The following supplement accompanies the article

Pufferfish mortality associated with novel polar marine toxins in Hawaii

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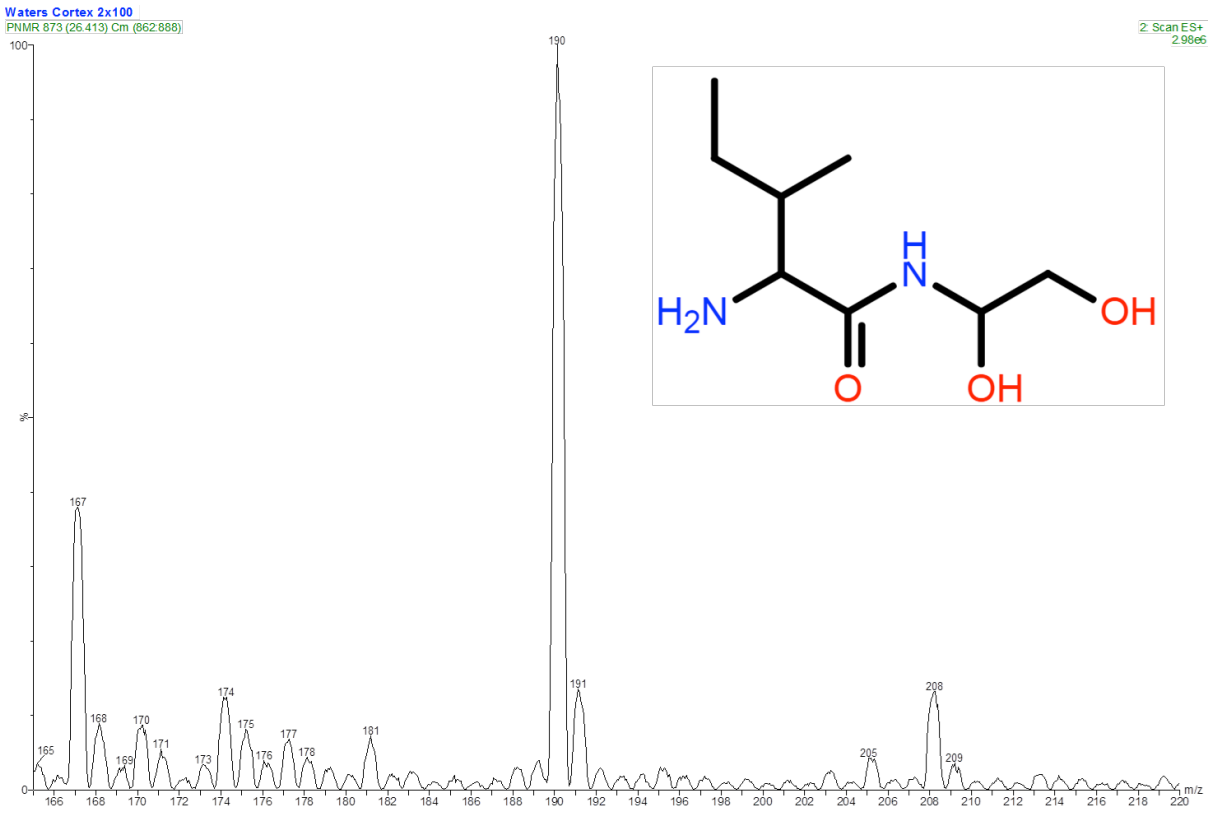

Fig. S1. Mass spectrometry of the major toxic fraction provided the predicted MS and a potential loss of H_2O. Running conditions: capillary 3.8 Kvcone 41 V, extractor 3 V, Rf lens 0.1 V, source Temp 120°C, desolvation Temp 350°C, desolvation gas 500 l hr^{-1}, cone gas 25 l hr^{-1}

