

Liver Lipids of Indian and Atlantic Ocean Spinner (*Carcharhinus brevipinna*) and Blacktip (*Carcharhinus limbatus*)

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Introduction

Shark liver oils have been reported to be rich in polyunsaturated fatty acids, especially the n3 moieties (1-4). In this study we report the results of analyses of the fatty acids found in the total lipid fractions from the livers of two shark species , spinner (*Carcharhinus brevipinna*) and blacktip (*Carcharhinus limbatus*), found in both the Atlantic and Indian oceans.



Blacktip (*Carcharhinus limbatus*)

Discussion

In agreement with published data (1-8) both shark species from both oceans showed comparatively high levels of total lipid and specifically n3 polyunsaturates. However, the Indian Ocean sharks showed greater total lipid concentrations in than those from the Atlantic. Both shark species examined are reported to exhibit similar prey selection patterns (9-11, 15-17). The most common prey were small pelagic shoaling fish. The lack of difference in liver total lipid between the two shark species within each ocean argues for similarity of diet within each locality, however the fatty acid differences suggest some differences in fatty acid provision. It is possible that the Indian Ocean sharks had been feeding to a greater extent on prey that had migrated from deeper and/or colder waters, as the continental shelf off South Africa's east coast is much narrower than that of the east coast of the USA, and that this had led to their higher levels of liver total lipids and polyunsaturated fatty acids.

Methods

Lipid and fatty acid extraction and quantitation were carried out using standard techniques (13-14).

Results

The livers from Indian Ocean sharks showed greater amounts of total lipids, while the individual fatty acids showed reduced amounts of saturates and slightly increased n3 polyunsaturates (Tables 1-3).

Table 2. Spinner liver fatty acid methyl esters .

Fatty Acids	Atlantic (n=15)		Indian (n=12)		p
	Mean	SD	Mean	SD	
TSFA	30.68	3.312	26.22	3.696	0.013
TMUFA	37.28	3.735	31.42	6.881	0.048
18:2n6	0.00	-	0.51	0.684	-
20:4n6	3.55	0.661	3.67	3.443	0.925
22:4n6	0.88	0.646	0.70	0.481	0.460
22:5n6	3.19	1.689	1.54	1.092	0.014
Tn6PUFA	8.59	2.146	6.51	5.328	0.316
18:3n3	1.26	0.633	1.91	0.906	0.099
20:5n3	3.71	1.148	11.78	3.698	0.002
22:5n3	1.86	0.976	2.68	1.937	0.288
22:6n3	16.05	2.609	17.21	2.169	0.277
Tn3PUFA	22.89	2.838	33.67	4.465	0.001
TPUFA	31.48	3.955	40.18	3.894	0.001

Table 3. Blacktip liver fatty acid methyl esters .

Fatty Acids	Atlantic (n=7)		Indian (n=14)		p
	Mean	SD	Mean	SD	
TSFA	35.14	2.545	28.94	4.570	0.007
TMUFA	39.56	3.852	42.32	11.263	0.533
18:2n6	0.23	0.404	0.32	0.491	0.765
20:4n6	3.07	0.359	2.40	1.945	0.365
22:4n6	0.86	0.239	0.66	0.940	0.577
22:5n6	1.94	0.871	1.23	1.575	0.300
Tn6PUFA	5.87	2.118	4.92	5.280	0.656
18:3n3	1.08	0.594	1.84	1.476	0.213
20:5n3	3.37	1.231	8.97	5.025	0.014
22:5n3	1.46	0.647	1.50	1.475	0.946
22:6n3	12.64	3.322	12.12	1.826	0.737
Tn3PUFA	18.68	4.508	24.77	4.377	0.025
TPUFA	25.09	4.979	29.70	6.267	0.148

Table 1. Liver total lipids per gram wet tissue mass.

lipid g/g wet mass	Mean	SD	p
Spinner Atlantic (n=15)	0.324	0.060	-
Spinner Indian (n=12)	0.431	0.098	-
Spinner Atlantic v Spinner Indian	-	-	0.004
Blacktip Indian v Spinner Indian	-	-	0.906
Blacktip Atlantic (n=7)	0.260	0.102	-
Blacktip Indian (n=14)	0.426	0.106	-
Blacktip Atlantic v Backtip Indian	-	-	0.004
Blacktip Atlantic v Spinner Atlantic	-	-	0.164



