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Authors: McCain, Stephanie, Sim, Richard R., Howerth, Elizabeth W., Aschenbroich, Sophie, Kirejczyk, Shannon G.M., et al.

Source: Journal of Zoo and Wildlife Medicine, 51(2) : 407-415

Published By: American Association of Zoo Veterinarians

URL: https://doi.org/10.1638/2019-0117

DOI: 10.1638/2019-0117 *Journal of Zoo and Wildlife Medicine* 51(2): 407–415, 2020

MYONECROSIS AND DEATH DUE TO PRESUMED MICROCYSTIN TOXICOSIS IN AMERICAN WHITE PELICANS (*PELECANUS ERYTHRORHYNCOS*)

Stephanie McCain, DVM, Dipl ACZM, Richard R. Sim, DVM, Dipl ACZM, Elizabeth W. Howerth, DVM, PhD, Dipl ACVP, Sophie Aschenbroich, DVM, PhD, Dipl ACVP, Shannon G.M. Kirejczyk, DVM, MPH, Dipl ACVP, Brittany McHale, DVM, MPH, Dipl ACVP, Carmen Jerry, Jack J. Kottwitz, DVM, Dipl ACVP, CertAqV, Alan E. Wilson, PhD, and Rita McManamon, DVM

Abstract: Over a period of 5 mo, seven out of eight American white pelicans (*Pelecanus erythrorhynchos*) housed on a spring-fed pond at a zoo died or were euthanized. Clinical signs included inability to stand, anorexia, and weight loss. Clinicopathologic findings included heterophilic leukocytosis and elevated creatine kinase and aspartate aminotransferase. Histopathologic findings on all pelicans demonstrated severe, chronic, diffuse rhabdomyofiber degeneration and necrosis, making vitamin E deficiency a differential diagnosis despite routine supplementation. Based on tissue and pond water assays for the cyanobacterial toxin, microcystin, toxicosis is suspected as the inciting cause of death in these cases. We hypothesize that vitamin E exhaustion and resultant rhabdomyodegeneration and cardiomyopathy were sequelae to this toxicosis.

Key words: cyanobacteria, microcystin, myonecrosis, myopathy, *Pelecanus*, pelican

INTRODUCTION

Rhabdomyolysis is a frequent finding in multiple species of pelican (*Pelecanus* sp.) and has a variety of inciting causes, including vitamin E deficiency.[11,20] Captive pelicans are routinely supplemented with oral vitamin E based on low levels in frozen fish diets, which are high in polyunsaturated fatty acids, and risk of rancidity.[6] In addition to rhabdomyolysis, vitamin E deficiency can result in cardiomyopathy, steatitis, and hemolytic anemia, while toxicity can result in a coagulopathy.[11,16,17,20]

Microcystins are a group of toxins produced by cyanobacteria (also called blue-green algae) that have been well documented as a cause of mortality in a variety of animal species worldwide.[3,5,7,15,19] The primary target organ is the liver, resulting in acute hepatic failure; however, toxins can be detected in multiple organs.[3,5] Mass mortalities have occurred in humans, livestock, dogs, turtles, and a variety of bird species.[15]

A mortality event occurred in 2016 in a captive group of American white pelicans (*Pelecanus erythrorhyncos*) during a pond algal bloom event with high levels of circulating microcystin concentrations. As vitamin E has been found to have a protective effect against microcystin toxicity,[5] the pelicans appear to have developed hypovitaminosis E in response to the microcystin exposure and died with severe rhabdomyolysis. To the best of the authors' knowledge, this is the second report associating a *Pelecanus* sp. mortality event and microcystin exposure[13] but the first to describe clinicopathologic findings in individual birds.

