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Mercury Concentrations in Pacific Angel Sharks (*Squatina californica*) and Prey Fishes from Southern Gulf of California, Mexico

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Abstract Concentrations of mercury (Hg) were quantified in muscle tissues of the Pacific angel shark, *Squatina californica* sampled from Southern Gulf of California, Mexico, considering total length, sex, diet and the dietary risk assessment. High Hg levels are typically associated with carnivorous fishes, however *S. californica* showed low Hg concentrations (<1.0 $\mu g\ g^{-1}$) in muscle (0.24 ± 0.27 $\mu g\ g^{-1}$ wet weight; n = 94). No effect of sex, total length and weight on Hg concentrations were observed in the shark ($p > 0.05$). Hg concentrations were highest in the darkedge mishipman: *Porichthys analis* (0.14 ± 0.08 $\mu g\ g^{-1}$) and red-eye round herring *Etrumeus teres* (0.13 ± 0.05 $\mu g\ g^{-1}$) relative to other prey species, which could suggest that Hg concentrations in *S. californica* were influenced by these species. Given the relatively low concentration of Hg across age-classes and sex, consumption of *S. californica*'s muscle tissue poses limited risk to humans.

Keywords Trace element · Biomagnification · Elasmobranch · Squatinidae · *Porichthys analis*

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Sharks and rays are traditionally used for food in Mexico, either fresh, frozen or more commonly salt-dried. Around 60 % of total shark catches reported in Mexico are made in the Pacific Ocean. From catches in the Mexican portion of the Pacific Ocean, 40 % correspond to small sharks (less than 150 cm total length) locally known as cazón (Bonfil 1994; Rose 1996). Although an important shark fishery has developed in the central Gulf of California, the southern portion supports captures of commercially important sharks like the Pacific angel shark *Squatina californica* (Villavicencio-Garayzar 1996).

Squatina californica is a demersal shark that feeds mainly on fishes, although it can also prey on crustaceans, and cephalopods (Escobar-Sánchez et al. 2006). This species is a long-lived (e.g., 25–35 years) carnivore, with an estimated trophic level of ~4.0 (Escobar-Sánchez et al. 2011). These characteristics suggest a high capacity for the bioaccumulation of mercury (Hg) in their tissues. Methylmercury is the most toxic form of the naturally occurring element and is considered a global pollutant of public health concern when its concentrations are above natural background level (Boening 2000).

In aquatic food webs, the primary source of Hg and anthropogenic contaminant exposure is from the consumption of contaminated prey, especially for carnivorous species. Hg transfer through the food chain is also the primary source of risk to human health due to the consumption of fish (Moore 2000) and metal accumulation can reach dangerous levels depending on the fish species and frequency of consumption (Ruelas-Inzunza et al. 2011). *Squatina californica* is commercially exploited in the Gulf of California and its meat is consumed largely by coastal communities. The rate of consumption may be an important factor associated to Hg transfer in these communities, representing a health risk, especially for children and

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pregnant women. In the present study, Hg was measured in muscle tissue of *S. californica* and in prey species sampled from stomach contents in order to determine the concentrations of Hg across age classes and sexes of *S. californica* and their prey species, and to assess human health risk.

Materials and Methods

Stomach and anterior-dorsal muscle samples from angel shark were sampled in the southern Gulf of California in the fishing camp El Portugués (24°44′N; 110°40′W). Sharks were captured by fishermen using multifilament gill nets of 10-30 cm mesh size. The nets were set a 1–10 km from the shoreline at sunset and inspected the following morning (Escobar-Sánchez et al. 2006). The Pacific angel shark migrates to deep waters during summer associated to reproduction (Galván-Magaña et al. 1989). Given sampling protocols sharks were collected in February, March, October and December of 2012, and in February of 2013.

Total length (T_L, cm), weight (w), sex, and state of maturity (juveniles and adults) were recorded for each shark captured. A total of 94 angel sharks were obtained with a range of 52–92 cm of T_L (77.1 ± 10.9 cm) and weight range of 1.0–6.1 kg (3.3 ± 1.6 kg) (Table 1). State of maturity was determined according to Romero-Caicedo (2013). Male sharks above 75.6 cm T_L and female individuals above of 77.7 cm T_L were considered as adults.

The choice of prey species for the Hg analyses was based on a previous study carried out by Escobar-Sánchez et al. (2006) in the Southern Gulf of California, in which the abundance, biomass, and frequency of occurrence were taken into account in order to identify the main prey species. These authors reported to fishes, *Porichthys analis*, *Etrumeus teres*, *Decapterus macrosoma*, and the crustacean, *Sicyionia penicillata*, as the most important prey in the Pacific angel shark's diet. From stomach contents, prey species that were intact and in good overall condition (without digestive effect) were used individually for analysis. These prey species were identified and processed. All the shark tissues and their prey were kept in polyethylene bags and transported in ice until the laboratory analysis.