Spinner (*Carcharhinus brevipinna*)

References

1. Nichols, P.D., Bakes, M.J. and Elliott, N.G. (1998) *Mar. Freshwater Res.* 49, 763-767.
2. Nichols, P.D., Mooney, B.D. and Elliott, N.G. (2001) *J. Chromatog.* 936, 183-191.
3. Nichols, P.D., Rayner, M. and Stevens, J. (2001) FRDC Project Report 99/369, CSIRO Marine Research and Fisheries Research and Development Corporation, Australia.
4. Wetherbee, B.M. and Nichols, P.D. (2000) *Comp. Biochem. Physiol. Part B.* 125B, 511-521.
5. Banjo, A.O. (1979) *J. Food Technol.* 14, 107-113.
6. Peyronel, D., Artaud, J., Iatrides, M.C., Rancurel, P. and Chevalier, J.L. (1984) *Lipids* 19, 643-648.
7. Davidson, B.C. and Cliff, G. (2003) *Fish Physiol. Biochem.* 26(2), 171-175.
8. Sargent, J.R., Gatten, R.R. and McIntosh, R. (1973) *J. Mar. Biol. Ass. U.K.* 53, 649-656.
9. Cliff, G. and Wilson, R.B. (1986) Natal Sharks Board, Umhlanga, South Africa.
10. Allen, B.R. and Cliff, G. (2000) *S. A. J. Mar. Sci.* 22, 199-215.
11. Dudley, S.F.J. and Cliff, G. (1993) *S. A. J. Mar. Sci.* 13, 237-254.
12. Cliff, G. and Dudley S.F.J. (1992) *Aust. J. Mar. Freshwater Res.* 43, 263-272.
13. Bligh, E.G. and Dyer, W.J., 1959. *Can. J. Biochem. Physiol.* 37, 911-917.
14. Moscatelli, E.A., 1972. *Lipids* 7, 268-271.
15. van der Elst, R. (1981) *The common sea fishes of Southern Africa*, pp36 and 38, Struiks, Cape Town.
16. Compagno, L., Dando, M. and Fowler, S. (2005) *Collins field guide to sharks*, pp293-4 and 300, HarperCollins, London.
17. Castro, J.I. (1996) *Bull. Mar. Sci.* 59, 508-522.

A comparison of the heart and muscle total lipid and fatty acid profiles of nine large shark species from the east coast of South Africa. Bruce Davidson¹, Jonathan Sidell¹, Jeffrey Rhodes¹ and Jeremy Cliff². 1. Saint James School of Medicine, Plaza Juliana #4, Kralendijk, Bonaire, Netherlands Antilles. 2. KwaZulu-Natal Sharks Board, Private Bag 2, Umhlanga Rocks, and Biomedical Resource Unit, University of KwaZulu-Natal, KwaZulu-Natal, South Africa.

Introduction. The east coast waters of South Africa are home to various species of shark, many of which are a bycatch in commercial fishing. They are dumped, wasting material of value for human nutrition. In this study we report the results of analysis of the fatty acids found in the total lipid fractions from the hearts and different muscle tissues of nine large shark species found in this coastal region (1) as an attempt to assess their potential lipid-related nutritional value. The species were *Carcharhinus limbatus* (blacktip), *Carcharhinus obscurus* (dusky), *Carcharhinus brevipinna* (spinner), *Carcharhinus leucas* (Zambezi/bull), *Galeocerdo cuvier* (tiger), *Sphyrna lewini* (scalloped hammerhead), *Sphyrna zygaena* (smooth hammerhead), *Carcharodon carcharias* (great white) and *Carcharias taurus* (raggedtooth/grey nurse/sand tiger).

Methods. Lipid and fatty acid extraction and quantitation were carried out using standard solvent, TLS and GC techniques (2-3).