CASE REPORTS

A group of eight American white pelicans (*P. erythrorhyncos*), two southern screamers (*Chauna torquata*), and one mute swan (*Cygnus olor*) were housed on an island surrounded by a spring-fed pond (Pond 1) at the Birmingham Zoo. A second pond (Pond 2) that was connected through a

From the Birmingham Zoo, Inc, 2630 Cahaba Road, Birmingham, AL 35223, USA (McCain and Sim); Zoo and Exotic Animal Pathology Service and Pathology Department, University of Georgia, Athens, GA 30602, USA (Aschenbroich, Howerth, Kirejczyk, McHale, and Jerry); Clinical Pharmacology Laboratory, Auburn University College of Veterinary Medicine, 1500 Wire Road, Auburn, AL 36849, USA (Kottwitz); School of Fisheries, Aquaculture, and Aquatic Sciences, 203 Swingle Hall, Auburn University, Auburn, AL 36849, USA (Wilson); Zoo and Exotic Animal Pathology Service and Infectious Diseases Laboratory, University of Georgia, Athens, GA 30602, USA (McManamon). Present address (Sim): Oregon Zoo, 4001 Southwest Canyon Road, Portland, OR 97221, USA. Present address (Aschenbroich): Department of Pathobiological Sciences, School of Veterinary Medicine, University of Wisconsin, Madison, Madison, WI 53706, USA. Present address (Kirejczyk): Yerkes National Primate Research Center, Emory University, 954 Gatewood Road NE, Atlanta, GA 30329, USA. Present address (Jerry): California Animal Health and Food Safety Lab System, Turlock Branch Lab, 1550 North Soderquist Road, P.O. Box 1522, Turlock, CA 95380, USA. Correspondence should be directed to Dr. McCain (smccain@birminghamzoo.com).

channel to the first pond housed two additional mute swans. A variety of fish and turtle species lived in both ponds. All of the pelicans were wild caught and had been housed at the zoo for 2–7 yr. Their diet consisted of capelin (*Mallotus villosus*) supplemented with a daily multivitamin (Mazuri Vita-Zu Large Bird tablet without vitamin A added; Mazuri, Richmond, IN 47374, USA; 1 tablet per kilogram of fish). Pelicans were hand-fed to ensure that each bird received one tablet daily, as birds consumed approximately 1 kg of fish each daily.

On 13 June 2016, an adult male pelican (Pelican 1) was swimming away from the group and refused his morning feeding (Table 1). He was caught and manually restrained for a physical examination, which revealed a thin body condition as well as maceration of the skin of both feet. He was alert and responsive but unable to stand or walk without assistance. The toes were curled under at rest, but he could straighten them during movement. The feathers on his ventrum were coated with algae, suggesting that he had not been out of the water for some time. An in-house complete blood count (CBC) and plasma biochemistry panel (VetScan VS2 Chemistry Analyzer, Abaxis Inc, Union City, CA 94587, USA) were unremarkable, including creatine kinase (CK), which was (erroneously) reported as 0 U/L. He was moved to an indoor holding area with no access to a pool.

That same day, another adult male pelican (Pelican 2) separated itself from the group. He was caught and restrained for a physical examination, which was unremarkable, but died before a blood sample could be obtained. Necropsy revealed mild pale tan streaking of the pectoral musculature and epicardium. The liver had scattered diffuse pinpoint, nonraised white foci on the capsular and cut surfaces. Samples of all tissues were sent for histopathology to the Infectious Disease Laboratory at the University of Georgia, College of Veterinary Medicine (Athens, GA 30602, USA).

A third adult male pelican (Pelican 3) presented later that day with similar signs as the first two. Physical examination findings were the same as Pelican 1 but less severe. A CBC showed a severe heterophilic leukocytosis (88×10^3 cells/μl).[14] On the biochemistry panel, aspartate aminotransferase (AST) reported as 0 U/L, and CK was 454 U/L. The bird was placed in the indoor holding with Pelican 1, and both birds were started on enrofloxacin (Putney, Inc, Portland, ME 04101, USA; 11.4–12.3 mg/kg p.o. b.i.d.) administered in a single force-fed capelin.