Teflon vessels and other utensils were previously washed with Milli-Q water and nitric acid (HNO_3) according to Moody and Lindstrom (1977). Tissues were also washed with Milli-Q water to remove residual blood, sand or other undesired material. All tissues were freeze-dried for 72 h at −52°C and 100×10^{-3} mbar in a Labconco Freeze Drying System. Samples were weighed before and after the lyophilization process in an analytical balance OHAUS (±0.001 g) to obtain the humidity percentage. Percentage of humidity was estimated (74.06 % ± 5.42 %) and conversion of Hg concentrations from dry to wet weight was made by using the equation Hg (ww) = Hg (dw)*(100-humidity)/100 (Magalhães et al. 2007).

Dry samples were pulverized in an agate mortar with pestle (Fischer Scientific) for homogenizing the samples. Pulverized samples (0.25 g) were placed in capped Teflon vessels (SAVILLEX) for digestion. Concentrated nitric acid (5 mL) was added to each sample vessel. The digestion process was conducted at 120°C for 3 h on a hot plate (Barnstead Thermolyne). Digested samples were placed in polyethylene recipients and then made up to 25 g with Milli-Q water.

Mercury readings were determined by cold vapor atomic absorption spectrophotometry (CV-AAS) in a Buck Scientific mercury analyzer. Blanks (n = 6) and Standard Reference Materials (DORM-3 fish protein) were used to assess the quality of the analytical process. Hg analysis was checked by running these blanks and reference materials with every set of muscle samples. Concentrations were expressed in μg g^{-1} wet weight (ww). Mean recovery was of 0.41 μg g^{-1} (107 %) for muscle (DORM-3: 0.382 ± 0.06 μg g^{-1}; n = 6). The limit of detection was 0.012 μg g^{-1} dry weight.

A generalized linear model (GLM) was used to test the effect of sex, total length, weight and state of maturity of individuals on Hg concentrations.

The hazard quotient (HQ) was calculated to assess the human health risk proposed by Newman and Unger (2002):

Table 1 Number of specimens (N), mean and standard deviation values of total length, weight, Hg concentrations (μg g^{-1}), hazard quotient values (HQ) and maximum possible consumption of fish meat (MPCF; g) of Pacific angel shark from southern Gulf of California, Mexico

Category	N	Muscle (Hg concentration)	TL Average (cm)	TW Average (kg)	HQ	MPCF		
						Man (70 kg)	Woman (60 kg)	Child (16 kg)
Overall	94	0.24 ± 0.28	77.2 ± 10.8	3.3 ± 1.6	0.004	1167	613	163
Males	35	0.19 ± 0.19	74.9 ± 10.4	2.60 ± 1.14	0.003	1474	774	206
Females	59	0.27 ± 0.31	78.3 ± 11.0	3.68 ± 1.68	0.005	1037	544	145
Juveniles	35	0.22 ± 0.20	64.7 ± 6.8	1.64 ± 0.86	0.004	1273	668	178
Adults	59	0.25 ± 0.31	84.36 ± 4.0	4.25 ± 1.01	0.005	1120	588	157

$HQ = E/RfD$

where E is the intake of Hg; RfD is the reference dose for Hg (0.5 μg kg^{-1} of body weight of a person day^{-1}). Hg intake was estimated as $E = C^* I/W$ (C is the concentration of Hg in muscle of sharks, I is the ingestion rate of sharks in Mexico = 0.63 g day^{-1} (CONAPESCA, 2013), and W is the weight of an average adult = 70 kg). If the quotient does not exceed 1 ($HQ \leq 1$), the human population is assumed to be safe.

The maximum possible consumption of fish meat (contained Hg) per week was calculated (MPCF; g of edible portion of fish per kg^{-1} of body weight) by means of the following formula: $MPCF = PTWI/THg_j$

Where: PTWI is the provisional tolerable weekly intake established by WHO (World Health Organization), THg_j is the average THg concentration in fish j). For THg, the PTWI value is 4.0 μg kg^{-1} body weight per week (equal to 0.57 μg/kg bw/day) (Joint FAO/WHO Expert Committee on Food Additives 2010). For pregnant or lactating women this value is restricted to 2.45 μg^{-1} kg^{-1} body weight per week. For adults men, women and children, the average weight of 70, 60 and 16 kg, respectively, were considered in this analysis.

