Stephen T. Abedon, Department of Microbiology, The Ohio State University, 1680 University Dr., Mansfield, Ohio 44906.

Submissions

The Cyanophage Literome

The following represents all of the cyanophage references that I am aware of. I make no claims as to completeness or accuracy of this list and will depend on those more familiar with these phages to point out any problems. Please send all corrections, additions, and comments to microdude+@osu.edu. I will make changes directly to this page, indicating when and where changes have been made. Also, please don't be shy about typing in and sending me any missing abstracts, **AND DON'T FORGET THAT YOU CAN REFERENCE THIS LIST IN YOUR PUBLICATIONS: Stephen T. Abedon (2004). Cyanophage Literome. *BEG News* 22 (www.phage.org/bgnws022.htm#submissions).**

Post-publication contributions:

- 9/14/04: [reference and abstract](#) added by [Matthew B. Sullivan](#)
- 9/14/04: [reference and abstract](#) added by [Matthew B. Sullivan](#)
- 9/20/04: [reference and abstract](#) added by [Matthew B. Sullivan](#)
- 9/20/04: [reference and abstract](#) added by [Matthew B. Sullivan](#)
- 4/12/05: [reference and abstract](#) added by [Matthew B. Sullivan](#)
- 4/12/05: [reference and abstract](#) added by [Matthew B. Sullivan](#)
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The Cyanophage Literome with Abstracts

The following represents all of the cyanophage references that I am aware of. I make no claims as to completeness or accuracy of this list and will depend on those more familiar with these phages to point out any problems. Please send all corrections, additions, and comments to microdude+@osu.edu. I will make changes directly to this page, indicating when and where changes have been made. Also, please don't be shy about typing in and sending me any missing abstracts, AND DON'T FORGET THAT YOU CAN REFERENCE THIS LIST IN YOUR PUBLICATIONS: Stephen T. Abedon (2004). Cyanophage Literome. *BEG News* 22 (www.phage.org/bgnws022.htm#submissions).

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195. **[Effect of 1,3,5-triazines on several prokaryote viruses and their hosts].** Stenz, E., Menzel, G. (1978). *Zeitsch. Allg. Mikrobiol.* 34:748-753. In the agar diffusion test 24 triazines were investigated with regard to their action on the multiplication of DNA phages (λ and LPP-1) and RNA phages (M12 and Q β). In several cases the amount of plaques was diminished or increased depending on the kind of triazine and virus. The investigations demonstrate the triazines to be able to interfere with the formation of plaques by virulent and temperate viruses of prokaryotes
196. **Photoreactivation of ultraviolet irradiated blue-green alga: *Anacystis nidulans* and cyanophage AS-1.** Amla, D. V. (1979). *Archives of Virology* 59:173-179. Ultraviolet (UV) inactivation and photoreactivation of *Anacystis nidulans* and cyanophage AS-1 was studied at different wavelengths. UV inactivation of free phage particles and one and two hour host-phage complexes (intracellular phages) were exponential. UV resistance of plaque forming units was attained at the latter phase of latent period. Black, blue and white lights were able to photoreactivate the UV irradiated *A. nidulans* whereas green, yellow and red lights were not. However, incubation of *A. nidulans* for more than 2 hours in black light resulted in loss of viability but shift to red light caused significant

recovery. This suggests the involvement of two types of photoactivation, i.e. of photoenzymatic repair of DNA and of the repair of the photosynthetic apparatus of *A. nidulans*

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199. **Characteristics of *Anabaena variabilis* influencing plaque formation by cyanophage N-1.** Currier, T. C., Wolk, C. P. (1979). *Journal of Bacteriology* 139:88-92. Phage N-1 grown in *Anabaena* strain 7120 [N-1. 7120] forms plaques on *A. variabilis* about 10⁻⁷ to 10⁻⁶ as efficiently as on *Anabaena* 7120. By manipulating different characteristics of the interaction between phage and host, it was possible to increase the relative efficiency of plaque formation to 0.38. Growth of *A. variabilis* at 40°C for at least three generations resulted in an increase in the rate of phage adsorption and a 10-fold increase in the efficiency of plaque formation. The efficiency of plaque formation was further increased about 42-fold, with little or no further increase in rate of adsorption, in a variant strain. *A. variabilis* strain FD, isolated from a culture of *A. variabilis* which had grown for more than 30 generations at 40°C. The low relative efficiency of plaque formation by N-1. 7120 on *A. variabilis* could be partially accounted for if *A. variabilis* contains a deoxyribonucleic acid restriction endonuclease which is absent from *Anabaena* 7120. Indirect evidence for such an endonuclease included the following: (i) phage N-1 grown in *A. variabilis* (N-1. Av) had approximately a 7 X 10³-fold higher relative efficiency of plaque formation on *A. variabilis* than had N-1. 7120; and (ii) the efficiency of plaque formation by N-1. 7120 on *A. variabilis* strain FD was increased by up to 146-fold after heating the latter organism at 51°C
200. **Lytic organisms and photooxidative effects: Influence of blue-green algae (cyanobacteria) in Lake Mendota, Wisconsin.** Fallon, R. D., Brock, T. D. (1979). *Applied and Environmental Microbiology* 38:499-505.
201. **Optimization kinetics and thermodynamics of cyanophage A-1 adsorption on algal cells.** Mendzhul, M. I. (1979). *Mikrobiologichnyi Zhurnal* 41:145-150. Optimization, kinetics and thermodynamics of cyanophage A-1 adsorption on algal cells. The extremely rapid adsorption of cyanophage A-1 on the alga *Anabaena variabilis* cells occurs in 0.01 M tris-HCl-buffer in the presence of 0.1 M MgCl₂ at pH 7.0 and 25°C. Kinetics of the cyanophage adsorption on the host cells is more complex than the 1st order reaction. Analysis of kinetic curves for the cyanophage adsorption and some other characteristics of the process showed that cyanophage A-1 adsorption on the cells occurred according to the competition model. Some thermodynamic potentials of the process are calculated; their values indicate an enzymic character of the reaction of the virion attachment to the algal cell
202. **[Effect of the detergent Metaupon on replication of various phages].** Menzel, G., Stenz, E. (1979). *Zeitschrift fur Allgemeine Mikrobiologie* 19:325-332. As several other surfactants do, the detergent Metaupon acts on the multiplication of bacteriophages. We investigated the influence of Metaupon on the phages ϕ and lambda, the cyanophage LPP-1, and the RNA-phages f 2, M 12, and Q beta by means of the agar diffusion test, pour plate test, adsorption test, and one-step growth test. The action of Metaupon on the free phages was also tested. Metaupon inhibits the formation of plaques by the phages with exception of lambda. With the phages f 2 and M 12 the substance increases the amount of plaques depending on concentration. The main mode of action of Metaupon was found to be the inhibition of the adsorption of the phages to the host cells. Only in the case of ϕ 105 free phages were inactivated
203. **Lysate effect of *Microcystis aeruginosa* infected with cyanophage AM-1 on survival of *Daphnia magna*.** Myslovich, V. O. (1979). *Gidrobiologicheskii Zhurnal* 15:67-70. Lysate effect of *Microcystis aeruginosa* infected with cyanophage AM-1 on survival of *Daphnia magna*. The behavior and survival of *Daphnia magna* juvenile influenced by lysates of *Microcystis aeruginosa* Keutz. emend. Elenk. culture infected by cyanophage AM-1 depend on dilution level and storage time of the lysates. Toxic effects are possible under natural conditions when lysis of cyanophage AM-1-infected algae occurs
204. **An ultraviolet light induced bacteriophage in *Beneckea gazogenes*.** Rambler, M., Margulis, L. (1979). *Origins of Life* 9:235-240. An ultraviolet light induced prophage has been discovered in the red pigmented marine vibrio *Beneckea gazogenes*. Two spontaneously derived pigment mutants, one forming pink colonies and one lacking pigment and forming white colonies, were also irradiated. The presence of pigment was not related to phage induction; uv-induced cell lysis occurred in wildtype and mutant strains at the same dosages. Lysis was not prevented or retarded by exposure after irradiation to visible light indicating the phenomenon was not photoreactivable. Electron micrographs of the 'T-like' *B. gazogenes* phage are shown. A second *Beneckea* was isolated from the anaerobic zone of cyanobacterial mats growing in the hypersaline environment of Laguna Mormona, Baja California. The *Baja beneckea* does not harbor a uv inducible prophage and is resistant to the *B. gazogenes* phage under all conditions tested
205. **The Practical Directory to the Phycovirus Literature.** Safferman, R. S., Rohr, M. E. (1979). EPA-600/9-79-013. The volume comprises a comprehensive survey of the phycovirus literature. It covers the period from their isolation to the present time.

206. **Host range plaque morphology studies of cyanophage LPP-1.** Kraus, M. P. (1980). *J. Phycol.* 16:186-191. Host-range, plaque-morphology studies of cyanophage LPP-1. Transduction by temperate cyanophage plays an important role in understanding the effects of environmental pollution on genetic function. Using a new isolate, the influence of contaminants and the rapid variations that result as a virus particle passes through successive hosts is illustrated. Host-range and plaque-morphology, using an extended range of genetically-differing hosts, compares archetype LPP-1 cyanophage cultured on microbially contaminated hosts with bacteria-free cyanophage cultured on pure host strains. Microbial contamination can alter the host-range and serology of the cyanophage produced. Bacteria are involved in the virus infection of cyanophycean hosts and the study of host-range and plaque-morphology can aid in the biological characterization and segregation of mutants illustrating mechanisms of intergeneric transfer of genetic material. Derivatives of archetype LPP-1, cultured on axenic hosts, possess a host-range, plaque-morphology and serology similar or identical to that of the temperate cyanophage, S3
207. **Isolation of characteristics of minute plaque forming mutant of cyanophage AS-1.** Amla, D. V. (1981). *Biochemie und Physiologie der Pflanzen* 176:83-89. Isolation of characteristics of minute plaque forming mutant of cyanophage AS-1. Minute plaque forming mutant (m) of cyanophage AS-1 infecting unicellular blue-green algae, *Anacystis nidulans*, was isolated spontaneously and after mutagenic treatment. Compared to wild type m mutant-formed small plaques, adsorption rate was slow and the burst-size was significantly decreased with prolonged eclipse and latent period. The plaque forming ability of mutant phage was sensitive to pH, heat, EDTA shock, distilled water and photosensitization with acriflavine; UV sensitivity of free and intracellular phage was identical to the parent. The spontaneous reversion frequencies of mutant phage to wild-type were between 10⁻⁵-10⁻³, and appeared to be clonal property. Reversion studies suggested possibilities of frame-shift or base-pair substitution for m mutation
208. **Chelating agent shock of cyanophage AS-1 infecting unicellular blue-green algae, *Anacystis nidulans*.** Amla, D. V. (1981). *Indian Journal of Experimental Biology* 19:209-211. Chelating agent shock of cyanophage AS-1 infecting unicellular blue-green algae (*Anacystis nidulans*). Three strains of free cyanophage AS-1 (wild, host-range h and minute plaque forming m) exposed to chelating agents were inactivated by chelating agent shock (CAS) when diluted rapidly in distilled water. The intracellular phage particles were comparatively resistant to CAS inactivation. Susceptibility of all the phage strains to CAS was enhanced with increases in concentration of chelating agents, time and temperature of the shocking water. Addition of monovalent or divalent salts but not the nonionic solutes to the shocking water resulted in protection of phage particles; addition of these salts to the shocking water after CAS treatment did not promote recovery of phage infectivity. Inactivation of cyanophages by CAS is probably due to interaction of the polyanionic chelating agents with the cations present in phage protein. In the course of rapid dilution the native structure of cyanophage particles is distorted, resulting in inactivation of phages
209. **Cyanobacteriophage interactions on the replication of cyanophage SM-2.** Barnet, Y. M., Daft, M. J., Stewart, W. D. P. (1981). *Journal of Applied Bacteriology* 51:541-552.
210. **Cyanobacteria-cyanophage interactions in continuous culture.** Barnet, Y. M., Draft, M. J., Stewart, W. D. P. (1981). *Journal of Applied Bacteriology* 51:541-552.
211. **Effects of photosynthetic inhibitors and light-dark regimes on the replication of cyanophage SM-2.** Benson, R., Martin, E. (1981). *Archives of Microbiology* 129:165-167.
212. **Cyanophages—are they potential biological control agents of nuisance blue-green algae?** Desjardins, P. R. (1981). *E-81-7:198-229*.
213. **New *Anabaena* and *Nostoc* cyanophages from sewage settling ponds.** Hu, N.-T., Thiel, T., Gidding, T. H., Jr., Wold, C. P. (1981). *Virology* 114:236-246.
214. **Sequence of morphological alterations in blue-green algae in the course of cyanophage infection.** Moisa, I., Sotropa, E., Velehorschi, V. (1981). *Virologie* 32:133-137. Electron microscopic studies were performed on the sequence of morphological alterations induced by the cyanophage PP-1 in the blue-green algae *Phormidium* sp. and *Plectonema boryanum*. The following phases of virus infection were made evident: virus adsorption onto the host cell; the presence of virus "ghosts", suggesting the penetration of viral DNA into the cell and its multiplication in the nucleoplasm; invagination of thylakoids and formation of the "virogenic stroma"; virus maturation within the "virogenic stroma"; cellular lysis at 48 hours post inoculation
215. **Investigation on the presence of cyanophages in fresh and sea waters of Romania.** Moisa, I., Sotropa, E., Velehorschi, V. (1981). *Virologie* 32:127-132. Investigations on the presence of cyanophages in the fresh and sea waters of Romania resulted in the isolation of 31 strains. The host range of the cyanophage isolates showed some particularities as compared with classical cyanophages types. The electron optic study of the cyanophage strains grown in *Phormidium* sp. revealed the presence of three types of virus particles, differing as regards their tail length, with a morphology similar to that of T-odd coliphages
216. **Collapsing *Aphanizomenon flos aquae* blooms: Possible contributions of photo-oxidation, oxygen toxicity and cyanophages.** Coulombe, A. M., Robinson, G. G. C. (1982). *Canadian Journal of Botany* 59:1277-1284. Triggering mechanisms for collapse of *A. flos-aquae* (L.) Ralfs blooms in 3 shallow eutrophic pothole lakes (L 885, L 958 and L 522), located within an aquaculture project study area in southwestern Manitoba, Canada, were examined. Three of the collapses observed (L 885, mid-July 1979; L 958, mid-Aug. 1979; and L 522, mid-July 1979) were initiated during periods of lake thermal stability when conditions conducive to photo-oxidation and/or death due to O₂ toxicity were operable. A 4th collapse (L 958, mid-Aug. 1973) was initiated during a period of lake thermal instability when photo-oxidation and O₂ toxicity could be dismissed as triggering mechanisms. The possibility of cyanophage-induced algal lysis causing bloom collapse was considered and morphological evidence

for the occurrence of viruslike particles (vlp) within Aphanizomenon cells from L 885 (1979) and L 958 (1978, 1979) are presented. Since transmission and isolation of the vlps was not substantiated, the verification of a virus infection of the Aphanizomenon populations studied is not yet possible. No single triggering mechanism can account for all of the algal collapses described.

218. **Pleiotrophic behavior of a cyanophage AS-1 resistant mutant of *Anacystis nidulans*. Kashyap, A. K., Gupta, S. L. (1982). *Molecular and General Genetics* 185:365-366.** Pleiotropic behavior of a cyanophage AS-1-resistant mutant of *Anacystis nidulans*. A cyanophage AS-1 resistant mutant strain of *A. nidulans* exhibited a slow rate of nutrient uptake compared to the wild type. The increased Ca^{2+} sensitivity of the mutant could be correlated with higher rates of Cu^{2+} uptake. The results are discussed in the light of alterations in the proteins involved in permeability of the outer membrane
219. **Peculiarities of a new cyanophage specific for the cyanobacterium *Synechococcus schmidlea*. Koz'yakov, S. Y. (1982). *Microbiology (New York)* 50:395-401.**
220. **AS1 cyanophage adsorption to liposomes. Oliveira, A. R., Mudd, J. B., Desjardins, P. R. (1982). *Journal of General Virology* 61:153-156.**
221. **The effect of light and temperature on the generation time, adsorption, and yield of the cyanophages AS-1. Olson, G. B., Desjardins, P. R. (1982). *Phytopathology* 72:937.**
222. **Metabolic aspects of cyanophage AS-1 replication and reproduction in cyanobacterium *Anacystis nidulans*. Amla, D. V., Saxena, P. N. (1983). *Biochemie und Physiologie der Pflanzen* 178:225-236.** The intracellular stages of the cyanophage AS-1 replication cycle were investigated under conditions that impair the metabolic functions of the host, *A. nidulans*. The reproductive cycle of the cyanophage consists of an eclipse period (3.5 h), latent period (7 h) and finally lysis of cells after 14 h with the release of 100-120 PFU/infected cell. Viral multiplication was inhibited in dark. Withdrawal of light before the eclipse period or incubation of the infected cells in the dark for 6 h followed by illumination, decreased the final yield of virus and prolonged the reproductive cycle. The inhibitor of Photosystem II, DCMU, prolonged the latent period and reduced the burst-size to 50-60% of the control. Inhibitor of electron transport, CCCP, abolished the viral growth completely. Treatment of infected cells with chloramphenicol up to 4 h during the latent period completely abolished the phage growth. These results demonstrated the dependent virulent nature of the cyanophage AS-1
223. **Aerosol release of cyanophages and coliforms from activated sludge basins. Cannon, R. E. (1983). *Journal Water Pollution Control Federation* 55:1070-1074.** Aerosol release of cyanophages and coliforms from activated sludge basins. Aerosol release of cyanophages and coliforms from activated sludge basins at 2 wastewater treatment plants in Greensboro, North Carolina [USA] was studied. One uses diffused aeration in the treatment process and the other, mechanical aerators. Detection methods consisted of mechanical air samplers and stationary sampling sites using petri dishes open to the air for varying times. Samples were taken weekly for 1 yr to ensure that virus dispersal was studied under a variety of weather conditions. There were considerably more aerosols when aeration was mechanical instead of diffused. Wind direction seemed to be an important environmental factor in the spread of viruses and coliforms from the basins. Cyanophages, which were found more readily than coliforms throughout the year, may serve as effective indicators for aerosol wastewater contamination
224. ***Viral Control of Nuisance Cyanobacteria (Blue-Green Algae). II. Cyanophage Strains, Stability on Phages and Hosts, and Effects of Environmental Factors on Phage-Host Interactions.* Desjardins, P. R., Olson, G. B. (1983). California Water Resource Center, University of California, Davis, CA.** Differentiation of phage strains in the AS cyanophage group was accomplished. Studies on *Anabaena* cyanophages (A-1, A-4, and AN-10), which originally were received from Russia, demonstrated that the A-4 preparation was actually a mixture of a lytic (AN-10) phage and a temperate (A-4) phage. An additional strain of *Anabaena variabilis* was shown to be a host of all three phages in the group. ¶ Antiserum to the LPP-1 cyanophage with a relatively high titer was prepared for later use in cyanophage detection. Storage by simply freezing in culture media permitted some cyanobacterial species to survive for several months. Failure of other species to survive under identical conditions indicates a need for additional research in this area. The adverse effects of freezing on virion structure and infectivity were characterized for the AS-1 and LPP-1 cyanophages. ¶ Bloom concentrations of *Plectonema boryanum* were established in outdoor pond facilities. Some control of this cyanobacterial species was effected with the LPP-1 cyanophage. Results suggest that the cyanophage is most effective when present before the bloom develops. ¶ Studies on the effect of temperature on the growth cycle of AS-1 cyanophage demonstrated that the length of the cycle varied inversely with temperature in the range 25-36°C. The importance of light quality in the growth cycle of this cyanophage has also been shown. Of special significance is the finding that the red/far red light ratio can greatly influence the yield of AS-1 in *Anacystis nidulans*.
225. **Cyanophage: History and likelihood as a control. Desjardins, P. R. (1983). pp. 242-248 *Lake Restoration, Protection, and Management*. Environmental Protection Agency, Washington, D.C.** It has been 20 years since the first cyanophage was discovered. Since then additional cyanophages and strains that infect both unicellular and filamentous cyanobacteria have been found. Cyanophages are similar to other bacteriophages in many physical, chemical and biological characteristics, but differ from them in their requirement of light for absorption to their hosts and their dependence upon the photosynthetic activity of their hosts for their replication. Light quality and the ratio of red to far-red light affect virus replication. Cyanophages play a distinctive role in the ecology of their hosts and probably are effecting some natural control. Certain factors (development of resistant host strains, specific ion requirements, environmental factors and lysogeny) may affect the potential of the cyanophages to control their hosts, but these have not been conclusively shown to completely destroy this potential. There is much need for additional research on the experimental control of nuisance species in natural water bodies. Preliminary studies suggest that the phages may be more effective in preventing blooms than in eliminating one already formed. An integrated approach involving several biological techniques is recommended for control of nuisance populations of cyanobacteria.
226. **Cyanophages. Gromov, B. V. (1983). *Ann. Microbiol. (Inst. Pasteur)* 134B:43-59.** The description of

cyanophages isolated in the USSR is given. The data presented here primarily concern cyanophages of A(L) and S (L) series developing in the cells of *Anabaena variabilis* strains of *Synechococcus* species strains, respectively

227. **Classification and nomenclature of viruses of cyanobacteria.** Safferman, R. S., Cannon, R. E., Desjardins, P. R., Gromov, B. V., Haselkorn, R., Sherman, L. A., Shilo, M. (1983). *Intervirology* 19:61-66.
228. **The effect of suspended particular material on cyanobacteria-cyanophage interactions in liquid culture.** Barnet, Y. M., Daft, M. J., Stewart, W. D. P. (1984). *Journal of Applied Bacteriology* 56:109-115. The effect of the lytic phage LPP-DUNI on the cyanobacterium *Plectonema borya* has been investigated in batch and in continuous cultures in the presence and absence of silt. In batch culture *Plectonema* without added phage grew normally: the presence of phage caused rapid lysis of the cyanobacterium and the addition of the prevented lysis of the cyanobacterium and the addition of the prevented lysis by the phage. In continuous culture the numbers of cyanobacterial cells and phage particles oscillated in a reciprocal manner, but the addition silt damped down the oscillations in *Plectonema* biomass without decreasing the numbers of phage particles isolated from the cultures
229. **Effect of toxicants on UV survival of cyanophage host virus systems.** Kraus, M. P. (1984). *Photochemistry & Photobiology* 39:97S.
230. **Effects of pesticides on cyanobacterium *Plectonema boryanum* and cyanophage LPP-1.** Mallison, S. M. III, Cannon, R. E. (1984). *Applied and Environmental Microbiology* 47:910-914. Cyanobacterium *Plectonema boryanum* IU 594 and cyanophage LPP-1 were used as indicator organisms in a bioassay of 16 pesticides. Experiments such as spot tests, disk assays, growth curves, and one-step growth experiments were used to examine the effects of pesticides on the host and virus. Also, experiments were done in which host or virus was incubated in pesticide solutions and then assayed for PFU. *P. boryanum* was inhibited by four herbicides: 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU), 1,1-dimethyl-3-(alpha, alpha,alpha-trifluoro-m-tolyl)urea (Fluometeron), 2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine (Atrazine), 2-(ethylamino)-4-(isopropylamino)-6-(methylthio)-s-triazine (Ametryn). One insecticide, 2-methyl-2-(methylthio)-propionaldehyde O-(methylcarbonyl)oxime (Aldicarb), also inhibited the cyanobacterium. Two insecticides inactivated LPP-1, O,O-dimethyl phosphorodithioate of diethyl mercaptosuccinate (malathion) and Isotox. Isotox is a mixture of three pesticides: S-[2-(ethylsulfinyl)ethyl]O,O-dimethyl phosphorothioate (Metasystox -R), 1-naphthyl methylcarbamate (Sevin) and 4,4'-dichloro-alpha-(trichloromethyl) benzhydrom (Kelthane). Two pesticide-resistant strains of *P. boryanum* were isolated against DCMU and Atrazine. These mutants showed resistance to all four herbicides, which indicates a relationship between these phototoxic chemicals. The results indicate that *P. boryanum* may be a useful indicator species for phototoxic agents in bioassay procedures
231. **Mutation to resistance for virus AS-1 in the cyanobacterium *Anacystis nidulans*.** Bisen, P. S., Audholia, S., Bhatnagar, A. K. (1985). *Microbiol. Lett.* 29:7-13.
232. **Host range of LPP cyanophages.** Johnson, D. W., Potts, M. (1985). *International Journal of Systematic Bacteriology [INT. J. SYST. BACTERIOL.]* 35:76-78. The authors determined the sensitivities of 33 strains and variants of cyanobacteria to infection by the cyanophage LPP-1 archaetype, five LPP-1 serotypes, six LPP-2 serotypes, and 8 new LPP isolates. The LPP-1 archaetype and LPP-1 serotypes have different host ranges on strains of LPP group B
233. **A survey for viruses from fresh water that infect a eukaryotic *Chlorella*-like green alga.** van Etten, J. L., van Etten, C. H., Johnson, J. K., Burbank, D. E. (1985). *Applied and Environmental Microbiology* 49:1326-1328.
234. **Evidence for lysogeny and viral resistance in the cyanobacterium *Phormidium uncinatum*.** Bisen, P. S., Audholia, S., Bhatnagar, A. K., Bagchi, S. N. (1986). *Current Microbiology* 13:1-5. Cyanophage LPP-1-induced lysogens and a resistant mutant of the cyanobacterium *Phormidium uncinatum* were isolated and characterized. In lysogens, spontaneous lysis occurred and increased with the growth of the host cyanobacterium. The virus-liberating property of the lysogens was not lost with the viricidal concentration of EDTA, and the titer obtained was > 3 plus or minus 10^3 PFU ml super⁻¹. Heat and UV treatment of lysogens failed to induce lysis, but mitomycin C induced lysis by fivefold. The adsorption rate of the virus on the lysogens was slower than on the sensitive parent host
235. **[The structure of cyanobacterial phycobilisomes and its change in viral infection].** Mendzhul, M. I., Averkiev, A. A. (1986). *Mikrobiologichnyi zhurnal* 48:89-101.
236. **[Role of temperate phage in bacterial dissociation].** Mil'ko, E. S., Egorov, N. S. (1986). *Nauchnye doklady vysshei shkoly Biologicheskie nauki* 6-19. The analysis of literary and own data testifies that the dissociants may appear in bacteria population from spontaneous mutations and transfer of genetic material (conjugation, transformation, transduction). The phage conversion and different DNA reorganizations within a cell where prophage plays an active role, probably introduce the largest contribution into the dissociative transitions of variants which occur with high frequency (about 10^{-2} - 10^{-4}). The dissociation of various bacteria has been studied with different degree. The role of temperate phage has been shown in splitting of bacteria into variants in the genera *Mycobacterium*, *Corynebacterium*, some *Bacillus*, *Clostridium*, *Staphylococcus*, some enterobacteria, *Yersinia*, *Vibrio Pseudomonas*, *Rhizobium*, *Nostoc*; the participation of prophage in dissociation of bacteria of the genera *Xanthomonas*, *Erwinia*, *Bacteroides* is proposed. A method for obtaining the nondissociating S-variants for stability of biologically active substances synthesized by cells has been suggested
237. **Cyanophage ecology.** Cannon, R. E. (1987). pp. 245-265 in Goyal, S. M., Gerba, C. P., Bitton, G. (eds.) *Phage Ecology*. John Wiley & Sons, New York.
238. **Isolation and characterization of a temperate cyanophage for a tropical *Anabaena* strain.** Franche, C. (1987). *Archives of Microbiology* 148:172-177. In this paper we describe the isolation and characterization of a temperate cyanophage N(S)1 of the genus cyanopodovirus which produces turbid plaques on the host *Anabaena* 77815 isolated from tropical soil. Its properties have been compared to those of other well-characterized

phages. In addition, two strains of *Anabaena* 77815 lysogenic for N(S)1 were isolated. N(S)1 seems to be integrated into the chromosome of the two lysogens, and a 2 kb plasmid present at a low copy number in the non-lysogenic strain is amplified significantly.

