

Animal Specimen Collection Guide for HAB Exposure

ALL SAMPLES

- 1) Double bag samples to prevent cross-contamination
 - 1) For liquids (blood, urine), make sure there is enough absorbent material placed between the primary and secondary packaging in case of leakage
- 2) Download and fill out our COC: <https://GreenWaterLab.com/COC.pdf>
- 3) Ship samples
 - a) Pack samples (tightly sealed and individually packed) on dry ice
 - b) Make sure samples are labeled with the sample ID and type of sample (e.g. urine)
 - c) Put the filled-out COC in a plastic bag in the cooler with samples
 - d) Ship overnight to the lab

SUMMARY

If you suspect poisoning due to harmful algae or cyanobacteria, save the **source** and any **stomach contents/vomit**. This can be used to look for intact cyanobacteria (microscopy), confirm exposure, and narrow down toxin testing. In mammalian intoxication events, the next specimen to collect is **urine** (antemortem/postmortem) and/or **liver** (postmortem). Additionally, bile, blood, and organs (kidneys, liver, brain) can be tested. For avian specimens, cloacal contents and liver are preferred. For fish and invertebrates, collect liver or whole organism. Specimens to be submitted are highly dependent on study goals and site history.

WHICH SAMPLES DO YOU COLLECT?

SOURCE

If at all possible, get the source of intoxication (e.g. lake water, surface scum). Do not freeze source water: keep refrigerated (< 1 week; 100 – 250 mL in a plastic bottle). For longer hold times (≤ 6 months) an aliquot (~10 mL) can be preserved (1:1 of 10% formalin; 5% final) and the remaining sample frozen. If the source is another animal (e.g. dead fish), freeze the specimen.

SPECIMEN COLLECTION

WE DO NOT CONDUCT DISSECTIONS

How the samples are submitted (e.g. individual organs, whole organism) is how they will be processed for testing

Stomach contents / Vomitus:	If available, collect all contents in a bag/bottle and <u>refrigerate</u>
Urine:	Collect urine (0.5 – 50 mL) and <u>freeze</u> (-20 °C)
Bile or Blood / Plasma / Serum:	Collect 0.5 – 10 mL and <u>freeze</u> (-20 °C)
Organs:	Excise organs (e.g. liver, kidney, brain), subsample as needed to acquire 1 – 10 grams, individually package each specimen, and <u>freeze</u> (-20 °C)
Fish / Mollusks / Crabs / Seafood:	If <u>human consumption</u> is of concern, send only the fillets/edible portion For <u>ecological</u> concerns or chronic exposure studies, the target organ (e.g. liver, kidney) or even the whole organism can be submitted

WHICH TEST DO YOU CHOOSE?

For **stomach contents** (or **source water** if available/accessible):

- Check "**PTOX cyanobacteria screen**" on the COC to identify cyanobacteria/algae
- This sample should be analyzed first so that toxin analysis recommendations can be made and to cut down on toxin testing costs. In the event this specimen is not available, or degraded, 'blind' toxin testing can be conducted.

All specimens - toxins listed in order of importance for 'Blind Testing'. Clinical signs and exposure history may also help determine initial choice of toxin test.

Toxin	Method	Detection Limit (ppb)	Hold Time (-20°C)
Microcystins/Nodularins	MMPB	10	≥ 1 year
Anatoxin-a Suite	LC-MS/MS	50	≤ 1 month ^a
Saxitoxins ^c	ELISA	50	≥ 6 months ^b
Cylindrospermopsin	LC-MS/MS	50	≥ 6 months
Guanitoxin (anatoxin-a(s))	ELISA	Qualitative	≤ 1 month ^a
Brevetoxins ^c	ELISA	250	≥ 6 months

^a = Little is known regarding the holding times of these analytes in animal specimens

^b = Interconversions may occur

^c = Predominantly a marine source of intoxication