CYANONEWS

Volume 4 Number 2 July 1988

CYANONEWS is intended to provide cyanobacteriologists with a forum for rapid informal communication, unavailable through journals. Everything you read in this newsletter is contributed by readers like yourself. The subscription rate is (at least) one communication every two years or so (your address label shows the date of your last communication). If you have a new result, if you know of an interesting meeting, if you have a post-doctoral opening, if you want strains, if you've published/submitted an article, if you have an insight or speculation into the cyanobacterial world,... why not tell us about it? It's news to us. At the very least, please send some message now and then, if only to tell us that the address is still current and you're still interested (but since you're writing anyway, a little news couldn't hurt).

INSIDE:

Please send all contributions to one of the addresses listed on the last page.

The name of the CORRESPONDENT for each item in this newsletter is capitalized, so you know who to write to for more information. The CORRESPONDENT'S ADDRESS appears at the end of the newsletter.

BULLETIN BOARD*BULLETIN BOARD*BULLETIN BOARD*BULLETIN BOARD*BULLETIN BOARD*BULLETIN BOARD*BULLETIN BOARD*

The THIRD WORKSHOP ON THE MOLECULAR BIOLOGY OF CYANOBACTERIA will be held in Toronto July 29-August 2, 1989, overlapping with the meeting of the American Society for Plant Physiology. It is hoped that some funds to cover travel expenses and lodging will be available. Contact NEIL STRAUS for further information.

The TWELFTH NORTH AMERICAN SYMBIOTIC NITROGEN FIXATION CONFERENCE is scheduled for July 30 - August 3, 1989, at Iowa State University. The scientific program will focus on the physiology, genetics, ecology, and inoculation of symbionts. Papers on all bacterial symbionts, including Rhizobium, Frankia, and cyanobacteria will be presented. Contact Steve Gendel, Dept. of Genetics, Iowa State University, Ames, IA 50011 U.S.A., (Tel) 515-294-3908.

ZVI COHEN has available a post-doc position to study environmental, biochemical, and genetic means for increased polyunsaturated fatty acid production. Send him a C.V. and three letters of recommendation.

TAN-CHI HUANG has post-doc positions available studying the molecular basis for the cyclical appearance of nitrogenase in Synechococcus RF-1. The positions come with 30,000 NT$ per month (1 US$ is currently about 28 NT$). [TCH was too polite to say so, but housing and living expenses are much lower in Taiwan than most post-doc locales, so 11,000 US$ goes a long way]

WAYNE CARMICHAEL is making available a "Directory to Toxic Cyanobacteria (blue-green algae) Literature". It attempts to list all references on toxic cyanobacteria and currently includes over 500 references. The directory is updated approximately every month.

Nguyen Hiu Thiok has recently published a monograph entitled "Photosynthesis and nitrogen fixation in the symbiotic system Azolla-Anabaena azollae" [in Russian], 148 pages. Topics include an anatomical description of both members of the symbiosis, optimization of growth, interrelationship between nitrogen fixation and photosynthesis, and economic uses of Azolla. (Contact BORIS GROMOV)

NEW PIGMENT PROTECTS NOSTOC AGAINST UV RADIATION

A new UV-B absorbing pigment with maxima at 312 nm and 330 nm, bound to a polysaccharide core, was found by S. Scherer, T.W. Chen, and PETER BÖGER. It was isolated from the cosmopolitan terrestrial cyanobacterium Nostoc commune. Colonies grown under solar UV-B radiation have high amounts of the pigment (up to 10% of dry weight), but only minor amounts are present in laboratory cultures illuminated by artificial light without UV-B. The pigment can experimentally induced by UV-B functions to efficiently protect Nostoc against UV-B radiation (submitted to Plant Physiol.).

ARRANGEMENT OF NIF GENES IN PLECTONEMA

SHREE KUMAR APTE reports that the structural genes (nifHDK) for nitrogenase are organized in a small, contiguous cluster of nearly 4 kb in the nonheterocystous cyanobacterium Plectonema boryanum-594. This arrangement differs from that of heterocystous cyanobacteria, which characteristically possess an 11 kb DNA sequence separating nifD from nifK. No DNA homologous to this sequence was detected in P. boryanum-594, nor was any found homologous to the xisA gene responsible for excision of the 11 kb sequence during heterocyst differentiation. The observed nifHDK gene arrangement resembles that found in mature heterocysts and in nitrogen-fixing unicellular cyanobacteria, such as Gloeothece.

P. boryanum-594 DNA isolated from repressed cultures (grown aerobically with nitrate) and from induced cultures (grown anaerobically without combined nitrogen) had identical hybridization patterns, when probed with fragments from nifH, D, and K from Anabaena 7120. This result indicates that there is no gene rearrangement in the vicinity of nifHDK accompanying the expression of nitrogenase activity in P. boryanum 594. In contrast, DNA from a related strain, Plectonema boryanum-581 (ATCC 27894/PCC 6306), has been reported to show different restriction patterns in the nifHDK region, depending on whether the DNA is isolated from repressed or induced cultures.