239. **Resistance of cultures of cyanobacteria *Synechococcus cedrorum* and *Synechococcus parvula* to AS-1K and S-8K cyanophages.** Goryushin, V. A., Shainskaya, O. A. (1987). *Mikrobiologichnyi Zhurnal* 48:74-78. Resistance of cultures of cyanobacteria *Synechococcus cedrorum* and *Synechococcus parvula* to AS-1K and S-8K cyanophages. Clones of unicellular cyanobacteria *Synechococcus cedrorum* and *S. parvula* having different susceptibilities to AS-1K and S-8K cyanophages were isolated, including clones with absolute resistance. Studies of age changes and culture conditions suggest that the resistance of the obtained cyanobacteria clones to virus infection is associated with a spontaneous mutation-induced modification in cell receptors
240. **Changes in sensitivity to cyanophage infection in axenic LPP cyanobacteria.** Johnson, D. W., Borovsky, D. (1987). *Microbios Letters* 35:105-112.
241. **An analysis of restriction endonuclease sites in cyanophages infecting the heterocystous cyanobacteria *Anabaena* and *Nostoc*.** Bancroft, I., Smith, R. J. (1988). *The Journal of general virology* 69 (Pt 3):739-743. An analysis of restriction endonuclease cleavage of DNA isolated from cyanophages that infect *Anabaena* and *Nostoc* species of cyanobacteria has provided evidence for counter-selection of restriction endonuclease sites. These include sites containing subsequences which are methylated by host (*Anabaena* PCC 7120) methylase(s) akin to the *dam* and *dcm* enzymes of *Escherichia coli*. Other sites which are counter-selected have no common sequence structure. The latter include those of the endogenous restriction endonucleases of the host, but other absent sequences are not attributable to isoschizomers of any known *Anabaena* or *Nostoc* restriction endonuclease. The cyanophages differ in their tolerance to DNA methylation. Isolates A-4L, AN-13 and AN-23 do not tolerate adenine methylation in the GATC sequence whereas two cyanophages, A-1L and AN-10 (which are related) do tolerate *dam*-like methylation of this sequence. In addition, A-1L allows cytosine methylation at GGCC sequences, but AN-10 has counter-selected these sequences and the remaining sites are not methylated. Analysis of native and cloned A-4L DNA suggests that counter-selection has occurred against all sequences which would be methylated by the host at either adenine or cytosine nucleotides
242. **Phages of cyanobacteria.** Martin, E. L., Benson, R. (1988). pp. 607-645 in Calendar, R. (ed.) *The Bacteriophages. Volume 2*. Plenum Press, New York.
243. **Sequence counter-selection in cyanophage.** Bancroft, I, Smith, R. J. (1989). p. 316 in Rogers, L. J., Gallon, J. R. (eds.) *Biochemistry of the Algae and Cyanobacteria*. An analysis of the cleavage of native and cloned DNA of five cyanophage which infect *Anabaena* 7120 by 34 restriction endonucleases provided evidence of sequence counter-selection similar to that present in T sub(7). One group are isoschizomers of *Anabaena* 7120 endogenous restriction endonucleases. Another group contain the subsequence GATC; and a third group contain dicytosine residues. The fourth group have no common sequence structure and may represent isoschizomers of restriction endonucleases present in the host range of the five cyanophage. Cyanophages AN-23, AN-13, and A-4L do not tolerate sequence methylation. A-1L and AN-10 tolerate adenine methylation, but differ in their tolerance of cytosine methylation. AN-10 appears able to prevent cytosine methylation by host enzymes. AN-10 and A-1L are closely related. Comparison of their restriction maps shows that counter-selection of some Hae III sites in AN-10 has occurred since divergence of the phage from their common ancestor
244. **Inhibitory effect of the extracts of *Zingiber* species on the adsorption and replication of phage LPP-1 in cyanobacterium.** Jido, E. P., Dhaliwal, A. S. (1989). Rhizome extracts from *Zingiber officinale* and *Zingiber zerumbet* were prepared by grinding in saline. Cyanobacterium was treated with each of the extracts (20% v/v). Extract treated cyanobacterium were inoculated with phage LPP-1. The extract of *Z. officinale* caused 59% inhibition of virus adsorption and 77% inhibition of burst size, whereas *Z. zerumbet* caused 67% inhibition of virus adsorption and 98% inhibition of burst size
245. **A new temperate cyanophage NP-1T lysogenizing cyanobacterial cultures belonging to the genera *Nostoc* and *Plectonema*.** Muradov, M., Cherkasov, G. V., Akhmedova, D. U., Khalmuradov, A. G. (1990). *Mikrobiologija (Microbiologia)* 59:1038-1045. A new temperate cyanophage NP-1T growing on *Nostoc* and *Plectonema cyanobacterial* cultures is described. Cyanophages of the NP type are widespread in Uzbekistan water basins. The cyanophage has a hexagonal head on a plane with a distance of 78 nm between the facets, but lacks a distinctly differentiated tail. Its adsorption takes 2.5 to 3 h. The lytic cycle takes 30 h with 14 h for the latent period. The phage yield is about 350 particles per infected cell. The DNA has 72 mol.% G + C. The mean contour length is 14.4.μm and the molecular mass of DNA calculated in terms of its length is 28 MDa. When the cyanophage interacts with the host culture, lysogenic clones resistant to the cyanophage appear.
246. **Comparative study of NP-IT cyanophages, which lysogenize nitrogen-fixing bacteria of the genera *Nostoc* and *Plectonema* (English).** Muradov, M. M., Cherkasova, G. V., Akhmedova, D. U., Kamilova, F. D., Mukhamedov, R. S., Abdukarimov, A. A., Khalmuradov, A. G. (1990). *Microbiology (translation of Mikrobiologiya)* 59:558-563. Comparative characteristics of NP-1T cyanophages causing lysis of nitrogen-fixing *Nostoc* and *Plectonema* cultures. The strain specificity of NP-1T cyanophages causing lysis of *Nostoc* and *Plectonema* cultures was exerted as differences in the time of formation and in the morphology of plaques. The specificity was confirmed by the data of restriction analysis using the EcoRV enzyme that hydrolysed the DNA of the cyanophages to yield a different number of fragments
247. **Comparative study of NP-IT cyanophages, which lysogenize nitrogen-fixing bacteria of the genera *Nostoc* and *Plectonema* (Russian?).** Muradov, M. M., Cherkasova, G. V., Akhmedova, D. U., Kamilova, F. D., Mukhamedov, R. S., Abdukarimov, A. A., Khalmuradov, A. G. (1990). *Mikrobiologija (Microbiologia)* 59:819-826. Comparative characteristics of NP-1T cyanophages causing lysis of nitrogen-fixing *Nostoc* and *Plectonema* cultures. The strain specificity of NP-1T cyanophages causing lysis of *Nostoc* and *Plectonema* cultures was exerted as differences in the time of formation and in the morphology of plaques. The specificity was confirmed by the data of restriction analysis using the EcoRV enzyme that hydrolysed the DNA of the cyanophages to yield a different

248. **Cyanophages which impact bloom-forming cyanobacteria.** Philips, E. J., Monegue, R. L., Aldridge, F. J. (1990). *J. Aquat. Plant. Manage.* **28:92-97**. Various mesotrophic and eutrophic freshwater environments in the state of Florida were surveyed for the existence of cyanophages. Cyanophages were discovered which infect and kill four common bloom-forming species of cyanobacteria, *Lyngbya birgei*, *Anabaena circinalis*, *Anabaena flos-aquae*, and *Microcystis aeruginosa*. These cyanophages are being maintained in the laboratory at titers around 10 super(7) PFU/ml. The potential use of these cyanophages to control blooms of these cyanobacteria is discussed
249. **Viral mortality of marine bacteria and cyanobacteria.** Proctor, L. M., Fuhrman, J. A. (1990). *Nature* **343:60-62**. Despite the importance of cyanobacteria in global primary productivity and of heterotrophic bacteria in the consumption of organic matter in the sea, the causes of their mortality, particularly the cyanobacteria, are poorly understood. Here the authors report not only high viral abundance in the ocean but also counts of bacteria and cyanobacteria in the final irreversible stage of lytic infection. The latter counts are necessary to evaluate mortality, because the sources, hosts, viability and ages of observed free viruses are unknown; even finding viruses attached to cells does not prove successful infection. Up to 7% of the heterotrophic bacteria and 5% of the cyanobacteria from diverse marine locations contained mature phage; interpretation via culture data indicates that up to 70% of the prokaryotes could be infected. These data demonstrate the existence of a significant new pathway of carbon and nitrogen cycling in marine food webs and have further implications for gene transfer between marine organisms
250. **Infection of phytoplankton by viruses and reduction of primary productivity.** Suttle, C. A., Chan, A. M., Cottrell, M. T. (1990). *Nature* **347:467-469**. Natural marine waters contain roughly 10^6 to 10^9 virus particles per ml, yet their role in aquatic ecosystems and the organisms that they infect remain largely unknown. Electron microscopy has been used to study interactions between viruses and their hosts, focusing mainly on pathogens to prokaryotic organisms. The authors demonstrate that viral pathogens infect a variety of important marine primary producers, including diatoms, cryptophytes, prasinophytes and chroococoid cyanobacteria. Also, addition to sea water of particles in the 0.002-0.2 μm size range, concentrated from sea water by ultrafiltration, reduced primary productivity (^{14}C -bicarbonate incorporation) by as much as 78%. Results indicate that in addition to grazing and nutrient limitation, infection by viruses could be a factor regulating phytoplankton community structure and primary productivity in the oceans.
251. **AS-1 cyanophage infection inhibits the photosynthetic electron flow of photosystem II in *Synechococcus* sp. PCC 6301, a cyanobacterium.** Teklemariam, T. A., Demeter, S., Deak, Z., Suranyi, G., Borbely, G. (1990). *FEBS Letters* **270:211-215**. In *Synechococcus* sp. cells AS-1 cyanophage infection gradually inhibits the photosystem II mediated photosynthetic electron flow whereas the activity of photosystem I is apparently unaffected by the cyanophage infection. Transient fluorescence induction and flash-induced delayed luminescence decay studies revealed that the inhibition may occur at the level of the secondary acceptor, Q(B) of photosystem II. In addition, the breakdown of D(1)-protein is inhibited, comparable to DCMU-induced protection of D(1)-protein turnover, in AS-1 infected cells.
252. **The effect of cyanophages on the growth and survival of *Lyngbya wollei*, *Anabaena flos aquae*, and *Anabaena circinalis*.** Monegue, R. L., Philips, E. J. (1991). *J. Aquat. Plant. Manage.* **29:88-93**.
253. **Roles of viral infection in organic particle flux.** Proctor, L. M., Fuhrman, J. A. (1991). *Marine Ecology Progress Series* **69:133-142**. Lack of information on the fate of particulate-associated microorganisms prompted this investigation of viruses (including bacteriophage or phage) and phage-infected cells in sinking particles from sediment traps. Sediment trap material from 30 to 400 m collected from the north Pacific Ocean during the 'VERTEX' cruises in 1980 to 1982 was examined by transmission electron microscopy. Viruses were present in all of the sinking particles examined except for those from one sample, of highly degraded algal cells or small fecal pellets, from 400 m. Viruses in the sinking particles often appeared aggregated. From 0.7 to 3.7% of the bacteria in sinking particles contained mature phage; from these data and limited information from pure cultures, we estimate that 2 to 37% of the particulate-associated bacteria may be killed by viral lysis. Many eukaryotic cells were also apparently infected with viruses, but none (100 cells observed) of the cyanobacteria or 'Chlorella-like' cells appeared infected. Viral lysis of bacteria associated with sinking particles and free-living bacteria may be causally linked and may play a role in dissolved organic carbon production and the dynamics of sinking particles. Viral lysis may have major implications for understanding cycling of material and energy in the ocean
254. **Analysis of marine picoplankton community by 16S ribosomal-RNA gene cloning and sequencing.** Schmidt, T. M., Delonge, E. F., Pace, N. R. (1991). *Journal of Bacteriology* **173:4371-4378**. The phylogenetic diversity of an oligotrophic marine picoplankton community was examined by analyzing the sequences of cloned ribosomal genes. This strategy does not rely on cultivation of the resident microorganisms. Bulk genomic DNA was isolated from picoplankton collected in the north central Pacific Ocean by tangential flow filtration. The mixed-population DNA was fragmented, size fractionated, and cloned into bacteriophage lambda. Thirty-eight clones containing 16S rRNA genes were identified in a screen of 3.2×10^4 recombinant phage, and portions of the rRNA gene were amplified by polymerase chain reaction and sequenced. The resulting sequences were used to establish the identities of the picoplankton by comparison with an established data base of rRNA sequences. Fifteen unique eubacterial sequences were obtained, including four from cyanobacteria and eleven from proteobacteria. A single eucaryote related to dinoflagellates was identified; no archaeobacterial sequences were detected. The cyanobacterial sequences are all closely related to sequences from cultivated marine *Synechococcus* strains and with cyanobacterial sequences obtained from the Atlantic Ocean (Sargasso Sea). Several sequences were related to common marine isolates of the gamma-subdivision of proteobacteria. In addition to sequences closely related to those of described bacteria, sequences were obtained from two phylogenetic groups of organisms that are not closely related to any known rRNA sequences from cultivated organisms. Both of these novel phylogenetic clusters are proteobacteria, one group within the alpha-subdivision and the other distinct from known proteobacterial subdivisions. The rRNA sequences of the alpha-related group are nearly identical to those of some Sargasso Sea picoplankton, suggesting a global distribution of these organisms.
255. **Promoter recognition by the RNA polymerase from vegetative cells of the cyanobacterium *Anabaena***

7120. Schneider, G. J., Lang, J. D., Haselkorn, R. (1991). *Gene* 105:51-60. The transcription start points (tsp) of seven genes of *Anabaena* 7120 were previously identified by S1 nuclease protection and primer extension experiments using RNA extracted from cells. In the present work, these tsp were confirmed, with one exception, by in vitro transcription using purified RNA polymerases of *Anabaena* 7120 and *Escherichia coli*, and crude extracts of *Anabaena* 7120 active in transcription. In all cases, the template for transcription consisted of closed circular plasmid DNA in which the putative promoter-containing fragment was cloned in front of a strong terminator, which resulted in defined 'pseudo-runoff' transcripts whose sizes correspond (with one exception) to those expected on the basis of the tsp determined for in vivo RNA. These results, together with others obtained with templates containing bacteriophage T4 or cyanophage N1 promoters, led to the conclusion that the principal *Anabaena* 7120 RNA polymerase prefers promoters whose sequence and spacing approximate that of the *E. coli* consensus promoter, and that the *Anabaena* 7120 genes expressed in vegetative cells, characterized to date, have relatively weak promoters

256. **Use of ultrafiltration to isolate viruses from seawater which are pathogens to marine phytoplankton.** Suttle, C. A., Chan, A. M., Cottrell, M. T. (1991). *Applied and Environmental Microbiology* 57:721-726. Viruses may be major structuring elements of phytoplankton communities, and hence important regulators of nutrient and energy flux in aquatic environments. In order to ascertain if viruses are potentially important in dictating phytoplankton community structure, it is essential to determine the extent to which representative phytoplankton taxa are susceptible to viral infection. We used a spiral ultrafiltration cartridge (30,000 MW cutoff) to concentrate viruses from seawater at efficiencies approaching 100 %. Natural virus communities were concentrated from stations in the Gulf of Mexico, a barrier island pass and a hypersaline lagoon (Laguna Madre), and added to cultures of potential phytoplankton hosts. By following changes in in-vivo fluorescence over time it was possible to isolate several viruses that were pathogens to a variety of marine phytoplankton, including a prasinophyte (*Micromonas pusilla*), a pennate diatom (likely *Navicula* sp.), a centric diatom (of unknown taxa), and a chroococcoid cyanobacterium (*Synechococcus* sp.). As well, we observed changes in fluorescence in cultures of a cryptophyte (*Rhodomonas* sp.) and a chlorophyte (*Nannochloropsis oculata*) which were consistent with the presence of viral pathogens. Although pathogens were isolated from all stations, all the pathogens were not isolated from every station. Filterability studies on the viruses infecting *Micromonas* and *Navicula* showed that the viruses were consistently infective after filtration through polycarbonate and glass-fiber filters, but were affected by most other filter types. Establishment of phytoplankton/pathogen systems will be important in elucidating the affect that viruses have on primary producers in aquatic systems.
257. **Circular dichroism studies of salt- and alcohol- induced conformational changes in cyanophage S-2L DNA which contains amino 2 adenine instead of adenine.** Vorlickova, M., Hejtmankova, I., Kypr, J. (1991). *Journal of biomolecular structure and dynamics* 9:81-85. DNA molecules containing AT pairs exhibit cesium cation specific conformational behavior. This specificity is shown to be cancelled with the title DNA, which not only concerns its conformational alterations in high-salt aqueous solutions but also the B-to-A transition induced by ethanol. S-2L DNA easily adopts the A-conformation in the presence of millimolar concentrations of CsCl which completely destabilize the A-conformation in calf thymus DNA. The present results demonstrate that the specific effects of cesium cations on DNA are connected with their binding to the AT pairs in the DNA minor groove
258. **The inhibitory effects of the extracts of Zingiber plants on the adsorption, growth, and replication of phage LPP-1 in cyanobacterium.** Jido, E. P. (1992). M.S. dissertation, Loyola University of Chicago.
259. **Effect of reproduction of cyanophages A-1, S-8K and LPP-3 on proteolysis processes in the cells of cyanobacteria.** Mendzhul, M. I., Koltukova, N. V., Lysenko, T. G., Shainskaya, O. A. (1992). *Mikrobiologichnyi Zhurnal* 54:90-95. The dynamics of proteolytic activity and formation of protein metabolism products (free aminoacids and peptides) in three virus-cell systems (*Anabaena variabilis* - A-1, *Synechococcus cedrorum*-S-8K, *Plectonema boryanum* - LPP-3) have been studied as affected by cyanophage infection. Proteolytic activity of the cell-free extracts of cyanobacteria is established to considerably change during the cyanophage development. Preparations isolated from the cells 1h after infection are the most active ones. Proteolytic activity of the cells 3h after the infection (period of intensive morphogenesis of virions) is almost commensurable with that of the noninfected cells. The level of proteolytic activity and the content of free aminoacids and peptides well correlate only in the system LPP-3-P. boryanum. Such an agreement was not revealed in two other-virus-cellular systems and this is, probably, connected with different specificity of proteinases, which perform degradation of cell proteins, and with the differences in the processes of the de novo synthesis of amino acids and proteins. Various ways and mechanisms, including proteolysis-performed by virus-induced proteinases, may be involved in formation of a pool of free aminoacids in different virus-cellular systems
260. **[The resistance of the DNA of cyanophage LPP-3 to the action of different restriction endonucleases].** Mendzhul, M. I., Syrchin, S. A., Rebutish, B. A., Averkiev, A. A., Busakhina, I. V. (1993). *Mikrobiologichnyi Zhurnal* 55:47-53. Data on the study of structure peculiarities of cyanophage LPP-3 DNA are presented in the work. The length of cyanophage DNA calculated by means of the enzymatic hydrolysis by restrictases is 40 ± 3.5 thou. pairs of bases. Cyanophage LPP-3 DNA was hydrolysed by more than 50 different restrictases. As a result of screening it was found out that the great number of restrictases, which recognized hexanucleotide sequences did not hydrolyze DNA of cyanophage LPP-3. A considerable deviation of the number of the observed sites of restriction from their theoretically expected number for restrictases Hae III and Cfr 131 was established. Restrictases-isoschisomeres with different sensitivity to the methylation of the recognition sites—Msp I, Hpa II and Sau 3A, MboI and DpnI were used to check the availability of methylated bases in LPP-3 DNA. Absence of methylated adenine in the site GATC and methylated cytosine in the second position of the site CCGG were established. The results obtained permit supposing that the expressed counterselection by the sites of recognition of many restriction endonucleases takes place in cyanophage LPP-3 DNA. It is supposed that apparently, this method of protection of its genome in LPP-3 is one of most important but the inconsiderable percentage of site-specific methylation of the virus DNA cannot be completely excluded
261. **Spontaneous and induced host range mutants of cyanophage N-1.** Sarma, T. A., Kaur, B. (1993). *Archives of Virology* 130:195-200. Optimal conditions for the induction of host-range mutants of cyanophage N-1 by acridine orange were established. Induced host-range mutants were isolated with a frequency of 0.1 to 4.0×10^{-5} over a

