Inorganic Constituents in Fresh and Processed Cannonball Jellyfish (*Stomolophus meleagris*)

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Cannonball jellyfish (*Stomolophus meleagris*) from the Gulf of Mexico has been under investigation as a potential source to meet the market demand for jellyfish food in Asia. Traditionally, jellyfish are processed by curing fresh jellyfish with salt and alum, and then the cured jellyfish are desalted in water before consumption. Very little is known about the inorganic constituents of jellyfish. In this study fresh and desalted processed cannonball jellyfish were examined for 22 elements using inductively coupled plasma optical emission spectrometry. The desalted processed tissues had significantly higher Al concentrations (leg, 688 μ g/g; umbrella, 271 μ g/g) than fresh jellyfish (leg, 0.29 μ g/g; umbrella,1.63 μ g/g). Concentrations of macro elements, such as Ca, Mg, K, and Na, were high in fresh jellyfish and rather low in desalted processed tissue. The results should be useful in providing biological and nutritional information about fresh and processed jellyfish.

Keywords: Jellyfish; ICP; mineral elements

INTRODUCTION

Cannonball jellyfish, *Stomolophus meleagris*, are found in abundance in the Atlantic from southern New England to Venezuela and in the Gulf of Mexico (Rudloe, 1988). Jellyfish shunned in Western countries are a multimillion dollar food commodity in Asia. Japan is one of the leading countries in consumption of jellyfish (Suelo, 1986). Interest in utilizing cannonball jellyfish from the United States, a huge untapped resource in the Gulf of Mexico and South Atlantic, has increased recently because of consumer demand and decreased production of jellyfish in Asia (Hsieh and Rudloe, 1994).

Because fresh jellyfish quickly spoil at ambient temperature, they are usually processed immediately after harvesting. The body of cannonball jellyfish consists of a hemispherical transparent umbrella and oral arms. The mouth is on the underside of the umbrella and is protected by four fused oral arms, commonly known as legs. Umbrella (U) and legs (L) are separated and separately processed. Traditional methods of processing jellyfish involve a stepwise removal of water (Subasinghe, 1992). For over a millennium, salt and alum have been used as components of curing solutions or as solids for the dehydration of jellyfish. This results in the elimination of compounds responsible for the sting, and, more importantly, for the achievement of the unique elastic, yet crispy, texture, which is the most desirable characteristic of prepared jellyfish. Processed jellyfish are then desalted and rehydrated in water overnight before shredding and flavoring for consumption.

Very little is known about the concentrations of inorganic constituents in cannonball jellyfish. Huang (1988) determined Ca, Mg, Zn, Fe, and Cu by atomic absorption spectrophotometry; however, results on desalted jellyfish were reported without regard to water content. Information on the effect of salt/alum process-

* Author to whom correspondence should be addressed [telephone (334) 844-2211; e-mail phsieh@ humsci.auburn.edu]. ing on the concentration of inorganic constituents in jellyfish is also lacking.

Elements in biological tissues can be simultaneously and accurately determined by inductively coupled plasma atomic emission spectrometry (ICP). ICP has been used for the detection of micrograms per kilogram levels of analytes in biological samples (Keliher, 1987). Sample preparation for ICP analysis by rapid microwave wet digestion further speeds analysis and reduces the potential for sample contamination (McCarthy and Ellis, 1990).

The objectives of this study were to determine the inorganic constituents in fresh and desalted processed cannonball jellyfish using ICP and microwave digestion and to determine the effect of salt/alum processing on the concentrations of the elements in cannonball jellyfish.

MATERIALS AND METHODS

All labware was washed with 50% (v/v) trace metal grade nitric acid (Fisher Scientific, Norcross, GA) prior to use and rinsed with 18 M Ω cm deionized water. Standard solutions were prepared from concentrate PE pure single and multielement atomic spectroscopy standards (PE XPRESS, Norwalk, CT). Deionized water (of 18 M Ω cm purity) was collected in a large container, and all water used in this study was drawn from the container. Cannonball jellyfish were caught in the Gulf of Mexico near Panacea, FL, and were processed at the Gulf Specimen Marine Laboratory (Panacea, FL) immediately after harvesting. Cured jellyfish along with freshly caught jellyfish were shipped in ice to our laboratory for analysis. Samples were prepared for analysis immediately upon receipt.