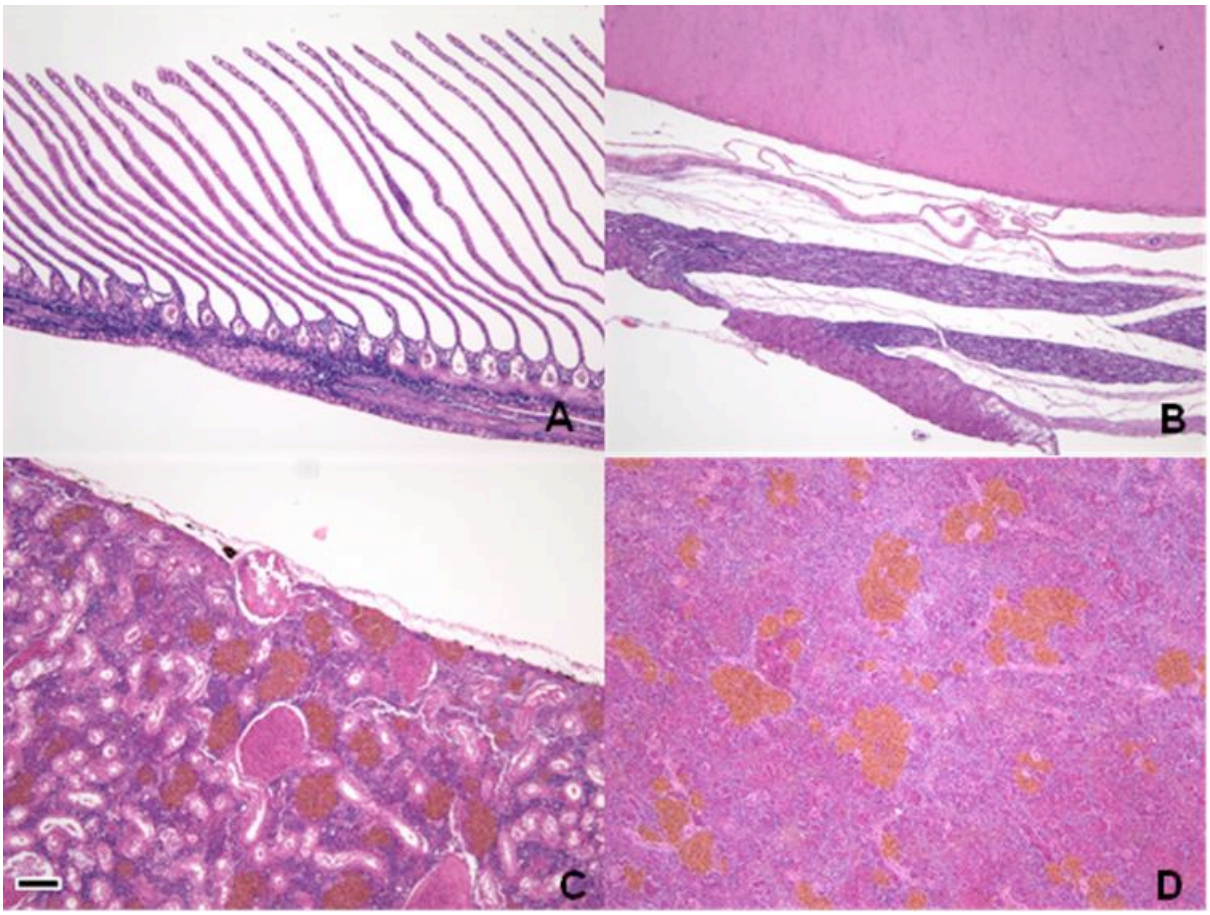

Fig. S2. *Arothron hispidus*, normal histology. (A) Gill; (B) Swim bladder with smooth muscle wall (top) and gas gland (bottom); (C) Caudal kidney; (D) Spleen with abundant melanized macrophage centers. Scale bar, for all panels = 100 μm