Results. The results for all species are shown in Table 1. Total lipid was significantly greater in heart samples compared to muscle samples in blacktip, scalloped hammerhead and Zambezi. Great white, smooth hammerhead, spinner and tiger showed greater, but not significant, levels of total lipid. In dusky the heart and muscle samples showed equal total lipid levels, while in raggedtooth the levels in muscle were marginally greater. Within the fatty acids, 20:5n3 was significantly greater in heart samples from blacktip, dusky, smooth hammerhead and spinner compared to muscle samples. 22:6n3 was only significantly greater in heart samples compared to muscle samples in dusky and spinner. Total polyunsaturates and total n3 polyunsaturates were significantly greater in heart compared to muscle in dusky and spinner.

Discussion. The much greater levels of n3 polyunsaturates in both heart and muscle of all species reflectis the preponderance of these fatty acids in the marine foodweb (4). The lack of interspecies variability in profiles in both heart and muscle shows the need for an optimal profile for tissue function. The differences between heart and muscle reflect structural, functional and metabolic differences between cardiac muscle and skeletal muscle (5). In humans, a low ratio of saturated fatty acids to polyunsaturated fatty acids (= <2:1) is considered beneficial (8), while long chain n3 moieties are present within the terrestrial food chain at much lower levels than the n6 (8). Shark muscle can provide a significant amount of polyunsaturated fatty acids and minimal saturated fatty acids. It would seem marine muscle may generally be a more healthy alternative to terrestrial commercially raised species, with the potential health benefits of high quality protein and both n6 and n3 polyunsaturates, compared to the highly saturated profiles of terrestrial commercial meats (9).

References

1. Cliff G and Dudley SFJ. 1992. Austral. J. Mar. Freshwater Res. 43: 263–272.
2. Bligh EG and Dyer WJ. 1959. Can. J. Biochem. Physiol. 37: 911–917.
3. Christie WW. 2003. Lipid Analysis, Oily Press, Bridgwater, England.
4. Nelson MM, Phleger CF, Mooney BD and Nichols PD. 2000. Lipids 35: 551–559.
5. Al-Khayat HA, Morris EP, Kensler RW and Squire JM. 2006. J. Structural Biol. 155: 202-217.
6. Mommsen TP. 2004. Comp. Biochem. Physiol. Part B. 139: 383-400.
7. Sinclair AJ and Mann NJ. 1996. J. Nutr. 126: 1110–1114.
8. Collier GR and Sinclair AJ. 1993. Ann. N.Y. Acad. Sci. 683: 322–330.



