A brief history of the molecular biology and genetics of cyanobacterial toxicity and its future in the age of OMICS

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The past ten years has witnessed major advances in our understanding of natural product biosynthesis, including the genetic basis for toxin production by a number of groups of cyanobacteria. Cyanobacteria produce an unparalleled array of bioactive secondary metabolites; including alkaloids, polyketides and non-ribosomal peptides, some of which are potent toxins. This paper addresses the molecular genetics underlying cyanotoxin production in fresh and brackish water environments. The major toxins that have been investigated include microcystin, cylindrospermopsin, nodularin, the paralytic shellfish poisons (PSP), including saxitoxin, and the anatoxins. Potentially toxic cyanobacteria in drinking water supplies pose a direct threat to public health.

Non-ribosomal peptide synthesis is achieved in prokaryotes and lower eukaryotes via the thiotemplate function of large, modular enzyme complexes, known collectively as peptide synthetases. Most non-ribosomal peptides from microorganisms are classified as secondary metabolites, that is, they rarely have a role in primary metabolism, growth, or reproduction but have evolved to somehow benefit the producing organism. Most cyanobacterial genera have either been shown to produce non-ribosomal peptides or have them encoded within their genomes. Early work on the genetics of cyanobacterial toxicity led to the discovery of one of the first examples of hybrid peptide-polyketide synthetases. This enzyme complex directed the production of the cyclic heptapeptide microcystin and as one of the largest known bacterial gene clusters, is encoded by more than 55 kb. Orthologs of microcystin synthetase have been found in several strains of *Microcystis* and other species of toxic cyanobacteria, including *Planktothrix*, *Nostoc*, *Anabaena*, *Nodularia*, *Phormidium*, and *Chroococcus*. The homologous gene cluster in *Nodularia* is predicted to be responsible for the synthesis of the pentapeptide nodularin, providing evidence of genetic recombination and possibly transfer during the evolution of these compounds. Genomic information related to microcystin and nodularin synthesis has also indicated their environmental and cellular regulators, as well as associated transport mechanisms. Light quality and quantity has been shown to regulate toxin gene expression in *Microcystis* and it appears that other cellular interactions, possibly involving nitrogen metabolism and quorum sensing also affect toxin levels.

More recently, hybrid peptide and polyketide synthetic pathways have been implicated in the production of the alkaloid cylindrospermopsin that possibly contradict previous feeding studies. Predicted biosynthetic pathways are also under scrutiny and are being used in the search for candidate gene loci involved in PSP and anatoxin production. The correlation between toxicity and salt tolerance may raise future concerns as these cyanobacteria could compromise the safety of recycled and desalinated drinking water supplies.

The pattern of acquisition of genes responsible for cyanobacterial toxicity is not, on the whole, related to the evolution of potentially toxic species and the global distribution of toxic strains has been the topic of several phylogeographical studies. The exceptions to this include Australian strains of *Anabaena circinalis* that produce saxitoxin and the species *Nodularia spumigena*. Toxin biosynthesis gene cluster-associated transposition and the natural transformability of certain species allude to a broader distribution of toxic taxa. The information gained from the discovery of these toxin biosynthetic pathways has also enabled the genetic screening of various environments for drinking water quality management. Understanding the role of these toxins in the producing microorganisms and the responses of their genes to the environment may suggest the means for controlling toxic bloom events.