LABORATORY TIP: ARSENATE HELPS TO CLEAN UP CONTAMINATED CULTURES

It is quite frustrating but all too common to find unwelcome bacterial or fungal visitors in our cyanobacterial cultures. TERRY THIEL tells us how she has used arsenate to facilitate purification of cyanobacteria from a contaminated culture, taking advantage of the fact that most cyanobacteria are resistant to killing by arsenate at concentrations up to 100 mM, if phosphate is also present [see J. Bacteriol. (1988) 170:1143-1147 and references within]. The contaminated culture is streaked on an agar medium containing arsenate (10 mM is generally sufficient). Although the cyanobacteria that grow will appear to be axenic, viable fungi (or perhaps fungal spores) may still be present. At least one more streaking on arsenate-containing plates is therefore advisable, and then a colony from the area of lightest growth should be subcultured on arsenate-free medium to verify that the strain is axenic.

FOREIGN DNA PUT INTO THE CHROMOSOME OF A FILAMENTOUS CYANOBACTERIUM

In the early 1970's it was found that certain unicellular cyanobacteria could be genetically transformed by exogenous DNA, and several years later it was reported that foreign DNA could be inserted into their chromosomes. Progress with filamentous cyanobacteria, however, has lagged about ten years behind. So while unicellular cyanobacteriologists may be excused for wondering what the fuss is all about, the rest of us will be pleased to learn that three groups in recent months have succeeded in demonstrating the insertion of foreign DNA into the chromosome of Anabaena 7120.

JIM GOLDEN inserted the streptomycin(Sm)/spectinomycin(Sp) drug resistance cassette from pHP45Ω [Prentki and Krisch, 1984. Gene 29:303-313] into the cloned xisA gene, believed to be required for the

11 kb rearrangement in the nifHDK region during heterocyst differentiation [Lammers et al. 1986. Cell 44:905-911]. The insertion was flanked by about 8 kb Anabaena DNA on both sides, all in a pBR322-derived vector conferring resistance to ampicillin (Ap). The resulting plasmid was transferred by conjugation into Anabaena 7120, selecting for simultaneous resistance to 2 μg/ml Sm and Sp. Southern blot analysis showed that all of the initial resistant exconjugants were the result of single recombination events, in which the entire plasmid was integrated into the chromosome at xisA. No double recombinants or non-homologous events were observed.

These strains were then screened for segregaton of colonies in which a second recombination event had removed the vector portion of the plasmid (as judged by their sensitivity to 25 μg/ml Ap). Southern analysis of single recombinants shows the presence of both the xisA$^+$ gene and the inactivated copy, while double recombinants show only the inactivated copy of xisA. The single-recombinant strain was wild type for all characteristics tested, but the double recombinant xisA mutant was unable to grow on N_2 as the sole source of nitrogen and showed no nitrogenase activity. Morphological heterocyst development and pattern formation were normal. The xisA mutant does not rearrange the 11 kb element during heterocyst differentiation, but it does rearrange the nifS-55 kb element.

TERRY THIEL complemented EF113 (a Nif$^-$ mutant of Anabaena 7120) with a plasmid containing Anabaena chromosomal DNA. The plasmid confers resistance to neomycin (Nm) and cannot replicate independently in Anabaena. 40% of the colonies selected for growth on dinitrogen were found to have also gained resistance to Nm. Southern analysis of DNA isolated from Nif$^+$Nm^r cells suggested that the entire plasmid, in single copy, had recombined by a single crossover event into the chromosome at the site homologous to the cloned Anabaena DNA. DNA from Nif$^+$Nm^S exconjugants was indistinguishible from DNA from the parent strain. Since the frequency of Nif$^+$ exconjugants was more than 1000-fold greater than spontaneous reversion of EF113, double recombination (or gene conversion) is a more likely explanation of the results than reversion.

JEFF ELHAI used cloned nifHD DNA to direct insertion of foreign DNA into the chromosome of Anabaena 7120. A drug resistance element (either a rubisco-Sm/Sp fusion or psbA/Nm fusion) was inserted into nifD, flanked by 2.8 kb and 0.7 kb Anabaena DNA. Exconjugants selected for the appropriate drug resistance were found to be Nif$^+$, and Southern analysis indicated that all five tested exconjugants had gained drug resistance by single homologous recombination at nifD. Chromosomes from single recombinants should have one good copy of nifHD, one inactivated copy, and the cloning vector. A Nif$^+$Nm^r strain was grown without selection for at least 20 generations, fragmented, plated, and screened for the loss of Nm^r or the ability to grow on dinitrogen. Three percent of the resulting colonies were Nm^r and a different three percent were Nif$^-$. Colony hybridization indicates that the Nif$^-$ strains had lost the cloning vector, suggesting that a second recombination event had removed the vector and the good copy of nifHD.

PANEL DISCUSSION*PANEL DISCUSSION*PANEL DISCUSSION*PANEL DISCUSSION*PANEL DISCUSSION*PANEL DISCUSSION*PAN

SPIRULINA - CAN LABORATORY CYANOBACTERIOLOGISTS SAVE THE WORLD FROM HUNGER?