262. **Cyanophages and sunlight: A paradox.** Suttle, C. A., Chan, A. M., Chen, F., Garza, D. R. (1993). pp. 303-307 in Guerrero, R., Pedros-Alio, C. (eds.) *Trends in Microbial Ecology*. Spanish Society Microbiology, Barcelona.
263. **Marine cyanophages infecting oceanic and coastal strains of *Synechococcus*: Abundance, morphology, cross-infectivity and growth characteristics.** Suttle, C. A., Chan, A. M. (1993). *Marine Ecology Progress Series* 92:99-109. Eight different phycoerythrin- and phycocyanin-containing strains of *Synechococcus* and one strain of *Anacystis* were screened against 29 natural virus communities taken from 3 locations in south Texas coastal waters, at different times of the year. In addition, one sample was screened from Peconic Bay, New York. Cyanophages were detected in all samples, but the frequency with which they were detected and their abundance depended upon the strain of *Synechococcus* that was screened. Viruses that infected red *Synechococcus* strains were particularly common. In some instances, concentrations infecting a single *Synechococcus* strain were in excess of 10^5 ml^{-1} . The abundances of cyanophages were weakly correlated with temperature ($r^2 = 0.53$ to 0.70), although they occurred at all of the temperatures ($12\text{-}30.4^\circ\text{C}$) and salinities ($18\text{-}70$ ppt) that were screened. The seven cyanophages that were cloned belonged to the same three families of viruses that have been observed to infect freshwater cyanobacteria, namely the *Siphoviridae* (formerly *Styloviridae*), *Myoviridae* and *Podoviridae*. The cyanophage clones varied in host-specificity. For example, one clone infected a single *Synechococcus* strain of 12 that were tested, whereas, another infected 4 of 9 strains tested. Growth characteristics of one of the virus clones was determined for a single host. Photosynthesis in *Synechococcus* was not affected until near the onset of cell lysis and the virus burst cycle was complete about 17 h post-infection. The burst size was approximately 250 infective particles. The high abundance of cyanophages in the natural environment provides further evidence that viruses are probably important regulators of phytoplankton dynamics in marine systems.
264. **Resistance to co-occurring phages enables marine *Synechococcus* communities to coexist with cyanophages abundant in seawater.** Waterbury, J. B., Valois, F. W. (1993). *Applied and Environmental Microbiology* 59:3393-3399. Recent reports documenting very high viral abundances in seawater have led to increased interest in the role of viruses in aquatic environments and a resurgence of the hypothesis that viruses are significant agents of bacterial mortality. *Synechococcus* spp., small unicellular cyanobacteria that are important primary producers at the base of the marine food web, were used to assess this hypothesis. We isolated a diverse group of *Synechococcus* phages that at times reach titers of between 10^3 and 10^4 cyanophages per ml in both inshore and offshore waters. However, despite their diversity and abundance, we present evidence in support of the hypothesis that lytic phages have a negligible effect in regulating the densities of marine *Synechococcus* populations. Our results indicate that these bacterial communities are dominated by cells resistant to their co-occurring phages and that these viruses are maintained by scavenging on the relatively rare sensitive cells in these communities
265. **Isolation and molecular characterization of five marine cyanophages propagated on *Synechococcus* sp. strain WH7803.** Wilson, W. H., Joint, I. R., Carr, N. G., Mann, N. H. (1993). *Applied and Environmental Microbiology* 59:3736-3743. Five marine cyanophages propagated on *Synechococcus* sp. strain WH7803 were isolated from three different oceanographic provinces during the months of August and September 1992: coastal water from the Sargasso Sea, Bermuda; Woods Hole harbor, Woods Hole, Mass.; and coastal water from the English Channel, off Plymouth Sound, United Kingdom. The five cyanophage isolates were found to belong to two families, Myoviridae and Styloviridae, on the basis of their morphology observed in the transmission electron microscope. DNA purified from each of the cyanophage isolates was restricted with a selection of restriction endonucleases, and three distinguishably different patterns were observed. DNA isolated from Myoviridae isolates from Bermuda and the English Channel had highly related restriction patterns, as did DNA isolated from Styloviridae isolates from Bermuda and the English Channel. DNA isolated from the Myoviridae isolate from Woods Hole had a unique restriction pattern. The genome size for each of the Myoviridae isolates was ca. 80 to 85 kb, and it was ca. 90 to 100 kb for each of the Styloviridae isolates. Southern blotting analysis revealed that there was a limited degree of homology among all cyanophage DNAs probed, but clear differences were observed between cyanophage DNA from the Myoviridae and that from the Styloviridae isolates. Polypeptide analysis revealed a clear difference between Myoviridae and Styloviridae polypeptide profiles, although the major, presumably structural, protein in each case was ca. 53 to 54 kDa.
266. **A New *Synechococcus* Cyanophage from a Reservoir in Korea.** Kim, M., Choi, Y.-K. (1994). *Virology* 204:338-342. A unicellular cyanobacterium (*Synechococcus*) and its cyanophage were both isolated from a reservoir in Korea. Although morphologically similar to AS-1, the cyanophage differs from cyanophage AS-1 in some respects. The burst size in the light is approximately 100 plaque-forming units (PFU)/cell. Replication of the virus also occurs in the dark, releasing about 10% of the virus particles observed in the light. Na^+ is not necessary for adsorption.
267. **[Aspartate kinase complex of *Anabaena variabilis* during the early period of development of cyanophage A-1].** Koltukova, N. V., Kadyrova, G. Kh, Lysenko, T. G., Mendzhul, M. I. (1994). *Ukrainskyi biokhimichnyi zhurnal* 66:41-48. Aspartate kinase activity in cells of *A. variabilis* has been studied in the dynamics of development of virus infection. An early period of reproduction of cyanophage A-1 has been determined to be conjugated with the increase of biosynthesis of amino acids from aspartate family. Five isoenzymes of aspartate kinase were isolated and purified from *A. variabilis* cells during early development period of cyanophage A-1. Physicochemical properties and influence of amino acids of aspartate family on the activity of homogeneous isoenzymes have been studied. Retroinhibition effect was not observed in infected cyanobacteria cells, which probably enables one to increase 2-7 times the concentration of amino acids in a cell. Such an increase of the amino acids pool is apparently necessary for realization of viral genome strategy
268. **Isolation and characterization of temperature-sensitive mutants of cyanophage N-1.** Sarma, T. A., Singh, R. (1994). *Acta virologica English ed* 38:11-16. Optimal conditions for the induction of temperature-sensitive (ts) mutants of cyanophage N-1 were established after mutagenesis with N-methyl-N'-nitro-N-nitrosoguanidine (MNNG).

A treatment with MNNG (400 micrograms/ml) for 2 hrs at pH 8.0 induced ts-mutants at a maximum frequency of 1.46×10^{-3} . A characterization of 10 such ts-mutants with regard to adsorption, one-step growth and temperature-shift experiments with *Nostoc muscorum* as host bacterium led to the identification of temperature-sensitive steps in the phage multiplication at the restrictive temperature (37°C). All the mutants were found to be conditionally lethal at 37°C since they resumed growth upon shifting to 28°C

269. **Energetics of cyanophage N-1 multiplication in the diazotrophic cyanobacterium *Nostoc muscorum*.** Singh, S., Bhatnagar, A., Kashyap, A. K. (1994). *Microbios* 78:259-265. Cyanophage N-1 multiplication was investigated during the latent period of the virus, when $^{14}\text{CO}_2$ fixation was inhibited whereas respiratory O_2 uptake increased similar to 67% at 4 h after infection. A simultaneous decrease (70%) in the glycogen content of infected cells indicated its catabolic involvement. A chloramphenicol-sensitive rise in glucose-6-phosphate dehydrogenase activity as a result of N-1 infection partly explained the increase in aerobic respiration. The total ATP pool declined to 53% of the control while Ca^{2+} -dependent ATPase activity also declined (25%). In contrast, Mg^{2+} -dependent ATPase activity increased (80%) in comparison with uninfected cells. Results suggest that oxidative phosphorylation was more crucial in the control of cyanophage N-1 development than photophosphorylation under photoautotrophic growth conditions
270. **Isolation of a marine cyanophage infecting the marine unicellular cyanobacterium, *Synechococcus* sp. NKBG 042902.** Sode, K., Oozeki, M., Asakawa, K., Burgess, J. G., Matsunaga, T. (1994). *Journal of Marine Biotechnology* 1:189-192. A marine cyanophage that infects the marine unicellular cyanobacterium, *Synechococcus* sp. NKBG 042902, was isolated from seawater. This marine cyanophage possesses a tail with a contractile sheath, which shows two distinct shapes, and is a temperate narrow host range phage that can be induced by mitomycin C. It is distinct from the cyanophage AS-1, which infects the freshwater strain *Anacystis nidulans* R2, with regard to host range and restriction enzyme pattern, and is designated as mS-1 (marine *Synechococcus* infecting)
271. **Dynamics and distribution of cyanophages and their effect on marine *Synechococcus* spp.** Suttle, C. A., Chan, A. M. (1994). *Applied and Environmental Microbiology* 60:3167-3174. Cyanophages infecting marine *Synechococcus* were frequently very abundant and were found in every seawater sample along a transect in the western Gulf of Mexico, and during a 28 month period in Aransas Pass, Texas. In Aransas Pass their abundance varied seasonally with the lowest concentrations coincident with cooler water and lower salinity. Along the transect, viruses infecting *Synechococcus* strains DC2 and SYN48 ranged in concentration from a few hundred ml^{-1} at 97 m depth and 83 km offshore, to ca. $4 \times 10^5 \text{ ml}^{-1}$ near the surface at stations within 18 km of the coast. The highest concentrations occurred at the surface where salinity decreased from ca. 35.5 to 34 ppt and concentrations of *Synechococcus* were greatest. Viruses infecting strains SNC1, SNC2 and 838BG were distributed in a similar manner, but were much less abundant (< 10 to $< 5 \times 10^3 \text{ ml}^{-1}$). When *Synechococcus* exceeded ca. 10^3 ml^{-1} , cyanophage concentrations increased markedly (ca. 10^2 to $< 10^5 \text{ ml}^{-1}$), suggesting that there was a minimum host density required for efficient viral propagation. Data on the decay rate of viral infectivity (d ; d^{-1}) as a function of solar radiation (I ; $\text{mmol quanta m}^{-2} \text{ s}^{-1}$) was used to develop a relationship ($d = 0.2610 I - 0.00718$; $r^2 = 0.69$) for conservatively estimating the destruction of infectious viruses in the mixed layer of two offshore stations. Assuming that virus production balances losses, and burst size is 250, ca. 5-7 % of *Synechococcus* would be infected daily by viruses. Calculations based on contact rates between *Synechococcus* and infectious viruses produce similar results (5-14 %). Moreover, balancing estimates of viral production with contact rates for the most offshore station required that most *Synechococcus* be susceptible to infection, that most contacts result in infection and that the burst size be about 324 viruses per lytic event. In contrast, in nearshore waters where ca. 80 % of *Synechococcus* would be contacted daily by infectious cyanophages, only ca. 1 % of the contacts would have to result in infection, in order to balance the estimated virus removal rates. These results indicate that cyanophages are an abundant and dynamic component of marine planktonic communities and are likely responsible for lysing a small but significant portion of the *Synechococcus* population on a daily basis.
272. **A study on cyanophages inhibiting the growth of algae producing musty odor.** Goto, Y., Kitayama, M. (1995). *Water Supply* 13:263-266. Recently blue-green algae have grown in large amount in the Lake Biwa. Authors have carried out a study on the cyanophages, which use *Phormidium tenue* (P. tenue), blue-green alga that produce musty odor in the Lake Biwa, as host and inhibit their growth. The samples used consisted of the surface-layer water in the Lake or the surface running water in Kizu River and Katsura River. A plate culture test by using the double-layered agar method was used in order to detect cyanophages, and the generation of plaque was observed. And, in order to confirm that the cyanophages inhibit the growth of *P. tenue*, authors also realized a liquid culture test, and observed the growth characteristics of the host. For the plate culture test, three samples presented the formation of plaque. In the liquid culture test, the growth of *P. tenue* was found to have been inhibited in the three samples. But for all these three samples, *P. tenue* was not completely killed; therefore, these cyanophages were believed to be temperate phages. By using these cyanophages is expected to be able to inhibit the growth of *P. tenue* alone without inhibiting the growth of other algae
273. **Fluorescently labeled virus probes show that natural virus populations can control the structure of marine microbial communities.** Hennes, K. P., Suttle, C. A., Chan, A. M. (1995). *Applied and Environmental Microbiology* 61:3623-3627. Fluorescently stained viruses were used as probes to label, identify and enumerate specific strains of bacteria and cyanobacteria in mixed microbial assemblages. Several marine virus isolates were fluorescently stained with YOYO-1 or POPO-1 (Molecular Probes, Inc.) and added to seawater samples that contained natural microbial communities. Cells to which the stained viruses adsorbed were easily distinguished from non-host cells; typically, there was undetectable binding of stained viruses to natural microbial assemblages containing $> 10^6$ bacteria ml^{-1} , but to which host cells were not added. Host cells that were added to natural seawater were quantified with 99 ± 2 % efficiency using fluorescently labeled virus probes (FLVPs). A marine bacterial isolate (strain PWH3a) was introduced into natural microbial communities that were either supplemented with nutrients or untreated, and changes in the abundance of the isolate were followed using FLVPs. Simultaneously, the concentration of viruses that infected strain PWH3a was monitored by plaque assay. Following the addition of

PWH3a, viruses infecting this strain increased from undetectable levels ($<1 \text{ ml}^{-1}$ to $2.9 \times 10^7 \text{ ml}^{-1}$ and $8.3 \times 10^8 \text{ ml}^{-1}$ for the untreated and nutrient-enriched samples, respectively. The increase in viruses was associated with a collapse in populations of strain PWH3a from ca. 30% to 2% and 43% to 0.01% of the microbial communities in untreated and nutrient-enriched samples, respectively. These results clearly demonstrate that FLVPs can be used to identify and quantify specific groups of bacteria in mixed microbial communities. As well, the data show that viruses which are present at low abundances in natural aquatic viral communities can control microbial community structure.

274. **Direct counts of viruses in natural waters and laboratory cultures by epifluorescence microscopy.** Hennes, K. P., Suttle, C. A. (1995). *Limnology and Oceanography* 40:1050-1055. Epifluorescent microscopy was used to determine the abundance of viruses in samples from marine and freshwater environments and in laboratory cultures that were filtered onto 0.02- μm pore-size filters and stained with a cyanine-based dye (Yo-Pro-1). Estimates of viral abundance based on Yo-Pro stained samples were 1.2 to 7.1 times greater than estimates obtained using transmission electron microscopy (TEM). Moreover, the precision of the Yo-Pro based method was much greater than that for TEM (coefficient of variation 7 % versus 20 %, respectively). DNase treatment of samples did not result in lower numbers of particles that could be stained by Yo-Pro, suggesting that the fluorescence was not the result of nucleic acids associated with the surface of particles. These results indicate that the concentration of viruses in natural waters may be higher than previously recognized and imply that the TEM-based method significantly underestimates virus abundance. Virus abundances ranged from 10^7 to $< 10^8 \text{ ml}^{-1}$ in surface waters along a transect in the western Gulf of Mexico to 10^9 ml^{-1} in water overlying a submerged cyanobacterial mat. High counting efficiency, ease of preparation, modest equipment requirements and the possibility of preparing specimens for long-term storage, make the Yo-Pro based method ideal for routine environmental analysis.
275. **Viral abundance in aquatic systems: a comparison between marine and fresh waters.** Maranger, R., Bird, D. F. (1995). *Marine Ecology Progress Series* 121:1-3. In order to investigate the factors controlling viral abundance, 22 lakes in Quebec were surveyed. We measured viral and bacterial abundance, bacterial production, chlorophyll *a*, total phosphorus and DOC (dissolved organic carbon) concentrations. Regression models built with these data were compared to models based on literature data, which to date have been collected largely from marine sites. Positive empirical relationships were found between viral abundance and (1) chlorophyll *a* concentrations, (2) bacterial abundances, (3) bacterial production, and (4) total phosphorus concentration. There was little to no trend in the virus-to-bacteria ratio with increasing trophity. Analysis of covariance revealed significant differences between relations in marine and freshwater systems. The virus-to-bacteria ratio was significantly higher in freshwater (mode = 22.5) than marine environments (mode = 2.5), and there were significantly more bacteria per unit chlorophyll in our freshwater samples. We suggest that this difference is related to the increased dependence of freshwater bacteria on allochthonous material relative to marine systems, as well as the increased relative importance of photosynthetic cyanobacteria in lakes.
276. **[Effect of reproduction of the LPP-3 cyanophage on glutamate dehydrogenase and glutamine synthetase activity in the cyanobacterium *Plectonema boryanum*].** Mendzhul, M. I., Koltukova, N. V., Lysenko, T. G., Shainskaia, O. A., Perepelitsa, S. I. (1995). *Ukrainskyi biokhimichnyi zhurnal* 67:33-37. The effect of cyanophage LPP-3 reproduction on glutamate dehydrogenase and glutamine synthetase (GS) in *P. boryanum* cells have been studied. It was determined that the both reactions are intensified by 135% and 220%, accordingly. Isoenzymes of GS were purified from native and infected cell of cyanobacteria. Their physical-and-chemical properties are different. The cyanophage development probably causes specific modification of the cell enzymes
277. **[Alanine dehydrogenase of the cyanobacterium *Plectonema boryanum* in the early period of cyanophage LPP-3 development].** Perepelitsa, S. I., Koltukova, N. V., Mendzhul, M. I. (1995). *Ukrainskyi biokhimichnyi zhurnal* 67:47-52. It has been studied how reproduction of LPP-3 in *Plectonema boryanum* cells influences the alanine dehydrogenase activity. It has been found that immediately after the virus adsorption the enzyme activity falls by 50% and the anabolic reaction is blocked. Physicochemical properties of the enzyme vary as well. An infected cell has one isoenzyme-octamer with $\text{pI } 9.1-9.2$, $\text{pH-optimum by action } 9-10$, molecular weight about 27 kDa
278. **Characterization of TS-mutants of cyanophage N-1 by their inactivation by physical and chemical agents.** Sarma, T. A., Singh, R. (1995). *Acta Virologica* 39:65-68. The effect of temperature, ultraviolet (UV) light and ethylenediaminetetraacetic acid (EDTA) on the stability of cyanophage N-1, infecting the cyanobacterium *Nostoc muscorum* was studied. Complete inactivation of the phage occurred at 60°C in 6 mins. All the temperature-sensitive (ts) mutants exhibited faster inactivation at 50°C than the wild type. UV light readily inactivated the particles of the wild giving a survival of 3.44% at a dose of 60 secs. All the ts-mutants were found to be more sensitive to UV light than the wild type. 10^{-4} mol/l EDTA inactivated 40% of the wild type in 60 mins. $5 \times 10^{-4} \text{ mol/l}$ EDTA inactivated the wild type nearly completely within 2 mins, while a similar inactivation of ts-mutants required only 90 secs
279. **The diversity of bacteria, eukaryotic cells and viruses in an oligotrophic lake.** Corpe, W. A., Jensen, T. E. (1996). *Applied Microbiology and Biotechnology* 46:622-630. An in situ transmission electron microscopic study of biomass samples concentrated from oligotrophic lake water revealed a variety of virus-infected microbial cells and many free viruses and virus-like particles. The most abundant group of microorganisms in screened and filtered water-column samples were 2 $\mu\text{-m}$ or less in diameter, and included representatives of several oligotrophic genera, *Prosthecomicrobium*, *Ancyclobacter*, *Caulobacter* and *Hyphomicrobium*. Among the prokaryotic host cells, which included both heterotrophs and autotrophs, on the basis of electron microscope observations, approximately 17% were infected with bacteriophage or bore adherent phage particles on their surfaces. Several bacterial morphotypes were observed among the prokaryotic hosts. Water samples passed through a 20- $\mu\text{-m}$ Nitex screen allowed us to concentrate and examine the larger host cells as well, including several species of single-celled algae and two amoeba species. The infected algal cells included those *Chlorella*-like in appearance, photosynthetic flagellates and others that could not be positively identified. About one-third of the eukaryotic cells were infected by viruses that were larger (150-200 nm) and structurally more complex than bacteriophages (50-60 nm). None of the viruses have been isolated, but when 0.2 $\mu\text{-m}$ filtrate from a biomass sample was spotted onto lawns of four representative heterotrophs and a *Chlorella*, the clearing observed was taken as evidence of lysis. Cyanobacterial lawns showed no plaques. Thin sections of two amoeba showed food vacuoles containing what appeared to be virus particles of a type seen in certain prokaryotic and eukaryotic cells in the biomass.

280. [Phagolysates of cyanobacteria: their biocidal properties and use]. Gol'din, E. B., Mendzhul, M. I. (1996). *Mikrobiologichnyi Zhurnal* 58:51-58. Data on the biological activity of cyanobacterial phagolysates were obtained in experiments. They concern the organisms of different evolutionary level, such as some conventional pathogenic bacteria, plant and root parasitic nematodes and phytophagous insects. The authors suppose the specific mechanism of biocidal and inhibitory action of cyanobacteria and their phagolysates in respect to different living systems. These facts are very important for the elaboration of practical aspects of algal metabolites employment in agriculture and medicine
281. **Evidence that the hanA gene coding for HU protein is essential for heterocyst differentiation in, and cyanophage A-4(L) sensitivity of, *Anabaena* sp. strain PCC 7120.** Khudyakov, I., Wolk, C. P. (1996). *J. Bacteriol.* 178:3572-3577. The highly pleiotropic, transposon-generated mutant *Anabaena* sp. strain PCC7120 exhibits slow growth, altered pigmentation, cellular fragility, resistance to phage A-4(L), and the inability to differentiate heterocysts. Reconstruction of the transposon mutation in the wild-type strain reproduced the phenotype of the original mutant. Sequencing of the flanking DNA showed that the transposon had inserted at the beginning of a gene, which we call hanA, that encodes *Anabaena* HU protein. Mapping of the transposon insertion by pulsed-field gel electrophoresis showed that hanA is located at ca. 4.76 Mb on the physical map of the chromosome and is transcribed clockwise. Repeated subculturing of filament fragmentation, presumably because of one or more compensatory mutations; however, the mutant retained its A-4(L) super(r) Het super(-) phenotype. The mutation in strain be complemented by a fragment of wild-type DNA bearing hanA as its only open reading frame
282. **Wastewater treatment and elimination of pathogens: new prospects for an old problem.** Lopez-Pila, J. M., Dizer, H., Dorau, W. (1996). *Microbiologia* 12:525-536. Although the development of wastewater treatment technology is more than one hundred years old, most wastewater treatment plants existing today do not eliminate pathogens satisfactorily. Even in highly developed nations, receiving waters, serving in many cases as drinking water resources, are contaminated with pathogens. Surface waters also contain large concentration of phosphate due to long lasting wastewater discharges. Cyanobacteria and algal overgrowth is the consequence. Present drinking water technology only partially overcomes the pollution; it can not be ruled out that drinking water originating from polluted resources contains pathogens. This situation frequently goes on unnoticed because current indicator organisms are not representative for all pathogens. As studies have shown that small concentrations of pathogens also pose a risk for the consumer health, this state of affairs is a matter of concern. Microfiltration technology is able to significantly eliminate bacteria and protists from wastewater. Viruses, although smaller than the pore size of the filters, are reduced too because, in wastewater, they are frequently bound to larger particles. If the microfiltration of wastewater is preceded by the addition of coagulants for the precipitation of phosphate, the precipitate will be retained by the filter. The effluent obtained contains very low concentrations of phosphate. As viruses also adsorb to the precipitate, the amount of viruses eliminated increases and with increasing amounts of coagulant they become undetectable
283. **Occurrence of a temperate cyanophage lysogenizing the marine cyanophyte *Phormidium persicinum*.** Ohki, K., Fujita, Y. (1996). *J. Phycol.* 32:365-370. A temperate cyanophage was found to lysogenize the marine cyanophyte *Phormidium persicinum* (Reinke) Gom. (Provasoli strain). The lytic cycle was induced by the addition of mitomycin C or by brief illumination with ultraviolet light. The lytic process observed under the electron microscope showed that phage particles appeared in a nucleoplasm region 15 to 24 h after the addition of mitomycin C. The induction of the lytic process occurred simultaneously in almost all cells of every trichome. Matured phage particles were released to the medium 30 to 50 h after the addition of mitomycin C. Phage particles isolated from algal lysates had a polyhedral head (about 40 nm in diameter) with a long (about 300 nm) and noncontractile tail. The most abundant protein, presumably a structural protein, had an apparent molecular mass of about 38 kDa. The genome size estimated from restriction analysis was about 50 kbp. Phage DNA was digested with several restriction endonucleases including Sau3AI and DpnI. However, MboI failed to digest the phage DNA, suggesting that the phage DNA is highly methylated. Southern blot analysis suggested that some part of the phage was in the lytic cycle in algal cells growing under normal conditions. A possible role of temperate cyanophages in the regulation of cyanophyte populations in the marine environment is discussed.
284. **Temporal and spatial dynamics of *Synechococcus* spp. and *Micromonas pusilla* host-viral systems.** Rodda, K. M. (1996). M.S. dissertation, University of Texas at Austin.
285. **Unusual contribution of 2-aminoadenine to the thermostability of DNA.** Sagi, J., Szakonyi, E., Vorlickova, M., Kypr, J. (1996). *Journal of biomolecular structure and dynamics* 13:1035-1041. The poly(dA-dU) and poly(dI-dC) duplexes have very similar thermostabilities (T_m). This similarity extends also to the pyrimidine 5-methyl group-containing poly(dA-dT) and poly(dI-m5dC). The differences between chemical structures of the A:U and I:C or the A:T and I:m5C base-pairs seem to be unimportant for the thermostability of the DNA. However, on the insertion of an amino group into position 2 of the purines the similarities disappear. Thermostabilities of poly(n2dA-dU) and poly(dG-dC) as well as the poly(n2dA-dT) and poly(dG-m5dC) are radically different. This is also the case with their other 5-substituted pyrimidine-containing derivatives, the 5-ethyl, 5-n-butyl and 5-bromo analogues. The G:C-based polynucleotides are more stable by an average of 40°C than the n2A.U-based ones. Poly(dA,n2dA-dT)-s containing various proportions of A and n2A as well as the natural DNA of S-2L cyanophage that contains n2A bases instead of A were also studied. It was found that dependence of T_m on the n2A-content was non-linear and that the lower T_m is not the consequence of a particular nucleotide sequence. The possible structural reasons for the lower thermostabilization of these B-DNAs by the n2A:T base-pair as compared to the G:C are discussed
286. **The effect of cyanophages on *Synechococcus* spp. during a bloom in the western Gulf of Mexico.** Suttle, C. A., Chan, A. M., Rodda, K. M., Short, S. M., Weinbauer, M. G., Garza, D. R., Wilhelm, S. W. (1996). *EOS* 76 (suppl.):OS207-OS208.
287. **The effects of nutrient limitation on the kinetics of cyanophage infection of the oceanic picoplankter *Synechococcus* sp. WH7803.** Wilson, S. H., Carr, N. G., Mann, N. H. (1996). *J. Phycol.* 32:506-516. Phycoerythrin-containing *Synechococcus* species are considered to be major primary producers in nutrient-limited gyres of subtropical and tropical oceanic provinces, and the cyanophages that infect them are thought to influence marine biogeochemical cycles. This study begins an examination of the effects of nutrient limitation on the dynamics of cyanophage/*Synechococcus* interactions in oligotrophic environments by analyzing the infection kinetics of cyanophage strain S-P1kf2 (Cyanomyoviridae isolated from coastal water off Plymouth, UK) propagated on

Synechococcus sp. WH7803 grown in either phosphate-deplete or phosphate-replete conditions. When the growth of *Synechococcus* sp. WH7803 in phosphate deplete medium was followed after injection with cyanophage, an 18-h [??] delay in cell lysis was observed when compared to a phosphate-replete control. *Synechococcus* sp. WH7803 cultures grown at two different rates (in the same nutritional conditions) both lysed 24 h postinfection, ruling out growth rate itself as a factor in the delay of cell lysis. One-step growth kinetics of S-PAV12 propagated on host *Synechococcus* sp. WH7803, grown in phosphate deplete and-replete media, revealed an apparent 80% decrease in burst size in phosphate-deplete growth conditions, but phage adsorption kinetics of S-PM2 under these conditions showed no differences. These results suggested that the cyanophages established lysogeny in response to phosphate-deplete growth of host cells. This suggestion was supported by comparison of the proportion of infected cells that lysed under phosphate-replete and-deplete conditions, which revealed that only 9.3% of phosphate-deplete infected cells lysed in contrast to 100% of infected phosphate-replete cells. Further studies with two independent cyanophage strains also revealed that only approximately 10% of infected phosphate-deplete host cells released progeny cyanophages. These data strongly support the concept that the phosphate status of the *Synechococcus* cell will have a profound effect on the eventual outcome of phage-host interactions and will therefore exert a similarly extensive effect on the dynamics of carbon flow in the marine environment.