On days 2 and 3, neither pelican would eat on their own, although both birds remained alert and responsive. Birds were initially tube fed 120 ml of a slurry made by blending capelin with water twice a day along with being force-fed a single whole capelin containing enrofloxacin. Multivitamin administration was continued once a day. An additional five capelin were force-fed to each pelican at each feeding starting on day 3.

On day 4, preliminary histopathology results on Pelican 2 showed extensive and severe myofiber necrosis and regeneration in the skeletal muscle suggestive of vitamin E deficiency. Following these results, Pelicans 1 and 3 were started on injectable vitamin E (Neogen Corporation, Lexington, KY 40511, USA; 113–115 IU/kg intramuscular [i.m.] once daily [s.i.d.]) for 3 days followed by additional oral vitamin E (CVS Pharmacy, Inc, Woonsocket, RI 02895, USA; 151–154 IU/kg p.o. s.i.d.). A sample of water from the pond was collected and frozen at –80°C until further analysis. On day 5, all the remaining pelicans on the pond were caught for a brief physical examination, weight, and blood draw to assess serum selenium and vitamin E (alpha-tocopherol) levels. These pelicans were also administered a single injection of 600 IU (106–138 IU/kg) vitamin E i.m. Serum vitamin E and selenium levels ranged from 11.16 to 23.80 μg/ml and 180 to 317 ng/ml, respectively. Reference intervals for alpha-tocopherol have not been reported in American white pelicans, but these levels were similar to historic values for this pelican flock at this institution (8.20–26.70 μg/ml). In free-ranging brown pelicans (*Pelecanus occidentalis*), these levels would not be considered low.[8] Supplemental oral vitamin E was initiated the following day (800 IU [142–184 IU/kg] p.o. s.i.d.). The skin on the feet of Pelicans 1 and 3 slowly improved, and they were allowed supervised swimming in a tank twice a day for 30–60 min starting on day 5.

On day 6, an adult female pelican (Pelican 4) presented with anorexia and a wing droop. She was restrained for a physical examination, which was unremarkable, and blood collection for a CBC and biochemistry panel, which revealed a marked heterophilic leukocytosis (50.29×10^3 cells/ul) and elevated AST (1,126 U/L). She was placed in the indoor holding with Pelicans 1 and 3 and began enrofloxacin therapy. All three pelicans received 120 ml of the slurry followed by eight capelin. Starting on day 9, the slurry was discon-

tinued, and each pelican received 10 capelin followed by 120 ml of water b.i.d.

A subsequent CBC and biochemistry panel on Pelican 1 on day 9 was unremarkable, except for the continued (erroneous) CK level of 0 U/L. Pelican 1 was found dead that evening. A necropsy performed the next day showed pale and soft skeletal muscles and a friable liver. A banked plasma sample from day 1 was sent to an outside reference laboratory (Antech Diagnostics, Vestavia Hills, AL 35243, USA) for CK, which was 24,222 U/L. Histopathology performed at the Infectious Disease Laboratory showed severe subacute widespread myonecrosis with mineralization as well as saponification of fat (Table 1).

On day 11, due to lack of clinical improvement, Pelican 3 was euthanized. A pre-euthanasia plasma sample was sent to the outside reference laboratory for a biochemistry panel. AST was 4,614 U/L, and CK was 33,303 U/L. Phosphorus was mildly elevated at 6.7 mg/dl, and glucose was moderately decreased at 84 mg/dl. Gross and histologic findings were similar to Pelicans 1 and 2. On day 12, a banked day 7 serum sample from Pelican 4 was sent to the reference laboratory, revealing a CK value of 21,247 U/L. All further biochemistry panels were sent to the outside reference laboratory.

The condition of Pelican 4 was static on day 15, and a follow-up CBC and biochemistry panel showed a normal leukogram and elevated AST and CK (4,485 U/L and 38,693 U/L, respectively). However, her condition deteriorated, with development of generalized weakness, ataxia, and unilateral foot knuckling prompting euthanasia on day 16. Gross and histologic findings were similar to the other three pelicans.