Most of us who work on cyanobacteria are content to study our favorite organism for its intrinsic beauty, but there are many other cyanobacteriologists intent on harnessing blue-greens' natural talents for very practical ends. As our ability to manipulate cyanobacteria increases, so does the value of communication between the pure and applied cyanobacteriologist, but in fact, there is now very little intercourse between the two. This is too bad, because the practical problems that seek answers may already have on laboratory benches academic answers that seek problems.

One blue-green that has had a long history of practical application is Spirulina, a filamentous, non-heterocystous cyanobacterium. Spirulina has been part of some human diets for centuries, providing a rich source of protein, vitamins (particularly B12), and the essential fatty acid gamma-linoleic acid. It can be grown on marginal land using saline water not suitable for conventional agriculture. The filamentous nature of the cyanobacterium permits harvesting simply by filtration through screens.

Despite these advantageous characteristics, Spirulina has not become a major food source. With this in mind, a panel of experts has been assembled (via the postal service) to look at the practical difficulties in cultivating Spirulina and what research directions might solve them.

Our panel: FRANCISCO AYALA (FA; Antafagasta, Chile), EIRIK DUERR (ED; Waimanalo, U.S.A.), CLARK LEE (CL; Honolulu, U.S.A.), ROBERT RAU (RR; Honolulu, U.S.A.), AMOS RICHMOND (AR; Sede Boqer, Israel), AVIGAD VONSHAK (AV; Sede Boqer, Israel)

Questions:

1. Spirulina costs 65 US$/kg at the local health food store. Why is it so expensive? What is the limiting step in production of Spirulina as food or animal feed?

Our panel was unanimous that only a small fraction of the ultimate cost of Spirulina is directly attributable to the cost of production, which ranges from $10-$20/kg for human food grade material to $1.75/kg for feed-grade, undried Spirulina. Nonetheless, as AR pointed, the cost is high in part because overall volume of Spirulina is small, and the volume will remain small so long as the cost of production is so much higher than that of Spirulina's competitors. So we return to the high cost of production, which must be substantially reduced -- another point made by all of our panelists. AV argued that economy measures, such as improvements in the nutritional supply, could reduce costs only by 20%. Only by increasing the output rates could the cost of production be reduced significantly. AR agreed, adding that very substantial improvements were possible on the low yields obtained in current reactors. AR summarized the answer to Question 1 this way: The limiting step in the end is our ignorance of how to grow microalgae efficiently on a large scale.

2. Is there any prospect that cultivation of Spirulina (or any other cyanobacterium) can develop into a major food crop? If so, in what area is the required breakthrough -- engineering? Physiology? Ecology?

Our panel split into two camps on this question. The pessimists are represented by ED who stated that there was little prospect of any use of microalgae as a food or feed source, at least in the U.S. Compared to a grain or soybean farmer, a prospective Spirulina farmer faces a considerably higher capital outlay, the absence of a support industry, and a public that already consumes more protein than it needs. He allowed, however, that the space industry might provide a special market for microalgae, and for third-world countries the scenario might be totally different, a point made by other panelists as well. If there were any major breakthrough, it would be some event that causes the price of soybeans to double for reasons that would not affect an algal farm [perhaps long-term drought?].

The optimists hoped for a major breakthrough in reactor technology. AR saw the essential problem as how to utilize efficiently solar irradiance, which impinges on the culture's surface at photon flux densities several times higher than required for light saturation of a single cell. He proposed that reactors of the future should achieve a much higher turbulence than that found in most current reactors. There are considerable advantages of closed systems over open ponds, he pointed out, particularly the ability of a closed system to cool the culture rapidly at sunset and heat it up rapidly at sunrise.

FA, CL, and RR all looked for breakthroughs in food technology, for example, the development of food products into which Spirulina can be appealingly incorporated. CL urged us to take the plunge and try eating the organism. We would find, he said, that the taste and odor is acceptable, but the color is a bit overpowering -- here is an area waiting for a breakthrough.

3. Is it reasonable to seek some expensive natural product produced by a cyanobacterium, thereby subsidizing its use as a food source? What kinds of natural products would be good candidates?

AR pointed out that the demand for an expensive natural product must be too small to subsidize the cultivation of the cyanobacterium as a major food source. ED agreed, but allowed that a small operation

might still profit by producing a fine chemical. Putting aside this dour assessment, our panelists had no qualms about offering a long list of candidate products. The list included phycobiliproteins, carotenoids, polysaccharides, antioxidants, antibiotics, and precursors to pharmaceuticals. In this regard, AV called to our attention a recent paper in Cancer and nutrition on the anti-carcinogenic effect of Spirulina-Dunaliella extracts.

4. What advice would you give to a naive cyanobacterial physiologist who wishes to study a problem related to the cultivation of cyanobacteria?

