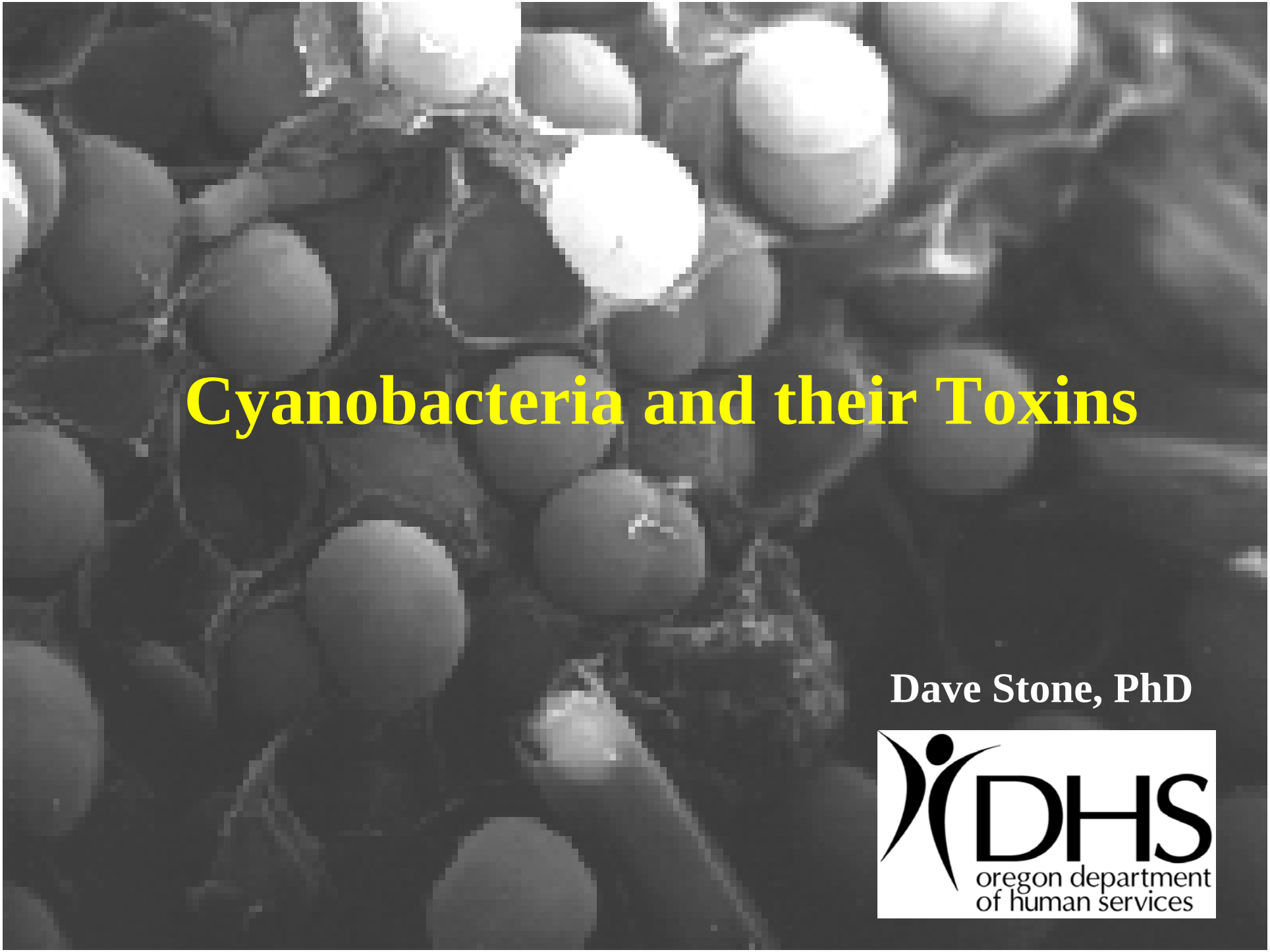

Cyanobacteria and their Toxins

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Cyanobacterial Toxins:

Cyanobacteria can produce a wide array of neurotoxins, liver toxins, cell toxins and skin irritants. In addition, many genera, such as *Anabaena*, can produce multiple toxins.