The water sample that was frozen on day 4 as well as a liver sample from Pelicans 1, 3, and 4 were submitted for microcystin levels (School of Fisheries, Aquaculture, and Aquatic Sciences, Auburn, AL 36849, USA). Microcystin was quantified in whole water samples (includes cell-bound and dissolved forms) after extraction using three freeze-thaw cycles using enzyme-linked immunosorbent assays according to the manufacturer's instructions (PN 520011, Abraxis Inc, Warminster, PA 18974, USA). Analyses performed revealed levels of 1.139 μg/L, 48.61 ng/g dry mass, 38.83 ng/g dry mass, and 17.33 ng/g dry mass in water and liver, respectively (Tables 2, 3). Zoo staff began incorporating city water into the pond every evening after closure of the zoo for 3–5 hr using a fire hose as a dilution measure.

The remaining four pelicans seemed to be doing well until day 32, when another adult male (Pelican 5) appeared lethargic. Due to the concern about the potential role of microcystin toxicosis, all of the pelicans were restrained and moved to a nearby pond that housed American flamingos (*Phoenicopterus ruber*). This pond was filled with city water. On day 35, Pelican 5 was unable to stand and was euthanized. Gross and histologic findings were consistent with those observed in all other pelicans.

On day 105, an adult female pelican (Pelican 6) presented for grade IV/V lameness on the left leg. The pelican was manually restrained for physical examination, which revealed a thin body condition with a 14% weight loss over a 2-mo period as well as moderate pododermatitis on the opposite foot. She was transported to the animal health center and mask induced with isoflurane (Piramal Critical Care, Inc, Bethlehem, PA 18017, USA) for two view radiographs of both hind limbs, which were unremarkable, and for blood collection. She was administered a single dose of meloxicam (Henry Schein Animal Health, Dublin, OH 43017, USA; 0.5 mg/kg i.m.) and returned to the habitat pending blood results. She had a marked heterophilic leukocytosis (114.08×10^3 cells/ul) as well as elevated AST (4,635 U/L) and CK (24,887 U/L). The remaining three pelicans were started back on increased oral vitamin E supplementation for 30 days. Pelican 6 refused food offered by staff and was found dead on day 107. Gross and histologic findings were consistent with those observed in all other pelicans.

Following the incorporation of city water into Pond 1, repeated water samples showed microcystin levels below the limit of detection beginning on day 46, and the remaining two pelicans were moved back to Pond 1 on day 122.

On day 154, an adult female pelican (Pelican 7) appeared to have a mass on the webbing of her left foot between digits 2 and 3. Both remaining pelicans were started back on increased vitamin E supplementation for 3 days prior to restraint of Pelican 7 on day 157. Pelican 7 was found to have a 3-cm-diameter, round, pedunculated black mass protruding from the plantar aspect of the middle of the second digit on the left foot. She was transported to the health center, mask induced with isoflurane, intubated, and maintained on isoflurane to perform radiographs. Two view radiographs of the hind legs revealed a pathologic fracture of P2 on the second digit of the left foot with marked soft tissue thickening around the base of the mass. The digit was surgically

Table 1. Clinical and pathologic characterization of microcystin exposure in captive, adult American white pelicans (*Pelecanus erythrorhyncos*) at the Birmingham Zoo in 2016.[a]