Species	Tissue	Stats	Lipid	TS	TM	Tn6P	Tn3P	TP
Blacktip	Heart	Mean	30.1	33.25	26.46	11.88	26.13	38.01
		SD	6.2	3.62	4.92	3.43	3.26	4.09
		th/M	0.01	0.58	0.83	0.06	0.47	0.53
	Muscle	Mean	5.9	32.01	25.84	15.67	24.27	39.94
		SD	2.5	5.64	7.51	3.79	4.30	5.33
		th/M	0.01	0.58	0.83	0.06	0.47	0.53
Dusky	Heart	Mean	9.5	32.14	34.67	10.02	19.10	29.12
		SD	2.5	2.94	4.59	3.32	5.23	5.78
		th/M	0.98	0.33	0.26	0.22	0.04	0.04
	Muscle	Mean	9.5	29.88	39.79	8.95	11.74	20.69
		SD	4.7	4.42	4.29	2.59	2.34	3.91
		th/M	0.98	0.33	0.26	0.22	0.04	0.04
Spinner	Heart	Mean	15.2	32.11	23.18	11.18	28.77	39.95
		SD	8.5	1.99	1.49	2.46	3.28	2.27
		th/M	0.49	0.21	0.11	0.11	0.01	0.01
	Muscle	Mean	6.6	29.35	35.27	8.29	14.23	22.52
		SD	7.1	2.83	9.77	2.74	4.29	5.80
		th/M	0.49	0.21	0.11	0.11	0.01	0.01
Zambezi	Heart	Mean	32.6	37.06	27.95	15.08	26.08	40.98
		SD	9.9	4.90	7.67	8.95	3.99	5.76
		th/M	0.04	0.08	0.81	0.45	0.68	0.70
	Muscle	Mean	5.4	30.92	29.05	14.41	27.20	34.57
		SD	1.7	1.94	4.99	5.09	4.99	7.29
		th/M	0.04	0.08	0.81	0.45	0.68	0.70
Tiger	Heart	Mean	6.5	34.56	32.20	14.25	28.44	38.01
		SD	4.6	3.97	7.98	3.71	13.85	4.27
		th/M	0.15	0.07	0.55	0.87	0.30	0.19
	Muscle	Mean	3.2	29.60	30.42	10.97	22.34	33.31
		SD	2.4	3.36	5.29	2.96	11.56	12.53
		th/M	0.15	0.07	0.55	0.87	0.30	0.19
Sc'H'head	Heart	Mean	19.0	33.69	25.27	9.57	37.03	47.56
		SD	7.8	6.80	6.54	2.20	5.50	3.71
		th/M	0.02	0.12	0.54	0.12	0.18	0.09
	Muscle	Mean	6.9	30.56	22.41	11.60	33.45	44.35
		SD	4.6	5.47	6.49	3.83	9.48	8.60
		th/M	0.02	0.12	0.54	0.12	0.18	0.09
Sm'H'head	Heart	Mean	14.8	32.91	16.34	10.53	28.77	39.95
		SD	2.7	3.87	4.15	3.08	3.28	2.27
		th/M	0.27	0.23	0.14	0.97	0.01	0.01
	Muscle	Mean	9.2	31.00	22.08	10.90	24.23	35.13
		SD	7.3	2.99	7.37	2.87	4.29	5.80
		th/M	0.27	0.23	0.14	0.97	0.01	0.01
G'White	Heart	Mean	8.9	34.25	28.34	10.99	13.89	28.14
		SD	3.1	4.22	3.33	2.74	3.84	6.85
		th/M	0.42	0.80	0.13	0.53	0.37	0.63
	Muscle	Mean	6.1	34.82	25.26	12.29	15.54	29.50
		SD	3.3	7.00	6.55	4.48	4.04	6.82
		th/M	0.42	0.80	0.13	0.53	0.37	0.63
Raggedt'th	Heart	Mean	12.0	30.90	26.03	14.90	15.73	30.81
		SD	3.9	5.99	6.66	3.21	5.30	11.17
		th/M	0.78	0.96	0.73	0.13	0.64	0.41
	Muscle	Mean	12.7	30.81	25.30	12.37	16.60	36.01
		SD	2.2	4.64	7.94	5.09	4.22	5.73
		th/M	0.78	0.96	0.73	0.13	0.64	0.41

The Quantitation of Mercury Concentrations in Two Species of Bonairean Predatory Fish.

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Introduction

Mercury is found in small amounts in the Earth's crust, most commonly in combination with other elements. Volcanic emission is a natural source of mercury, and in areas where volcanic eruptions are common, there is a higher incidence of mercury. Environmental mercury also results from human activities. Coal-powered factories are by far the largest contributor of atmospheric mercury deposits, followed by gold mining. The hydrogeochemical release of mercury from gold-mines in the United States is the biggest source of mercury into the waters surrounding the country. Other sources include metal smelting, waste disposal and cement production.

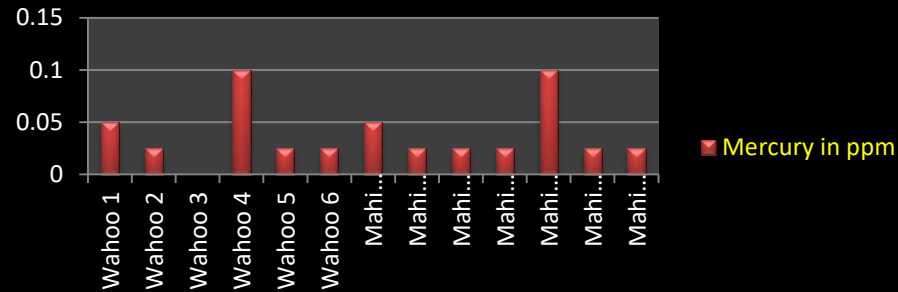
Burning coal releases mercury into the atmosphere and it then is carried back to the surface by rain. In aquatic sediments it is converted by sulphate producing bacteria into methyl-mercury, which is the most lethal form. Methyl-mercury is readily absorbed by dinoflagellates, worms, and other aqueous invertebrates. These are consumed by their natural predators, and the chemical concentrates as it passes through the food web, reaching high concentrations in the top predators (Marine, 2010).