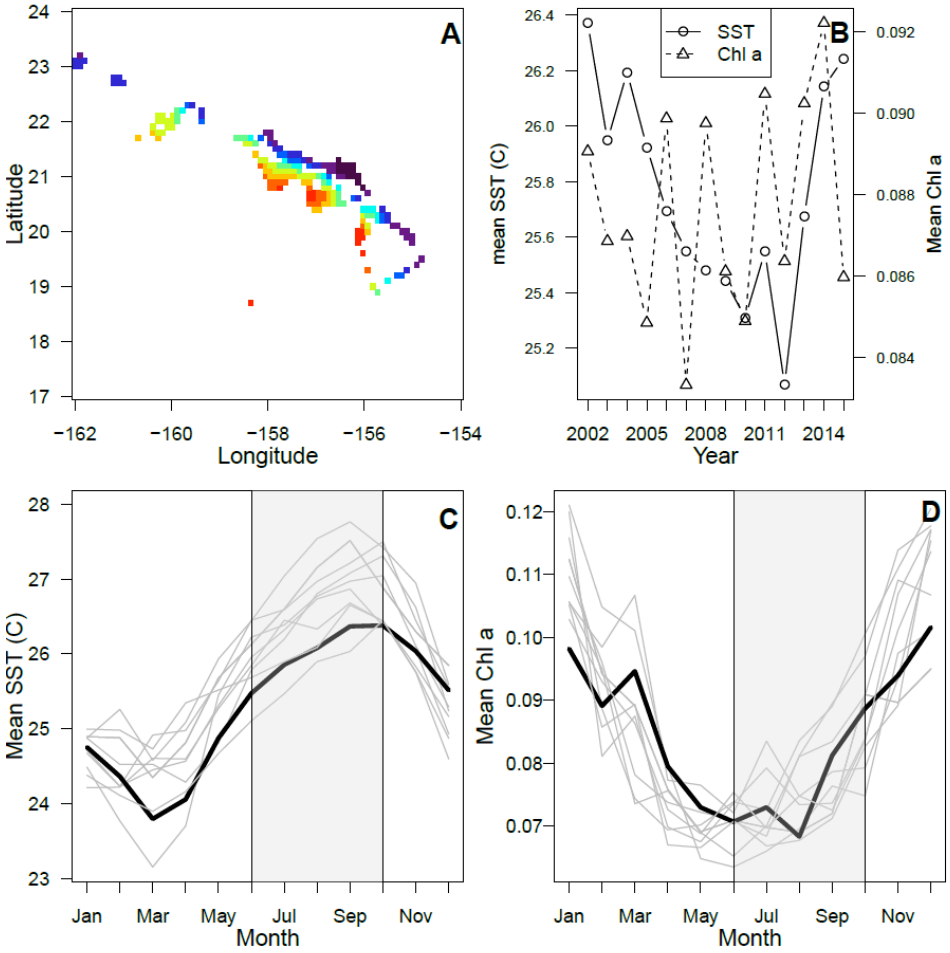

Fig. S3. The pufferfish mortality outbreak occurred during a time period where sea surface temperatures (SST) were high and chlorophyll *a* (Chl *a*) levels were low. (A) Coastal bathymetry 4 km grid (1000 m blue, 100 m red) around main Hawaiian Isands. (B) Mean SST and Chl *a* by year within region highlighted in (A). (C) Mean SST in same region by month for 2002 through 2015 (grey lines) with 2010 (black line) and months of peak epizootic (grey rectangle). (D) Same as C for Chl *a*

Table S1. Location and sex of *A. hispidus* submitted for necropsy

Island	Female	Male	Unknown	Total
Hawaii	7	4		11
Oahu	53	17	16	86
Kauai	1			1
Maui	10	5	1	16
Molokai	1	5		6
Total	72	31	17	120

Table S2. Number and percentage of fish showing gross lesions at different organ systems for 120 necropsied *A. hispidus*

Organ system	n	%
Liver	112	93
Gas bladder	107	89
Skin	81	68
Pouch inflation	59	49
Musculoskeletal	33	28
Gills	30	25
Abdomen	13	11
Eyes	12	10
Cloaca	2	2