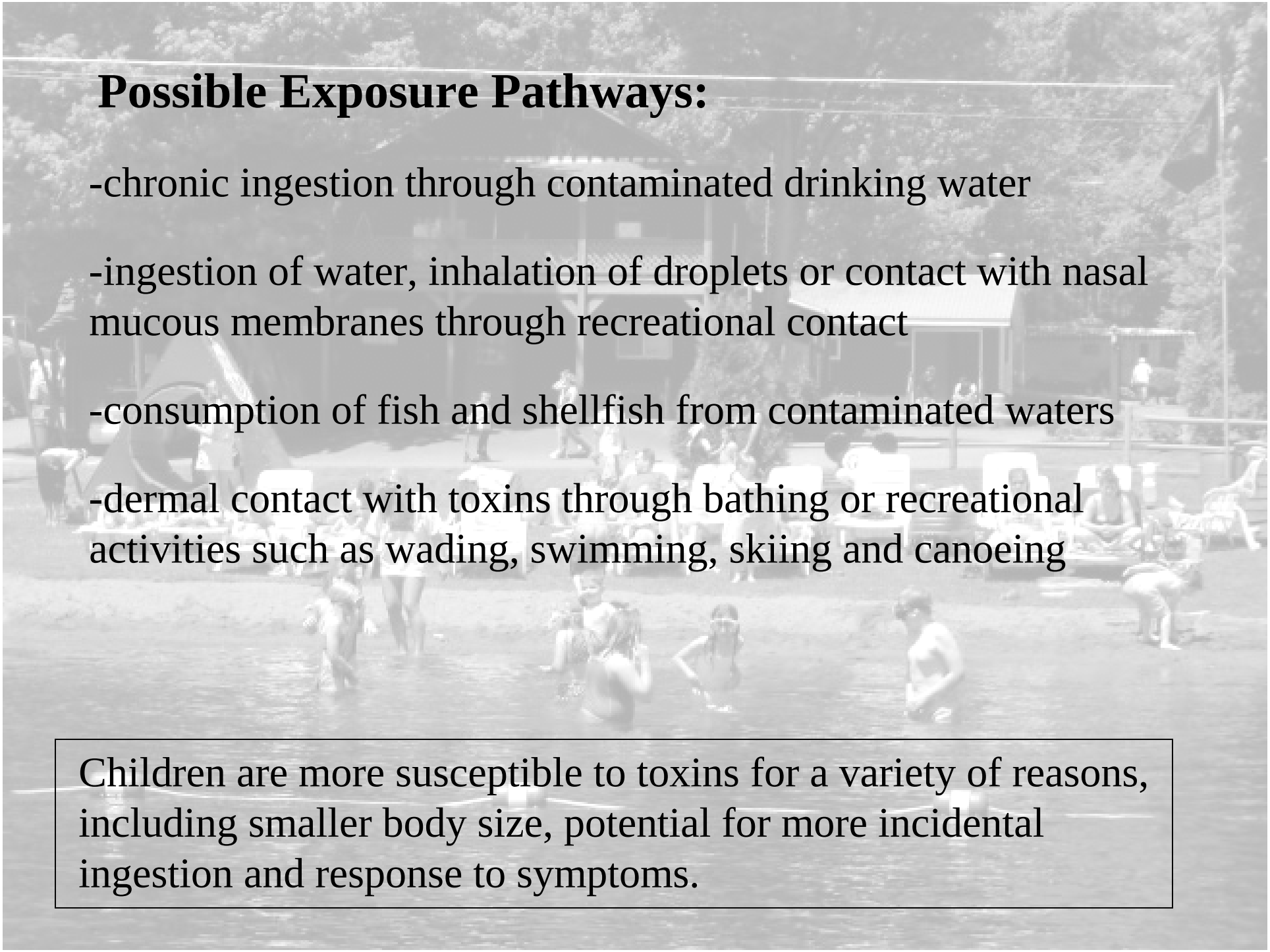
Toxin	Cyanobacteria genera
Microcystin	*Microcystis*, *Anabaena*, *Oscillatoria*, *Nostoc*, *Anabaenopsis*, & more
Anatoxin-a	*Anabaena*, *Aphanizomenon*
Clyindrospermopsin	*Cylindrospermopsis*, *Aphanizomenon*
Saxitoxins	*Anabaena*, *Cylindrospermopsis*, *Aphanizomenon*, & more
Lipopolysaccharides	All

Toxin Production:

- Most toxins in cyanobacteria are found in the cell when conditions are favorable. Older and senescing blooms tend to release toxins into the water as the cells break open (or via treatment with copper sulphate).