Results and Discussion

A total of 94 muscle samples were analyzed. Shark muscles of all specimens showed a mean and standard deviation of 0.24 ± 0.27 μg g^{-1} w.w. In comparison with other elasmobranchs distributed in Mexican waters, *S. californica* showed Hg values more similar to batoids (<0.5 μg g^{-1} w. w.) (Ruelas-Inzunza et al. 2013; Escobar-Sánchez et al. 2014) than to other sharks (>1.0 μg g^{-1} w.w.) that are mainly pelagic (Maz-Courrau et al. 2012). Even though the consumption of fishes by predators is related with high Hg levels (de Pinho et al. 2002), it was not reflected in *S. californica*, which is an ichthyophagous shark that prey on species that live in the water column (*Decapterus macrosoma*, *Etrumeus teres*) and other fish associated to ocean floor (*Porichthys analis*, *Ophidion iris*).

Some variables such as total length and sex of sharks can have a significant effect on the concentration of Hg (Adams et al. 2003; Storelli et al. 2005). However, Hg levels were not affected by sex, total length, weight or state of maturity in the Pacific angel shark (Table 2). In different sharks species (e.g. *Carcharhinus limbatus*), embryos shown certain Hg levels due to the pass of Hg through the maternal fluid exchange (Adams et al. 2003), therefore the mature females could have a lower amount of Hg respect to the males (Lyons et al. 2013). However, Hg transfer to embryos differs in shark species due to differences by the reproduction mode (Le Bourg et al. 2014). In comparison with placental viviparous, the aplacental species have a minor Hg transfer (Pethybridge et al. 2010). Likewise, *S. californica* is an aplacental viviparous, where not placental connection exist between mother and the embryo, it which are successively nourished by external and internal yolk-sac reserves. Furthermore diet composition can vary ontogenetically between size classes and therefore different patterns of Hg bioaccumulation could be observed. However, there are no ontogenetic diet shifts in relation with angel shark total length's (Escobar-Sánchez et al. 2006), and therefore the Hg levels did not show that differences by total length.

Feeding habit is an important factor related to expected Hg concentrations in fish (Ruelas-Inzunza et al. 2011), because fish absorb Hg (in methylated form) mostly from their food. Knowledge of prey selection enables mapping the flow of persistent chemical contaminants through aquatic food webs (Murphy 2004). Therefore main prey reported for *S. californica* in the Gulf of California (Escobar-Sánchez et al. 2006) were considered in this study to know the potential prey items that provide the major amount of Hg to this predator. The prey with the highest Hg concentration were fish as the darkedge midshipman *P. analis* (0.14 ± 0.08 μg g^{-1} w.w.), followed by the red-eye round herring, *E. teres* (0.13 ± 0.05 μg g^{-1} w.w.). The crustacean *Sicyonia penicillata* (peanut rock shrimp) was also analyzed but their Hg concentration was below the method Hg detection limit (<0.012 μg g^{-1} dry weigh) (Table 3).

Although the uptake through from the water column also provide certain Hg, the main transference of mercury is through the food. In spite of the trophic transference of Hg was based on a few prey, the Hg concentration in the prey indicated different levels of bioaccumulation, i.e. fish accumulated more Hg than crustacean and the Pacific angel shark showed higher concentrations of chemical contaminant than their prey. Most of the prey analyzed are carnivorous, but the flathead grey mullet *Mugil cephalus* is an omnivorous fish, that fed on periphyton or microphytobenthic organisms. Overall, the piscivorous fish contain more Hg than the non-piscivorous (Ruelas-Inzunza et al. 2011). It which is observed among *P. analis* (piscivorous) and *M. cephalus* (detritivorous). Furthermore it is necessary to know the consumption rate per day of *S. californica* to determine the Hg uptake per individual, e.g. if shark consume four specimens of *P. analis* the total Hg transfer would be 0.56 μg g^{-1} per day. However it is necessary known the specific assimilation of the metal by sharks and the chemical form of Hg, due to that organics ones being efficiently assimilated whereas inorganics ones are poorly assimilated.

Elevated Hg levels in fishes represent both an ecological and human health concern (Houserova et al. 2006). Human exposure to Hg is primarily through the consumption of

Table 2 Results of the GML analysis of shark Hg concentrations (μg g^{-1}, w.w.)