Our panel generously gave the naive cyanobacterial physiologist enough advice for several lifetimes of research projects. AR suggested a general but basic question worthy of study: What is the highest output rate of biomass attainable per unit area or per given irradiance? It is difficult to know what yields to aim for outdoors if the limits are not known in the test tube. AV put forth more specific questions. He pointed out that almost 30% of biomass produced by Spirulina during the day can be lost during hot nights by dark respiration and that photoinhibition may account for 20-40% loss of Spirulina's photosynthetic potential. Understanding these phenomena might help us find the means of selecting strains resistant to high photon flux densities and less active in the dark. Other panelists advised that studies on nutrient uptake and its relationship with turbulence would be useful. Another interesting question suggested by the panel is characteristics of culture medium used for months on end. More information here might result in less frequent changes of expensive nutrient medium.

The panel also suggested research directions that would draw the physiologist away from the lab bench. ED thought it might be profitable to screen strains for their nutritional suitability for larval shrimp, finfish, and mollusc culture. Here, algae have a natural advantage over soybeans. He also suggested that further work should be down investigating the possibility that mass algal cultures can perform water treatment functions not practical by current mechanical or chemical means. Panelists also noted the practical importance of studies on algal/bacterial interactions and species-specific competition between algal strains.

5. Suppose the time came when genes of Spirulina could be manipulated as easily as genes of E.coli. Can you think of any traits in the organism that might be improved by genetic means?

Before we went too far on our genetic flight of fancy, CL wanted to inject a note of reality. He pointed out that so far as he is aware, Spirulina has not been shown to accept DNA by transformation, transfection, or conjugation, nor are any auxotrophic mutants available, nor will the cyanobacterium stay put on plates as single colonies, nor (despite advertisements) will they make your teeth brighter and breath fresher. Clearly basic genetic work is necessary.

Despite this reminder, our panelists had a deep bag of wishes. AV advised starting with the basic processes of photosynthesis and carbon dioxide fixation. Besides lower levels of dark respiration and photoinhibition, AR wished for a strain that could use more incident light. Accoring to one model, a three-fold increase in the value for the saturating light intensity would result in almost a doubling of algal yield.

AR and ED both stressed the value of a strain that could simply grow faster. A mixotrophic Spirulina could have much higher yields as well, pointed out AR, since dense cultures could continue growing at night with the addition of a cheap carbon source, such as acetate. RR desired a strain that could grow optimally at 20 deg, but AR, still pushing the merits of a closed system wanted a strain with an optimum temperature of 55 deg, since such a system would otherwise have to be cooled during the hot hours of the day.

Several panelists asked for a strain that was more easily harvestable, with longer, stronger filaments. RR added another desired characteristic: lower water retention, to facilitate drying. Greater tolerance to variation in growth conditions -- e.g., temperature, pH, salinity, water quality -- was mentioned by CL and other panelists, and ED also suggested that an algicide-resistant Spirulina would be valuable where algal contaminants are a problem.

6. Alternatively (combining Q.3 and Q.5), is it reasonable to clone into Spirulina foreign genes that would allow it to produce some exogenous product? (This is probably attainable now). What genes or product would you suggest?

This was the key question for ED. He asked himself what can an alga do better than E.coli, on one hand, and soybeans, on the other. Not much. Forget food and feed, he advised (for reasons listed in answer to Question 2), and energy as well. The answer is to look for high volume products, since algae are cheaper to mass culture than bacteria once the quantity exceeds one ton per day. Perhaps lubricants or specialty oils are such products. CL's favorite fantasy was another potential high volume product. He proposed site directed mutagenesis of cyanophycin polymerase to produce poly asp-phe, as a precursor to aspartame.

Most of the other panelists found it more appealing to build on Spirulina's strong points, by introducing genes that would overexpress products already made by the cyanobacterium. Gamma-linolenic acid, phycoerythrin, Vitamin B12, and ß-carotene were popular suggestions, and AR added astaxanthin and eicosapentaenoic acid to the list. AV, who had held himself aloof from this speculative question, turned to the moderator of this discussion and asked whether he, a professed student of the genetics of nitrogen-fixing cyanobacteria, would take the challenge and construct a nitrogen-fixing Spirulina. This left the moderator quite speechless, and the panel discussion was abruptly terminated.

Addendum

In January 1990 AV will host the 5th SAA meeting on Recent Advances in Algal Biotechnology. He hopes to have a session on the application of genetic engineering in this field.

ED would be glad to share mass outdoor culture protocols with anyone who can honestly benefit from it.

PUBLICATIONS*PUBLICATIONS*PUBLICATIONS*PUBLICATIONS*PUBLICATIONS*PUBLICATIONS*PUBLICATIONS*P

TAXONOMY AND ECOLOGY

ANAGNOSTIDIS, K.; Economou-Amilli, A.; Makris, K. (1988). On the morphotypes of Phromidium boryanum (Bory ex Gom.) Anagn. et Kom. and Phormidium janthiphorum (Fior.-Mazz. ex Gom.) Elenk. A taxonomic consideration. Arch.Hydrobiol./Suppl. 80, Algological Studies 50-53:71-91.