- Studies have shown that in some strains, toxin content is highest in cells at temperatures between 18-25 ^{0}C; low (10 ^{0}C) or very high temperatures (30 ^{0}C) decreased toxin content. Cells were also found to be have higher toxin levels when grown at extremes of pH.

- In high phosphorus containing waters, hepatotoxic strains produced more toxins (no effect on anatoxin-a production). Non-nitrogen fixing species, such as *Microcystis*, produce more toxins under nitrogen-rich conditions.



Possible Exposure Pathways:

-chronic ingestion through contaminated drinking water

-ingestion of water, inhalation of droplets or contact with nasal mucous membranes through recreational contact

-consumption of fish and shellfish from contaminated waters

-dermal contact with toxins through bathing or recreational activities such as wading, swimming, skiing and canoeing

Children are more susceptible to toxins for a variety of reasons, including smaller body size, potential for more incidental ingestion and response to symptoms.

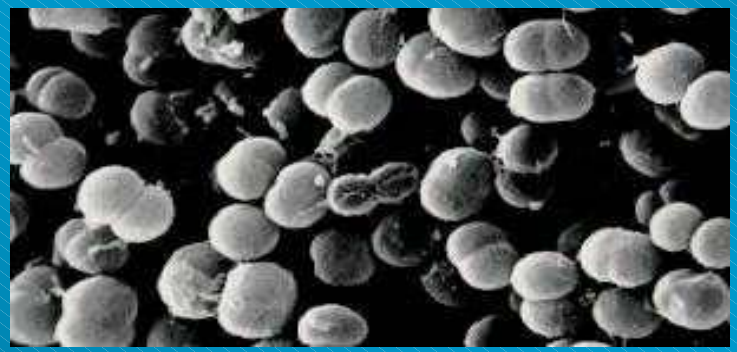
Mirocystins:

Globally, microcystins are the most commonly detected cyanobacterial toxin in fresh & brackish waters.

The toxins are water soluble and most microcystins cannot penetrate directly through plant or animal cell membranes. Uptake into cells occurs through membrane transporters. The liver is the ultimate target organ for toxic effects.

Microcystins are highly toxic with low doses required for lethal effects. The dose-response curve is steep – acute damage as threshold is approached.

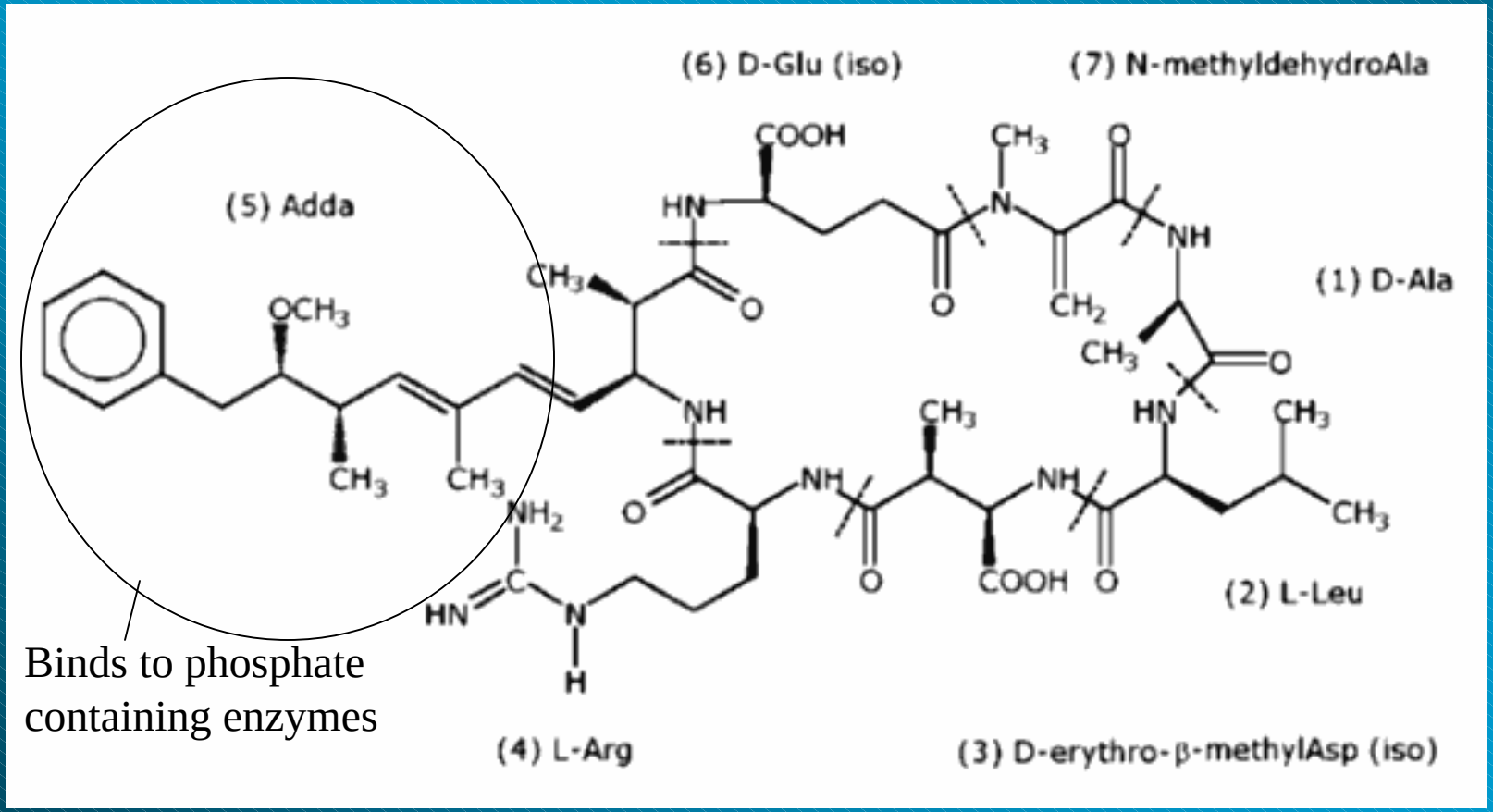
There is some evidence that microcystins can act as tumor promoters as well.