		Presenting							Hepatic vitamin E concentration at time of death (μg/g dry tissue)
Animal	Sex	Day	Clinical signs	WBC ($\times 10^3$ cells/μl)	CK (U/L) (analyzer)[b]	AST (U/L) (analyzer)[b]	Outcome	Pathology	
Pelican 1	M	1	Separation, anorexia, thin, unable to stand, toes curled under, algae on feathers	23.4	0 (Ab) 24,222 (An)	7 (Ab)	Died d9	*Gross:* pale and soft skeletal muscles and friable liver *Histology:* severe subacute myonecrosis with mineralization and moderate acute myodegeneration; fat degeneration with saponification	2,651.18
Pelican 2	M	1	Separation	NT	NT	NT	Died d1	*Gross:* mild pale streaking of skeletal muscle and heart; diffuse pinpoint, nonraised white foci of the liver *Histology:* severe chronic myonecrosis with regeneration and lymphohistiocytic myositis; extramedullary hematopoiesis of the liver	NT
Pelican 3	M	1	Separation, anorexia, thin, unable to stand, toes curled under	88	d1: 454 (Ab) d11: 33,303 (An)	d1 0 (Ab) d11 4,614 (An)	Euth d11	*Gross:* diffuse paleness to pale streaking of skeletal muscles; spleen was diffusely pale and soft *Histology:* severe chronic-active myonecrosis and degeneration with mineralization and regeneration; splenic lymphoid depletion	691.52
Pelican 4	F	6	Anorexia, wing droop	50.3	d6: 21,247 (An) d15: 38,693 (An)	d6: 1,126 (Ab) d15: 4,485 (An)	Euth d16	*Gross:* diffuse paleness to pale streaking of skeletal muscles *Histology:* severe chronic-active myonecrosis and degeneration with mineralization, regeneration, and severe fatty infiltration; splenic lymphoid depletion; hepatic lipidosis	871.34

Table 1. Continued.

Animal	Sex	Presenting			CK (U/L) (analyzer)[b]	AST (U/L) (analyzer)[b]	Outcome	Pathology	Hepatic vitamin E concentration at time of death (μg/g dry tissue)
		Day	Clinical signs	WBC ($\times 10^3$ cells/μl)					
Pelican 5	M	32	Lethargic	NT	NT	NT	Euth d35	*Gross:* diffuse paleness to pale streaking of skeletal muscles; pale spleen; nodular fat in coelom *Histology:* severe chronic multifocal myonecrosis and regeneration; splenic lipidosis; chronic fat necrosis and steatitis	1,025.22
Pelican 6	F	105	Thin, lameness	114	24,887 (An)	4,635 (An)	Died d107	*Gross:* diffuse paleness to pale streaking of skeletal muscles; spleen was diffusely pale and soft *Histology:* severe chronic myonecrosis and degeneration with regeneration and fatty infiltration; moderate multifocal cardiomyofiber degeneration; splenic steatosis	444.85
Pelican 7	F	154	Pododermatitis	55.2	799 (An)	256 (An)	Died d158	*Gross:* pododermatitis *Histology:* myofascial steatosis; splenic lymphoid depletion; hepatic extramedullary hematopoiesis	301.1

[a] WBC, white blood count; CK, creatinine kinase; AST, aspartate aminotransferase; M. male; Ab, Abaxis VetScan VS2 Chemistry Analyzer; d, day; An, Antech Diagnostics; Euth, euthanized; F, female; M, male; NT, not tested.

[b] CK and AST values listed are from day of presentation unless noted otherwise.

Table 2. Microcystin concentrations in water samples from a pond (Pond 1) housing American white pelicans (*Pelecanus erythrorhyncos*) that died of suspected microcystin toxicosis and an adjacent connected pond (Pond 2).[a]

	Microcystin concentration (μg/L) in water samples	
Date	Pond 1	Pond 2
2016		
16 Jun	1.139	NT
13 Jul	1.004	0.203
15 Jul	0.303	BLD
29 Jul	BLD	BLD
5 Aug	0.229	BLD
12 Aug	0.382	0.206
26 Aug	0.162	NT
1 Sep	BLD	NT
16 Sep	BLD	BLD
23 Sep	BLD	BLD
14 Oct	BLD	BLD
28 Oct	BLD	BLD
2017		
24 Mar	NT	BLD
5 May	BLD	0.242
19 May	BLD	BLD
7 Jul	0.445	0.841
14 Jul	0.270	0.268
21 Jul	0.533	0.458
4 Aug	0.529	0.437
17 Aug	BLD	0.469
24 Aug	BLD	0.262
1 Sep	BLD	BLD
7 Sep	BLD	BLD

