Supplementary Information

Anatoxins from Benthic Cyanobacteria Responsible for Dog Mortalities in New Brunswick, Canada

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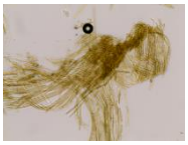
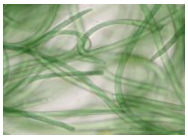
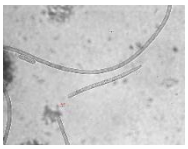
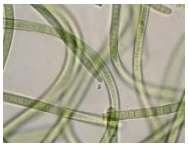
Table S1. Results from taxonomic, toxin and genetic analysis of isolates obtained from samples collected at the Wolastoq following 2018 dog poisonings.

Isolate ID (original sample)	**Tentative ID (Microscopy)**	**Culture Conditions**	**Species affiliation** (16S rRNA)	**Clonal** (microscopic observations)	**Total ATX (ng/g)**	**AnaC Gene**	**Micrographs * Scale bar = 20 µm**
W4-A	*Microcoleus* sp.	5 mL tube liquid BG11	*Tychonema* sp. *Oscillatoria amoena* *Microcoleus vaginatus* *Microcoleus acremanii*	NO 3+ species	40	-	
W4-B	*Phormidium* sp.	5 mL tube liquid Z8	*Phormidium autumnale* similar to CAWBG638, 637, 636, 618	NO >1 species	600	-	
W5-A	*Phormidium* sp.	5 mL tubes liquid Z8	*Geitlernema carotinosum* *Phormidium. autumnale*	NO >1 species	<LOD	-	
W6-A	*Leptolyngbya* sp.	5 mL tubes liquid Z8	*Wilmottia sp* similar to CAWBG522; uncultured cyanobacterium P19.2.3	>1 species	<LOD	-	
W6-B	*Leptolyngbya* sp.	5 mL tubes liquid BG11	*Phormidium autumnale* similar to CAWBG646; CAWBG638	YES	<LOD	+	
W3-H	*Microcoleus* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG638, 637, 636, 618	YES	<LOD	-	
W3-J	*Microcoleus* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG646, 645, 644, 643, 642, 641, 639, 635	YES	230	+	
W3-C	*Microcoleus* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG646, 645, 644, 643, 642, 641, 639, 635	YES	1800	+	
W3-D	*Microcoleus* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG646, 645, 644, 643, 642, 641, 639, 635	YES	3000	+	

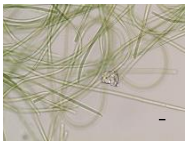
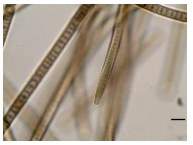
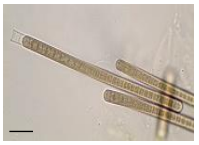
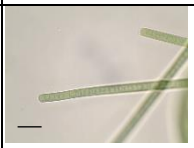
W3-E	*Leptolyngbya* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG646, 645, 644, 643, 642, 641, 639, 635	YES	>10000	+	
W3-A	*Phormidium* sp.	Z8 agar Petri plate to 5 mL tube Z8	*Phormidium autumnale* (CAWBG646)	YES	100	+	
W3-B	*Phormidium* sp.	Z8 agar Petri plate to 5 mL tube Z8	*Phormidium autumnale* (CAWBG646)	YES	600	+	
W3-F	*Phormidium* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG638, 637, 636, 618	YES	<LOD	-	
W3-G	*Leptolyngbya* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG646, 645, 644, 643, 642, 641, 639, 635	YES	3000	+	
W3-I	*Phormidium* sp.	Z8 agar Petri plate to 5 mL tube Z8	Identical to *Phormidium autumnale* CAWBG646, 645, 644, 643, 642, 641, 639, 635	YES	1100	+	

Table S2. Samples collected in August 2018 in other New Brunswick water systems following the poisonings on the Wolastoq in July 2018. Samples were collected in association with dog mortalities, but were not linked to anatoxins.

