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Relationship between Cyanobacteria, Cyanotoxins, and Toxin Synthetase Genes in the Environment

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Cyanobacteria, aka blue-green algae, are widespread in global freshwater bodies including important water resources for animals such as livestock and wildlife. They can proliferate sharply and finally form harmful algal blooms (HABs) in eutrophic waters where microalgal nutrients are overly enriched including nitrates and phosphates in hot seasons. Although HABs can bring disastrous impacts on aquatic ecosystems, cyanobacterial threats to terrestrial animals concern us more because they can produce potently lethal toxins namely cyanotoxins, and animals as well as humans that drink water contaminated by the toxins are poisoned to acquire compromised health condition or even death. In the worst-case scenario, animals enhance their water consumption due to a quick loss of internal water in hot days, considerably increasing the risk of cyanotoxin poisoning. Therefore, monitoring of toxic cyanobacteria and cyanotoxins should be taken into action for the wellness of animals involved in problematic waters.

Not all cyanobacteria can produce cyanotoxins, and a single species may have toxic and non-toxic strains. Nevertheless, the differentiation between toxic and non-toxic strains is impossible to be visibly noticed due to their identical morphologies. As a consequence, observation of certain species which are reportedly toxic is not a reliable measure for the confirmation of toxic cyanobacteria. On the other hand, it has been found that production of cyanotoxins is realized by the enzymatic synthetic reactions catalyzed by a group of proteins that are the expressed products of toxin synthetase genes in toxic cyanobacteria. Toxins are secreted by active transportation of cyanobacteria or released when cyanobacterial cells are broken apart due to apoptosis or external forces. However, whether toxin production is consistent as housekeeping gene expression or regulated by certain intracellular or extracellular factors is still a mystery. Then presence of toxin genes may not directly implicate the presence of toxins. Here comes up the question: what are their relationships in fact?.

The author studied Microcystis spp., microcystin, and microcystin synthetase genes (mcy) and investigated the relationships in sixty field samples collected from four farm ponds utilized to provide drinking water to four nearby swine facilities in the Midwestern United States in 2015. While microscopic biomass of Microcystis spp. and concentration of microcystin determined by liquid chromatography – mass spectrometry (LC-MS) were used for a quantitatively descriptive purpose, mcy genes were only reported as presence or absence via their detection by polymerase chain reaction (PCR). Finally, 56 out of 60 samples contained Microcystis spp., and 17 were microcystin positive, and mcy genes were found in 34 samples. Please note that the small-numbered positive samples were not totally incorporated in large-numbered ones, meaning a few of them were only positive for Microcystis spp., microcystin, mcy genes, or two out of the three. The subsequent investigation of one-on-one correlation using statistical tools revealed the following facts:

A. The biomass of Microcystis spp. could not predict the presence of mcy genes because they did not have any correlation in logistic regression test ($p = 1$). This implies that the existence of toxic cyanobacteria cannot be guaranteed merely by seeing the cells microscopically. Detection of toxin genes is a must now that "what you see is not what you get".

B. The microcystin levels could not be estimated using the biomass of Microcystis spp. because the two factors did not have a good linear correlation in Pearson's r ($r2 = 0.184$).

It is easily understood that different Microcystis populations may comprise different proportions of toxic cells, so the production of microcystin is discrepant.

C. Presence of mcy genes could be used as a predictive measurement for the presence of microcystin, or vice versa as they showed significant interdependency in McNemar's test ($p << 0.05$). This might help prove the proposal that toxin synthetase genes be consistently expressed like the housekeeping genes, whereas the detected toxins cannot directly reflect the gene expression because they are influenced by many intracellular and extracellular factors including secretory mechanism and environmental degradation.

In summary, cyanobacteria, cyanotoxins, and toxin synthetase genes have no correlative relationships except the presence of toxins and genes. However, it cannot be so easily taken for granted that their relationships are as such because there are many environmental factors affecting the measurement which cannot be controlled or assessed regarding their contributions to the conclusion. Therefore, further studies under more stringent and controllable conditions should be conducted to disclose their relationships as profoundly as possible, for instance, whether the toxin production is consistent or regulated, and what is the quantitative correlation between toxins and genes. All these efforts will no doubt facilitate the development of robust diagnostic and appraisal methods for toxic cyanobacteria and cyanotoxins so as to secure the water usage of animals.

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