Independent variable	Coefficient	Standard error	T value	Probability level
Intercept	0.433	0.523	0.828	0.410
Total length	−0.005	0.009	−0.555	0.581
Sex	−0.143	0.072	−0.199	0.843
Weight	0.078	0.056	1.400	0.165
State of maturity	−0.083	0.131	−0.632	0.529

Table 3 Hg concentrations (μg g^{-1}, w.w.) in the main prey of *Squatina californica* caught from the Southern Gulf of California, Mexico

Group	Family	Item prey	Common name	n	THg $\dot{x} \pm$ SD	% diet contribution (by IRI)*
Fishes	Batrachoididae	*Porichthys analis*	Darkedge midshipman	3	0.14 ± 0.08	22.71
	Carangidae	*Decapterus macrosoma*	Shortfin scad	4	0.06 ± 0.04	29.69
	Dussumieriidae	*Etrumeus teres*	Red-eye round herring	5	0.13 ± 0.05	4.23
	Mugilidae	*Mugil cephalus*	Flathead grey mullet	1	0.04	0.02
	Ophidiidae	*Ophidion iris*	Rainbow cusk eel	2	0.06 ± 0.04	1.31
	Scombridae	*Scomber japonicus*	Chub mackerel	12	0.09 ± 0.06	0.02
	N/A	Pleuronectiformes	Flatfish	1	0.12	0.02
Crustacean	Sicyonidae	*Sicyonia penicillata*	Peanut rock shrimp	1	BDL	5.24

* Values obtained by Escobar-Sánchez et al. (2006) using the Index of Relative Importance (IRI), it which incorporate the occurrence, mass and abundance; N/A: not applicable <; BDL: Below detection limit

fish. Recently, there has been an increase in the consumption of *S. californica* in the market stalls from Baja California Sur, and also the product is exported mainly to Distrito Federal, Jalisco and Baja California, Mexico. Therefore this is a product that should be monitored to assess the risks derived from its consumption. With the exception of one sample of shark muscle tissue, all the sharks have mercury levels below the 1.0 μg g^{-1} (Limit permissible; NOM-027-SSA1-1993). The HQ calculated for *S. californica* was of 0.004, while the average amount of meat consumed maximum possible consumption of fish meat (contained Hg) per week were of 1167 g (adults men), 613 g (women) and 163 g (children) (Table 1). Therefore, based on the tissue concentrations and risk analysis exercises completed in this study, the Hg concentrations determined in Pacific Angel Shark from the Gulf of California (Mexico) present low risk for human consumption.

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References

Adams DH, McMichael RH, Henderson GE (2003) Mercury levels in marine and estuarine fishes of Florida 1989–2001. 2nd edition revised. St. Petersburg, FL, Florida Marine Research Institute, (Florida Marine Research Institute. Technical Report, TR-9). http://aquaticcommons.org/120/1/TR9.pdf

Boening DW (2000) Ecological effects, transport, and fate of mercury: a general view. Chemosphere 40:1335–1351. doi:10.1016/S0045-6535(99)00283-0

Bonfil R (1994) Overview of world elasmobranch fisheries. FAO Fisheries Technical Paper No. 341. Rome

CONAPESCA (2013) Anuario Estadístico de Acuacultura y Pesca. Secretaría de Agricultura, Ganadería y Desarrollo Rural, Pesca y Alimentación. Comisión Nacional de Pesca. Mazatlán. http://www.conapesca.sagarpa.gob.mx/wb/cona/cona_anuario_estadistico_de_pesca

De Pinho AP, Guimarães JRD, Martins AS, Costa PAS, Olavo G, Valentin J (2002) Total mercury in muscle tissue of five shark species from Brazilian offshore waters: effects of feeding habit, sex, and length. Environ Res 89(3):250–258. doi:10.1006/enrs.2002.4365

Escobar-Sánchez O, Abitia-Cárdenas LA, Galván-Magaña F (2006) Food habits of the Pacific angel shark *Squatina californica* in the southern Gulf of California. Cybium 30(4):91–97

Escobar-Sánchez O, Galván-Magaña F, Abitia-Cárdenas LA (2011) Trophic level and isotopic composition of $\delta^{13}C$ and $\delta^{15}N$ of Squatina californica in the southern Gulf of California, Mexico. J Fish Aquat Sci 6(2):141–150. doi:10.3923/jfas.2011.141.150

Escobar-Sánchez O, Ruelas-Inzunza J, Patrón-Gómez JC, Corro-Espinosa D (2014) Mercury levels in myliobatid stingrays (Batoidea) from the Gulf of California: tissue distribution and health risk assessment. Environ Monit Assess 186(3):1931–1937. doi:10.1007/s10661-013-3506-7

Galvan-Magaña F, Nienhuis HJ, Klimley AP (1989) Seasonal abundance and feeding habits of sharks of the lower Golf of California, México. Calif Fish Game 75(2):74–84