[a] BLD, below limit of detection, which is 0.15 μg/L; NT, not tested.

amputated, and the pelican received perioperative vitamin E (126 IU/kg i.m.), meloxicam (2 mg/kg i.m.), and ceftiofur crystalline free acid (Zoetis, Inc, Kalamazoo, MI 49007, USA; 15 mg/kg s.c.). The pelican recovered from anesthesia without complications but was found dead the following morning. Gross and histologic findings for this bird showed steatosis and rhabdomyofiber regeneration consistent with a chronic healing response to the same disease process as in the other pelicans. The single remaining adult female pelican, the screamers, and the mute swans all continue to appear healthy 3 yr later.

Water samples were collected from Ponds 1 and 2 weekly throughout the summer and fall of 2016 and 2017 and frozen at −80°F until further analysis for microcystin levels. Frozen tissue samples from deceased pelicans were also submitted for microcystin assays. Based on tissue and water assays for microcystin, toxicosis is suspected as the inciting cause of death in these cases. Vitamin E exhaustion and resultant rhabdomyodegeneration and cardiomyopathy were sequelae to this toxicosis.

Table 3. Tissue microcystin concentrations from American white pelicans (*Pelecanus erythrorhyncos*) that died of suspected microcystin toxicosis.

Animal	Tissue	Microcystin concentration (ng/g dry mass)
Pelican 1	Liver	48.61
Pelican 2	Liver	207.09
	Small intestine	156.42
	Pancreas	40.76
Pelican 3	Liver	38.83
Pelican 4	Liver	17.33
Pelican 5	Liver	136.07
	Small intestine	33.59
	Pancreas	84.48
Pelican 6	Liver	63.69
Pelican 7	Liver	140.28

Microcystin was quantified in lyophilized animal tissues after two hour-long extractions using acidified 75% aqueous methanol. To remove lipids prior to analysis, pooled extracts were dried, redissolved in 40% methanol and 60% hexane, and mixed vigorously; the hexane fraction was removed after allowing enough time for the fractions to separate; and then another cycle of hexane addition, mixing, separation, and removal was performed. Dried tissue extracts were redissolved in phosphate buffer immediately prior to analysis[1,7,18] according to the manufacturer's instructions (PN 520011, Abraxis Inc).

DISCUSSION

The pond that housed the pelicans was initially the third pond in a series of four connected ponds that were fed by a spring in the first pond. During the autumn prior to the deaths, the first two ponds were filled in, and the water from the spring was rerouted to a cistern that then filled the remaining ponds. This change in hydrology affected the flow through the remaining ponds, allowing the water to become more stagnant, thus better supporting cyanobacterial growth and thus microcystin production. Cyanobacterial blooms resulting in high microcystin levels in water features at a zoological institution have previously been reported.[7]

The microcystin concentration in the first evaluated water sample was above the human drinking water guidelines set by the World Health Organization (1 μg/L),[4] and this water was the

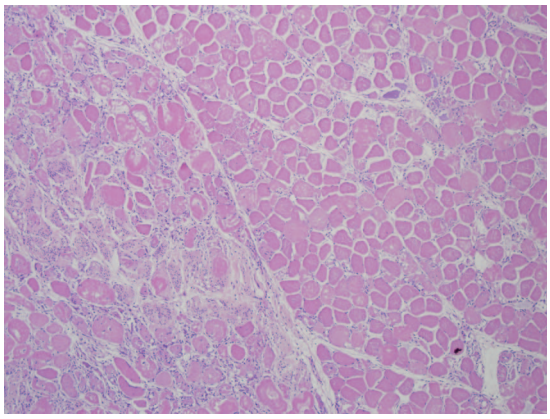

Figure 1. Photomicrograph of severe subacute rhabdomyofiber degeneration in an American white pelican (*Pelecanus erythrorhyncos*). Myofiber degeneration (swelling, vacuolation, flocculation), coagulative necrosis (hypereosinophilia, loss of cross striations, mineralization), and attempts at regeneration (sarcoplasmic basophilia, perisarcolemmal or internalized satellite cells) are present. hematoxylin and eosin, ×100.