288. **The effect of phosphate status on the kinetics of cyanophage infection in the oceanic cyanobacterium *Synechococcus* sp. WH7803.** Wilson, W. H., Carr, N. G., Mann, N. H. (1996). *J. Phycol.* 32:506-516. Phycoerythrin-containing *Synechococcus* species are considered to be major primary producers in nutrient-limited gyres of subtropical and tropical oceanic provinces, and the cyanophages that infect them are thought to influence marine biogeochemical cycles. This study begins an examination of the effects of nutrient limitation on the dynamics of cyanophage/*Synechococcus* interactions in oligotrophic environments by analyzing the infection kinetics of cyanophage strain S-PM2 (*Cyanomyoviridae* isolated from coastal water off Plymouth, UK) propagated on *Synechococcus* sp. WH7803 grown in either phosphate-deplete or phosphate-replete conditions. When the growth of *Synechococcus* sp. WH7803 in phosphate-deplete medium was followed after infection with cyanophage, an 18-h delay in cell lysis was observed when compared to a phosphate-replete control. *Synechococcus* sp. WH7803 cultures grown at two different rates (in the same nutritional conditions) both lysed 24 h postinfection, ruling out growth rate itself as a factor in the delay of cell lysis. One-step growth kinetics of S-PM2 propagated on host *Synechococcus* sp. WH7803, grown in phosphate-deplete and-replete media, revealed an apparent 80% decrease in burst size in phosphate-deplete growth conditions, but phage adsorption kinetics of S-PM2 under these conditions showed no differences. These results suggested that the cyanophages established lysogeny in response to phosphate-deplete growth of host cells. This suggestion was supported by comparison of the proportion of infected cells that lysed under phosphate-replete and-deplete conditions, which revealed that only 9.3% of phosphate-deplete infected cells lysed in contrast to 100% of infected phosphate-replete cells. Further studies with two independent cyanophage strains also revealed that only approximately 10% of infected phosphate-deplete host cells released progeny cyanophages. These data strongly support the concept that the phosphate status of the *Synechococcus* cell will have a profound effect on the eventual outcome of phage-host interactions and will therefore exert a similarly extensive effect on the dynamics of carbon flow in the marine environment
289. **Growth and phage resistance of *Anabaena* sp. strain PCC 7120 in the presence of cyanophage AN-15.** Mole, R., Meredith, D., Adams, D. G. (1997). *Journal of Applied Phycology [J. Appl. Phycol.]* 9:339-345. The cyanophage AN-15 was found to have a requirement for either 1 mM calcium or 1 mM magnesium ions to maintain viral stability, whereas 1 mM calcium ions alone were essential for the infection process to proceed in *Anabaena* sp. strain PCC 7120. Following prolonged incubation, phage-resistant cells were detected at a high frequency (approximately 10^{-5}) in lysates, as either renewed growth in liquid cultures, or as colonies in confluent lysed lawns. Southern hybridisation failed to detect AN-15 DNA in any of the resistant strains, implying that resistance is unlikely to be due to the presence of temperate phages. A high rate of spontaneous mutation is therefore likely to be the cause of resistance. Two classes of resistant cells were identified; those in which AN-15 failed to attach to host cells, and those in which attachment occurred, but subsequent replication was defective. However, it was possible to overcome phage resistance by the isolation of spontaneous mutants of AN-15, capable of infecting phage-resistant cells. These observations imply that if cyanophages are to be assessed as a means of controlling cyanobacterial blooms in freshwater bodies, the ionic (notably calcium) concentration of the water must be considered, together with the possible need to employ alternative cyanophage strains if resistance to the original one arises
290. **Characterization of host-range mutants of cyanophage N-1.** Sarma, T. A., Kaur, B. (1997). *Acta Virologica* 41:245-250. Fifteen host-range (h) mutants of cyanophage N-1 were characterized with reference to their efficiency of plating, time of appearance, morphology and size of plaques on *Nostoc muscorum* and its three phage-resistant (Nm 1/N-1, Nm 2/N-1 and Nm 8/N-1) mutants. While phage N-1 did not adsorb to the three phage-resistant mutants, the h mutants differed one from the other in having lower or higher adsorption rate constants on *N. muscorum* or the phage-resistant mutants. The inability of majority of h mutants isolated on Nm 1/N-1 to grow in Nm 8/N-1 was shown to be due to a failure of adsorption. The h mutants also differed one from the other in their reversion (back mutation) frequencies. The lethal doses (LD_{37}) required to kill 37% of free phage particles after UV-irradiation, heating and ethylenediamine tetraacetate (EDTA) treatment greatly varied. Most of the h mutants were found to be considerably more sensitive to UV and thermic inactivation than N-1 while they were resistant to EDTA. The h mutants except five of them were unable to multiply at 40°C. The significance of these features is discussed
291. **Viruses in aquatic ecosystems. A review.** Sime-Ngando, T. (1997). *Annee Biologique* 36:181-210. Even though the contribution of water ecosystems for disseminating enteric viral pathogens has been known for decades, the importance of wild virions in structuring aquatic communities and food webs has only come to light relatively recently. Evidences of viral infections in both pro- and eukaryotic phytoplankton, as well as in heterotrophic bacterio- and protozooplankton, have recently brought marine biologists to question the impact of viroplankton on processes such as (1) the mortality of microorganisms, (2) the nutrition of heterotrophic protists, (3) the promotion of genetic material exchanges among microbial populations, (4) the maintenance of species diversity, (5) the induction of planktonic aggregates, and (6) the cycling of organic matter in aquatic ecosystems. In this paper, all these points are reviewed and discussed, in the light of recent contributions to the ecology of aquatic viruses, for evidence of the impact of viruses on both steady and non-steady state processes in fresh- and salt-waters. ¶ Viruses are ubiquitous, abundant and dynamic components of pelagic ecosystems. They are, undoubtedly, more diversified than the phage-like morphotypes that are generally characterized by the presence of an icosahedral head and a tail, via

observations under electron microscope. The diversity of bacteriophage viruses is further enhanced by the genetic viewpoint, and likely implies the diversity of sensible hosts. Genetically related marine phages are likely widely distributed in the space (i. e. without significant geographical segregation), suggesting prevalence of a reduced competition among viral "populations", and that the main biotic limiting factor for a viral "species" production is the density of the sensible host. Some viral "species", known from marine systems, typically harbor knob-like projections and long spines (i. e. previously not noted from non-aquatic habitats), which are suggested to increase the efficiency of hitting a specific host, especially in oligotrophic waters. Despite the general scarcity of viral isolates that lyse ciliated protozooplankton and metazoan zooplankton, it is becoming increasingly evident that most of the pelagic pro- and eukaryotic organisms are subject to infectious attacks from ambient "free-living" viruses. ¶ Quantitatively, recent total counts from the plankton generally fall in between 10^4 and 10^8 viruses ml^{-1} , with seasonal high densities in spring and summer, and a lowering tendency in abundance from the coastal to the open marine systems, and from the surface to the depth waters, likely in relation to temperature and the organic matter load. It was recently shown that lytic infection, rather than induction of lysogeny, is responsible for the majority of bacteriophage production in the plankton, especially in the coastal marine and surface waters and during blooming events, where the threshold-product level of virus x bacteria numbers of greater than or equal to 10(12) for the start of a viral-lytic activity is generally achieved. Closed linear relationships have been reported between viroplankton dynamics and bacteria, algae and nutrients. Because of the preponderance of allochthonous organic matter and cyanobacterial cells in lakes as compared to oceanic systems, the virus-to-bacteria ratio in lakes are significantly higher than in marine systems, although there is little trend in the virus-to-bacteria ratio with increasing trophicity, and despite the occurrence of more bacteria per unit chlorophyll in lake samples. ¶ The functional impact of virions on the structure and metabolism of planktonic communities is more important than their quantitative importance, as viruses represent only a minor fraction of the total planktonic biomass. The viral-induced mortality of microbial communities in marine systems is estimated to represent about 30 and < 10 percent of the mortality of bacterio- and phytoplankton, respectively. Based on one study, the contribution of viruses to bacterial mortality in lakes seems considerably lower than in marine systems. The greatest impact of viruses on aquatic communities is likely through hazardous (i.e. non-steady state) processes which are difficult to quantify, such as the promotion of genetic material exchanges among populations and the maintenance of species diversity. The lytic pressure from virulent viruses may act as a "nonstop" inductor of modifications in the genetic heritage of host-organisms, thereby increasing the potential of these hosts to share their habitat with homologous species, i.e. with similar nutritional requirements. ¶ It has recently been shown that lysates resulting from phage infection can cause a significant increase in metabolic activity of noninfected bacterioplankton community, but the growth efficiency of these noninfected hosts decreased in the presence of viruses, likely because of the increase in bacterial energy demand associated with extracellular degradation of polymers that are prevalent in viral lysates. This seems to verify the hypothesis on a substantial contribution of the lytic activity from viruses, to the cycling of organic matter in aquatic systems. Viral lytic production may indeed (1) reduce the bacterial biomass contribution to the transfer of metabolic energy on to higher-order consumers, (2) result in an increase of bacterial secondary production in the absence of an increase in the ambient primary production, and (3) increase competition between bacterial exo- or ectoenzyme activity and the feeding activity of protozoa on high molecular weight polymers (including viruses), although ingestion of viruses by protists seems to be of less importance in the carbon flows through the microbial food web in pelagic systems. ¶ However, almost all studies on the ecology of pelagic viruses are done during a limited period of year, mainly in marine waters situated in temperate zones. The data discussed in this paper are thus to be considered as preliminary data. Nevertheless, viruses undoubtedly influence to various degrees the biological processes in aquatic ecosystems. The quantitative assessment of their functional impact is thus required for incorporation into models that simulate flows of matter, nutrients and energy in aquatic systems. This task is to be included on the agenda of both marine and freshwater biologists, as a high priority concern for the near future.

292. **Induction of a temperate marine cyanophage by heavy metal. Sode, K., Oonari, R., Oozeki, M. (1997). *Journal of Marine Biotechnology* 5:178-180.** The activity of a temperate marine cyanophage, ms-1, of *Synechococcus* sp. NKBG 042902, was induced by Cu^{2+} . This induction was specific to Cu^{2+} and dependent upon Cu^{2+} concentration. Cr, Pb, Co, and Zn were not effective as inducers. These results suggested that Cu^{2+} is a significant inducer for lysogenic cyanobacterial cells and consequently will be a potential trigger for changes in the cyanobacterial population in the marine environment.
293. **Lipopolysaccharide dependence of cyanophage sensitivity and aerobic nitrogen fixation in *Anabaena* sp. strain PCC 7120. Xu, X, Khudyakov, I, Wolk, C. P. (1997). *J. Bacteriol.* 179:2884-2891.** Fox super(-) mutants of *Anabaena* sp. strain PCC 7120 are unable to fix dinitrogen in the presence of oxygen. A fragment of the DNA of *Anabaena* sp. was cloned by complementation of a spontaneous Fox super(-), cyanophage-resistant mutant, R56, and characterized. Random insertion of transposon Tn5 delimited the complementing DNA to a 0.6-kb portion of the cloned fragment. Sequencing of this region and flanking DNA showed one complete open reading frame (ORF) similar to the gene rfbP (undecaprenyl-phosphate galactosephosphotransferase) and two partial ORFs similar to genes rfbD (GDP-D-mannose dehydratase) and rfbZ (first mannosyl transferase), all of which are active in the synthesis of the O antigen unit of the lipopolysaccharide (LPS) component of the outer membrane of gram-negative bacteria. In a transposon (Tn5-1087b)-induced, Fox super(-), cyanophage-resistant mutant, B14, the transposon was found within the same rfbP-like ORF. The three ORFs were insertionally inactivated with the omega cassette or with Tn5::omega. Only the insertions in the rfbZ- and rfbP-like ORFs led to resistance to cyanophages A-1(L) and A-4(L) and to a Fox super(-) phenotype. Electrophoretic analysis showed that interruption of the rfbZ- and rfbP-like ORFs resulted in a change in or loss of the characteristic pattern of the lengths of the LPS, whereas interruption of the rfbD-like ORF merely changed the distribution of the lengths of the LPS to one with a greater prevalence of low molecular weights. According to electron microscopy, interruption of the rfbP-like ORF may have led to aberrant deposition of the layers of the heterocyst envelope, resulting in increased leakage of oxygen into the heterocyst. The results suggest that modified LPS may prevent cyanophage infection of *Anabaena* sp. vegetative cells and the formation of a functional heterocyst envelope.
294. **Dissolved esterase activity as a tracer of phytoplankton lysis: Evidence of high phytoplankton lysis rates in the northwestern Mediterranean. Agustí, S., Satta, M. P., Mura, M. P., Benavent, E. (1998). *Limnology and Oceanography* 43:1836-1849.** Phytoplankton cell lysis is perceived to be an important loss process in the sea, although a quantification of this process has proved elusive. A recently developed method, based on the measurement of dissolved esterase activity (EA), was used to estimate the release of esterases following phytoplankton cell lysis in an effort to evaluate the importance of this process as a loss factor in the summer

phytoplankton of the northwestern Mediterranean Sea. Implicit in this method was the assumption that the lysis of phytoplankton cells caused these enzymes to be released to the medium. This assumption was tested by analyzing the presence and release of esterases by marine bacteria, heterotrophic flagellates, and heterotrophic ciliates, all isolated from the Blanes Bay (northwestern Mediterranean, Spain), and by phytoplankton grown in culture (*Synechococcus elongatus*, *Dunaliella* sp., *Chlorella* sp., *Phaeodactyllum tricornutum*, and *Chaetoceros decipiens*). The dissolved EA found during the growth, stationary, and decay phases of microheterotrophs (bacteria, flagellate, and ciliate) was negligible when compared to that found for phytoplanktonic cultures. Differences in cell volume explained the differences in cell EA among the organisms, but heterotrophs showed lower cell EA (10-50-fold) than phytoplanktonic cells of similar cell size. These results support the assumption that microheterotrophs do not contribute significant amounts of EA to the dissolved pool, allowing the use of the method to estimate phytoplankton lysis. Independent estimates of cell loss in phytoplankton cultures, derived from cell cycle analysis, confirmed the estimates of cell lysis obtained from the measurement of dissolved EA. ¶ During the study conducted in the Mediterranean Sea, the water column was strongly stratified, showing a deep (40-55 m) chlorophyll a (Chl a) maximum (DCM; $1.25 \pm 0.09 \mu\text{g liter}^{-1}$) and low surface Chl a concentrations ($0.09 \pm 0.008 \mu\text{g liter}^{-1}$).

Phytoplankton lysis rates ranged between 0.026 d^{-1} and 1.9 d^{-1} , and they declined significantly with depth; the fastest rates were found in surface waters and the slowest ones at the DCM. Despite the fast gross growth rates of surface phytoplankton (as calculated from phytoplankton biovolume and oxygen production), the calculated lysis rates represented a considerable proportion of gross phytoplankton growth rate (50%) at the surface, whereas they were comparatively less important at the DCM (7%). These results provide strong evidence that phytoplankton lysis can be an important loss factor in the surface waters of this stratified, oligotrophic sea. Phytoplankton lysis could provide the loss factor needed to explain the low phytoplankton biomass despite fast growth and low grazing rates in the northwestern Mediterranean surface waters. The high lysis rate of phytoplankton in surface waters represents an important path by which primary production may fuel the growth of microheterotrophic organisms, consistent with the high respiration rate of the surface community examined. The conclusion that phytoplankton lysis rates can occur at rates high enough to influence food web dynamics and biogeochemical cycles in the oligotrophic ocean should stimulate research on this largely neglected loss factor in phytoplankton ecology.

295. **Occurrence of a sequence in marine cyanophages similar to that of T4 gp20 and its application to PCR-based detection and quantification techniques.** Fuller, N. J., Wilson, W. H., Joint, I. R., Mann, N. H. (1998). *Applied and Environmental Microbiology* **64**:2051-2060. Viruses are ubiquitous components of marine ecosystems and are known to infect unicellular phycoerythrin-containing cyanobacteria belonging to the genus *Synechococcus*. A conserved region from cyanophage genome was identified in three genetically distinct cyanomyoviruses, and a sequence analysis revealed that this region exhibited significant similarity to a gene encoding a capsid assembly protein (gp20) from the enteric coliphage T4. The results of a comparison of gene 20 sequences from three cyanomyoviruses and T4 allowed us to design two degenerate PCR primers, CPS1 and CPS2, which specifically amplified a 165-bp region from the majority of cyanomyoviruses tested. A competitive PCR (cPCR) analysis revealed that cyanomyovirus strains should be accurately enumerated, and it was demonstrated that quantification was log-linear over ca. 3 orders of magnitude. Different calibration curves were obtained for each of the three cyanomyovirus strains tested; consequently, cPCR performed with primers CPS1 and CPS2 could lead to substantial inaccuracies in estimates of phage abundance in natural assemblages. Further sequence analysis of cyanomyovirus gene 20 homologs would be necessary in order to design primers which do not exhibit phage-to-phage variability in priming efficiency. It was demonstrated that PCR products of the correct size could be amplified from seawater samples following 100x concentration and even directly without any prior concentration. Hence, the use of degenerate primers in PCR analysis of cyanophage populations should provide valuable data on the diversity of cyanophages in natural assemblages. Further optimization of procedures may ultimately lead to a sensitive assay which can be used to analyze natural cyanophage populations both quantitatively (by cPCR) and qualitatively following phylogenetic analysis of amplified products.
296. **The effect of cyanophages on the mortality of *Synechococcus* spp. and selection for UV resistant viral communities.** Garza, D. R., Suttle, C. A. (1998). *Microbial Ecology* **36**:281-292. Viruses that cause lysis of *Synechococcus* spp. are present throughout the year in the western Gulf of Mexico. The effect of sunlight on loss rates of cyanophage infectivity was determined by incubating natural cyanophage communities and cyanophage isolates (strains S-PWM1 and S-PWM3) in UV-transparent bags at the surface, and at depth, on several occasions throughout the year. Decay rates of infectivity of natural cyanophage communities at the surface, at Port Aransas, Texas, USA, ranged from undetectable to 0.335 h^{-1} , with the highest rates occurring during the summer. During the spring and winter, decay rates of cyanophage isolates and natural cyanophage communities were generally similar, but during summer, decay rates of isolates were as much as twofold higher than the natural communities. In situ incubations at two offshore stations during a bloom of *Synechococcus* spp. produced decay rates of 0.53 and 0.75 d^{-1} , integrated over the mixed layer and averaged over 24 h. Based on a burst size of 81 viruses produced per lysed cell (measured for natural cyanobacterial communities in the Gulf of Mexico), cyanophages imposed mortality rates of 1 and 8%, respectively, on *Synechococcus* spp. In contrast, in nearshore incubations in the winter and spring, cyanophages were responsible for removing <1% of the *Synechococcus* cells on a daily basis. Only an estimated 2 to 3% of contacts led to viral infections (based on theoretical contact rates between host cells and cyanophages, and estimates of cyanophage mortality), regardless of the time of year or concentrations of viruses and hosts. These results indicate that natural cyanophage communities tolerate damage by solar radiation better in summer than in winter. Moreover, net decay rates of cyanophage infectivity in sunlight were similar, whether host cells were present or not, indicating that detectable cyanophage production did not occur during daytime in situ incubations.
297. **[Key enzymes of biosynthesis of amino acids of the glutamic series in the virus-cell system *Anabaena variabilis* + A-1].** Mendzhul, M. I., Lysenko, T. G., Koltukova, N. V. (1998). *Ukrainskyi biokhimichnyi zhurnal* **70**:16-22. The influence of cyanophage A-1 reproduction on glutamate dehydrogenase (GDG) and glutamine synthetase (GS) in *A. variabilis* cells was studied. It was determined that the both enzymes are intensified by 70% and 30%, accordingly. Isoenzymes of GDG and GS were isolated from native and infected cells of cyanobacteria, they had various physicochemical properties. It is concluded that cyanophage development causes the specific modification of cell enzymes
298. **Principles of virus-directed regulation of formation of the dynamic system virus-cell (problems, methodology and prospects of cyanophagia).** Mendzhul, M. I., Lysenko, T. G., Koltukova, N. V., Syrchin, S.