Table S3. Number of tissue sections by organs examined histologically (N) and percent (%) with microscopic lesions for 120 necropsied *Arothron hispidus*. Note that not all fish had all organs examined, and in some cases, 2 sections were examined for a particular fish

Organ	Lesion	n	%
Liver	113	138	82
Spleen	72	93	77
Gill	88	120	73
Kidney	101	145	70
Gas bladder	51	129	40
Brain	41	110	37
Adrenal	1	3	33
Intestine	67	207	32
Eye	34	112	30
Skeletal muscle	17	112	15
Urinary bladder	2	26	8
Integument	7	125	6
Heart	6	112	5
Gonad	1	97	1
TOTAL	601	1529	

Table S4. Number of tissue sections with particular microscopic lesion for 120 necropsied *A. hispidus*. Note that not all fish had all organs examined, and in some cases, 2 sections were examined for a particular fish

Lesion	n
Liver	
Vacuolar degeneration	130
Hepatocellular necrosis	87
Intracytoplasmic pigment deposition	22
Encapsulated metazoa	33
Hemorrhage	10
Hepatocellular atrophy	3
Bacterial necrosis	2
Congestion	3
Non-suppurative inflammation	1
Total	291
Spleen	
Depletion of red and white pulp	50
Depletion of red pulp	15
Depletion of white pulp	8
Multifocal necrosis	5
Encapsulated metazoan	6
Non-suppurative inflammation	2
Total	86
Gill	
Epithelial sloughing and necrosis	76
Epithelial hypertrophy	57
Telangiectasia	40
Rosette formation	30
Lamellar collapse	13
Trematodes	12
Myxosporea	9
Non-suppurative inflammation	8
Epithelial pigment deposition	4
Epithelial vacuolation	3
Epithelial atrophy	1
Total	253
Kidney	
Hemorrhage	77
Acute proximal tubular necrosis	32
Depletion of hematopoietic elements	30
Glomerulopathy	14
Non-suppurative inflammation	7
Vacuolation of proximal tubules	5
Encysted metazoa	6
Thrombosis	3
Fragmentation	1
Total	175
Gas bladder	
Intramural hemorrhage	22

Lesion	n
Encysted metazoa	16
Necrosis of gas gland cells	13
Non-suppurative inflammation	7
Vacuolation of gas gland cells	7
Intraluminal and intramural pigment deposition	6
Hypertrophy of gas gland cells	5
Intramural fibrosis	5
Gas gland cell atrophy	2
Vascular engorgement	1
Intramural cleft formation	1
Total	85
Brain	
Congestion	18
Gliosis	16
Neuronal vacuolation	12
Hemorrhage	9
Neuronal necrosis	6
Total	61
Intestines	
Encysted metazoa	29
Non-suppurative inflammation submucosal	27
Focal necrosis and inflammation smooth muscle	20
Ablation of mucosa	17
Hemorrhage of muscularis	9
Submucosal vascular engorgement	1
Suppurative inflammation submucosa	1
Total	104
Eye	
Hemorrhage periocular	12
Engorgement of rete mirabile	10
Lenticular bodies	8
Necrosis of rete mirabile	3
Encysted metazoa	3
Non-suppurative inflammation sclera	2
Thrombosis rete mirabile	1
Total	39
Skeletal muscle	
Rhabdomyolysis	18
Hemorrhage	9
Encysted metazoa	2
Non-suppurative inflammation	1
Total	30
Integument	
Encysted metazoa dermis	5
Dermal hemorrhage	1
Non-suppurative inflammation dermis	1
Thrombosis	1
Total	8
Heart	

Lesion	n
Myocardial necrosis	3
Myocardial hemorrhage	2
Encysted metazoa	1
Non-suppurative inflammation	1
Thrombosis	1
Total	8
Urinary bladder	
Intramural hemorrhage	2
Gonad	
Focal necrosis	1
Intracellular pigment deposition	1
Total	2
Adrenal	
Hemorrhage	1
Intracellular pigment deposition	1
Total	2