ANAGNOSTIDIS, K.; Economou-Amilli, A.; Overbeck, J. (1988). Anabaena azollae Strassb., and the periphyton of Azolla filiculoides Lam. in lake Trichonis and the Lagoon of Aetolikon, Hellas (Greece). Arch.Hydrobiol. (In press).

ANAGNOSTIDIS, K.; Economou-Amilli, A.; Tafas, T. (1988). Aphanizomenon sp. from Lake Trichois, Hellas (Greece) - A taxonomic consideration in relation to morphological and ecological parameters. Arch.Hydrobiol./Suppl. 80, Algological Studies 50-53:529-543.

ANAGNOSTIDIS, K.; Pantazidou, A. (1988). Endolithic cyanophytes from the saline thermal springs of Aedipsos, Hellas (Greece). Arch.Hydrobiol./Suppl. 80, Algological Studies 50-53:555-559.

ANAGNOSTIDIS, K.; Pantazidou, A. (1988). Cyanosaccus atticus, new marine euendolithic chroococcoid cyanophyte in relation to the epilithic Podocapsa Erceg. Arch.Hydrobiol./Suppl. 78, Algological Studies 48:279-302.

ANAGNOSTIDIS, K.; Roussomoustakaki, M. (1988). Cyanophytes from metal burdened substrates. Arch.Hydrobiol./Suppl. 80, Algological Studies 50-53:561-564.

ANAGNOSTIDIS, K; Komarek, J. (1988). Modern approach to the classicfication system of cyanophytes. 3 - Oscillatoriales. Arch.Hydrobiol./Suppl.80, Algological Studies 50-53:327-472.

PHYSIOLOGY

APTE, S.K., Reddy, B.R.; Thomas, J. (1987). Relationship between Na+ influx and salt tolerance of nitrogen-fixing cyanobacteria. Appl.Env.Microbiol. 53:1934-1939.

Ernst, A.; Chen, T.W.; BÖGER, P. (1987). Carbohydrate formation in rewetted terrestrial cyanobacteria. Oecologia 72:574-576.

GROBBELAAR, N.; Hattingh, W.; Marshall, J. (1986). The occurrence of coralloid roots on the South African species of the Cycadales and their ability fo fix nitrogen symbiotically. S.Afr.J.Bot. 52:467-471.

GROBBELAAR, N.; Scott, W.E.; Hattingh, W.; Marshall, J. (1987). The identification of the coralloid root endophytes of the southern African cycads and the ability of the isolates to fix dinitrogen. S.Afr.J.Bot. 53:111-118.

Gusev, M.V.; NIKITINA, K.A.; Grot, A.V.; Vshivtsev, V.S. (1980). Age-related changes in fatty acid composition of lipids in the cyanobacterium Anabaena variabilis. Fiziol.Rastyenii 27:1083-1087.

Scherer, S.; Ernst, A.; Chen, T.W.; BÖGER, P. (1984). Rewetting of drought-resistant blue-green algae: time course of water uptake and reappearance of respiration, photosynthesis, and nitrogen fixation. Oecologia 62:418-423.

Kae, T.D.; NIKITINA, K.A. (1980). The cell contents of cyanobacteria Anabaena variabilis in dependence of the age of culture [Russian]. Vestnik MGU, Seria Biologichiskaya 16:27-31.

ALMON, H.; Böger, P. (1988). Hydrogen metabolism of the unicellular cyanobacterium Chroococcidiopsis thermalis ATCC 29380. FEMS Microbiol.Lett. 49:445-449.

APTE, S.K.; Thomas, J. (1987). Nitrogen fixation genes (nifK,D,H) in the filamentous, nitrogen-fixing cyanobacterium Plectonema boryanum do not rearrange. J.Genet.66:101-110.

Chen, T.H.; HUANG, T.C.; Chow, T.J. (19). Calcium requirement in nitrogen fixation in the cyanobacterium Synechococcus RF-1.

Helber, J.T.; Johnson, T.R.; Yarbrough, L.R.; HIRSCHBERG, R. (1988). Regulation of nitrogenase gene expression in anaerobic cultures of Anabaena variabilis. J.Bacteriol. 170:5525-557.

Helber, J.T.; Johnson, T.R.; Yarbrough, L.R.; HIRSCHBERG, R. (1988). Effect of nitrogenous compounds on nitrogenase gene expression in Anaerobic cultures of Anabaena variabilis. J.Bacteriol. 170:558-563.

HUANG, T.C.; Chow, T.J.; Hwang, I.S. (1988). The cyclic synthesis of the nitrogenase of Synechococcus RF-1 and its control at the transcription level. FEMS Microbiol.Lett. 50:127-130.

Reich, S.; ALMON, H.; BÖGER, P. (1987). Comparing short-term effects of ammonia and methylamine on nitrogenase activity in Anabaena variabilis (ATCC 29413). Z.Naturforsch. 42c:902-906.

Reich, S.; ALMON, H.; BÖGER, P. (1986). Short-term effect of ammonia on nitrogenase activity of Anabaena variabilis (ATCC 29413). FEMS Microbiol.Lett. 34:53-56.