In the United States in 2007, 40% of human exposure to methyl-mercury came from one genus of top predators, tuna. It has been predicted that within the next 20 years that may rise to as much as 50% as known sources experience lax regulations on emissions and other sources of contamination continue to increase (Cone, 2009). Within the last twenty years there has been a 30% increase of mercury levels in the Pacific Ocean. According to the CDC, chronic exposure to mercury can cause severe disability and disease affecting the central and peripheral nervous system. Manifestations in children and adults may differ, with more serious consequences presenting in children.

In 2004, the FDA and EPA issued a joint advisory aimed at protecting pregnant women, women considering becoming pregnant, nursing mothers, and young children (CDC, 2010). The advisory recommended that these populations avoid four types of large predatory fish: swordfish, shark, tilefish and king mackerel. Mahi mahi (dolphinfish) and wahoo are not found in most areas of US coastal waters, but are common top predators in the tropical waters of the Caribbean, as are several species of tuna and barracuda.



Mercury concentrations in Bonairean fish



According to the EPA, chronic amounts of mercury greater than 1 ppm may cause serious health consequences, and excessive amounts of mercury have been linked to several central nervous system disorders, hearing and vision loss, and autism (CDC, 2010). Mercury has a multisystemic toxic effect, divided into long term and short term effects, and dependent on the amount of mercury exposure or ingestion. A study in 2008 showed that the Cuyuni river basin in Venezuela had levels of mercury three times higher than the WHO recommended value (Garcia-Sanchez, 2008), as result of illegal gold mining which had tainted the water with high levels of mercury. Because of Bonaire's location, adjacent to Venezuela, it is possible that indigenous fish populations may have been contaminated by the mercury run off from the gold mines.

Materials and Methods

Samples were obtained from a local restaurant, immediately post cleaning of the fish. Samples were taken from the muscle remaining between the bones of the spinal column of the fish, were heated in sealed containers using microwave energy for three minutes each. Subsequently the fluid exuding from the samples was stored frozen prior to analysis. Samples were analysed using semi-quantitative techniques, according to methods approved by the FDA, and using kits supplied by Osumex Natural Alternatives (Toronto, Canada).

Results

The mean measure of mercury found in the Wahoo samples of fish was 0.045 ppm. The mean measure of mercury found in the Mahi Mahi samples was 0.026 ppm. Overall, the average amount of mercury in the total sample population was 0.038 ppm. This explicitly demonstrates that both species of Bonaire fish should be considered acceptable for consumption, falling below the recommended mercury level of 1 ppm.

Discussion

According to the World Resource Institute, in 2000, the per capita food supply from fish and fishery products was 21 kg/person in the Netherland Antilles. This compared to 8 kg/person for the rest of the Caribbean and the 16 kg/person for the rest of the world. In 2007, 49% of the Bonairean population was female, 25% between the ages of 0-17, 40% within child-bearing age, 25% between the ages of 31-45 and 15% of the population was between ages of 18-30. Therefore a total of 65% of the population of Bonaire would be classified in the group with high risk potential for mercury toxicity (Department of Economic Affairs, 2007).

A study similar to this has been done by Oceana, a Marine conservationist group in the United States, and they demonstrated an average mercury concentration of 0.82 ppm for Wahoo and 0.14 ppm for Mahi Mahi (Oceana, 2009). In their study, they also tested 94 samples of tuna and other types of sushi from grocery stores and other restaurants. High levels of mercury were found in over half of the samples they collected. In our study, potentially toxic mercury levels were not demonstrated, indicating that Bonairean fish may well be comparatively safe for human consumption. However, sampling only two species may not give a holistic view, and other species need to be investigated (eg. tuna, barracuda). Also, the exposure of the fish may vary with time of year, so further work needs to be done in different seasons. Finally, independent corroboration of the results by another laboratory should be done to validate the results obtained.