(Microcystis aeruginosa)

Microcystin toxin:

-large, cyclic peptide with several structural variants
-the linear form (break-down or precursor products) is 100x less toxic compared with the cyclic toxin



Microcystin Disease Outbreaks:

- 1988 (Brazil): Severe gastro-enteritis epidemic developed with over 2000 cases reported (88 deaths). Results pointed to a massive bloom of *Anabaena and Microcystis* in reservoir.
- Epidemiological link to liver cancer in rural Chinese populations infected with Hepatitis-B and drinking water contaminated with microcystins.
- In Brazil, 117 patients developed liver disease (50+ deaths) attributed to dialysis with microcystin-contaminated water (also occurred in the U.S. in 1975)
- Atlantic salmon reared in B.C. and Washington dieing of progressive liver disease from an unidentified organism producing microcystins; severe economic losses

Fate and Breakdown of Microcystins:

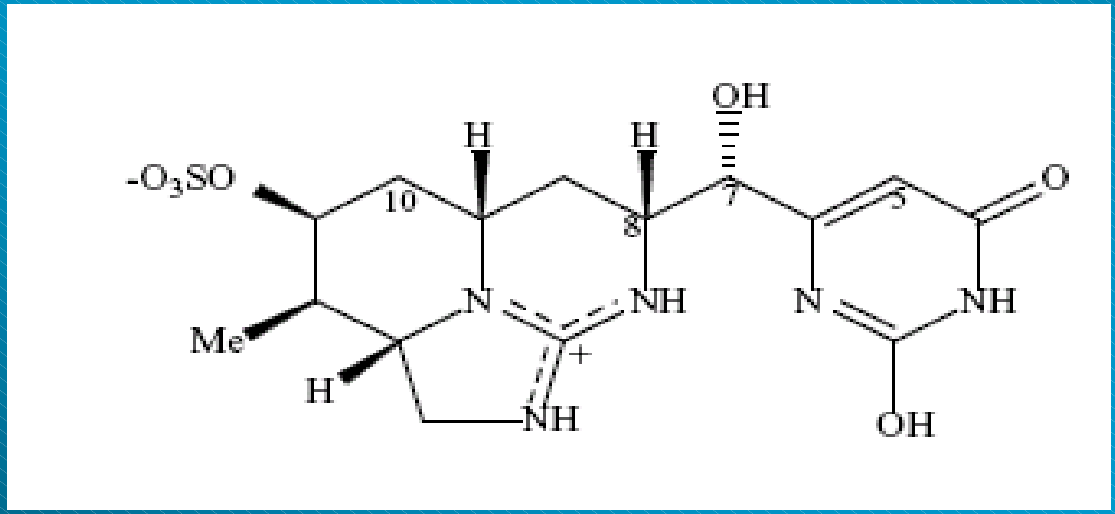
- In dark waters, microcystins can persist for months or even years. Scums of microcystins that dry on the shores may contain high amounts of toxins for months.