A., Sukhanov, S. N. (1998). *Mikrobiologichnyi Zhurnal* 60:66-78. Dynamics of virus-directed regulation of formation of the complex virus - cell has been studied on the example of the system cyanophage-cyanobacterium. It is shown that in the process of virus reproduction the host-cell loses its own genetic apparatus, system of regulation of biosynthetic processes, reproductive ability and other functions of vital importance. As a matter of fact the formed virus - cell complex turns into powerful generator of nucleotides and amino acids for nonlimited synthesis of virus nucleic acids, proteins and morphogenesis of virions. The question is discussed concerning the possibility of the use of the system cyanophage cyanobacterium as the experimental models for development of functional unified model of productive infection, effective methods of prophylaxis and therapy of virus infections as well as the decision of various biotechnological problems

299. **The role of sunlight in the removal and repair of viruses in the sea.** Wilhelm, S. W., Weinbauer, M. G., Suttle, C. A., Jeffrey, W. H. (1998). *Limnology and Oceanography* 43:586-592. We investigated the in situ destruction rates of marine viral particles as well as the decay rates of infectivity for viral isolates along an similar to 400-km transect from oligotrophic offshore waters to productive coastal waters in the Gulf of Mexico. Light-mediated decay rates of viral infectivity averaged over the solar day ranged from 0.7 to 0.85 h super⁻¹ in surface waters at all stations and decreased with depth in proportion to the attenuation of UVB (305 nm). The destruction rates of viral particles also decreased with depth, although the rates of particle destruction were only 22-61% of infectivity when integrated over the mixed layer. The rates of viral particle destruction indicated that at three of four stations 6-12% of the daily bacterial production would have to be lysed in order to maintain ambient viral concentrations. At the fourth station, where there was a dense bloom of *Synechococcus* spp. and the mixed layer was shallower, 34-52% of the daily bacterial production would have to be lysed. A comparison of the difference between destruction rates of viral particles and infectivity integrated over the depth of the mixed layer implies that host-mediated repair must have restored infectivity to 39-78% of the sunlight-damaged viruses daily. The calculated frequency of contacts between viral particles and bacterial cells that resulted in infection (contact success) ranged from similar to 18 to 34% in offshore waters, where the frequency of contacts between viruses and bacteria was much lower, to similar to 1.0% at the most inshore station, where contact rates are much higher. This suggests that in offshore waters bacterial communities are less diverse, and that there is less selection to be resistant to viral infection. This paper provides a framework for balancing viral production, destruction, and light-dependent repair in aquatic viral communities.
300. **Population dynamics of phytoplankton and viruses in a phosphate-limited mesocosm and their effect on DMSP and DMS production.** Wilson, W. H., Turner, S., Mann, N. H. (1998). *Estuarine, Coastal and Shelf Science* 46:49-59. The effect of phosphate limitation on viral abundance, phytoplankton bloom dynamics and production of dimethylsulphoniopropionate (DMSP) and dimethyl sulphide (DMS) was investigated in seawater mesocosm enclosures, in a Norwegian fjord, during June 1995. Daily estimates of viral concentrations, based on transmission electron microscope (TEM) counts, varied on an apparently random basis in each of the enclosures. A large *Synechococcus* spp. bloom developed in an enclosure which was maintained at a high N:P ratio, simulating phosphate-deplete growth conditions. Following phosphate addition to this enclosure, there was a large increase in estimated virus numbers shortly before an apparent collapse of the *Synechococcus* bloom. It is tentatively suggested that lysogenic viruses were induced following phosphate addition to the phosphate-limited enclosures, and that these observations add to a growing body of evidence which supports the hypothesis that nutrient availability may be responsible for the switch between lysogeny and lytic production. High DMS concentrations and viral numbers were observed on the demise of the flagellate (predominantly *Emiliania huxleyi*) and diatom blooms, but overall there was no significant correlation. Highest concentrations of DMSP were associated with blooms of *E. huxleyi*, for which an intracellular concentration of 0.5 pg cell⁻¹ (SD, 0.06) was calculated. Good correlation of DMSP with *Synechococcus* spp. cell numbers was observed, suggesting that these species of picoplankton may be significant producers of DMSP. No effects of phosphate limitation on DMS and/or DMSP production were evident from the data.
301. **Seasonal changes in densities of cyanophage infectious to *Microcystis aeruginosa* in a hypereutrophic pond.** Manage, P., Kawabata, Z., Nakano, S. (1999). *Hydrobiologia*. 411:211-216. Seasonal changes in densities of cyanophages infectious to *Microcystis aeruginosa* were studied in a hypereutrophic pond from March 1997 to January 1998 to elucidate the potential impact of the cyanophage on *M. aeruginosa* mortality. Densities of *M. aeruginosa* ranged between 1.8 X 10⁴ and 9.4 X 10⁵ cells ml⁻¹, while those of the cyanophages were between 2.0 X 10² and 4.2 X 10⁴ PFU ml⁻¹. Sharp decreases in densities of *M. aeruginosa* were detected on 10 June and 24 September, as densities of the cyanophages increased, suggesting release of the cyanophages due to the lysis of infected *M. aeruginosa*. Thus, infection by cyanophages may have a substantial effect on cyanobacterial succession in the pond. Densities of cyanophages became undetectable when those of *M. aeruginosa* were at low levels during winter. We suggest that there is a tight host-pathogen relationship between *M. aeruginosa* and the cyanophage in the pond
302. **Cyanophages.** Martin, E. L., Kokjohn, T. A. (1999). pp. 324-332 in Granoff, A., Webster, R. G. (eds.) *Encyclopedia of Virology second edition*. Academic Press, San Diego. The cyanobacteria with their prokaryotic structure and oxygenic photosynthesis occupy a unique position among living organisms. These characteristics closely relate them not only to the eubacteria, but also to eucaryotic algae and plants. Over the last 5 or 6 years research has continued to investigate the genetics, physiology, and metabolism, primarily for cyanophages which replicate in both unicellular and filamentous freshwater cyanobacteria; however, the major impetus of the latest research has shifted to the role cyanophages occupy in marine ecosystems. Cyanophages are of critical importance in that they can cause substantial lysis of primary producer organisms. This in turn can exert an extensive effect on the dynamics of carbon flow in many of the marine environments studied. It is hoped that many of the aspects of the research described here can continue to come together to give a more interconnected and complete understanding of the interactions between cyanophages and cyanobacteria.
303. **First report of a putative cyanophage, MC-1 of *Microcoleus* sp.** Rosowski, J. R., Shaffer, J. J., Martin, E. L. (1999). *Microsc. Microanalysis* 5:1142-1143.
304. **Phycodnaviridae.** van Etten, J. L. (1999). pp. 183-193 *Virus Taxonomy - Seventh Report*.
305. **Changes in bacterial and eukaryotic community structure after mass lysis of filamentous cyanobacteria**

associated with viruses. van Hanne, E. J., Zwart, G., van Agterveld, M. P., Gons, H. J., Ebert, J., Laanbroek, H. J. (1999). *Applied and Environmental Microbiology* 65:795-801. During an experiment in two laboratory-scale enclosures filled with lake water (130 liters each) we noticed the almost-complete lysis of the cyanobacterial population. Based on electron microscopic observations of viral particles inside cyanobacterial filaments and counts of virus-like particles, we concluded that a viral lysis of the filamentous cyanobacteria had taken place. Denaturing gradient gel electrophoresis (DGGE) of 16S ribosomal DNA fragments qualitatively monitored the removal of the cyanobacterial species from the community and the appearance of newly emerging bacterial species. The majority of these bacteria were related to the Cytophagales and actinomycetes, bacterial divisions known to contain species capable of degrading complex organic molecules. A few days after the cyanobacteria started to lyse, a rotifer species became dominant in the DGGE profile of the eukaryotic community. Since rotifers play an important role in the carbon transfer between the microbial loop and higher trophic levels, these observations confirm the role of viruses in channeling carbon through food webs. Multidimensional scaling analysis of the DGGE profiles showed large changes in the structures of both the bacterial and eukaryotic communities at the time of lysis. These changes were remarkably similar in the two enclosures, indicating that such community structure changes are not random but occur according to a fixed pattern. Our findings strongly support the idea that viruses can structure microbial communities.

306. **Sunlight-induced DNA damage and resistance in natural viral communities.** Weinbauer, M. G., Wilhelm, S. W., Suttle, C. A., Pledger, R. J., Mitchell, D. L. (1999). *Aquatic Microbial Ecology* 17:111-120. Using a highly specific radioimmunoassay, the sunlight-induced formation of cyclobutane pyrimidine dimers (CPDs) and pyrimidine (6-4) pyrimidone photoproducts ([6-4] PPs) in viral DNA was investigated for natural virus communities in offshore and coastal waters of the western Gulf of Mexico as well as for clonal viral isolates. Concentrations of (6-4) PPs were consistently lower than CPD concentrations, and ranged from 1.5 to 17.0% of total measured photodamage. The accumulation of photoproducts varied among the natural viral community, the marine *Vibrio* phage PWH3a-P1 and the *Synechococcus* sp. DC2 (WH7803) cyanophage SYN-M3, which were deployed *in situ* from dawn until dark. Natural viral communities were more resistant to DNA damage than the cyanophage isolate SYN-M3, which was more resistant to damage than bacteriophage PWH3a-P1. Moreover, depth profiles revealed that photodamage in viral isolates deployed in the water column accumulated more rapidly at offshore stations than at coastal stations. In natural virus communities collected from offshore surface waters, photodamage accumulated during the solar day with maximum damage occurring between 15:00 and 18:00 h. Depth profiles obtained during calm seas showed that photodamage concentrations were high in surface waters at the offshore stations and at 1 coastal station. Results at other coastal stations undergoing significant mixing demonstrated no photoproduct accumulations. Results demonstrate that natural virus communities were more tolerant to DNA damaging radiation than the laboratory isolates used in this study. Consequently, laboratory isolates can be poor proxies for UV impacts on natural viral communities.
307. **Analysis of cyanophage diversity and population structure in a south-north transect of the Atlantic ocean.** Wilson, W. H., Fuller, N. J., Joint, I. R., Mann, N. H. (1999). *Bulletin de l'Institut Océanographique (Monaco)* 0:209-216. Cyanophages (viruses which infect cyanobacteria) are abundant in the marine environment and are thought to be a significant factor in determining the dynamics of *Synechococcus* spp. populations. In an effort to use molecular techniques to characterise cyanophage populations, we designed cyanophage-specific (CPS) PCR primers based on a gene found in three genetically distinct marine cyanophages (Fuller et al., 1998). CPS primers were used to amplify cyanophage DNA extracted from viral communities concentrated from sea-water samples obtained during a cruise transect between the Falkland Islands, in the south Atlantic ocean, to the UK. Following phylogenetic analysis of cloned and sequenced PCR products, it was revealed that genetic diversity of marine cyanophage clones within a single water sample was as great as clones and cyanophage isolates collected between different oceans. Denaturing gradient gel electrophoresis (DGGE) analysis confirmed this high diversity. DGGE analysis also revealed changes in cyanophage population structure in surface seawater over the south-north transect and throughout depth profiles in the water column. Maximum *Synechococcus* spp. concentrations, in a stratified water column, correlated with maximum cyanophage diversity
308. **Blue-green algal viruses (cyanophages).** Zhao, Y., Shi, Z., Huang, G., Wang, X. (1999). *Virologica Sinica* 14:100-105.
309. **Genomic sequence of a lytic cyanophage of *Synechococcus* spp.** Lu, J. R., Chen, F., Hodson, R. E. (2000). *Abstracts of the General Meeting of the American Society for Microbiology* 100:465.
310. **Ecology of bacteriophages in nature.** Paul, J. H., Kellogg, C. A. (2000). pp. 211-246 in Hurst, C. J. (ed.) *Viral Ecology*. Academic Press, San Diego. [first paragraph] The role of bacteriophages (viruses that infect bacteria) in the environment has been the subject of intense investigation over the past several years. The development of techniques to study natural viral populations *in situ* has progressed tremendously. Various aspects of bacteriophage ecology in nature - including abundance, role in microbial mortality and water column trophodynamics, viral decay rates, repair mechanisms, and lysogeny - are now becoming or are nearly understood. However, most of these studies have been performed in aquatic environments. Thus, this review will mainly be limited to a discussion of aquatic environments. For reviews of the earlier literature, the reader is referred to Moebus (1987), Goyal *et al.* (1987), Fuhrman and Suttle (1993), Ackermann and DuBow (1987), and Proctor (1997).
311. **Cyanophages and their role in the ecology of cyanobacteria.** Suttle, C. A. (2000). pp. 563-589 in Whitton, B. A., Potts, M. (eds.) *The Ecology of Cyanobacteria: Their Diversity in Time and Space*. Kluwer Academic Publishers, Boston. Cyanophages belong to three recognized families of double-stranded DNA viruses; Myoviridae (contractile tails); Styloviridae (long non-contractile tails); and Podoviridae (short tails). They have a complex pattern of host ranges, are widely distributed, and can be readily isolated from marine and fresh waters. Although cyanophages are related to other bacteriophages, it is likely that they evolved more than 3 billion years ago when cyanobacteria diverged from other prokaryotes. In marine waters, genetically-diverse Myoviridae which infected *Synechococcus* spp. are the most abundant cyanophages; Styloviridae and Podoviridae are most commonly isolated from fresh waters. Morphological evidence also suggests that freshwater and marine myoviruses are more closely related to each other than they are to other bacteriophages. Cyanophages that infect phycoerythrin-rich *Synechococcus* spp. can be extremely abundant in coastal marine environments where they can occur at titers in

excess of 10^6 ml⁻¹ and 10^5 g⁻¹ of sediment. In surface waters abundance varies over orders of magnitude on a seasonal basis. Abundance follows that of *Synechococcus*, with evidence for a threshold in *Synechococcus* of ca. 10^3 to 10^4 mL⁻¹ beyond which cyanophage abundance increases greatly. In nearshore waters the high concentrations of cyanophages and *Synechococcus* result in high encounter frequencies and selection for *Synechococcus* communities that are largely resistant to infection. Encounters are much less frequent offshore and this leads to the appearance of a community that appears to have low resistance to infection. In freshwaters, viruses which infect filamentous cyanobacteria appear to be most abundant and also show strong seasonal dynamics however; even in the most eutrophic environments titers are orders of magnitude less than in productive coastal waters. Little effort was made to screen freshwaters for cyanophages that infect phycoerythrin-rich *Synechococcus*. In marine surface waters turnover times for cyanophage populations range from hours to days. Solar radiation has a major effect on cyanophage infectivity and results in the selection of cyanophage communities that are more resistant to destruction by sunlight during summer. In contrast to surface waters, infectious cyanophages can persist in sediments for at least 100 years. Although the effect of cyanophages on the mortality of cyanobacterial communities is likely to be variable, current estimates suggest that cyanophages are responsible for the removal of approximately 3% of marine *Synechococcus* on a daily basis. In addition to the lytic infection, lysogenic associations were clearly demonstrated in filamentous and unicellular cyanobacteria, but the ecological implications of lysogeny remain unexplored. Environmental factors and the physiological state of cyanobacteria clearly affect cyanophage-cyanobacterial interactions but remain poorly understood.

312. **The ecology, evolutionary and geochemical consequences of viral infection of cyanobacteria and eukaryotic algae.** Suttle, C. A. (2000). pp. 248-286 in Hurst, C. J. (ed.) *Viral Ecology*. Academic Press, New York. [first paragraph] More than 35 years ago, Safferman and Morris (1963) reported the isolation of a virus (cyanophage) that infected a freshwater filamentous "blue-green alga." This discovery stimulated research that led to isolation of many viruses that infect freshwater cyanobacteria. Although the potential for viruses to control cyanobacterial blooms was recognized (Safferman and Morris, 1964; Shilo, 1971), much of this work focused on the biology rather than the ecology of cyanophages (reviewed in Brown, 1972; Padan and Shilo, 1973; Safferman, 1973; Stewart and Daft, 1977; Sherman and Brown, 1978; Martin and Benson, 1988). At about the same time, evidence for viral infection of eukaryotic algae was beginning to emerge with reports by Zavarzina (1961, 1964) of lysis of *Chlorella pyrenoidosa* cultures. However, it was not until a decade later that observations of viruslike particles (VLPs) in eukaryotic algae began to appear in the literature (e.g., Lee, 1971; Chapman and Lang, 1973; Lemke, 1976; Dodds, 1979, 1983), and shortly thereafter a virus (CCV) was isolated that infected the macroalga *Cham corullina* (Gibbs *et al.*, 1975; Skotnicki *et al.*, 1976). This was followed a few years later by isolation of a virus (MPV) that infected the marine photosynthetic flagellate *Micromonas pusilla* (Mayer, 1978; Mayer and Taylor, 1979). Despite the ecological implications of viruses infecting major primary producers in aquatic environments, interest in viruses that infect eukaryotic algae was slow to gather. In fact, after the work by Waters and Chan (1982), there was little further interest in MPV, and the virus was lost from culture (F. J. R. Taylor, personal communication). Furthermore, by the 1980s ecological interest in cyanophages also began to wane as cyanobacterial blooms were brought under control by regulations reducing nutrient inputs to lakes. The decline in interest was exacerbated by the lack of appreciation by many algal and aquatic ecologists of the ecological importance of microbes in general and of viruses in particular. Although there was little interest in the ecological aspects, there were major advances in our understanding of the biology of a group of viruses that infect *Chlorella*-like algae that are symbionts of *Hydra viridis* and *Paramecium bursaria*. These viruses were isolated in the early 1980s and possess a number of unusual features (reviewed in Van Etten *et al.*, 1991; Reisser, 1993; Van Etten, 1995; Van Etten and Meints, 1999) that led to the creation of a new family of viruses, Phycodnaviridae (Van Etten and Ghabrial, 1991). As well, work has progressed on a group of widely distributed viruses that infect filamentous brown algae belonging to order Ectocarpales (Oliveira and Bisalputra, 1978; Muller, 1991; Henry and Meints, 1992; Muller and Frenzer, 1993; Muller, 1996; Van Etten and Meints, 1999).
313. **Transmission electron microscope analysis of viruses in the freshwater lakes of Signy Island, Antarctica.** Wilson, W. H., Lane, D., Pearce, D., Ellis-Evans, J. C. (2000). *Polar Biology* 23:657-660.
314. **Analysis of cyanophage diversity in the marine environment using denaturing gradient gel electrophoresis.** Wilson, W. H., Fuller, N. J., Jount, I. R., Mann, N. H. (2000). pp. 565-570 in Bell, C. R., Brylinsky, M., Johnson-Green, P. (eds.) *Microbial Biosystems: New Frontiers*. Atlantic Canada Society for Microbial Ecology, Halifax, Canada.
315. **Distribution of virus-like particles in an oligotrophic marine environment (Alboran Sea, Western Mediterranean).** Alonso, M. C., Jimenez-Gomez, F., Rodriguez, J., Borrego, J. J. (2001). *Microbial Ecology* 42:407-415. Viruses are abundant in a variety of aquatic environments, often exceeding bacterial abundance by one order of magnitude. In the present study, the spatial distribution of viruses in offshore waters of the Alboran Sea (Western Mediterranean) have been studied to determine the relationships between viruses and host communities in this oligotrophic marine environment. Viral abundance was determined using two methods: (i) epifluorescence light microscopy using the dsDNA binding fluorochrome DAPI, and (ii) direct counts by transmission electron microscopy (TEM). The results obtained were significantly different; the highest viral counts were obtained by mean of TEM analyses. In all the samples tested the number of viruses was exceeded by the bacterial concentrations, with a ratio between viral and bacterial titers varying between 1.4 and 20. VLP (virus-like particle) counts were not significantly correlated ($p > 0.001$) with chlorophyll a concentration or the abundance of cyanobacteria. However, there was a positive and significant correlation with bacterial abundance ($p < 0.001$). The analysis of size and morphology of viral particles by TEM and the correlation obtained between the numbers of VLP and bacteria suggest that the majority of the viral particles in the Alboran Sea are bacteriophages. None of the indirect evidence suggested that eukaryotic algae or cyanobacteria were important host organisms in these waters.
316. **Use of octyl β -thioglucopyranoside in two-dimensional crystallization of membrane proteins.** Chami, M., Pehau-Arnaudet, G., Lambert, O., Ranck, J. L., Levy, D., Rigaud, J. L. (2001). *Journal of Structural Biology* 133:64-74. A great interest exists in producing and/or improving two-dimensional (2D) crystals of membrane proteins amenable to structural analysis by electron crystallography. Here we report on the use of the detergent n-octyl beta-d-thioglucopyranoside in 2D crystallization trials of membrane proteins with radically different structures including FhuA from the outer membrane of *Escherichia coli*, light-harvesting complex II from *Rubrivivax gelatinosus*,

and Photosystem I from cyanobacterium *Synechococcus* sp. We have analyzed by electron microscopy the structures reconstituted after detergent removal from lipid-detergent or lipid-protein-detergent micellar solutions containing either only n-octyl beta-d-thioglucoopyranoside or n-octyl beta-d-thioglucoopyranoside in combination with other detergents commonly used in membrane protein biochemistry. This allowed the definition of experimental conditions in which the use of n-octyl beta-d-thioglucoopyranoside could induce a considerable increase in the size of reconstituted membrane structures, up to several micrometers. An other important feature was that, in addition to reconstitution of membrane proteins into large bilayered structures, this thioglycosylated detergent also was revealed to be efficient in crystallization trials, allowing the proteins to be analyzed in large coherent two-dimensional arrays. Thus, inclusion of n-octyl beta-d-thioglucoopyranoside in 2D crystallization trials appears to be a promising method for the production of large and coherent 2D crystals that will be valuable for structural analysis by electron crystallography and atomic force microscopy

317. **A conserved genetic module that encodes the major virion components in both the coliphage T4 and the marine cyanophage S-PM2.** Hambly, E., Tétart, F., Desplats, C., Wilson, W. H., Krisch, H. M., Mann, N. H. (2001). *Proceedings of the National Academy of Sciences, USA* 98:11411-11416. Sequence analysis of a 10-kb region of the genome of the marine cyanomyovirus S-PM2 reveals a homology to coliphage T4 that extends as a contiguous block from gene (g)18 to g23. The order of the S-PM2 genes in this region is similar to that of T4, but there are insertions and deletions of small ORFs of unknown function. In T4, g18 codes for the tail sheath, g19, the tail tube, g20, the head portal protein, g21, the prohead core protein, g22, a scaffolding protein, and g23, the major capsid protein. Thus, the entire module that determines the structural components of the phage head and contractile tail is conserved between T4 and this cyanophage. The significant differences in the morphology of these phages must reflect the considerable divergence of the amino acid sequence of their homologous virion proteins, which uniformly exceeds 50%. We suggest that their enormous diversity in the sea could be a result of genetic shuffling between disparate phages mediated by such commonly shared modules. These conserved sequences could facilitate genetic exchange by providing partially homologous substrates for recombination between otherwise divergent phage genomes. Such a mechanism would thus expand the pool of phage genes accessible by recombination to all those phages that share common modules.
318. **Distribution, isolation, host specificity, and diversity of cyanophages infecting marine *Synechococcus* spp. in river estuaries.** Lu, J., Chen, F., Hodson, R. E. (2001). *Applied and Environmental Microbiology* 67:3285-3290. The abundance of cyanophages infecting marine *Synechococcus* spp. increased with increasing salinity in three Georgia coastal rivers. About 80% of the cyanophage isolates were cyanomyoviruses. High cross-infectivity was found among the cyanophages infecting phycoerythrin-containing *Synechococcus* strains. Cyanophages in the river estuaries were diverse in terms of their morphotypes and genotypes
319. **Fingerprinting viral assemblages by pulsed field gel electrophoresis.** Steward, G. F. (2001). pp. 85-102 in Paul, J. H. (ed.) *Marine Microbiology*. Academic Press, London. Viruses are the most abundant microorganisms in marine and freshwater environments and perhaps the most genetically diverse (Fuhrman and Suttle, 1993). Counting viruses in aquatic samples is now a routine matter, but assessing the diversity and dynamics within complex assemblages is still a challenge. DNA-based fingerprinting approaches which rely on amplification of rRNA gene fragments by PCR have facilitated analyses of bacterial community composition. These approaches have more restricted application when analyzing viral assemblages, because of the extreme genetic diversity among viruses. Unlike in bacteria, there are no gene sequences conserved in all viruses which can serve as universal primer sites for PCR amplification. PCR-based analyses of viral assemblages must therefore target specific subsets of the total viral assemblage. For example, PCR amplification of specific genes has recently been used to examine the genetic diversity among cyanophages (Fuller *et al.*, 1998) and among phycodnaviridae (Chen *et al.*, 1996; Short and Suttle, 1999). A more general fingerprinting approach, which encompasses the total viral assemblage, is a valuable complement to these more specific, higher resolution analyses. The approach described here uses variation in genome size as the basis for obtaining a fingerprint of a viral assemblage (Klieve and Swain, 1993). A whole genome fingerprinting approach is possible, because viral genomes can vary greatly in length (a few thousand to hundreds of thousands of base pairs) yet they fall within a range that is easily resolved using pulsed field gel electrophoresis (PFGE). The PFGE fingerprinting technique provides a quick and relatively simple means of visualizing differences in the composition of viral assemblages (Swain *et al.*, 1996; Wommack *et al.*, 1999a; Steward and Azam, 2000). As a supplement to the more specific treatment of PFGE provided in this chapter, the reader is encouraged to consult the excellent introductory text to PFGE by Birren and Lai (1993).
320. **Genomic sequence and evolution of marine cyanophage P60: a new insight on lytic and lysogenic phages.** Chen, F., Lu, J. (2002). *Applied and Environmental Microbiology* 68:2589-2594. The genome of cyanophage P60, a lytic virus which infects marine *Synechococcus* WH7803, was completely sequenced. The P60 genome contained 47,872 bp with 80 potential open reading frames that were mostly similar to the genes found in lytic phages like T7, fYeO3-12, and SIO1. The DNA replication system, consisting of primase-helicase and DNA polymerase, appeared to be more conserved in podoviruses than in siphoviruses and myoviruses, suggesting that DNA replication genes could be the critical elements for lytic phages. Strikingly high sequence similarities in the regions coding for nucleotide metabolism were found between cyanophage P60 and marine unicellular cyanobacteria
321. **Prokaryotic and viral diversity patterns in marine plankton.** Fuhrman, J. A., Griffith, J., Schwalbach, M. (2002). *Ecological Research* 17:183-194. Prokaryotes and viruses play critical roles in marine ecosystems, where they are both highly abundant and active. Although early work on both prokaryotes and viruses revealed little of their diversity, molecular biological approaches now allow us to break apart these 'black boxes.' The most revealing methods have been cloning and sequencing of 16S rRNA genes, community fingerprinting (such as terminal restriction fragment length polymorphism; TRFLP), and fluorescent *in situ* hybridization. Viral diversity can now be analyzed by pulsed field gel electrophoresis (PFGE) of viral genomes. The present paper summarizes recent advances in bacterial and virus diversity studies, and presents examples of measurements from polar, tropical, and temperate marine waters. Terminal restriction fragment length polymorphism shows that many of the same operationally defined prokaryotic taxa are present in polar and tropical waters, but there are also some unique to each environment. By one measure, a sample from over a Philippine coral reef had about 100 operationally defined taxa, whereas one from the open tropical Atlantic had about 50 and from the icy Weddell Sea, about 60. Pulsed field gel electrophoresis of two depth profiles, to 500 m, from Southern California, measured 2 months apart, shows

striking similarities in viral genome length diversity over time, and some distinct differences with depth. The euphotic zone samples had extremely similar apparent diversity, but samples from 150 m and 500 m were different. An obvious next step is to compare the bacterial and viral diversity patterns, because theory tells us they should be related.