References

- A Garcia-Sanchez (2008). International Journal of Environment and Pollution: Mercury contamination of surface water and fish in a gold mining region. Volume 33, Number 2-3
- Agency for Toxic Substances and Disease Registry. <http://www.atsdr.cdc.gov/csem/pediatric/diagnosis.html>
- Waterland Village, Bonaire. <http://www.waterlandsvillage.com/aboutbonaire.html>
- Centers for Disease Control and Prevention. Retrieved from: <http://www.cdc.gov/>
- U.S. Department of Health and Human Services (1999). Toxicology Profile: Mercury. Retrieved from: <http://www.atsdr.cdc.gov/toxprofiles/tp46.pdf>
- National Marine Fisheries Service (2010). Retrieved from: <http://www.nmfs.noaa.gov/fishwatch/species/dolphinfish.html>
- Department of Economic and Labour Affairs Bonaire Economy 2007. Retrieved from: http://www.bonaireeconomy.org/stats_pdfs/Economic%20Note%202007%20pdf.pdf
- Doan, T., Melvold, R., Vessili, S., Waltenbaugh, C. (2007). Lippincott's Illustrated Reviews: Immunology. Philadelphia, PA: Lippincott Williams & Wilkins
- United States Environmental Protection Agency (2001). Mercury Update: Impact on Fish Advisories. Retrieved from: <http://epa.gov/osf/fishadvice/mercupd.pdf>
- Lipske, M. (2006). On a Remote Canadian Lake, Scientists Track Mercury's Path Through the Food Chain. Smithsonian Institute Insider. Retrieved from http://www.si.edu/cpa/insiderresearch/articles/V14_Mercury.html
- World Resource Institute (2000). Coastal and Marine Ecosystems: Netherlands Antilles. Retrieved from: <http://earthtrends.wri.org/text/coastal-marine/country-profile-132.html>
- Hightower, J. (2009) A Call for Tougher Standards on Mercury Levels in Fish. Retrieved from: <http://e360.yale.edu/content/feature.msp?id=2113>
- Fish Base Website (2010). Retrieved from: www.fishbase.org
- Wener, K. (2009). Oceana: Mercury levels in Hair of Coastal Alabama Anglers and Residents. Retrieved from: http://na.oceana.org/sites/default/files/0/fileadmin/oceana/uploads/mercury/Rodeo_Hair_Report_Final.pdf
- Methylmercury (MeHg) (CASRN 22967-92-6) | IRIS | US EPA. US Environmental Protection Agency. <http://www.epa.gov/iris/subst/0073.htm>.

Mercury Concentrations in Four Species of Bonairean Predatory Fish.

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Introduction

Burning coal releases mercury into the atmosphere and it then is carried back to the surface by rain. Gold mining increases mercury contamination of rivers and the oceans. In aquatic sediments it is converted by sulphate-producing bacteria into methyl-mercury, which is the most lethal form. Methyl-mercury is readily absorbed by dinoflagellates, worms, and other aqueous invertebrates. These are consumed by their natural predators, and the chemical concentrates as it passes through the food web, reaching high concentrations in the top predators (Marine, 2010).

In the United States in 2007, 40% of human exposure to methyl-mercury came from one genus of top predator, tuna. It has been predicted that within the next 20 years that may rise to as much as 50% as known sources experience lax regulations on emissions and other sources of contamination continue to increase (Cone, 2009). Within the last twenty years there has been a 30% increase of mercury levels in the Pacific Ocean. According to the CDC, chronic exposure to mercury can cause severe disability and disease affecting the central and peripheral nervous system. Manifestations in children and adults may differ, with more serious consequences presenting in children.

In 2004, the FDA and EPA issued a joint advisory aimed at protecting pregnant women, women considering becoming pregnant, nursing mothers, and young children (CDC, 2010). The advisory recommended that these populations avoid four types of large predatory fish: swordfish, shark, tilefish and king mackerel. Mahi mahi (dolphinfish) and wahoo are not found in most areas of US coastal waters, but are common top predators in the tropical waters of the Caribbean, as are several species of tuna and barracuda.