Receipt date/Analysis date	Sample #	Description (provided by S. Scott)	ATX (ng/L)
Received Friday, Aug 3rd	#10	Mackins Lake water, July 27, 2018	-
Received Friday, Aug 3rd	#11	Douglas Lake water, July 27, 2018	4.1
Received Friday, Aug 10th	#12	Kennebecasis River water "near shore"	-
Received Friday, Aug 10th	#13	Kennebecasis River water "east dock"	-
Received Friday, Aug 10th	#14	Aquatic vegetation near shore	-
Received Friday, Aug 10th	#15	Aquatic vegetation near shore	-
Received Friday, Aug 10th	#16	Kennebecasis Dog Stomach Contents*	-
Received Tuesday, Aug 23rd	#17	Thornes Brook water Aug 16th	-
Received Thursday, Aug 30th	#18	Treadwell Lake Dog Stomach Contents*	-

* high background in HRMS

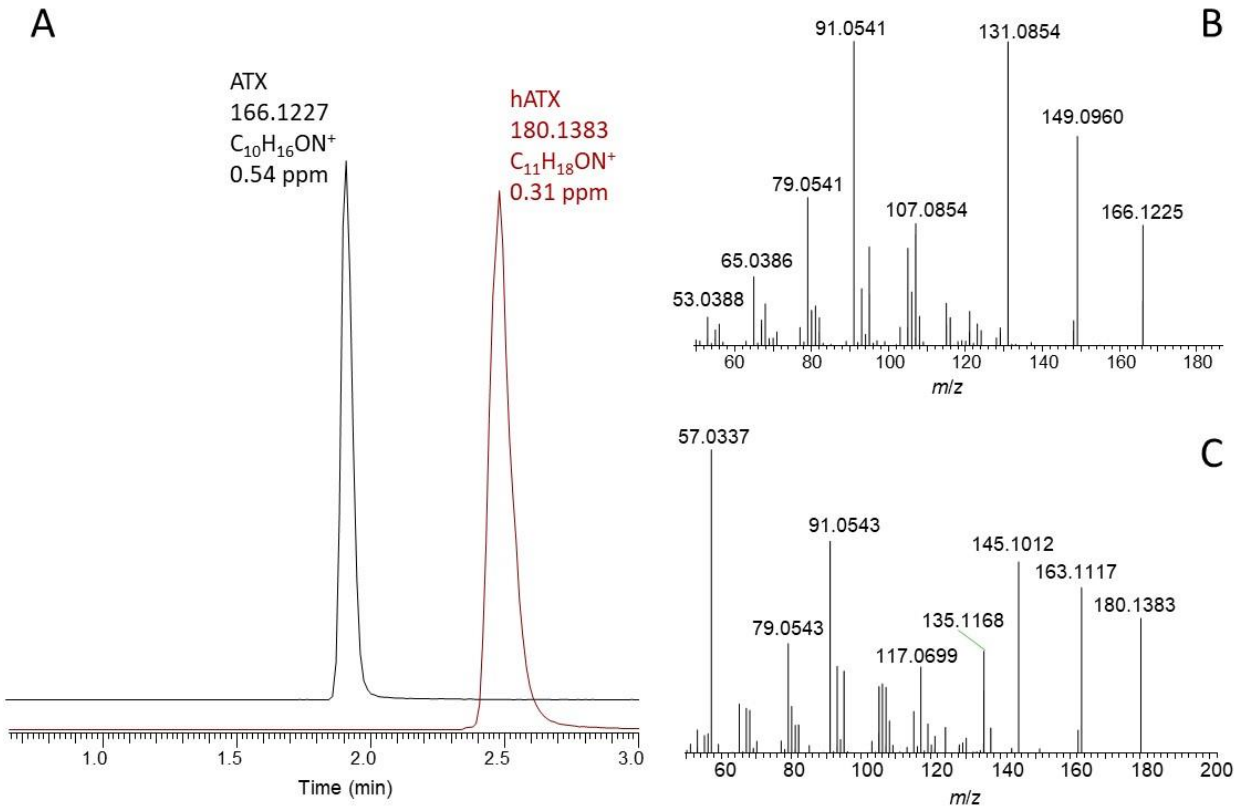


Figure S1. LC–HRMS extracted ion chromatograms of ATX-a (*m*/*z* 166.1226 ± 5 ppm) and hATX-a (*m*/*z* 180.1383 ± 5 ppm) in a mixed standard (A) and associated collision induced dissociation spectra of ATX-a (B) and hATX-a (C).