Sample Preparation. Fresh jellyfish samples were separated into umbrellas and legs, cut to remove gonads and viscera, and rapidly rinsed under running deionized water (10 s for each sample) to minimize possible loss of analytes. Processed jellyfish were rinsed with running tap water to remove excess amount of dry salt and then soaked in deionized water (sample:water weight ratio was 1:20) for 24 h in a covered plastic container. The water was changed 12 times on an hourly basis and left overnight to achieve an adequate desalting of the processed jellyfish. All samples were blotted on white Bounty paper towels before tissue homogenization, which was done in a Waring Blendor for a total of 60 s (20 s on, 10 s off, repeated twice). Umbrellas and legs were pooled

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Table 1. Instrument Conditions for the ICP

parameter	setting	parameter	setting		
RF power (W)	1100	measurement processing mode	area		
nebulizer flow (L/min)	0.85	autointegration (s)	20 min-50 max		
auxiliary flow (L/min)	1.0	autointegration threshold (ms)	1000		
plasma flow (L/min)	15	read delay (s)	45		
sample flow (mL/min)	1.5	rinse delay (s)	45		
source equilibration time (s)	15	no. of replicates	3		
background correction	manual selection of points	Hg recalibration time (h)	1.0		

Table 2. Element Concentrations of Processed^a and Fresh Cannonball Jellyfish Samples (n = 3)

	processed leg		processed umbrella		fresh leg		fresh umbrella		IDL^d	MDL ^e	spike recovery
element, λ	mean ^b (μ g/g)	RSD (%)	mean (µg/g)	RSD (%)	mean (µg/g)	RSD (%)	mean (µg/g)	RSD (%)	(µg/L)	(µg/g)	(%)
Ag, 328.068	ND^{c}		ND		ND		ND		0.8	0.004	
Al, 396.152	688 a	1.1	271 b	1.9	0.29 с	5.7	1.6 c	17.1	1.4	0.007	97.9
As, 193.696	ND		ND		ND		ND		24.7	0.124	
Ba, 233.527	0.22 a	0.9	ND		0.05 c	2.4	0.09 b	20.8	0.9	0.0045	
Be, 234.861	ND		ND		ND		ND		0.1	0.0005	
Ca, 317.933	11.6 с	2.3	0.90 d	1.8	146 b	0.2	173 a	1.2	2.4	0.012	92.7
Cd, 214.438	ND		ND		ND		ND		1.7	0.009	
Co, 238.892	0.06 a	6.5	0.02 b	16.3	0.06 a	2.9	0.06 a	1.2	1.7	0.009	
Cr, 205.552	0.45 a	0.7	0.08 b	1.4	<mdl< td=""><td></td><td><mdl< td=""><td></td><td>1.6</td><td>0.008</td><td></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td>1.6</td><td>0.008</td><td></td></mdl<>		1.6	0.008	
Cu, 324.754	0.56 a	1.8	0.10 a	7.7	0.13 a	2.6	0.20 a	0.1	0.4	0.002	
Fe, 238.204	40.8 a	0.2	10.4 b	0.7	ND		0.34 c	29.8	1.5	0.008	92.0
K, 766.491	3.34 c	1.8	1.41 c	0.4	234 b	0.2	251 a	0.7	1.0	0.005	
Mg, 279.553	ND		ND		300 b	0.5	313 a	0.5	1.1	0.006	94.9
Mn, 257.610	<mdl< td=""><td></td><td>ND</td><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td>0.3</td><td>0.002</td><td></td></mdl<></td></mdl<></td></mdl<>		ND		<mdl< td=""><td></td><td><mdl< td=""><td></td><td>0.3</td><td>0.002</td><td></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td>0.3</td><td>0.002</td><td></td></mdl<>		0.3	0.002	
Na, 589.626	7.36 b	9.6	5.46 b	5.3	3800 a	0.6	3823 a	0.2	2.1	0.01	101.3
Ni, 221.647	<mdl< td=""><td></td><td>ND</td><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td>3.7</td><td>0.02</td><td></td></mdl<></td></mdl<></td></mdl<>		ND		<mdl< td=""><td></td><td><mdl< td=""><td></td><td>3.7</td><td>0.02</td><td></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td>3.7</td><td>0.02</td><td></td></mdl<>		3.7	0.02	
Pb, 220.353	<mdl< td=""><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td>10.9</td><td>0.05</td><td></td></mdl<></td></mdl<></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td>10.9</td><td>0.05</td><td></td></mdl<></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td><mdl< td=""><td></td><td>10.9</td><td>0.05</td><td></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td>10.9</td><td>0.05</td><td></td></mdl<>		10.9	0.05	
Se, 196.026	<mdl< td=""><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td>17.0</td><td>0.09</td><td></td></mdl<></td></mdl<></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td><mdl< td=""><td></td><td><mdl< td=""><td></td><td>17.0</td><td>0.09</td><td></td></mdl<></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td><mdl< td=""><td></td><td>17.0</td><td>0.09</td><td></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td>17.0</td><td>0.09</td><td></td></mdl<>		17.0	0.09	
Si, 251.605	40.0 a	1.7	7.95 b	1.3	2.50 c	20.8	3.12 с	2.5	17.0	0.09	
Ti, 190.800	1.99 a	0.9	0.48 b	3.7	<mdl< td=""><td></td><td><mdl< td=""><td></td><td>12.1</td><td>0.06</td><td></td></mdl<></td></mdl<>		<mdl< td=""><td></td><td>12.1</td><td>0.06</td><td></td></mdl<>		12.1	0.06	
V, 310.230	1.03 a	8.7	0.54 b	4.0	0.16 c	5.8	0.15 с	33.3	9.4	0.05	
Zn, 213.856	4.06 a	11.7	<mdl< td=""><td></td><td>2.68 b</td><td>0.4</td><td>2.73 b</td><td>1.0</td><td>1.0</td><td>0.005</td><td></td></mdl<>		2.68 b	0.4	2.73 b	1.0	1.0	0.005	