- Microcystins remain potent after boiling and have a high degree of chemical stability. Sunlight will slowly breakdown microcystins. Photopigments and humic acid can accelerate this process.

- Despite their chemical stability, microcystins are susceptible to bacterial breakdown. These types of bacteria are found in lake water and sediment, river water and sewage effluent.

Cylindrospermopsis:

As more bodies of water are analyzed for the presence of cylindrospermopsin, these bacteria are found on a global scale. Certain strains of *Cylindrospermopsi*, *Anabaena*, *Aphanizomenon* and others have been determined capable of producing this toxin.



Main target of toxicity is the liver. Other organs such as the thymus, kidneys, lungs, intestinal tract and heart may be affected.

Characteristics of Cylindrospermopsin Toxin:

- Highly water soluble
- Stable in extreme temperatures and pH
- No degradation of toxin after 15 minutes of boiling
- Toxin will degrade more rapidly under natural light compared to short-wave UV light
- Often, the level of cylindrospermopsin produced extracellularly will exceed the level of toxin inside the cells

Health effects of cylindrospermopsin:

-Cylindrospermopsin was reported to be a possible tumor initiator

-No reproductive or teratrogenic studies were located

-Symptoms of cylindro-spermopsin toxicity in cattle have been reported as weakness, anorexia, palor of mucous membranes and diarrhea. Several incidences of death in livestock have been attributed to this toxin.

(Cylindrospermopsis raciborskii)

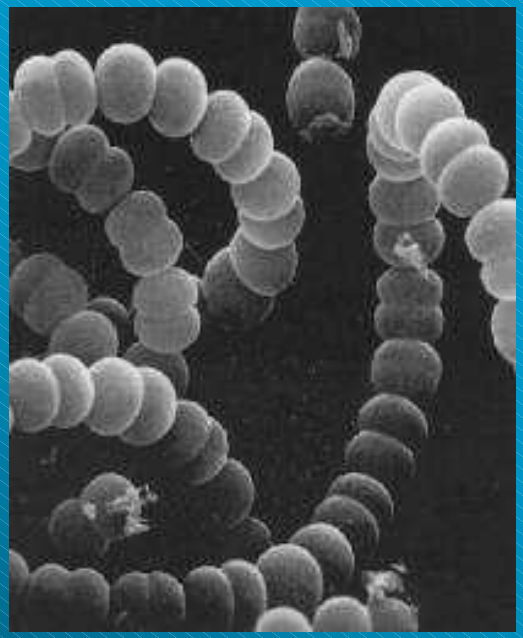
Palm Island Mystery:

In 1979, a major bloom occurred in a reservoir of Palm Island, Australia (water was chlorinated but unfiltered). Residents complained of a bad taste and smell of drinking water, so the water was treated with copper sulfate to kill the bloom.

Shortly after, 139 children and 10 adults complained of hepatitis-like symptoms including malaise, anorexia, vomiting, tender hepatomegaly, headache and stomach pain. Kidney malfunction, bloody diarrhea and urine were also reported. In a few cases, the loss of electrolytes was so severe that patients suffered from hypovolemic shock.

A culture of the reservoir water revealed the presence of *Cylindrospermopsis raciborskii*.

Anatoxins:



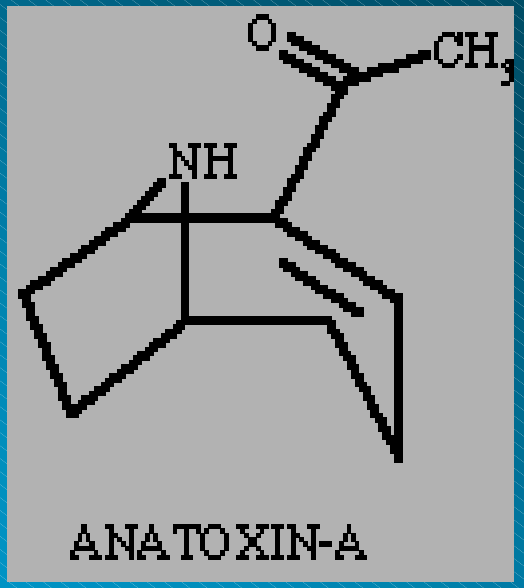
(Anabaena flos-aquae)

Anatoxins are neurotoxic agents produced by a variety of cyanobacterial organisms. They are alkaloids, which are capable of transformations into toxic by-products. Anatoxins are stable in dark waters, but are susceptible to photo-chemical degradation. Breakdown is further accelerated by alkaline conditions.