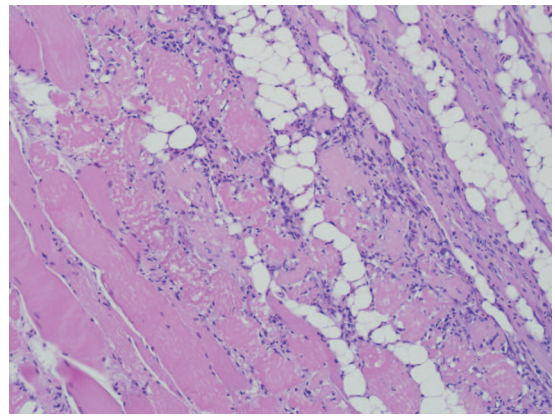

Figure 2. Photomicrograph of skeletal muscle of an American white pelican (*Pelecanus erythrorhyncos*). In addition to the changes noted in Figure 1, there is scattered mineralization of necrotic myofibers and marked fatty infiltration and replacement. hematoxylin and eosin, ×200.

sole source of drinking water for the pelicans. It is also possible that the pelicans could have been consuming fish from within this pond. The microcystin concentration decreased with the addition of city water to the ponds, although several pelican deaths happened after the microcystin level had decreased. It is unknown if the levels were higher prior to the first sample or for how long they were elevated; however, given the warm climate in Birmingham, temperatures were high enough to support cyanobacterial growth for several months prior to the index case; therefore, chronic exposure is possible.

In certain bird species, microcystin levels bioaccumulate at higher concentrations in the intestine than in liver but can be found in multiple tissues.[3] Mortality events involving free-ranging Dalmatian pelicans (*Pelecanus crispus*) and Lesser flamingos (*Phoeniconaias minor*) in Greece and Tanzania, respectively, were associated with microcystin, which bioaccumulated in the liver most significantly and in kidney, lung, and skeletal muscle to a lesser degree.[12,13] In the Dalmatian pelican report, microcystin levels were nearly as high in the spleen as they were in the liver.[13] In the mortality event of this report, the small intestine and pancreas were tested in two pelicans in addition to the liver. In both pelicans, among the submitted tissues, the liver sample had the highest concentration, suggesting that submission of liver may most accurately reflect microcystin levels. However, given the small sample size of pelicans in the present study, further evaluation in a larger cohort of pelicans would be required for the establishment of recommendations on preferred tissue submission for analysis in American white pelicans.

While the liver is considered the primary target organ of microcystin,[3,5] there was no evidence of acute hepatic failure or hemorrhage in any of the pelicans. Histopathologic findings in the livers of these pelicans were limited to extramedullary hematopoiesis ($n = 4$), lymphoplasmacytic infiltration ($n = 2$), and hepatic lipidosis ($n = 2$). An additional predominant finding noted in all cases was severe, chronic, diffuse rhabdomyofiber degeneration and necrosis, which was suggestive of vitamin E deficiency, as previously reported in pelicans.[2,6,11] Initially, infectious causes of myonecrosis and exertional myopathy (capture myopathy) were considered in the differential diagnosis for chronic rhabdomyonecrosis with regeneration. However, the clinical history did not support repeated capture attempts, and no infectious organisms, including bacteria and protozoa such as *Sarcocystis* spp., were observed in the tissues and on Gram tissue stain. These possibilities were thus considered less likely than hypovitaminosis E as the cause of skeletal muscle degeneration and necrosis.