^{*a*} Desalted and rehydrated in water for 24 h. ^{*b*} Means within rows followed by same letter are not significantly different ($\alpha = 0.01$). ^{*c*} ND, not detectable; concentration below IDL. ^{*d*} IDL, instrument detection limit; 3 standard deviations of 10 reps of the blank analyzed as a sample. ^{*e*} MDL, method detection limit; IDL × 0.005 in $\mu g/g$.

separately, and from each pool three composite samples were procured for moisture determination, digestion, and elemental analysis.

Microwave Digestion. Fresh and desalted processed jellyfish samples were digested using a rapid microwave sample preparation system (MDS 2000, CEM Co., Matthews, NC) equipped with Teflon PFA lined digestion vessels (LDVs). About 10 g of wet composite sample tissue (approximately 0.4 g dry tissue equivalence) was accurately weighed to 0.1 mg into each vessel. Ten milliliters of trace metal grade nitric acid was added to each sample, mixed well, and capped according to the manufacturer's direction for digestion. The microwave oven (power level 630 + 50 W) was programmed to run alternating hot-cold cycles (50% power for 10 min followed by a 6 min cooling time at 0% power) with an increasing pressure of 50, 70, 90, 110, and 130 psi for five running cycles, respectively. The pressure was held constant during each of the five 10-min heating cycles. The digests were then cooled to room temperature and transferred to 50-mL volumetric flasks, and deionized water was added to volume. Umbrella and legs of jellyfish were examined separately. Each sample was digested in triplicate.

Determination of Moisture and Elements. The water in the samples was determined with the official oven-drying method (AOAC, 1990). Approximately 10 g of each wet sample was accurately weighed to 0.1 mg into a Al weighing dish. The samples were then dried in the oven (Thelco Model 16, Precision Scientific) at 100 °C for 24 h, cooled in a desiccator to room temperature, and weighed. Three replicate moisture determinations for each of the leg and umbrella samples were made.

Elements were determined using an Optima 3000XL inductively coupled plasma optical emission spectrometer (Perkin-Elmer, Wilton, CT) equipped with an axial torch, Scott-type spray chamber, and cross-flow nebulizer with gemtips. The plasma conditions are summarized in Table 1. The wavelengths used and the instrument and method detection limits for each element are listed in Table 2. An autosampler (Perkin-Elmer AS-91) was used for the introduction of the solutions into the nebulizer. Standards, analytical blanks, and rinse blanks were matrix matched to the sample, so that all solutions contained 20% concentrated HNO₃ by volume. Spiked standards were prepared at 10 μ g/mL Na, 12 μ g/mL Al, 30 μ g/mL Mg, 50 μ g/mL Fe, and 60 μ g/mL Ca from a PE pure multielement standard (PE XPRESS). Additional quality control was performed by analyzing the PE pure multielement standard as unknowns following analysis of the samples. Three determinations were made for all standards and samples.