322. **Observations on cyanobacterial population collapse in eutrophic lake water.** Gons, H. J., Ebert, J., Hoogveld, H. L., van den Hove, L., Pel, R., Takkenberg, W., Woldringh, C. J. (2002). *Antonie van Leeuwenhoek* 81:319-326. In two laboratory-scale enclosures of water from the shallow, eutrophic Lake Loosdrecht (the Netherlands), the predominating filamentous cyanobacteria grew vigorously for 2 weeks, but then their populations simultaneously collapsed, whereas coccoid cyanobacteria and algae persisted. The collapse coincided with a short peak in the counts of virus-like particles. Transmission electron microscopy showed the morphotype Myoviridae phages, with isometric heads of about 90 nm outer diameter and > 100-nm long tails, that occurred free, attached to and emerging from cyanobacterial cells. Also observed were other virus-like particles of various morphology. Similar mass mortality of the filamentous cyanobacteria occurred in later experiments, but not in Lake Loosdrecht. As applies to lakes in general, this lake exhibits high abundance of virus-like particles. The share and dynamics of infectious cyanophages remain to be established, and it is as yet unknown which factors primarily stabilize the host-cyanophage relationship. Observations on shallow, eutrophic lakes elsewhere indicate that the cyanophage control may also fail in natural water bodies exhibiting predominance of filamentous cyanobacteria. Rapid supply of nutrients appeared to be a common history of mass mortality of cyanobacteria and algae in laboratory and outdoor enclosures as well as in highly eutrophic lakes
323. **[Action of *Spirulina platensis* on bacterial viruses].** Gorobets, O. B., Blinkova, L. P., Baturo, A. P. (2002). *Zh. Mikrobiol. Epidemiol. Immunobiol.* 18-21. The impact of the biomass of the blue-green microalga (cyanobacterium) *S. platensis* on bacteriophage T4 (bacterial virus) has been evaluated. The study revealed that the addition of *S. platensis* biomass into the agar nutrient medium, followed by sterilization with 2% chloroform and thermal treatment, produced an inhibiting or stimulating effect on the reproduction of the bacteriophage in *Escherichia coli* B cells, depending on the concentration of *S. platensis* and the multiplicity of phage infection, as well as on the fact whether the microalgae were added during the first cycle of the development of the virus. The reproduction of the bacteriophage in *E. coli* B was influenced by the method and duration of the sterilization of the nutrient medium with *S. platensis*
324. **Use of signal-mediated amplification of RNA technology (SMART) to detect marine cyanophage DNA.** Hall, M. J., Wharam, S. D., Weston, A., Cardy, D. L. N., Wilson, W. H. (2002). *BioTechniques* 32:604-611. Here, we describe the application of an isothermal nucleic acid amplification assay, signal-mediated amplification of RNA technology (SMART), to detect DNA extracted from marine cyanophages known to infect unicellular cyanobacteria from the genus *Synechococcus*. The SMART assay is based on the target-dependent production of multiple copies of an RNA signal, which is measured by an enzyme-linked oligosorbent assay. SMART was able to detect both synthetic oligonucleotide targets and genomic cyanophage DNA using probes designed against the portal vertex gene (g20). Specific signals were obtained for each cyanophage strain (S-PM2 and S-BnMI). Nonspecific genomic DNA did not produce false signals or inhibit the detection of a specific target. In addition, we found that extensive purification of target DNA may not be required since signals were obtained from crude cyanophage lysates. This is the first report of the SMART assay being used to discriminate between two similar target sequences
325. **Plankton blooms: Lysogeny in marine *Synechococcus*.** McDaniel, L., Houchin, L. A., Williamson, S. J., Paul, J. H. (2002). *Nature* 415:496. Viral infection of bacteria can be lytic, causing destruction of the host cell, or lysogenic, in which the viral genome is instead stably maintained as a prophage within its host. Here we show that lysogeny occurs in natural populations of an autotrophic picoplankton (*Synechococcus*) and that there is a seasonal pattern to this interaction. Because lysogeny confers immunity to infection by related viruses, this process may account for the resistance to viral infection seen in common forms of autotrophic picoplankton. We undertook a seasonal study in Tampa Bay, Florida, of prophage induction in cyanobacteria over the year ending in October 2000 to find out whether lysogeny occurs in natural *Synechococcus* populations and, if so, how it is affected by changing environmental conditions
326. **Lysogeny and lytic viral production during a bloom of the cyanobacterium *Synechococcus* spp.** Ortmann, A. C., Lawrence, J. E., Suttle, C. A. (2002). *Microbial Ecology* 43:225-231. Lytic viral production and lysogeny were investigated in cyanobacteria and heterotrophic bacteria during a bloom of *Synechococcus* spp. in a pristine fjord in British Columbia, Canada. Triplicate seawater samples were incubated with and without mitomycin C and the abundances of heterotrophic bacteria, cyanobacteria, total viruses and infectious cyanophage were followed over 24 h. Addition of mitomycin C led to increases in total viral abundance as well as the abundance of cyanophages infecting *Synechococcus* strain DC2. Given typical estimates of burst size, these increases were consistent with 80% of the heterotrophic bacteria and 0.6% of *Synechococcus* cells being inducible by the addition of mitomycin C. This is the highest percentage of lysogens reported for a natural microbial community and demonstrates induction in a marine *Synechococcus* population. It is likely that the cyanophage production following the addition of mitomycin C was much higher than that titered against a single strain of *Synechococcus*; hence this estimate is a minimum. In untreated seawater samples, lytic viral production was estimated to remove ca. 27% of the gross heterotrophic bacterial production, and a minimum of 1.0% of the gross cyanobacterial production. Our results demonstrate very high levels of lysogeny in the heterotrophic bacterial community, outside of an oligotrophic environment, and the presence of inducible lysogens in *Synechococcus* spp. during a naturally occurring bloom. These data emphasize the need for further examination of the factors influencing lytic and lysogenic viral infection in natural microbial communities.
327. **Marine phage genomics.** Paul, J. H., Sullivan, M. B., Segall, A. M., Rohwer, F. (2002). *Comparative Biochemistry and Physiology* 133:463-476. Marine phages are the most abundant biological entities in the oceans. They play important roles in carbon cycling through marine food webs, gene transfer by transduction and conversion of hosts by lysogeny. The handful of marine phage genomes that have been sequenced to date, along with prophages in marine bacterial genomes, and partial sequencing of uncultivated phages are yielding glimpses of the tremendous diversity and physiological potential of the marine phage community. Common gene modules in diverse phages are providing the information necessary to make evolutionary comparisons. Finally, deciphering

328. **[Some peculiarities of DNA structure of cyanophage LPP-3]. Syrchin, S. A., Mendzhul, M. I. (2002). *Mikrobiologichnyi Zhurnal* 64:35-43.** The efficiency of radioactive labeling of 3'- and 5'-ends of cyanophage LPP-3 DNA by polynucleotide kinase T4 and terminal transferase under various reaction conditions has been investigated. The obtained data prove that cyanophage LPP-3 DNA has the protruding 3'-ends. The experiments on ligation of native molecules of LPP-3 DNA evidence that the virus genome ends do not display any complementarity. Separate fragments of LPP-3 DNA were cloned. The restriction analysis of the cloned fragments has confirmed a supposition on the absence of LPP-3 cyanophage of GGGCC and GGCCC sequences in the genome. A hypothesis has been suggested about similar site-specificity of the virus. Counterselection of the genome LPP-3 cyanophage allows it to be considered a promising one in the construction of new cloning vectors in cyanobacterium
329. **[Physical mapping of DNA of cyanophage LPP-3]. Syrchin, S. A., Mendzhul, M. I. (2002). *Mikrobiologichnyi Zhurnal* 64:24-30.** Restrictases fit for the purposes of physical mapping of cyanophage LPP-3 DNA have been selected as a result of the restriction analysis. The use of the methods of mutual hydrolysis, restriction of the fragment isolated from gel and terminal labeling allowed formation a physical map of LPP-3 cyanophage DNA with the complete scheme of allocation of 14 sites for 8 restrictases: Alw44I, Bsp119I, BsuRI, Eco147I, EheI, NcoI, Kpn2I and PvuI as well as the position of certain sites for restrictases HindIII, KpnI and Sau3A
330. **Phylogenetic diversity of marine cyanophage isolates and natural virus communities as revealed by sequences of viral capsid assembly protein gene g20. Zhong, Y., Chen, F., Wilhelm, S. W., Poorvin, L., Hodson, R. E. (2002). *Applied and Environmental Microbiology* 68:1576-1584.** In order to characterize the genetic diversity and phylogenetic affiliations of marine cyanophage isolates and natural cyanophage assemblages, oligonucleotide primers CPS1 and CPS8 were designed to specifically amplify ca. 592-bp fragments of the gene for viral capsid assembly protein g20. Phylogenetic analysis of isolated cyanophages revealed that the marine cyanophages were highly diverse yet more closely related to each other than to enteric coliphage T4. Genetically related marine cyanophage isolates were widely distributed without significant geographic segregation (i.e., no correlation between genetic variation and geographic distance). Cloning and sequencing analysis of six natural virus concentrates from estuarine and oligotrophic offshore environments revealed nine phylogenetic groups in a total of 114 different g20 homologs, with up to six clusters and 29 genotypes encountered in a single sample. The composition and structure of natural cyanophage communities in the estuary and open-ocean samples were different from each other, with unique phylogenetic clusters found for each environment. Changes in clonal diversity were also observed from the surface waters to the deep chlorophyll maximum layer in the open ocean. Only three clusters contained known cyanophage isolates, while the identities of the other six clusters remain unknown. Whether or not these unidentified groups are composed of bacteriophages that infect different *Synechococcus* groups or other closely related cyanobacteria remains to be determined. The high genetic diversity of marine cyanophage assemblages revealed by the g20 sequences suggests that marine viruses can potentially play important roles in regulating microbial genetic diversity
331. **NEW Encapsidation of host DNA by bacteriophages infective marine *Synechococcus* strains. Clokie, M. R., Millard, A. D., Wilson, W. H., Mann, N. H. (2003). *FEMS Microbiology Ecology* 46:349-352.** It has been speculated that horizontal gene transfer might be important in the evolution of strains of the marine cyanobacterium *Synechococcus* and that phages might mediate this process, but until now there has been no direct evidence to support this idea. We have rigorously purified bacteriophages (cyanomyoviruses) from their *Synechococcus* host and performed a series of experiments on phageencapsidated DNA to reveal the presence of chromosomal *Synechococcus* DNA. Quantitative polymerase chain reaction has shown that V1 in 105 *Synechococcus* phage particles contain a host marker gene in their capsids. This is the first study that has shown that phages infecting marine *Synechococcus* strains can package host DNA and this provides evidence for the potential importance of these phage in horizontal gene transfer. (added Monday, September 20, 2004 by Matt Sullivan)
332. **The physical environment affects cyanophage communities in British Columbia inlets. Frederickson, C. M., Short, S. M., Suttle, C. A. (2003). *Microbial Ecology* 46:348-357.** Little is known about the natural distribution of viruses that infect the photosynthetically important group of marine prokaryotes, the cyanobacteria. The current investigation reveals that the structure of cyanophage communities is dependent on water column structure. PCR was used to amplify a fragment of the cyanomyovirus gene (g) 20, which codes for the portal vertex protein. Denaturing gradient gel electrophoresis (DGGE) of PCR amplified g20 gene fragments was used to examine variations in cyanophage community structure in three inlets in British Columbia, Canada. Qualitative examination of denaturing gradient gels revealed cyanophage community patterns that reflected the physical structure of the water column as indicated by temperature and salinity. Based on mobility of PCR fragments in the DGGE gels, some cyanophages appeared to be widespread, while others were observed only at specific depths. Cyanophage communities within Salmon Inlet were more related to one another than to communities from either Malaspina Inlet or Pendrell Sound. As well, surface communities in Malaspina Inlet and Pendrell Sound were different when compared to communities at depth. In the same two locations, distinct differences in community composition were observed in communities that coincided with depths of high chlorophyll fluorescence. The observed community shifts over small distances (only a few meters in depth or inlets separated by less than 100 km) support the idea that cyanophage communities separated by small spatial scales develop independently of each other as a result isolation by water column stratification or land mass separation, which may ultimately lead to changes in the distribution or composition of the host community
333. **Phages of the marine cyanobacterial picophytoplankton. Mann, N. H. (2003). *FEMS Microbiology Reviews* 27:17-34.** Cyanobacteria of the genera *Synechococcus* and *Prochlorococcus* dominate the prokaryotic component of the picophytoplankton in the oceans. It is still less than 10 years since the discovery of phages that infect marine *Synechococcus* and the beginning of the characterisation of these phages and assessment of their ecological impact. Estimations of the contribution of phages to *Synechococcus* mortality are highly variable, but there is clear evidence that phages exert a significant selection pressure on *Synechococcus* community structure. In turn, there are strong selection pressures on the phage community, in terms of both abundance and composition. This review focuses on the factors affecting the diversity of cyanophages in the marine environment, cyanophage interactions with their hosts, and the selective pressures in the marine environment that affect cyanophage evolutionary biology.

334. **Bacterial photosynthesis genes in a virus.** Mann, N. H., Cook, A., Millard, A., Bailey, S., Clokie, M. (2003). *Nature* 424:741. A bacteriophage may protect itself and its host against a deadly effect of bright sunlight.
335. **Genetic diversity and temporal variation in the cyanophage community infecting marine *Synechococcus* species in Rhode Island's coastal waters.** Marston, M. F., Sallee, J. L. (2003). *Applied and Environmental Microbiology* 69:4639-4647. The cyanophage community in Rhode Island's coastal waters is genetically diverse and dynamic. Cyanophage abundance ranged from over 10^4 phage ml⁻¹ in the summer months to less than 10^2 phage ml⁻¹ during the winter months. Thirty-six distinct cyanomyovirus g20 genotypes were identified over a 3-year sampling period; however, only one to nine g20 genotypes were detected at any one sampling date. Phylogenetic analyses of g20 sequences revealed that the Rhode Island cyanomyoviral isolates fall into three main clades and are closely related to other known viral isolates of *Synechococcus* spp. Extinction dilution enrichment followed by host range tests and PCR restriction fragment length polymorphism analysis was used to detect changes in the relative abundance of cyanophage types in June, July, and August 2002. Temporal changes in both the overall composition of the cyanophage community and the relative abundance of specific cyanophage g20 genotypes were observed. In some seawater samples, the g20 gene from over 50% of isolated cyanophages could not be amplified by using the PCR primer pairs specific for cyanomyoviruses, which suggested that cyanophages in other viral families (e.g., Podoviridae or Siphoviridae) may be important components of the Rhode Island cyanophage community.
336. **[Development of cyanobacterial phages at the Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine (History and perspectives)].** Mendzhul, M. I., Lysenko, T. G., Syrchin, S. A. (2003). *Mikrobiologichnyi Zhurnal* 65:133-140. The paper deals with the basic trends of fundamental investigations of the Department of Algae Viruses in the field of cyanophagia-ecology, biological and physico-chemical properties of cyanophages as well as interrelation with the host cells. Such problems as a possibility to use the system cyanophage-cyanobacteria as the experimental model for development of the unified functional model of productive infection, efficient methods of prophylaxis and therapy of virus infections as well as the solution of various biotechnological problems are discussed
337. **[Comparative characteristics of native proteinases of the cyanobacteria *Plectonema boryanum* and *Anabaena variabilis* and those induced by cyanophages].** Mendzhul, M. I., Perepelytsia, S. I. (2003). *Mikrobiologichnyi zhurnal* 65:21-28. Physico-chemical and catalytic properties of proteinases of native and induced cells of cyanobacteria *Plectonema boryanum* have been comparatively studied. It has been established that at early stages of reproduction of cyanophage LPP-3 in cyanobacteria *P. boryanum* is formed de novo proteinase complex consisting at least of five enzymes. Proteinases induced by the virus are distinguished from those of native cells by a series of physico-chemical characteristics and possess higher catalytic activity. Analogous virus-induced changes in proteinase complex also occur in the system cyanobacterium *Anabaena variabilis*—cyanophage A-1. Possible functions of certain enzymes of proteinase complex in the virus pathology of cyanobacteria cells are discussed in the paper
338. **Cyanophages infecting the oceanic cyanobacterium *Prochlorococcus*.** Sullivan, M. B., Waterbury, J. B., Chisholm, S. W. (2003). *Nature* 424:1047-1051. *Prochlorococcus* is the numerically dominant phototroph in the tropical and subtropical oceans, accounting for half of the photosynthetic biomass in some areas. Here we report the isolation of cyanophages that infect *Prochlorococcus*, and show that although some are host-strain-specific, others cross-infect with closely related marine *Synechococcus* as well as between high-light- and low-light-adapted *Prochlorococcus* isolates, suggesting a mechanism for horizontal gene transfer. High-light-adapted *Prochlorococcus* hosts yielded Podoviridae exclusively, which were extremely host-specific, whereas low-light-adapted *Prochlorococcus* and all strains of *Synechococcus* yielded primarily Myoviridae, which has a broad host range. Finally, both *Prochlorococcus* and *Synechococcus* strain-specific cyanophage titres were low ($< 10^3$ ml⁻¹) in stratified oligotrophic waters even where total cyanobacterial abundances were high ($> 10^5$ cells x ml⁻¹). These low titres in areas of high total host cell abundance seem to be a feature of open ocean ecosystems. We hypothesize that gradients in cyanobacterial population diversity, growth rates, and/or the incidence of lysogeny underlie these trends.
339. **Cyanophage diversity, inferred from g20 gene analyses, in the largest natural lake in France, Lake Bourget.** Dorigo, U., Jacquet, S., Humbert, J. F. (2004). *Applied and Environmental Microbiology* 70:1017-1022. The genetic diversity of the natural freshwater community of cyanophages and its variations over time have been investigated for the first time in the surface waters of the largest natural lake in France. This was done by random screening of clone libraries for the g20 gene and by denaturing gradient gel electrophoresis (DGGE). Nucleotide sequence analysis revealed 35 distinct cyanomyovirus g20 genotypes among the 47 sequences analyzed. Phylogenetic analyses showed that these sequences fell into seven genetically distinct operational taxonomic units (OTUs). The distances between these OTUs were comparable to those reported between marine clusters. Moreover, some of these freshwater cyanophage sequences were genetically more closely related to marine cyanophage sequences than to other freshwater sequences. Both approaches for the g20 gene (sequencing and DGGE analysis) showed that there was a clear seasonal pattern of variation in the composition of the cyanophage community that could reflect changes in its biological, chemical, and/or physical environment.
340. **NEW Transfer of photosynthesis genes to and from *Prochlorococcus* viruses.** Lindell, D., Sullivan, M. B., Johnson, Z. I., Tolonen, A. C., Rohwer, F., Chisholm, S. W. (2004). *Proceedings of the National Academy of Sciences, USA* 101:11013-11018. Comparative genomics gives us a new window into phage-host interactions and their evolutionary implications. Here we report the presence of genes central to oxygenic photosynthesis in the genomes of three phages from two viral families (Myoviridae and Podoviridae) that infect the marine cyanobacterium *Prochlorococcus*. The genes that encode the photosystem II core reaction center protein D1 (psbA), and a high-light-inducible protein (HLIP) (hli) are present in all three genomes. Both myoviruses contain additional hli gene types, and one of them encodes the second photosystem II core reaction center protein D2 (psbD), whereas the other encodes the photosynthetic electron transport proteins plastocyanin (petE) and ferredoxin (petF). These uninterrupted, full-length genes are conserved in their amino acid sequence, suggesting that they encode functional proteins that may help maintain photosynthetic activity during infection. Phylogenetic analyses show that phage D1, D2, and HLIP proteins cluster with those from *Prochlorococcus*, indicating that they are of cyanobacterial origin. Their distribution among several *Prochlorococcus* clades further suggests that the genes encoding these proteins were transferred

times. Phage HLIPIs cluster with multicopy types found exclusively in *Prochlorococcus*, suggesting that phage may be mediating the expansion of the hli gene family by transferring these genes back to their hosts after a period of evolution in the phage. These gene transfers are likely to play a role in the fitness landscape of hosts and phages in the surface oceans. (added Tuesday, September 14, 2004 by [Matt Sullivan](#))