Anatoxins have been implicated in numerous animal and wildfowl poisonings. Symptoms, including death, can occur in minutes. No information could be found on chronic exposure to anatoxins.

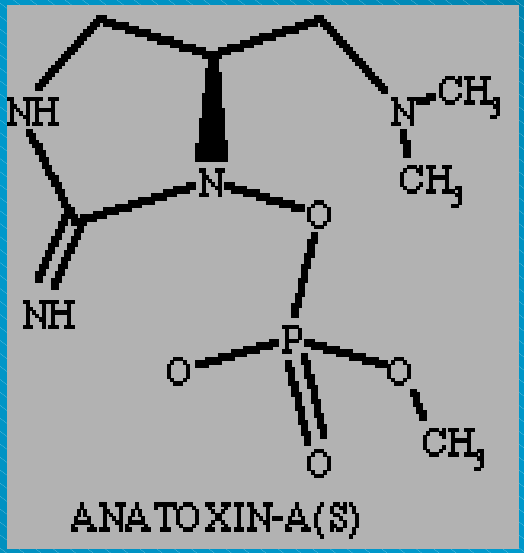
Health Effects of Anatoxins:

Anatoxin-a: sufficient exposure can lead to paralysis, asphyxiation and death; oral LD_{50} (mice) ~ 5 ppm; repeated exposures to toxin caused fetal malformations and stunted growth in hamster litters; no maternal toxicity noted



ANATOXIN-A

Anatoxin-a(S): potent organophosphate produced by *A. flos-aquae*; this toxin blocks acetylcholinesterase activity; no oral toxicity studies could be found; symptoms include muscle weakness, respiratory distress and convulsions



ANATOXIN-A(S)

Case history of anatoxin-a exposure:

In July 2002, five teenagers went swimming in a pond at a golf course in Dane County, Wisconsin. The pond was described as “scummy” and “dirty.” The boys splashed around and two had their head submerged underwater.

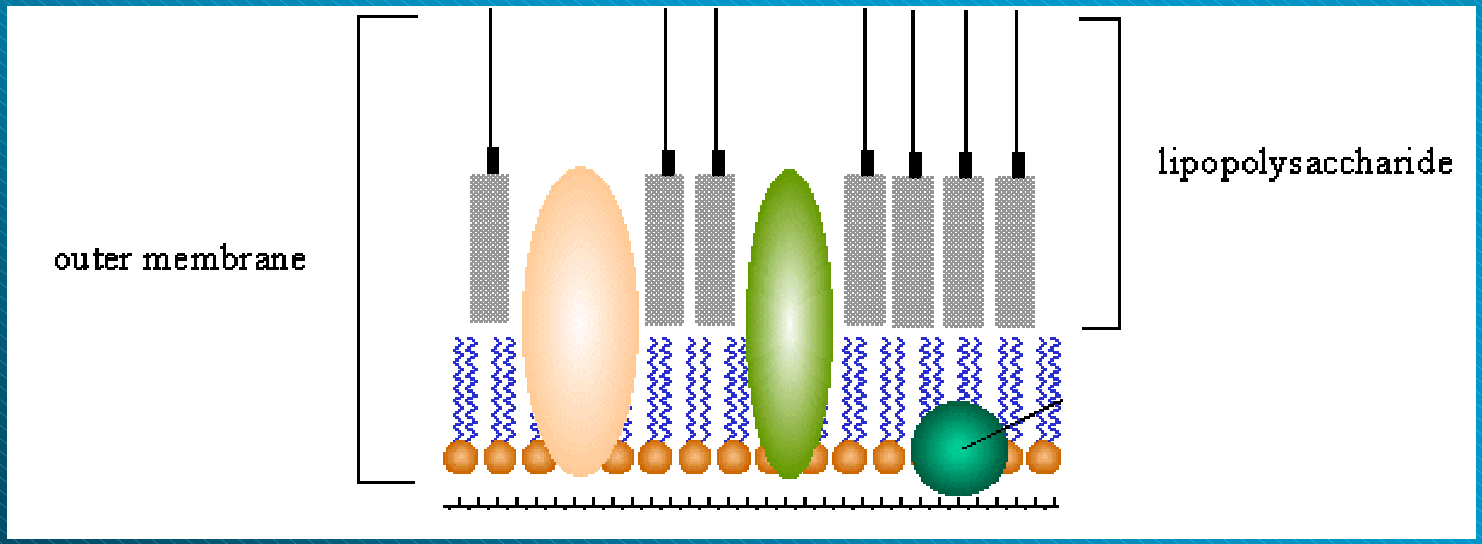
Of the two who went underwater, one boy died of acute heart failure 48 hours later and the other became ill with acute diarrhea and abdominal pain. Blood tests on the boys confirmed the presence of *A. flos aquae* and anatoxin-a.