Houserova P, Kuban V, Spurny P, Habarta P (2006) Determination of total mercury and mercury species in fish and aquatic ecosystems of Moravian rivers. Vet Med-US 51(3):101–110

JECFA (Joint FAO/WHO Expert Committee on Food Additives) 2010. Joint FAO/WHO Food Standards Programme, Committee of the Codex Alimentarius Commission, Thirty-third Session. http://www.fsis.usda.gov/PDF/2010-CAC/cac33_15e.pdf. Accessed 20 July 2010

Le Bourg B, Kiszka J, Bustamante P (2014) Mother-embryo isotope ($\delta^{15}N$, $\delta^{13}C$) fractionation and mercury (Hg) transfer in aplacental deep-sea sharks. J Fish Biol 84:1574–1581. doi:10.1111/jfb.12357

Lyons K, Carlisle A, Preti A, Mull C, Blasius M, O'Sullivan J, Winkler C, Lowe CG (2013) Effects of trophic ecology and habitat use on maternal transfer of contaminants in four species of young of the year lamniform sharks. Mar Environ Res 90:27–38. doi:10.1016/j.marenvres.2013.05.009

Magalhães MC, Costa V, Menezes GM, Pinho MR, Santos RS, Monteiro LR (2007) Intra- and inter-specific variability in total and methylmercury bioaccumulation by eight marine fish species from the Azores. Mar Pollut Bull 54:1654–1662. doi:10.1016/j.marpolbul.2007.07.006

Maz-Courrau A, López-Vera C, Galván-Magaña F, Escobar-Sánchez O, Rosiles-Martínez R, Sanjuán-Muñoz A (2012) Bioaccumulation and biomagnification of total mercury in four exploited shark species in the Baja California peninsula, Mexico. Bull Environ Contam Toxicol 88(2):129–134. doi:10.1007/s00128-011-0499-1

Moody JR, Lindstrom PM (1977) Selection and cleaning of plastic containers for storage of trace element samples. Anal Chem 49:2264–2267. doi:10.1021/ac50022a039

Moore CJ (2000) A review of mercury in the environment (its occurrence in marine fish). Office of Environmental Management. Marine Resources Division, South Carolina Department of Natural Resources. http://www.dnr.sc.gov/marine/img/mm_paper.pdf

Murphy GW (2004) Uptake of mercury and relationship to food habits of selected fish species in the Shenandoah river basin, Virginia. Dissertation, Virginia Polytechnic Institute

Newman MC, Unger MA (2002) Fundamentals of ecotoxicology. Lewis Publishers, Boca Raton, FL

NOM-027-SSA1-1993. Norma Oficial Mexicana (1993) Bienes y Servicios. Productos de la pesca. Pescados frescos-refrigerados y congelados. Especificaciones sanitarias. Published: June 17 1994

Pethybridge H, Cossa D, Butler ECV (2010) Mercury in 16 demersal sharks from southeast Australia: biotic and abiotic sources of variation and consumer health implications. Mar Environ Res 69:18–26. doi:10.1016/j.marenvres.2009.07.006

Romero-Caicedo A (2013). Biología reproductiva del tiburón angelito (Ayres, 1859) en el suroeste del Golfo de California. Dissertation. IPN-CICIMAR

Rose DA (1996) Shark fisheries and trade in the Americas, vol 1. North America. TRAFFIC, Cambridge

Ruelas-Inzunza J, Páez-Osuna F, Ruíz-Fernández C, Zamora-Arellano N (2011) Health risk associated to dietary intake of mercury in selected coastal areas of Mexico. Bull Environ Contam Toxicol 86:180–188. doi:10.1007/s00128-011-0189-z

Ruelas-Inzunza J, Escobar-Sánchez O, Patrón-Gómez JC, Moreno-Sánchez XG, Murillo-Olmeda A, Spanopoulos-Hernández M, Corro-Espinosa D (2013) Mercury levels in muscle and liver of selected ray species from Northwest Mexico. Mar Pollut Bull 77:434–436. doi:10.1016/j.marpolbul.2013.09.010

Storelli MM, Busco P, Marcotrigiano GO (2005) Mercury and arsenic speciation in the muscle tissue of *Scyliorhinus canicula* from the Mediterranean Sea. Bull Environ Contam Toxicol 75:81–88. doi:10.1007/s00128-005-0721-0

Villavicencio-Garayzar CJ (1996) Aspectos poblacionales del angelito, *Squatina californica*, AYRES, en Baja California, México. Rev Inv Cient Ser Cienc Mar 1:15–21