341. **NEW Genetic organization of the *psbAD* region in phages infecting marine *Synechococcus* strains.** Millard, A., Clokie, M., Shub, D. A., Mann, N. H. (2004). *Proceedings of the National Academy of Sciences, USA* 101:11007-11012. The discovery of the genes *psbA* and *psbD*, encoding the D1 and D2 core components of the photosynthetic reaction center PSII (photosystem II), in the genome of the bacteriophage S-PM2 (a cyanomyovirus) that infects marine cyanobacteria begs the question as to how these genes were acquired. In an attempt to answer this question, it was established that the occurrence of the genes is widespread among marine cyanomyovirus isolates and may even extend to podoviruses. The phage *psbA* genes fall into a clade that includes the *psbA* genes from their potential *Synechococcus* and *Prochlorococcus* hosts, and thus, this phylogenetic analysis provides evidence to support the idea of the acquisition of these genes by horizontal gene transfer from their cyanobacterial hosts. However, the phage *psbA* genes form distinct subclades within this lineage, which suggests that their acquisition was not very recent. The *psbA* genes of two phages contain identical 212-bp insertions that exhibit all of the canonical structural features of a group I self-splicing intron. The different patterns of genetic organization of the *psbAD* region are consistent with the idea that the *psbA* and *psbD* genes were acquired more than once by cyanomyoviruses and that their horizontal transfer between phages via a common phage gene pool, as part of mobile genetic modules, may be a continuing process. In addition, genes were discovered encoding a high-light inducible protein and a putative key enzyme of dark metabolism, transaldolase, extending the areas of host-cell metabolism that may be affected by phage infection. (added Tuesday, September 14, 2004 by [Matt Sullivan](#))
342. **NEW Genetic diversity and population dynamics of cyanophage communities in the Chesapeake Bay.** Wang, K., Chen, F. (2004). *Aquatic Microbiology Ecology* 34:105-116. In order to understand the genetic diversity and population dynamics of cyanophages in estuarine waters, the viral capsid assembly (*g20*) gene was used as a gene marker to monitor genetic variations of natural cyanomyovirus communities in the Chesapeake Bay, USA. Unique and diverse *g20* sequences were found. Only 1 of 15 *g20* genotypes was closely related to the known cyanomyovirus isolates. Most of the *g20* genotypes in the bay were not related to the *g20* clonal sequences recovered from open-ocean waters. Terminal-restriction fragment length polymorphism (T-RFLP) based on the *g20* gene was developed to investigate spatial and temporal distribution of cyanomyovirus communities in the bay. The T-RFLP profiles of the *g20* gene demonstrated that the cyanomyovirus population structures in the bay were more dynamic seasonally than spatially. Seasonal variation in the cyanophage community appeared to correspond to changes in host-cell density, which in turn was mainly affected by water temperature. This study represents the first effort to monitor both cyanophage titer and genetic diversity over time and space. The results of our study suggest that cyanophages could play a significant role in regulating *Synechococcus* biomass and population structure in the Chesapeake Bay. (added Tuesday, September 14, 2004 by [Matt Sullivan](#))
343. **NEW Genetic diversity of marine *Synechococcus* and co-occurring cyanophage communities: evidence for viral control of phytoplankton.** Muhling, M., Fuller, N.J., Millard, A., Somerfield, P.J., Marie, D., Wilson, W.H., Scanlan, D.J., Post, A.F., Joint, I., Mann, N.H. (2005). *Environmental Microbiology* 7:499-508. Unicellular cyanobacteria of the genus *Synechococcus* are a major component of the picophytoplankton and make a substantial contribution to primary productivity in the oceans. Here we provide evidence that supports the hypothesis that virus infection can play an important role in determining the success of different *Synechococcus* genotypes and hence of seasonal succession. In a study of the oligotrophic Gulf of Aqaba, Red Sea, we show a succession of *Synechococcus* genotypes over an annual cycle. There were large changes in the genetic diversity of *Synechococcus*, as determined by restriction fragment length polymorphism analysis of a 403-bp *rpoC1* gene fragment, which was reduced to one dominant genotype in July. The abundance of co-occurring cyanophage capable of infecting marine *Synechococcus* was determined by plaque assays and their genetic diversity was determined by denaturing gradient gel electrophoresis analysis of a 118-bp *g20* gene fragment. The results indicate that both abundance and genetic diversity of cyanophage covaried with that of *Synechococcus*. Multivariate statistical analyses show a significant relationship between cyanophage assemblage structure and that of *Synechococcus*. These observations are consistent with cyanophage infection being a major controlling factor in picophytoplankton succession. (added Tuesday, April 12, 2005 by [Matt Sullivan](#))
344. **NEW Nearly identical bacteriophage structural gene sequences are widely distributed in both marine and freshwater environments.** Short, C.M., Suttle, C.A. (2005). *Applied and Environmental Microbiology* 71:480-486. Primers were designed to amplify a 592-bp region within a conserved structural gene (*g20*) found in some cyanophages. The goal was to use this gene as a proxy to infer genetic richness in natural cyanophage communities and to determine if sequences were more similar in similar environments. Gene products were amplified from samples from the Gulf of Mexico, the Arctic, Southern, and Northeast and Southeast Pacific Oceans, an Arctic cyanobacterial mat, a catfish production pond, lakes in Canada and Germany, and a depth of ca. 3,246 m in the Chuckchi Sea. Amplicons were separated by denaturing gradient gel electrophoresis, and selected bands were sequenced. Phylogenetic analysis revealed four previously unknown groups of *g20* clusters, two of which were entirely found in freshwater. Also, sequences with >99% identities were recovered from environments that differed greatly in temperature and salinity. For example, nearly identical sequences were recovered from the Gulf of Mexico, the Southern Pacific Ocean, an Arctic freshwater cyanobacterial mat, and Lake Constance, Germany. These results imply that closely related hosts and the viruses infecting them are distributed widely across environments or that horizontal gene exchange occurs among phage communities from very different environments. Moreover, the amplification of *g20* products from deep in the cyanobacterium-sparse Chuckchi Sea suggests that this primer set targets bacteriophages other than those infecting cyanobacteria. (added Tuesday, April 12, 2005 by [Matt Sullivan](#))
345. **NEW Three *Prochlorococcus* cyanophage genomes: Signature features and ecological interpretations.** Sullivan, M.B., Coleman, M., Weigele, P. Rohwer, F., Chisholm, S.W. (2005). *PLoS Biology* 3:e144. The oceanic cyanobacteria *Prochlorococcus* are globally important, ecologically diverse primary producers. It is thought that their viruses (phages) mediate population sizes and affect the evolutionary trajectories of their hosts. Here we present an analysis of genomes from three *Prochlorococcus* phages: a podovirus and two myoviruses. The morphology, overall genome features, and gene content of these phages suggest that they are quite similar to T7-like

(P-SSP7) and T4-like (P-SSM2 and P-SSM4) phages. Using the existing phage taxonomic framework as a guideline, we examined genome sequences to establish "core" genes for each phage group. We found the podovirus contained 15 of 26 core T7-like genes and the two myoviruses contained 43 and 42 of 75 core T4-like genes. In addition to these core genes, each genome contains a significant number of "cyanobacterial" genes, i.e., genes with significant best BLAST hits to genes found in cyanobacteria. Some of these, we speculate, represent "signature" cyanophage genes. For example, all three phage genomes contain photosynthetic genes (psbA, hliP) that are thought to help maintain host photosynthetic activity during infection, as well as an aldolase family gene (talC) that could facilitate alternative routes of carbon metabolism during infection. The podovirus genome also contains an integrase gene (int) and other features that suggest it is capable of integrating into its host. If indeed it is, this would be unprecedented among cultured T7-like phages or marine cyanophages and would have significant evolutionary and ecological implications for phage and host. Further, both myoviruses contain phosphate-inducible genes (phoH and pstS) that are likely to be important for phage and host responses to phosphate stress, a commonly limiting nutrient in marine systems. Thus, these marine cyanophages appear to be variations of two well-known phages T7 and T4 but contain genes that, if functional, reflect adaptations for infection of photosynthetic hosts in low-nutrient oceanic environments. (added Tuesday, April 12, 2005 by [Matt Sullivan](#))

346. **NEW Identification of cyanophage Ma-LBP and infection of the cyanobacterium *Microcystis aeruginosa* from an Australian subtropical lake by the virus. Tucker, S., Pollard, P. (2005). *Applied and Environmental Microbiology* 71:629-635.** Viruses can control the structure of bacterial communities in aquatic environments. The aim of this project was to determine if cyanophages (viruses specific to cyanobacteria) could exert a controlling influence on the abundance of the potentially toxic cyanobacterium *Microcystis aeruginosa* (host). *M. aeruginosa* was isolated, cultured, and characterized from a subtropical monomictic lake Lake Baroon, Sunshine Coast, Queensland, Australia. The viral communities in the lake were separated from cyanobacterial grazers by filtration and chloroform washing. The natural lake viral cocktail was incubated with the *M. aeruginosa* host growing under optimal light and nutrient conditions. The specific growth rate of the host was 0.023 h⁻¹; generation time, 30.2 h. Within 6 days, the host abundance decreased by 95%. The density of the cyanophage was positively correlated with the rate of *M. aeruginosa* cell lysis (r² = 0.95). The cyanophage replication time was 11.2 h, with an average burst size of 28 viral particles per host cell. However, in 3 weeks, the cultured host community recovered, possibly because the host developed resistance (immunity) to the cyanophage. The multiplicity of infection was determined to be 2,890 virus-like particles/cultured host cell, using an undiluted lake viral population. Transmission electron microscopy showed that two types of virus were likely controlling the host cyanobacterial abundance. Both viruses displayed T7-like morphology and belonged to the *Podoviridae* group (short tails) of viruses that we called cyanophage Ma-LBP. In Lake Baroon, the number of the cyanophage Ma-LBP was 5.6 x 10⁴ cyanophage ml⁻¹, representing 0.23% of the natural viral population of 2.46 x 10⁷ ml⁻¹. Our results showed that this cyanophage could be a major natural control mechanism of *M. aeruginosa* abundance in aquatic ecosystems like Lake Baroon. Future studies of potentially toxic cyanobacterial blooms need to consider factors that influence cyanophage attachment, infectivity, and lysis of their host alongside the physical and chemical parameters that drive cyanobacterial growth and production. (added Tuesday, April 12, 2005 by [Matt Sullivan](#))

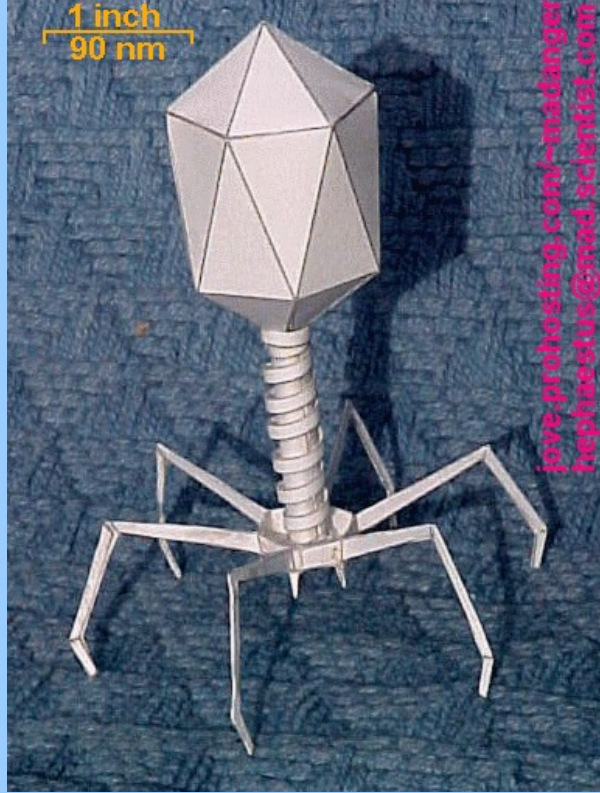
Submissions Archive

- [On an Invisible Microbe Antagonistic to the Dysentery Bacillus by Felix d'Herelle](#)
- [Obituary: Hansjürgen Raettig - Collector of Bacteriophage References \(October 12, 1911 - December 1, 1997\)](#)
- [Some Quotations](#)
- [Bacteriophages: A Model System for Human Viruses](#)
- [How Big is 10³⁰?](#)
- [Selling Phage Candy](#)
- [A List of Phage Names](#)
- [An Expanded Overview of Phage Ecology](#)
- [Rendering Phage Heads](#)
- [The Contractile-Tail Sheath, In Three Dimensions](#)
- [Eye On The Needle: Phage T4 Puncturing Point May Answer Penetrating Questions](#)
- [Pioneering genetic researcher Gisela Mosig dies](#)
- [Updated Eiserling T4 Virion](#)
- [Some Recent Phage and Phage-Related U.S. Patents \(1976-present\)](#)
- [Some Images of BEG Members](#)
- [Early Phage References, pre-1950](#)
- [Zooming Through the Tail Tube - A Steve McQuinn Perspective on Phage T4](#)
- [The T4 Prolate Head](#)
- [The Cyanophage Literome](#)

Submissions are non-editorial items describing or highlighting some aspect of bacteriophage ecology including news pieces, historical pieces, reviews, and write-ups of research. Peer review of submissions is possible and a desire for peer review should be indicated. Send all submissions to microdude+@osu.edu or to "Submissions", Bacteriophage Ecology Group News, care of Stephen T. Abedon, Department of Microbiology, The Ohio State University, 1680 University Dr., Mansfield, Ohio 44906. Please send all submissions as Microsoft Word documents, if possible (I'll let you know if I have trouble converting any other document formats), and in English.

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Phage Images



Yes, these phages are made out of paper! They are courtesy of [Masamune Washington](#). Click [here](#) for a PDF kit for making your own paper T4 phage and [here](#) for additional directions.

Phage Image Archive

- [BEG Phage Images Page](#)
- [The Face of the Phage](#)
- [Bacteriophage T2](#)
- [SSV1-Type Phage](#)
- [Saline Lake Bacteriophage](#)
- [Coliphage LG1](#)
- [Bacteriophage HK97](#)
- [Phage T4 \(art\)](#)
- [Phage T4 on the pedestal outside of Barker Hall at Berkeley](#)
- [Electron micrograph of phage P22](#)
- [Thin section of T4 phages hitting a microcolony of *E. coli* K-12](#)
- [T4 phage v1](#)
- [T4 Tail Model](#)
- [Gingerbread phage](#)
- [T4 adsorbing en mass](#)
- [Lysis of *E. coli* O157](#)
- [Homologous Recombination - 2000 by Jake McKinlay](#)
- [X-Ray Structure of Bacteriophage HK97 by William R. Wikoff](#)
- [Balloon Phage T4 by Celeste O'Neil and Larry Goodridge](#)

- Image from the 2004 ASM Conference on the New Phage Biology
- Siphovirus pin by Jutta Loeffler
- Paper T4 by Masamune Washington

Please send any phage images that you would like to present in this section to "Phage Images," *The Bacteriophage Ecology Group*, care of Stephen T. Abedon, Department of Microbiology, The Ohio State University, 1680 University Dr., Mansfield, Ohio 44906. Alternatively, you may scan the images yourself and send them as an attachment to microdude+@osu.edu. Please save all scans in gif or jpg formats and preferably with an image size (in terms of width, height, and kbytes) that will readily fit on a standard web page. No copyrighted material without permission, please!

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New Publications

New bacteriophage publications are listed below. Each quarter not-yet-listed publications from the previous two years will be presented along with their abstracts. The indicator "???" denotes, of course, that specific information is not yet in the [BEG Bibliography](#). Please help in the compilation of the [BEG Bibliography](#) by supplying any updated information, correcting any mistakes, and, of course, e-mailing with the references to your bacteriophage ecology publications, as well as the references to any bacteriophage ecology publications that you know of but which are not yet in the bibliography or to point out references that are not appropriate for the bibliography (send to microdude+@osu.edu or to "BEG Bibliography," *Bacteriophage Ecology Group News*, care of Stephen T. Abedon, Department of Microbiology, The Ohio State University, 1680 University Dr., Mansfield, Ohio 44906). This list is also present with available abstracts at the [end](#) of *BEG News*.

1. Micro-organism re-growth in wastewater disinfected by UV radiation and ozone: a micro-biological study. Alonso, E., Santos, A., Riesco, P. (2004). *Environmental Technology* 25:433-441. [\[PRESS FOR ABSTRACT\]](#)
2. Global distribution of nearly identical phage-encoded DNA sequences. Breitbart, M., Miyake, J. H., Rohwer, F. (2004). *FEMS Microbiology Letters* 236:249-256. [\[PRESS FOR ABSTRACT\]](#)
3. Treating cocaine addiction with viruses. Carrera, M. R., Kaufmann, G. F., Mee, J. M., Meijler, M. M., Koob, G. F., Janda, K. D. (2004). *Proceedings of the National Academy of Sciences, USA* 101:10416-10421. [\[PRESS FOR ABSTRACT\]](#)
4. In vitro and in vivo bacteriolytic activities of *Escherichia coli* phages: implications for phage therapy. Chibani-Chennoufi, S., Sidoti, J., Bruttin, A., Kutter, E., Sarker, S., Brüssow, H. (2004). *Antimicrobial Agents and Chemotherapy* 48:2558-2569. [\[PRESS FOR ABSTRACT\]](#)
5. Phage-host interaction: an ecological perspective. Chibani-Chennoufi, S., Bruttin, A., Dillmann, M. L., Brüssow, H. (2004). *Journal of Bacteriology* 186:3677-3686. [\[no abstract\]](#)
6. Cloning *Serratia entomophila* antifeeding genes—a putative defective prophage active against the grass grub *Costelytra zealandica*. Hurst, M. R. H., Glare, T. R., Jackson, T. A. (2004). *Journal of Bacteriology* 186:5116-5128. [\[PRESS FOR ABSTRACT\]](#)
7. Bacteriophage lambda is a highly stable DNA vaccine delivery vehicle. Jepson, C. D., March, J. B. (2004). *Vaccine* 22:2413-2419. [\[PRESS FOR ABSTRACT\]](#)
8. Abundance, distribution, and diversity of viruses in alkaline, hypersaline Mono Lake, California. Jiang, S., Steward, G., Jellison, R., Chu, W., Choi, S. (2004). *Microbial Ecology* 47:9-17. [\[PRESS FOR ABSTRACT\]](#)
9. Involvement of colicin in the limited protection of the colicin producing cells against bacteriophage. Lin, Y. H., Liao, C. C., Liang, P. H., Yuan, H. S., Chak, K. F. (2004). *Biochemical and biophysical research communications* 318:81-87. [\[PRESS FOR ABSTRACT\]](#)
10. Development of a bacteriophage phage replication assay for diagnosis of pulmonary tuberculosis. Mc Nerney, R., Kambashi, B. S., Kinkese, J., Tembwe, R., Godfrey-Faussett, P. (2004). *Journal of Clinical Microbiology* 42:2115-2120. [\[PRESS FOR ABSTRACT\]](#)
11. Nodulation competitiveness between contrasting phage phenotypes of pigeonpea rhizobial strains. Mishra, A., Dhar, B., Singh, R. M. (2004). *Indian Journal of Experimental Biology* 42:611-615. [\[PRESS FOR ABSTRACT\]](#)
12. Assessment of the microbial integrity, sensu G.S. Wilson, of piped and bottled drinking water in the condition as ingested. Mossel, D. A. A., Struijk, C. B. (2004). *International Journal of Food Microbiology* 92:375-390. [\[PRESS FOR ABSTRACT\]](#)
13. Bacteriophages and diffusion of β -lactamase genes. Muniesa, M., Garcia, A., Miro, E., Mirelis, B., Prats, G., Jofre, J., Navarro, F. (2004). *Emerging Infectious Diseases* 10:1134-1137. [\[PRESS FOR ABSTRACT\]](#)
14. Abundance in sewage of bacteriophages infecting *Escherichia coli* O157:H7. Muniesa, M., Jofre, J. (2004). *Methods in Molecular Biology* 268:79-88. [\[PRESS FOR ABSTRACT\]](#)
15. Evaluation of a cocktail of three bacteriophages for biocontrol of *Escherichia coli* O157:H7. O'Flynn, G., Ross, R. P., Fitzgerald, G. F., Coffey, A. (2004). *Applied and Environmental Microbiology* 70:3417-3424. [\[PRESS FOR ABSTRACT\]](#)

ABSTRACT]

16. Comparison of coliforms and coliphages as tools for assessment of viral contamination in river water. Skraber, S., Gassilloud, B., Gantzer, C. (2004). *Applied and Environmental Microbiology* 70:3644-3649. [PRESS FOR ABSTRACT]
17. Viral abundance and a high proportion of lysogens suggest that viruses are important members of the microbial community in the Gulf of Trieste. Stopar, D., Cerne, A., Zigman, M., Poljsak-Prijatelj, M., Turk, V. (2004). *Microbial Ecology* 47:1-8. [PRESS FOR ABSTRACT]
18. Development and evaluation of a reflective solar disinfection pouch for treatment of drinking water. Walker, D. C., Len, S. V., Sheehan, B. (2004). *Applied and Environmental Microbiology* 70:2545-2550. [PRESS FOR ABSTRACT]
19. Prophage contribution to bacterial population dynamics. Bossi, L., Fuentes, J. A., Mora, G., Figuero-Bossi, N. (2003). *Journal of Bacteriology* 185:6467-6471. [PRESS FOR ABSTRACT]
20. Strategies for analysis of the evolution of bacteriophages. Huang, S., Hayes, S. J., Lieman, K., Griess, G. A., Serwer, P. (2001). *Recent Res. Dev. Virol.* 3:1-12. [no abstract]
21. Impage of viruses on bacterial processes. Fuhrman, J. (2000). pp. 327-350 in Kirchman, D. (ed.) *Microbial Ecology of the Oceans*. Wiley-Liss,
22. Analysis of cyanophage diversity in the marine environment using denaturing gradient gel electrophoresis. Wilson, W. H., Fuller, N. J., Jount, I. R., Mann, N. H. (2000). pp. 565-570 in Bell, C. R., Brylinsky, M., Johnson-Green, P. (eds.) *Microbial Biosystems: New Frontiers*. Atlantic Canada Society for Microbial Ecology, Halifax, Canada. [no abstract]
23. Mycobacteriophages. Hatfull, G. F. (1999). pp. 38-58 in Ratledge, C., Dale, J. (eds.) *Mycobacteria: Molecular Biology and Virulence*. Chapman and Hall, London. [no abstract]
24. The complete genome sequence of the *Streptomyces* temperate phage ϕ C31: evolutionary relationships to other viruses. Smith, M., Burns, R., Wilson, S., Gregory, M. (1999). *Nucleic Acids Res.* 27:2145-2155. [PRESS FOR ABSTRACT]
25. Marine Virus Ecology. Proctor, L. M. (1998). pp. 113-130 in Cooksey, S. E. (ed.) *Molecular Approaches to the Study of the Ocean*. Chapman & Hall, London. [no abstract]

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New Publications with Abstracts

For your convenience, a list of new publications without associated abstracts (but with links to abstracts) is found [above](#). The list presented below is identical to the [above list](#) except that abstracts are included.

1. **Micro-organism re-growth in wastewater disinfected by UV radiation and ozone: a micro-biological study.** Alonso, E., Santos, A., Riesco, P. (2004). *Environmental Technology* 25:433-441. A series of disinfection experiments using UV radiation and ozone was performed on the secondary effluent from a wastewater treatment plant at a pilot plant scale. The microbial population in the inflowing wastewater and the treated outflow water were quantified for each of the treatment modules (fecal coliforms, fecal streptococci, *Salmonella* spp. (presence/absence), *Clostridium* Sulphite-reducers, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, coliphages, nematodes, intestinal nematodes and pathogenic fungi). Treated water was stored in opaque tanks at a temperature between 20 and 22°C, after which, a one-month study of the regrowth of the bacterial flora, nematodes and fungi was carried out. *Clostridium* Sulphite-reducers, pathogenic fungi and nematodes were the micro-organisms showing a greatest degree of resistance to UV- and Ozone-treatment. It was only concerning *Clostridium* and *Pseudomonas* abatement that significant elimination results were achieved with both technologies.
2. **Global distribution of nearly identical phage-encoded DNA sequences.** Breitbart, M., Miyake, J. H., Rohwer, F. (2004). *FEMS Microbiology Letters* 236:249-256. Phages, the most abundant biological entities on the planet, play important roles in biogeochemical cycling, horizontal gene transfer, and defining microbial community composition. However, very little is known about phage diversity or biogeography, and there has not yet been a systematic effort to compare the phages found in different ecosystems. Here, we report that T7-like Podophage DNA polymerase sequences occur in every major biome investigated, including marine, freshwater, sediment, terrestrial, extreme, and metazoan-associated. The majority of these sequences belong to a unique clade that is only distantly related to cultured isolates. Some identical T7-like phage-encoded DNA polymerase genes from this clade were >99% conserved at the nucleotide level in multiple different environments, suggesting that these phages are moving between biomes in recent evolutionary time and that the global genomic pool for T7-like phages may be smaller than previously hypothesized.
3. **Treating cocaine addiction with viruses.** Carrera, M. R., Kaufmann, G. F., Mee, J. M., Meijler, M. M., Koob, G. F., Janda, K. D. (2004). *Proceedings of the National Academy of Sciences, USA* 101:10416-10421. Cocaine addiction continues to be a major health and social problem in the United States and other countries. Currently used

pharmacological agents for treating cocaine abuse have proved inadequate, leaving few treatment options. An alternative is to use protein-based therapeutics that can eliminate the load of cocaine, thereby attenuating its effects. This approach is especially attractive because the therapeutic agents exert no pharmacodynamic action of their own and therefore have little potential for side effects. The effectiveness of these agents, however, is limited by their inability to act directly within the CNS. Bacteriophage have the capacity to penetrate the CNS when administered intranasally. Here, a method is presented for engineering filamentous bacteriophage to display cocaine-binding proteins on its surface that sequester cocaine in the brain. These antibody-displaying constructs were examined by using a locomotor activity rodent model to assess the ability of the phage-displayed proteins to block the psychoactive effects of cocaine. Results presented demonstrate a strategy in the continuing efforts to find effective treatments for cocaine addiction and suggest the application of this protein-based treatment for other drug abuse syndromes.