An algal toxin expert was quoted as saying the toxin was present in amounts that could cause symptoms & death based on animal studies, but was puzzled by the amount of time that had elapsed prior to death (Milwaukee Journal Sentinel, Sept. 5, 2003). Typically, neurological toxins act in minutes or hours versus days.

Irritant toxins- Lipopolysaccharides

Lipopolysaccharides (LPS) are found in the outer cell wall of Gram negative bacteria. They are pyrogenic and can be toxic. They generally consist of a sugar and a fatty acid component. Generally, the fatty acid portion elicits an irritant or allergenic response in humans and animals.

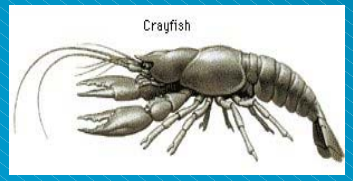
A few studies have shown that LPS endotoxins from cyanobacteria are less toxic than other bacterial LPS toxins, such as *Salmonella*.



Algal toxins in Fish/Shellfish Tissue:

Microcystins: can bioaccumulate in shellfish and the liver of fish. Some studies have found microcystin in fish fillet (range: 16- 300 ppb microcystin in fillet). Based on a RfD of 0.04 ug/kg/day, this would not present a health hazard to the recreational angler. Current recommendations from DHS are for the removal of organs during moderate blooms and no consumption guidelines for severe blooms that exceed 1,000,000 cells/mL.

Anatoxins: no information could be located.



Cylindrospermopsin: distribution of toxin in crayfish was 5:1 between viscera:muscle. No cylindrospermopsin was detected in the muscle of rainbow fish, but was detected in the liver.

Examples of Risk Levels & Standards:

Drinking water (provisional): 1 ug/L microcystin-LR (WHO)
3 ug/L anatoxin-a (Australia)

BGA dietary supplements: 1 ppm microcystin (ODA)

Tolerable Daily Intake (provisional): 0.04 ug/kg/day (WHO)

Recreational Bathing Waters (WHO):

Relatively **low-risk** of adverse effects: 20,000 cells/ml
(4 ug/L microcystin)
Moderate probability of adverse effects: 100,000 cells/ml
(20 ug/L microcystin)
High probability of adverse effects: scums

Special considerations:

- Scums can increase local cell density and toxin concentration in hours. This has numerous implications for public health and presents a challenge for routine water monitoring schedules.
- During bloom die-offs, the water may look more inviting, but toxin levels may be at their highest.
- The incidence of low-level symptoms (nausea, vomiting, diarrhea) associated with recreational exposure to algal toxins is most likely under-reported
- Most likely, not all toxic cyanobacteria have been identified and not all possible toxins have been discovered
- Children and people with pre-existing medical conditions should be considered as susceptible risk groups.

Additional Information Sources

Books:

Toxic Cyanobacteria in Water, Chorus & Bartram, ed. World Health Organization, published by E & FN Spon (1999)

Toxic Microcystis, Watanabe, Harada, Carmichael, Fujiki, ed., published by CRC Press (1996)

Guidelines for safe recreational water environments, Volume 1 Coastal & Fresh Waters, World Health Organization (2003)

Websites:

www.aims.gov.au/arnat - Australian Research Network for Algal Toxins

www.dhs.state.or.us/publichealth/esc/docs/maadvisories.cfm - Oregon Department of Human Services