

DEVELOPMENT OF SENSITIVE IMMUNOASSAY FORMATS FOR ALGAL TOXIN DETECTION

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Cyanobacteria (blue-green-algae) are known to produce various toxins. Cyanobacteria live in terrestrial, fresh, brackish, or marine water. Some of the toxins they produce can be highly toxic, others can cause severe taste and odor problems in drinking water supplies. Cyanobacterial toxins can make drinking water and recreational use of water unsafe. Animals die each year as a result of cyanotoxins, and though human death is not common, many people experience symptoms indicative of cyanotoxin exposure. Very little is known about the long-term side effects of the ingestion of cyanotoxins, although there is a guideline set by WHO for safe concentrations, these minimal concentrations could have an effect over time.

Concerns about contamination of lakes and reservoirs with algal toxins have led to the need for more rapid, sensitive, and selective methods of analysis. To protect populations from adverse health effects due to microcystins (MC), the WHO has established an upper limit for MC-LR of 1 ng/mL (ppb) in drinking water and 20 ng/mL in recreational waters. However, effective consumer protection should require the sensitive and effective detection of the whole spectrum of congeners, many of which are as toxic as MC-LR. This requires that methods of analysis be able to quantify the various congeners at concentrations below WHO limits.

Immunoassays (ELISAs) have proven to be rapid, sensitive, accurate, and cost-effective. Microtiter plate ELISAs have previously been described and widely applied to the detection of pesticides and other environmental contaminants in various sample matrices including water, soil, produce, and fish tissue. Other immunoassay formats utilizing immunochromatography are often simple devices, where a test sample is analyzed for the presence of certain analytes. For example, a specified volume of the sample is added to a tube containing a pre-dispensed antibody-gold conjugate and allowed to react. The solution is then contacted with one end of a test strip containing discrete reactive zones. As the sample is wicked up the test strip, the analyte in the sample continues to react with the antibody. As the reaction mixture flows up the strip, any reaction between the antigens and the analyte, if present, may be observed by the appearance or absence of color in the discrete zones.

This paper describes the development and assay performance of ELISAs for microcystins, cylindrospermopsin, and saxitoxin in water samples, and an immunochromatographic device for the quick analysis of MC in recreational waters. The results show that the ELISAs exhibit parts per trillion (ng/L, ppt) sensitivity in water samples. The sensitivity of the immunochromatographic device allows the detection of MC at 1 and 10 ng/mL. Average recoveries in water were between 85-115% and within and between assay precision of < 12%. Data obtained as well as sample comparison using a newly developed cell lysis procedure will also be presented.

KEYWORDS

Microcystin, Cylindrospermopsin, Saxitoxin, ELISA, immunochromatography, cell lysis