4. **In vitro and in vivo bacteriolytic activities of *Escherichia coli* phages: implications for phage therapy.** Chibani-Chennoufi, S., Sidoti, J., Bruttin, A., Kutter, E., Sarker, S., Brüßow, H. (2004). *Antimicrobial Agents and Chemotherapy* 48:2558-2569. Four T4-like coliphages with broad host ranges for diarrhea-associated *Escherichia coli* serotypes were isolated from stool specimens from pediatric diarrhea patients and from environmental water samples. All four phages showed a highly efficient gastrointestinal passage in adult mice when added to drinking water. Viable phages were recovered from the feces in a dose-dependent way. The minimal oral dose for consistent fecal recovery was as low as 10^3 PFU of phage per ml of drinking water. In conventional mice, the orally applied phage remained restricted to the gut lumen, and as expected for a noninvasive phage, no histopathological changes of the gut mucosa were detected in the phage-exposed animals. *E. coli* strains recently introduced into the intestines of conventional mice and traced as ampicillin-resistant colonies were efficiently lysed *in vivo* by phage added to the drinking water. Likewise, an *in vitro* phage-susceptible *E. coli* strain freshly inoculated into axenic mice was lysed *in vivo* by an orally applied phage, while an *in vitro*-resistant *E. coli* strain was not lysed. In contrast, the normal *E. coli* gut flora of conventional mice was only minimally affected by oral phage application despite the fact that *in vitro* the majority of the murine intestinal *E. coli* colonies were susceptible to the given phage cocktail. Apparently, the resident *E. coli* gut flora is physically or physiologically protected against phage infection.
5. **Phage-host interaction: an ecological perspective.** Chibani-Chennoufi, S., Bruttin, A., Dillmann, M. L., Brüßow, H. (2004). *Journal of Bacteriology* 186:3677-3686. [first three paragraphs] Nearly 100 years ago, Felix d'Herelle, the codiscoverer of bacteriophages, used bacteria to control insect pests and used phages against bacterial disease. His approaches reflected ecological insights before this branch of biology became an established scientific discipline. In fact, one might have predicted that phage research would become the springboard for biotechnology and ecology. However, d'Herelle was ahead of his time, and the zeitgeist in the 1930s pushed physicists into the question "What is life?" Phages as the simplest biological systems were the logical choice for this question, and phage research became the cradle of molecular biology. ¶ Now many researchers speak of a "new age of phage research." It is now realized that phages play an important role in ecology (e.g., phage impact on the cycling of organic matter in the biosphere at a global level) (27), that phages influence the evolution of bacterial genomes (most obviously in the development of bacterial pathogenicity) (7), and that phages might provide potential tools to face the antibiotic resistance crisis in medicine (59). With this new trend, we now see a clear shift from the reductionist approach, focusing on a handful of phages in carefully controlled laboratory conditions, towards the study of many different phages in the complexity of real-life situations. ¶ In contrast to the molecular biology-oriented phage research where the interaction of molecules took center stage, ecology focuses on the interactions between organisms and their physical environment. Much of ecology is therefore about the evolution of biological diversity in space and time. In contrast to many branches of biology, ecology attributes a great importance to quantitative relationships and numbers and aims at a mathematical formulation of its observations. It is thus appropriate to start this review with an overview of phage titers encountered in the biosphere. Next, we ask how a parasite targets its host if the latter is scarce or not in an appropriate physiological state. Finally, we report on research that tries to bridge phage ecology and genomics and cell biology approaches. It is concluded that the integration of phages into complex networks of interacting biological systems, and analysis by molecular techniques, could give phage research a model character in biology again.
6. **Cloning *Serratia entomophila* antifeeding genes—a putative defective prophage active against the grass grub *Costelytra zealandica*.** Hurst, M. R. H., Glare, T. R., Jackson, T. A. (2004). *Journal of Bacteriology* 186:5116-5128. *Serratia entomophila* and *Serratia proteamaculans* (Enterobacteriaceae) cause amber disease in the grass grub *Costelytra zealandica* (Coleoptera: Scarabaeidae), an important pasture pest in New Zealand. Larval disease symptoms include cessation of feeding, clearance of the gut, amber coloration, and eventual death. A 155-kb plasmid, pADAP, carries the genes *sepA*, *sepB*, and *sepC*, which are essential for production of amber disease symptoms. Transposon insertions in any of the *sep* genes in pADAP abolish gut clearance but not cessation of feeding, indicating the presence of an antifeeding gene(s) elsewhere on pADAP. Based on deletion analysis of pADAP and subsequent sequence data, a 47-kb clone was constructed, which when placed in either an *Escherichia coli* or a *Serratia* background exerted strong antifeeding activity and often led to rapid death of the infected grass grub larvae. Sequence data show that the antifeeding component is part of a large gene cluster that may form a defective prophage and that six potential members of this prophage are present in *Photobacterium luminescens* subsp. *laumondii* TTO1, a species which also has *sep* gene homologues.
7. **Bacteriophage lambda is a highly stable DNA vaccine delivery vehicle.** Jepson, C. D., March, J. B. (2004). *Vaccine* 22:2413-2419. The stability of whole bacteriophage lambda particles, used as a DNA vaccine delivery system has been examined. Phage were found to be highly stable under normal storage conditions. In liquid suspension, no decrease in titre was observed over a 6-month period at 4 and -70°C, and phage stability was unaffected by freeze/thawing. The measured half life of phage in suspension was 36 days at 20°C, 3.4 days at 37°C and 2.3 days at 42°C. Freeze drying of a phage suspension (with or without the stabilizers dry skim milk or trehalose) resulted in 5-20% residual viability. Following desiccation (with or without stabilizers), measured half lives ranged from 20 to 100 days at 20°C, 2.6 to 38 days at 37°C, 2.1 to 26 days at 42°C, 7 to 33 h at 70°C, and 1.3 to 6m at 100°C. In all cases the addition of trehalose significantly increased the stability of the desiccated phage. When stored at -70°C, desiccated phage appeared to be stable in the absence of stabilizers. When phage lambda was diluted into water, a marginal loss in titre was observed over a 2-week period. Over a 24 h period, liquid phage suspensions were stable within the pH range pH 3-11, therefore oral administration of bacteriophage DNA vaccines

via drinking water may be possible.

8. **Abundance, distribution, and diversity of viruses in alkaline, hypersaline Mono Lake, California.** Jiang, S., Steward, G., Jellison, R., Chu, W., Choi, S. (2004). *Microbial Ecology* 47:9-17. Mono Lake is a large (180 km (2)), alkaline (pH approximately 10), moderately hypersaline (70-85 g kg⁻¹) lake lying at the western edge of the Great Basin. An episode of persistent chemical stratification (meromixis) was initiated in 1995 and has resulted in depletion of oxygen and accumulation of ammonia and sulfide beneath the chemocline. Although previous studies have documented high bacterial abundances and marked seasonal changes in phytoplankton abundance and community composition, there have been no previous reports on the occurrence of viruses in this unique lake. Based on the high concentrations and diversity of microbial life in this lake, we hypothesized that planktonic viruses are also abundant and diverse. To examine the abundance and distribution of viruses and bacteria, water samples were collected from four stations along 5 to 15 vertical depths at each station. Viral abundance ranged from 1 x 10⁸ to 1 x 10⁹ mL⁻¹, among the highest observed in any natural aquatic system examined so far. Increases (p < 0.1) in viral densities were observed in the anoxic bottom water at multiple stations. However, regression analysis indicated that viral abundance could not be predicted by any single environmental parameter. Pulsed field gel electrophoresis revealed a diverse viral community in Mono Lake with genome sizes ranging from approximately 14 to >400 kb with most of the DNA in the 30 to 60 kb size range. Cluster analysis grouped the anoxic bottom-water viral community into a unique cluster differentiating it from surface and mid-water viral communities. A hybridization study using an indigenous viral isolate as a probe revealed an episodic pattern of temporal phage distribution with strong niche stratification between oxic and anoxic waters.
9. **Involvement of colicin in the limited protection of the colicin producing cells against bacteriophage.** Lin, Y. H., Liao, C. C., Liang, P. H., Yuan, H. S., Chak, K. F. (2004). *Biochemical and biophysical research communications* 318:81-87. The restriction/modification system is considered to be the most common machinery of microorganisms for protection against bacteriophage infection. However, we found that mitomycin C induced *Escherichia coli* containing ColE7-K317 can confer limited protection against bacteriophage M13K07 and lambda infection. Our study showed that degree of protection is correlated with the expression level of the ColE7 operon, indicating that colicin E7 alone or the colicin E7-immunity protein complex is directly involved in this protection mechanism. It was also noted that the degree of protection is greater against the single-strand DNA bacteriophage M13K07 than the double-strand bacteriophage (lambda). Coincidentally, the K(A) value of ColE7-Im either interacting with single-strand DNA (2.94x10⁵M⁻¹) or double-strand DNA (1.75x10⁵M⁻¹) reveals that the binding affinity of ColE7-Im with ssDNA is 1.68-fold stronger than that of the protein complex interacting with dsDNA. Interaction between colicin and the DNA may play a central role in this limited protection of the colicin-producing cell against bacteriophages. Based on these observations, we suggest that the colicin exporting pathway may interact to some extent with the bacteriophage infection pathway leading to a limited selective advantage for and limited protection of colicin-producing cells against different bacteriophages.
10. **Development of a bacteriophage phage replication assay for diagnosis of pulmonary tuberculosis.** McNerney, R., Kambashi, B. S., Kinkese, J., Tembwe, R., Godfrey-Faussett, P. (2004). *Journal of Clinical Microbiology* 42:2115-2120. Successful infection and replication of bacteriophages is indicative of the presence of viable bacteria. We describe here the development of a bacteriophage replication assay for the detection of *Mycobacterium tuberculosis* by using mycobacteriophage D29. Optimization of phage inoculate and incubation times allowed highly sensitive detection of *M. bovis* BCG. Fewer than 10 CFU (100 CFU/ml) were detected. No false-positive results were observed in negative samples. Application of the assay to 496 sputum specimens in the National Reference Laboratory of Zambia produced sensitivity, specificity, and positive and negative predictive values of 44.1, 92.6, 82.2, and 67.5%, respectively, compared to culture on Lowenstein-Jensen medium. The equivalent corresponding results for direct fluorescent smear microscopy were 42.3, 96.8, 91.2, and 67.6%. The small increase in sensitivity over that of direct microscopy does not justify the introduction of this technique for routine diagnosis of pulmonary tuberculosis at this time.
11. **Nodulation competitiveness between contrasting phage phenotypes of pigeonpea rhizobial strains.** Mishra, A., Dhar, B., Singh, R. M. (2004). *Indian Journal of Experimental Biology* 42:611-615. Competitiveness between (I) lysogenic vs. phage-indicator strains, (II) phage-resistant vs phage-sensitive strains, and (III) large plaque vs. small plaque developing strains was examined under laboratory and field conditions in order to study the involvement of these crucial phage sensitivity patterns in the competition for nodule occupancy of pigeonpea rhizobia. The phage-indicator strain (A039) exhibited higher competitiveness over the lysogenic strain (A025 Sm_r); the phage sensitive strain (IHP-195) over the phage resistant strain (IHP 195 Sm_rV_r); and the large plaque developing strain (A059) over the small plaque developing strain (IHP195 Sm_r) in association with pigeonpea cv. bahar both under laboratory and field conditions. Dual inoculation of A025 Sm_r + A039 and A059 + IHP195 Sm_r (mixed in equal proportion just before treatment) improved the nodule occupancy by inoculant strains against native rhizobia and resulted into higher plant dry weight and yield as compared to their application as single inoculum. The phage-resistant mutant IHP195 Sm_rV_r showed reduced competitiveness against native rhizobia, compared to its parental strain. The dual inoculation of parental strain and phage-resistant mutant gave the same result as the inoculation of parental strain alone.
12. **Assessment of the microbial integrity, sensu G.S. Wilson, of piped and bottled drinking water in the condition as ingested.** Mossel, D. A. A., Struijk, C. B. (2004). *International Journal of Food Microbiology* 92:375-390. The second half of the 20th century witnessed substantial progress in the assurance and verification of microbiological integrity, i.e., safety and sensory quality, of drinking water. Enteropathogenic agents, such as particular viruses and protozoa, not previously identified as transmitted by industrially provided water supplies, were demonstrated to cause disease outbreaks, when ingested with piped water. The potential harm posed by carry-over of orally toxic metabolites of organisms, producing 'algal' (cyanophytic) blooms, was considered. In addition, earlier observations on the colonization of attenuated drinking water bodies by a variety of oligotrophic Gram-negative bacteria were confirmed and extended. This new evidence called for updating both water purification technologies and analytical methodology, serving to verify that goals had been attained. For the former purpose, the hazard analysis empowering control of critical practices (HACCP) strategy, introduced about 1960 in industrial food

processing was successfully adopted. Elimination, deactivation, or barrier technologies for the more recently identified water-borne pathogens were elaborated, taking account of the hazard of production of chlorinated compounds with alleged adverse health effects. Biofilm formation throughout water distribution networks was brought under control by strict limitation of concentrations of compounds, assimilable by oligotrophic bacteria. Upon acknowledging that direct detection tests for pathogens were futile, because of their most sporadic and erratic distribution, Schardinger's marker organism concept was anew embraced, rigorously revised and substantially enlarged. Misleading designations, like searches for 'faecal coliforms' were replaced by boundary testing for *Escherichia coli* and appropriate *Enterococcus* spp. In addition, though still to be perfected, detection protocols for relevant bacteriophages or index viruses and, to a certain extent, also for spores of aerobic and anaerobic sporing rods were also elaborated. In all monitoring account was taken of sublethally injured target organisms, surviving purification technologies, though not deprived of their ecological significance. A need remains for a rigorously standardized operating procedure (SOP) for colony counts of psychrotrophic, oligotrophic Gram-negative rod-shaped bacteria ('heterotrophic plate count'), which constitute a useful criterion of indicator value. As in the contemporary HACCP approach to food safety, guidelines for assessing success or failure in control of integrity (Water Safety Objectives) were empirically elaborated. These rely on surveys on water samples, originating from drinking water supplies, previously verified as complying with longitudinally integrated HACCP-based purification technologies. Structured Academic dissemination of these innovations, through professional microbiologists to operator and executive levels, is recommended. Web based Distance Learning MSc Programmes, like the one, since the academic year 2003-2004, offered by the University of Hertfordshire, Hatfield, UK, may contribute to such endeavours. Though the complete Course is centered around Food Safety, the Modules in-Residence Practicals and Science and Technology of Drinking Water can be studied as an entity while being employed.

13. **Bacteriophages and diffusion of β -lactamase genes. Muniesa, M., Garcia, A., Miro, E., Mirelis, B., Prats, G., Jofre, J., Navarro, F. (2004). *Emerging Infectious Diseases* 10:1134-1137.** We evaluated the presence of various β -lactamase genes within the bacteriophages in sewage. Results showed the occurrence of phage particles carrying sequences of bla_{OXA-2}, bla_{PSE-1} or bla_{PSE-4} and bla_{PSE}-type genes. Phages may contribute to the spread of some β -lactamase genes.
14. **Abundance in sewage of bacteriophages infecting *Escherichia coli* O157:H7. Muniesa, M., Jofre, J. (2004). *Methods in Molecular Biology* 268:79-88.** Bacterial virulence factors such as toxins are often encoded by bacteriophages. Among other examples, factors encoded by phages have been described in some of the emerging or re-emerging pathogens, including the pyrogenic exotoxin A production in group A streptococci, the cholera toxin in *Vibrio cholerae*, or enterotoxin production in enterohemorrhagic (EHEC) strains of *E. coli*. Most described virulence factors in Shiga toxin (Stx)-producing *E. coli* strains are located in mobile genetic elements such as plasmids and bacteriophages. Stx, which are one of the most important virulence elements in Shiga toxin-producing *E. coli* (STEC), are encoded in the genome of temperate bacteriophages infecting *E. coli* and other Enterobacteriaceae. Studies on Stx phages indicate that they are transmitted between different bacteria *in vivo* and *in vitro*. Phages could also be transmitted extraintestinally, hence the observed presence of infectious Shiga toxin phages in sewage and in fecally contaminated rivers. Stx phages also show a higher persistence under natural inactivation and disinfectant treatments in aquatic environments. This background shows that phages or lysogenic strains carrying Stx2 phages might be the natural reservoir of Stx2 genes and that lysogenization could be the main cause of the emergence of STEC strains, as suggested by several authors. It has also been suggested that lysogenization/conversion processes could take place in food and water and probably inside the human and animal gut. Ingestion of Stx2 phages could produce conversion of non-Stx2-*E. coli* strains, present inside the gut and producing new pathogenic strains. To control these phenomena, it is first necessary to gain more information about the distribution of Stx phages in the environment. For this purpose, a method of detecting Stx2 phages present in environmental water samples has been developed. The particularity of this method is that it allows detection of all (infectious and noninfectious) Stx2 phages in a water sample; in a second stage, the method allows detection of those phages able to infect and replicate on *E. coli* O157:H7. Although this method has been applied to Stx2 phages able to infect *E. coli* O157:H7, it is also applicable to detection in the natural environment of other genes carried by other bacteriophages and other bacteria.
15. **Evaluation of a cocktail of three bacteriophages for biocontrol of *Escherichia coli* O157:H7. O'Flynn, G., Ross, R. P., Fitzgerald, G. F., Coffey, A. (2004). *Applied and Environmental Microbiology* 70:3417-3424.** *Escherichia coli* O157:H7 is an endemic pathogen causing a variety of human diseases including mild diarrhea, hemorrhagic colitis, hemolytic-uremic syndrome, and thrombotic thrombocytopenic purpura. This study concerns the exploitation of bacteriophages as biocontrol agents to eliminate the pathogen *E. coli* O157:H7. Two distinct lytic phages (e11/2 and e4/1c) isolated against a human strain of *E. coli* O157:H7, a previously isolated lytic phage (pp01), and a cocktail of all three phages were evaluated for their ability to lyse the bacterium *in vivo* and *in vitro*. Phage e11/2, pp01, and the cocktail of all three virulent phages resulted in a 5-log-unit reduction of pathogen numbers in 1 h at 37°C. However, bacteriophage-insensitive mutants (BIMs) emerged following the challenge. All tested BIMs had a growth rate which approximated that of the parental O157 strain, although many of these BIMs had a smaller, more coccoid cellular morphology. The frequency of BIM formation (10⁻⁶ CFU) was similar for e11/2, pp01, and the phage cocktail, while BIMs insensitive to e4/1c occurred at the higher frequency (10⁻⁴ CFU). In addition, BIMs commonly reverted to phage sensitivity within 50 generations. In an initial meat trial experiment, the phage cocktail completely eliminated *E. coli* O157:H7 from the beef meat surface in seven of nine cases. Given that the frequency of BIM formation is low (10⁻⁶ CFU) for two of the phages, allied to the propensity of these mutants to revert to phage sensitivity, we expect that BIM formation should not hinder the use of these phages as biocontrol agents, particularly since low levels of the pathogen are typically encountered in the environment.
16. **Comparison of coliforms and coliphages as tools for assessment of viral contamination in river water. Skraber, S., Gassilloud, B., Gantzer, C. (2004). *Applied and Environmental Microbiology* 70:3644-3649.** The aim of the study was to evaluate the presence of pathogenic viruses in the Moselle River and to compare the usefulness of thermotolerant coliforms and somatic coliphages as tools for river water quality assessment in terms of viral contamination. Thermotolerant coliforms and somatic coliphages were enumerated by standardized methods in 170 samples of river water drawn from five sampling sites along the Moselle River (eastern France). BGM cell culture and integrated cell culture-reverse transcription-PCR DNA enzyme immunoassay were used to determine the presence of pathogenic viral genome (Enterovirus and Norovirus genogroup II [GGII]) and infectious Enterovirus spp.

in 90 1-liter samples. No infectious Enterovirus spp. were isolated, but Norovirus and Norovirus GGII genomes were detected in 38% of the samples. Norovirus GGII genome was mostly detected in winter, whereas Enterovirus genome was mostly detected in summer and fall. Somatic coliphages appeared to be less sensitive to higher river water temperature than thermotolerant coliforms. Furthermore, the number of river water samples positive for pathogenic viral genome increased with increasing concentration of somatic coliphages, whereas coliform concentration was unrelated to viral genome contamination. Consequently somatic coliphages, which are less sensitive to environmental factors than thermotolerant coliforms in river water, would provide a promising tool for assessment of river water quality in terms of fecal and viral pollution.

17. **Viral abundance and a high proportion of lysogens suggest that viruses are important members of the microbial community in the Gulf of Trieste.** Stopar, D., Cerne, A., Zigman, M., Poljsak-Prijatelj, M., Turk, V. (2004). *Microbial Ecology* 47:1-8. Epifluorescence microscopy and transmission electron microscopy were applied to study viroplankton community in the Gulf of Trieste (northern Adriatic Sea). The total viral abundance was in a range between $2.5 \times 10^9/L$ and $2.9 \times 10^{10}/L$ and was positively correlated with trophic status of the environment. Viruslike particles were significantly correlated with bacterial abundance in all samples studied. Correlations with other physicochemical or biological parameters were not significant. The data suggest that, because of the substantial fraction of tailed viruses present (26%), bacteriophages are an important component of the viroplankton community in the Gulf of Trieste. The abundance of viruslike particles in the seawater changed at hour intervals in a range from $1.3 \times 10^9/L$ to $5.1 \times 10^9/L$. A significant fraction (71%) of the bacterial isolates was inducible in vitro by mitomycin C, and a high occurrence (51%) of lysogenic isolates with more than one phage morphotype present in the lysate was detected. The presence of lysogenic bacteria in the seawater was confirmed in situ with a mitomycin C induction experiment on the natural bacterial population. Results suggest that viroplankton is an abundant component of the microbial community in the Gulf of Trieste.
18. **Development and evaluation of a reflective solar disinfection pouch for treatment of drinking water.** Walker, D. C., Len, S. V., Sheehan, B. (2004). *Applied and Environmental Microbiology* 70:2545-2550. A second-generation solar disinfection (SODIS) system (pouch) was constructed from food-grade, commercially available packaging materials selected to fully transmit and amplify the antimicrobial properties of sunlight. Depending upon the season, water source, and challenge organism, culturable bacteria were reduced between 3.5 and 5.5 log cycles. The system was also capable of reducing the background presumptive coliform population in nonsterile river water below the level of detection. Similar experiments conducted with a model virus, the F-specific RNA bacteriophage MS2, indicated that the pouch was slightly less efficient, reducing viable plaques by 3.5 log units in comparison to a 5.0 log reduction of enterotoxigenic *Escherichia coli* O18:H11 within the same time period. These results suggest that water of poor microbiological quality can be improved by using a freely available resource (sunlight) and a specifically designed plastic pouch constructed of food-grade packaging materials.
19. **Prophage contribution to bacterial population dynamics.** Bossi, L., Fuentes, J. A., Mora, G., Figuero-Bossi, N. (2003). *Journal of Bacteriology* 185:6467-6471. Cocultures of *Salmonella* strains carrying or lacking specific prophages undergo swift composition changes as a result of phage-mediated killing of sensitive bacteria and lysogenic conversion of survivors. Thus, spontaneous prophage induction in a few lysogenic cells enhances the competitive fitness of the lysogen population as a whole, setting a selection regime that forces maintenance and spread of viral DNA. This is likely to account for the profusion of prophage sequences in bacterial genomes and may contribute to the evolutionary success of certain phylogenetic lineages.
20. **Strategies for analysis of the evolution of bacteriophages.** Huang, S., Hayes, S. J., Lieman, K., Griess, G. A., Serwer, P. (2001). *Recent Res. Dev. Virol.* 3:1-12.
21. **Impact of viruses on bacterial processes.** Fuhrman, J. (2000). pp. 327-350 in Kirchman, D. (ed.) *Microbial Ecology of the Oceans*. Wiley-Liss,
22. **Analysis of cyanophage diversity in the marine environment using denaturing gradient gel electrophoresis.** Wilson, W. H., Fuller, N. J., Jount, I. R., Mann, N. H. (2000). pp. 565-570 in Bell, C. R., Brylinsky, M., Johnson-Green, P. (eds.) *Microbial Biosystems: New Frontiers*. Atlantic Canada Society for Microbial Ecology, Halifax, Canada.
23. **Mycobacteriophages.** Hatfull, G. F. (1999). pp. 38-58 in Ratledge, C., Dale, J. (eds.) *Mycobacteria: Molecular Biology and Virulence*. Chapman and Hall, London.
24. **The complete genome sequence of the *Streptomyces* temperate phage ϕ C31: evolutionary relationships to other viruses.** Smith, M., Burns, R., Wilson, S., Gregory, M. (1999). *Nucleic Acids Res.* 27:2145-2155. The completed genome sequence of the temperate *Streptomyces* phage ϕ C31 is reported. ϕ C31 contains genes that are related by sequence similarities to several other dsDNA phages infecting many diverse bacterial hosts, including *Escherichia*, *Arthrobacter*, *Mycobacterium*, *Rhodobacter*, *Staphylococcus*, *Bacillus*, *Streptococcus*, *Lactobacillus* and *Lactococcus*. These observations provide further evidence that dsDNA phages from diverse bacterial hosts are related and have had access to a common genetic pool. Analysis of the late genes was particularly informative. The sequences of the head assembly proteins (portal, head protease and major capsid) were conserved between ϕ C31, coliphage HK97, staphylococcal phage straight fPVL, two *Rhodobacter capsulatus* prophages and two *Mycobacterium tuberculosis* prophages. These phages and prophages (where non-defective) from evolutionarily diverse hosts are, therefore, likely to share a common head assembly mechanism i.e. that of HK97. The organisation of the tail genes in ϕ C31 is highly reminiscent of tail regions from other phage genomes. The unusual organisation of the putative lysis genes in ϕ C31 is discussed, and speculations are made as to the roles of some inessential early gene products. Similarities between certain phage gene products and eukaryotic dsDNA virus proteins were noted, in particular, the primase/helicases and the terminases (large subunits). Furthermore, the complete sequence clarifies the overall transcription map of the phage during lytic growth and the positions of elements involved in the maintenance of lysogeny.
25. **Marine Virus Ecology.** Proctor, L. M. (1998). pp. 113-130 in Cooksey, S. E. (ed.) *Molecular Approaches to*

Acknowledgements

Thank you!

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