Thomas, J.; APTE, S.K.; Bhagwat, A.A.; Vacchani, A.K.; Fernandes, T.; Tuli, R (1987). Aspects of cyanobacterial nitrogen fixation, early interactions in legume-rhizobium symbiosis and prospects for genetic manipulations. In: Biotechnology in Agriculture (S.Natesh, V.L.Chpora, S.Ramachandran, eds.). Oxford & IBH Publ. Co., New Delhi, pp.47-59.

TOXICOLOGY

Botes, D.P.; Wessels, L.; Kruger, H.; RUNNEGAR, M.T.C.; Santikarn, S.; Smith, R.J.; Barna, J.C.J.; Williams, D.H. (1985). Structural studies on cyanoginosins-LR, -YR, -YA, and -YM, peptide toxins from Microcystis aeruginosa. J.Chem.Soc.Perkin Trans. 2747-2748.

CARMICHAEL, W.W. (1988). Toxins of freshwater algae. In: Handbook of Natural Toxins, Vol.3 Marine Toxins and Venoms. (A.T.Tu, ed.). Marcel Dekker, Inc., pp.121-147.

DABHOLKAR, A.S.; Carmichael, W.W. (1987). Ultrastructural changes in the mouse liver induced by hepatotoxin from the freshwater cyanobacterium Microcystis aeruginosa strain 7820. Toxicon 25:285-292.

DABHOLKAR, A.S.; Carmichael, W.W.; Berg, K.; Wyman, J. (1987). Ultrastructural changes in isolated rat liver perfused with cyclic heptapeptide toxins from Norwegian freshwater cyanobacteria (Blue-green algae). Proc.45th Ann.Meeting Electron Microsc.Soc.of America, pp.858-859.

FALCONER, I.R.; RUNNEGAR, M.T.C. (1987). Effects of the peptide toxin from Microcystis aeruginosa on intracellular calcium, pH and membrane integrity in mammalian cells. Chemico-Biol.Interactions 63:215-225.
Falconer, I.R.; RUNNEGAR, M.T.C.; Buckley, T.H.; Huyn, V.L.; Bradshaw, P. (1988). Use of carbon to remove blue-geen algal toxins from drinking water. (Submitted).
Falconer, I.R.; Smith, J.V.; Jackson, A.R.B.; Jones, A.; RUNNEGAR, M.T.C. (1988). Oral toxicity of a bloom of the cyanobacterium Microcystis aeruginosa administered to mice over peirods up to one year. (In press).
Gorham, P.R.; CARMICHAEL, W.W. (1988). Hazards of freshwater blue-greens (cyanobacteria). In: Algae and Human Affairs (C.A. Lembi, L.R. Waaland, eds.). Cambridge Univ. Press, p.403-31.
Jackson, A.R.B.; RUNNEGAR, M.T.C.; Cumming, R.B.; Brunner, J.F. (1986). Experimental acute intoxication of young layer and broiler chickens with the cyanobacterium (blue green alga) Microcystis aeruginosa. Avian Patho. 15:741-748.
RUNNEGAR, M.T.C.; Andrews, J.; Gerdes, R.G.; FALCONER, I.R. (1987). Injury to hepatocytes induced by a peptide toxin from the cyanobacterium Microcystis aeruginosa. Toxicon 25:1235-1239.
RUNNEGAR, M.T.C.; Jackson, A.R.B.; Falconer, I.R. (1988). Toxicity to mice and sheep of a bloom of the cyanobacterium (blue-green alga) Anabaena circinalis. Toxicon (In press).
RUNNEGAR, M.T.C.; Jackson, A.R.B.; FALCONER, I.R. (1988). Toxicity of the cyanobacterium Nodularia spumigena Mertens. Toxicon 26:143-151.