Statistical Analysis. Data were analyzed by analysis of variance using the General Linear Model (GLM) procedures in the SAS statistical software package (SAS Institute Inc., 1991). Tukey's Studentized range test was used to determine significant differences between means. All comparisons were made at a 1% level of significance.

RESULTS AND DISCUSSION

Moisture in the umbrella and leg tissues of fresh jellyfish (FJ) accounted for 96.1% and 95.8% of the mass, respectively. Moisture in the umbrella and leg tissues of desalted/rehydrated processed jellyfish (DPJ) samples accounted for 96.3% and 96.0% of the mass, respectively. The DPJ took up a substantial amount of water after 24 h of soaking in water, yet did not look watery. Huang (1988) previously reported that commercial and laboratory processed dry salted jellyfish contained 66-69% water.

The concentrations of 22 elements in FJ and DPJ are listed in Table 2. Generally, no significant differences in elemental composition between umbrella and leg tissues of FJ were observed. In DPJ, however, significantly higher concentrations of most elements were found in the legs than in the umbrellas.

The macro elements, Ca, Mg, K, and Na, which were very high in FJ, were greatly reduced in DPJ (Table 2). In FJ tissue, the concentrations of these elements were influenced by composition of sea water. Sodium concentration of FJ was 3800 μ g/g and reduced to only 5–7 $\mu g/g$ in DPJ. The low sodium concentration in DPJ suggests that although processed jellyfish are cured with salt and sold as a dry salted product, they can be used in a low sodium diet after an adequate desalting procedure. There was no significant difference in sodium concentration between the DPJ leg and DPJ umbrella tissues. Apparently, the macro elements were removed with water during the desalting procedures. Results indicated that these elements are present in the FJ mainly as exchangeable or soluble forms rather than in tissue-bound forms. In a nonlaboratory setting, the hardness of water used for soaking and the number of water changes for desalting may slightly affect the residual amount of these elements.

Other elements determined in FJ were at very low levels ranging from below method detection limits to 3.12 μ g/g (Si). However, DPJ had a noticeably higher Al concentration (leg, 688 μ g/g; umbrella, 271 μ g/g) than FJ (leg, 0.29 μ g/g; umbrella, 1.6 μ g/g). Apparently, Al in DPJ was remarkably increased by the curing procedure, which utilizes alum (KAl[SO₄]₃·12H₂O). Generally, the crispiness of cured jellyfish is attributed to the protein precipitation of the firming agent, alum, and processed legs have a crispier and firmer texture than the processed umbrella. The Al concentration was found to be 2.5 times higher in the legs (688 μ g/g) than in the umbrellas (271 μ g/g). Therefore, a higher Al concentration in legs could be associated with the firmer and crispier texture of the legs compared to the umbrella.

Whereas Fe was undetectable in legs and found at very low level (0.34 μ g/g) in the umbrella of FJ, it was significantly higher in DPJ umbrella (10.4 μ g/g) and in DPJ leg (40.8 μ g/g). Si concentrations in processed samples (leg, 40.0 μ g/g; umbrella, 7.95 μ g/g) were also significantly higher than in fresh samples (leg, 2.50 μ g/g; umbrella, 3.12 μ g/g). Cr, Ti, and V were barely detectable in FJ and present at low concentrations in DPJ. Increased concentrations of Fe, Si, Cr, Ti, and V in processed jellyfish could be attributed to the impurities in the curing chemicals, tap water, and processing equipment during processing.

Elements such as Ba, Co, Zn, and Cu were present at very low levels in both fresh and processed jellyfish. Other elements including Ag, As, Be, Cd, Pb, Mn, Ni, and Se were not detectable or below the method detection limit. Heavy metal contamination appears not to be a problem in the fresh and processed cannonball jellyfish.

Three multielement standards (Na, Al, Mg, Fe, and Ca) were analyzed as unknowns after sample analysis to provide quality control. The results of this test (not shown) indicated that no instrument drift and no contamination of the sample introduction system (nebulizer, spray chamber, injector, or torch) occurred. Recoveries of spikes ranged from 92.0% to 101.3% for all elements in the standards (Table 2). The methodology used in this study can be efficiently employed to study a wide range of elements in biological samples. Results are important in providing biological and nutritional information on the inorganic constituents of fresh and processed cannonball jellyfish.

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