Vitamin E has been found to have a protective effect against microcystin toxicity.[5,10] In a study evaluating multiple chemoprotectants against microcystin toxicity in mice, vitamin E consistently prevented deaths in 50% of mice treated with a

lethal dose of microcystin. As Silymarin provided complete protection whereas hypdrophilic antioxidants such as vitamin C offered no protection, the protective effect of antioxidants appears to be dependent on the lipophilicity of the antioxidant.[10] The authors suspect that the pelicans mobilized vitamin E stores in response to the toxin exposure, resulting in vitamin E deficiency despite routine supplementation. Due to the severity of the muscle damage seen histopathologically, the pelicans that survived the initial period likely had irreversible damage. Treatment of microystin toxicity is largely unrewarding, and prophylaxis is critical.[5] While additional vitamin E supplementation was initiated, it was likely too late. Additionally, the dose of supplemental vitamin E administered to the pelicans (165–210 IU/kg) was much lower than the dose found to be protective in mice (7,000–17,000 IU/kg), and it is unknown what dose would be protective in pelicans.[10]

In this cohort of pelicans, serum concentrations of vitamin E were within expected ranges for avian piscivores, as has been seen in previous reports of rhabdomyolysis due to hypovitaminosis E in pelicans,[6,20] suggesting that serum vitamin E concentration may not be a reliable indicator of deficiency. While coagulopathy has been seen in cases of vitamin E toxicosis in pelicans on a daily vitamin E supplement,[16,20] there was no evidence of this in any of the pelicans in this report. Unfortunately, liver samples were analyzed for vitamin E analysis only at the time of death, after supplemental vitamin E had been initiated (Table 1). All values were high compared to values reported for domestic chickens (45–120 μg/g dry tissue; Michigan State University Diagnostic Laboratory, Lansing, MI 48910, USA) but similar to values seen in other pelicans on vitamin E supplementation (59–705 μg/g dry tissue).[16] Hepatic vitamin E levels at the time of presentation may have provided additional information.

Initial biochemistry panels were analyzed on an in-house analyzer (Abaxis VetScan), and neither CK nor AST was reported as elevated in multiple pelicans. Gross and histologic findings suggestive of skeletal muscle damage prompted sending samples to an outside reference laboratory that reported marked increases in both analytes in all birds sampled. A study in Hispanolian parrots found the Abaxis VetScan to be reliable for both analytes in healthy individuals.[9] This case series suggests that using this instrument may not be accurate in cases with extensive muscle damage in American white pelicans.

Neither the screamers nor the mute swans that resided in Ponds 1 and 2 ever developed any clinical signs consistent with muscle damage or microcystin toxicosis. It is possible that the levels were not high enough to cause hepatic failure, so only the pelicans were affected due to their sensitivity to vitamin E deficiency. This is supported by the lack of histologic findings consistent with acute hepatic failure in the pelicans. While supplementation failure was considered, all the birds were hand-fed to ensure adequate vitamin intake for each individual. Additionally, no other piscivorous birds that were housed in an adjacent habitat were affected, including white storks (*Ciconia ciconia ciconia*), wood storks (*Mycteria americana*), and Roseate spoonbills (*Ajaia ajaja*), making rancid fish an unlikely cause unless they could have ingested rancid fish that resided in the pond.

Microcystin toxicosis should be considered in cases of muscle weakness or acute death of pelicans with rhabdomyolysis. Given that vitamin E has been found to have a protective effect against microcystin toxicosis, this case series suggests that vitamin E exhaustion may be a notable sequela even in the face of daily supplementation. The relationship between microcystin exposure and vitamin E levels warrants further investigation, and measuring both serum and hepatic vitamin E levels in cases of exposure would be helpful.

Acknowledgments: The authors would like to thank the bird department and veterinary staff at the Birmingham Zoo for their assistance in the care of these birds. In addition, much appreciation to Edna Fernadez-Figueroa and Riley Buley for providing comments that improved the final version of the manuscript. This project was supported by the School of Fisheries, Aquaculture, and Aquatic Sciences and the College of Agriculture at Auburn University.

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Accepted for publication 4 January 2020