BIOENERGETICS

Grunwaldt, G.; NIKITINA, K.A. (1982). Uptake and efflux of fluoresceine in Anacystis nidulans. Biol.Plantarum 24:270-274.
Johnson, T.R.; Haynes II, J.I.; Wealand, J.L.; Yarbrough, L.R.; HIRSCHBERG, R. (1988). Structure and regulation of genes encoding phycocyanin and allophycocyanin from Anabaena variabilis ATCC 29413. J.Bacteriol. 170:1858-1865.
KALLA, R. (1988). Acclimation of light-harvesting antennae in the cyanobacterium Synechococcus 6301: Transcriptional organization and coordination of gene expression of the phycobilisome rod operon. Thesis, University of Umeå.
KALLA, R.; Gustafsson, P (1988). Genetic analysis of phycobilisome mutants in the cyanobacterium Synechococcus sp. PCC 6301. Molecular Microbiology.
KALLA, R.; Lind, L.; Lidholm, J.; Gustafsson, P (1988). Transcriptional organization of the phycocyanin subunit gene clusters in the cyanobacterium Anacystis nidulans sp. UTEX 625. (submitted).
KALLA, S.R.; Lönneborg, A.; Öquist, G.; Gustafsson, P. (1986). Light-modulated antennae acclimation in the cyanobacterium Anacystis nidulans: effects of transcriptional and translational inhibitors. J.Gen.Microbiol. 132:3195-3200.
KALLAS, T.; Spiller, S.; Malkin, R. (1988). Primary structure of cotranscribed genes encoding the Rieske-FeS and cytochrome-f proteins of the cyanobacterium Nostoc PCC 7906. Proc.Natl.Acad.Sci.USA (In press).
Kuwabara, T.; Reddy, K.J.; SHERMAN, L.A. (1987). Nucleotide sequence of the gene from the cyanobacterium Anacystis nidulans R2 encoding the Mn-stabilizing protein involved in photosystem II water oxidation. Proc.Natl.Acad.Sci.USA 84:8230-8234.
NIKITINA, K.A.; Gusev, M.V. (1980). Interaction of Anabaena variabilis and Anacystis nidulans with methylviologen. Mikrobiol. 49:483-487.
PAPAGEORGIOU, G.C.; Lagoyanni, T. (1985). Photosynthetic properties of rapidly permeabilized cells of the cyanobacterium Anacystis nidulans. Biochim.Biophys.Acta 807:230-237.
PAPAGEORGIOU, G.C.; Sotiropoulou, G. (1987). Functional interactions of anions and cations with the outer surface of intact thylakoids: studies with the blue-green bacterium Anacystis nidulans. In: Prog.Photosynth.Res.Vol II (J.Biggens, ed.). Martinus Nijhoff Pub., Dordrecht, pp.785-788.
Scherer, S.; Almon, H.; BÖGER, P. (1988). Interaction of photosynthesis, respiration and nitrogen fixation in cyanobacteria. Photosynth.Res. 15:95-114.
Scherer, S.; Häfele, U.; Krüger, G.H.J.; BÖGER, P. (1988). Respiration, cyanide-insensitive oxygen uptake and oxidative phosphorylation in cyanobacteria. Physiol.Plantarum 72:379-384.
Scherer, S.; Sadowski, H.; BÖGER, P. (1987). Bioenergetics studies of the cyanobacterium Anabaena variabilis. Z.Naturforsch. 42c:1280-1284.

Sotiropoulou, G.; PAPAGEORGIOU, G.C. (1986). Stimulation and inhibition of photosystem II electron transport in cyanobacteria by ions interacting with the cytoplasmic face of thylakoids. Photosynth.Res. 10:445-450.

APPLIED CYANOBACTERIOLOGY

PAPAGEORGIOU, G.C. (1987). Immobilized photosynthetic microorganisms. Photosynthetica 21:367-383.
PAPAGEORGIOU, G.C.; Lagoyanni, T. (1986). Immobilization of photosynthetically active cyanobacteria in glutaraldehyde-crosslinked albumin. Appl.Microbiol.Biotechnol. 23:417-423.
SHI, D.J.; Brouers, M.; Hall, D.O.; Robins, R.J. (1987). The effects of immobilization on the biochemical physiological and morphological features of Anabaena azollae. Planta 172:298-308.
SHI, D.J.; Hall, D.O. (1988). Azolla and immobilized cyanobacteria (blue-green algae): from traditional agriculture to biotechnology. Plants Today Jan/Feb.
SHI, D.J.; Hall, D.O.; Tang, P.S. (1987). Photosynthesis, nitrogen fixation, ammonia photoproduction and structure of Anabaena azollae immobilized in natural and artificial systems. In: Progress in Photosynth.Res. II (J.Biggens,ed.), pp.641-644.
COHEN, Z.; Vonshak, A.; Richmond, A. (1987). Fatty acid composition of Spirulina strains grown under various environmental conditions. Phytochem. 26:2255-2258.
VONSHAK, A. (1987). Biological limitations in developing the biotechnology for algal mass cultivation. Sciences de L'eau 6:99-103.
VONSHAK, A. (1987). Strain selection of Spirulina suitable for mass production. Hydrobiol. 151:75-77.
VONSHAK, A.; Guy, R. (1988). Photoinhibition as a limiting factor in outdoor cultivation of Spirulina platensis. Recent Advances in Algal Biotechnology.
VONSHAK, A.; Richmond, A. (1988). Mass production of the blue-green alga Spirulina: An overview. Biomass (In press).

ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRESSES*ADDRE

Anagnostidis, Konstantinos	Institute of Systematic Botany, Section Ecology & Systematics, University of Athens, Panepistimiopolis, GR- 157 84 Athens, Hellas, GREECE
Apte, Shree Kumar	Dept. Molecular Genetics & Cell Biology, University of Chicago, 920 E 58th Street, Chicago, IL 60637 U.S.A.
Ayala, Francisco	P.O.Box 691, Antafagasta, CHILE
Böger, Peter	Lehrstuhl für Physiologie und Biochemie der Pflanzen, Universität Konstanz, D-7750 Konstanz, FRG - W.GERMANY
Carmichael, Wayne W.	Dept. of Biological Sciences, Wright State University, Dayton, Ohio 45435 U.S.A.
Cohen, Zvi	Algal Biotechnology Lab, Ben-Gurion University of the Negev, Jacob Blaustein Inst. for Desert Research, Sede Boqer Campus 84990 ISRAEL
Dabholkar, Arun S.	535 N. Michigan Ave., Unit 402,Chicago, Illinois 60611 U.S.A.
Duerr, Eirik O.	The Oceanic Institute, Waimanalo, HI 96795 U.S.A.
Falconer, I.R.	Department of Biochemistry, Microbiology and Nutrition, University of New England, Armidale, N.S.W. 2351,, AUSTRALIA
Golden, Jim	Department of Biology, Texas A&M University, College Station, TX 77843 U.S.A.
Grobbelaar, Nathanael	Department of Botany, Universiteit van Pretoria, Faculty of Mathematics & Sci., 0002 Pretoria, SOUTH AFRICA
Gromov, Boris V.	Biological Institute of Leningrad University, Oranienbaumskoye sch.2, Stary Peterhof, Leningrad, 198904 USSR
Hirschberg, Rona	School of Basic Life Sciences, University of Missouri, Kansas City, Missouri 64110 U.S.A.
Huang, Tan-Chi	Institute of Botony, Academia Sinica, Nankang, Taipei, Taiwan, R.O.CHINA
Kalla, Roger	Institute for Applied Cell and Molecular Biology, Umeå University, S-901 87 Umeå, SWEDEN
Kallas, Toivo	Div. of Molecular Plant Biol., Hilgard Hall, Univ. California, Berkeley, CA 94720 U.S.A.

Lee, Clark	University of Hawaii-Manoa, Dept. Biochem-Biophys., 1960 E.W. Rd., Honolulu, HI 96822 U.S.A.
Nikitina, Kamilla A.	Department of Plant Physiology, Biological Faculty, M.V. Lomonosov State University, 119899 Moscow, U.S.S.R.
Papageorgiu, George C.	NRC Demokritos - Biology, Aghia Paraskevi, Athens, 15310 GREECE
Rau, Robert C.	Rau Botanical Laboratories, 2590 La-i Rd., Honolulu 96816 U.S.A.
Richmond, Amos	Micro-Algal Biotechnology Lab., Jacob Blaustein Inst. for Desert Research, Ben Gurion Univ. of the Negev, Sede Boker Campus, 84993 ISRAEL
Runnegar, Maria	University of California, Department of Microbiology, 5304 Life Sciences Building, College of Letters and Sci., 405 Hilgard Avenue, Los Angeles, California 90024 U.S.A.
Sherman, Louis	University of Missouri, Div. of Biological Sci., Tucker Hall, Columbia, MO 65211 U.S.A.
Shi, Ding Ji	Institute of Botony, Academia Sinica, Beijing, P.R.CHINA
Thiel, Terry	Biology Dept., University of Missouri, St. Louis, MO 63121 U.S.A.
Vonshak, Avigad	Jacob Blaustein Institute for Desert Research, Sede Boqer Campus, 84990 ISRAEL

Send CONTRIBUTIONS to one of the addresses listed below. If you wish to be included in the mailing list, send your name, address, telephone number, and a brief description of your research interests.

AUSTRALIA/NEW ZEAL./SE.ASIA	Steve Delaney	Department of Biotechnology, The University of New South Wales, P.O. Box 1, Kensington, New South Wales, AUSTRALIA 2033
AUSTRIA	Georg Schmetterer	Institut für Physikalische Chemie, Währingerstrasse 42, A-1090 WIEN
CANADA	Neil Strauss	Dept. of Botany, University of Toronto, Toronto, Ontario M5S 1A1
P.R.CHINA	Shang-Hao Li	Laboratory of Phycology, Institute of Hydrobiology, Academia Sinica, Wuhan
FRANCE	Nicole Tandeau de Marsac	Physiologie Microbienne, Institut Pasteur, 29 rue du Dr. Roux, 75724 Paris Cedex 15
FRG-W.GERMANY	Wolfgang Lockau	Institut für Allgemeine Biochemie, Universitätsstr. 31, 8400 Regensburg,
GDR-E.GERMANY /CZECH.	J.-G. Kohl	Section Biology at Humboldt University, Department Ecology, Invalidenstraße 43, Berlin 1040, DDR-GERMANY
INDIA	Joe Thomas	Biology and Agriculture Div., Modular Labs, Bhabha Atomic Research Centre, Trombay, Bombay 400 085
ISRAEL	Elisha Tel-Or	Dept. of Agricultural Botany, The Hebrew University, Rehovot 76100
NETHERLANDS	Luuc Mur	Laboratorium voor Microbiologie, Universiteit voor Amsterdam, Nieuwe Achtergracht 127, 1018 WS Amsterdam
NORWAY	Olav Skulberg	Norwegian Institute for Water Research, P.B. 333, Blindern, N-0314, Oslo 3
U.K.	Tony Walsby	Dept. of Botany, University of Bristol, Bristol BS8 1UG
ANYWHERE ELSE	Jeff Elhai	MSU/DOE Plant Research Laboratory, Michigan State University, East Lansing, MI 48824, U.S.A.