Mahi Mahi



Wahoo



Barracuda



Tuna

Chronic amounts of mercury greater than 1 ppm have serious health consequences, and excessive amounts of mercury have been linked to central nervous system disorders, hearing and vision loss, and autism (CDC, 2010). Mercury has a multisystemic toxic effect dependent on the amount and type of mercury exposure. A recent study showed that the Cuyuni river basin in Venezuela had levels of mercury three times higher than the WHO recommended value (Garcia-Sanchez, 2008), as result of illegal gold mining. Because of Bonaire's proximity to Venezuela, it is possible that indigenous fish populations may have been contaminated by the mercury run off from the gold mines.

Materials and Methods

Samples were obtained from both a local restaurant and fishermen, immediately post cleaning of the fish. Samples were taken from the muscle remaining between the bones of the spinal column of the fish, were heated in sealed containers using microwave energy for three minutes each. Subsequently the fluid exuding from the samples was stored frozen prior to analysis. Samples were analysed using semi-quantitative techniques, according to methods approved by the FDA, and using kits supplied by Osumex Natural Alternatives (Toronto, Canada). Some samples were also analysed by a private laboratory using atomic absorptiometry and confirmed the data reported here.

Hg ppm	Wahoo	Mahi Mahi	Tuna	Barracuda
1	0.025	0.025	0.050	0.000
2	0.000	0.025	0.000	0.000
3	0.050	0.025	0.100	0.050
4	0.000	0.025	0.050	0.025
5	0.000	0.050		0.025
6	0.025	0.100		0.000
7	0.025	0.025		
8	0.100	0.050		
9	0.025	0.025		
10	0.050	0.025		
11	0.025	0.025		
12		0.025		

Results

The mean mercury found in the Wahoo samples was 0.029 ppm. The mean mercury found in the Mahi Mahi samples was 0.035 ppm. The mean mercury found in the Tuna samples was 0.050 ppm. The mean mercury found in the Barracuda samples was 0.017 ppm. Overall, the average amount of mercury in the total sample population was 0.033 ppm. This explicitly demonstrates that all four species of Bonaire fish should be considered acceptable for consumption, falling well below the recommended maximum mercury level of 1 ppm.

Discussion

According to the World Resource Institute, in 2000, the per capita food supply from fish and fishery products was 21 kg/person in the Netherland Antilles. This compared to 8 kg/person for the rest of the Caribbean and the 16 kg/person for the rest of the world. In 2007, 49% of the Bonairean population was female, 25% between the ages of 0-17, 40% within child-bearing age, 25% between the ages of 31-45 and 15% of the population was between ages of 18-30. Therefore a total of 65% of the population of Bonaire would be classified in the group with high risk potential for mercury toxicity (Department of Economic Affairs, 2009).

A study similar to this has been done by Oceana, a Marine conservation group in the United States, and they demonstrated an average mercury concentration of 0.82 ppm for Wahoo and 0.14 ppm for Mahi Mahi (Oceana, 2009). In their study, they also tested 94 samples of tuna and other types of sushi from grocery stores and other restaurants. High levels of mercury were found in over half of the samples they collected. In our study, potentially toxic mercury levels were not demonstrated, indicating that Bonairean fish may well be comparatively safe for human consumption. However, sampling only two species may not give a holistic view, and other species need to be investigated (eg. tuna, barracuda). Also, the exposure of the fish may vary with time of year, so further work needs to be done in different seasons. Finally, independent corroboration of the results by another laboratory should be done to validate the results obtained.

References

- Garcia-Sanchez (2008), International Journal of Environment and Pollution: Mercury contamination of surface water and fish in a gold mining region. Volume 33, Number 2-3
- Agency for Toxic Substances and Disease Registry. <http://www.atsdr.cdc.gov/csem/pediatric/diagnosis.html>
- Waterland Village, Bonaire. <http://www.waterlandsvillage.com/aboutbonaire.html>
- Centers for Disease Control and Prevention. Retrieved from: <http://www.cdc.gov/>
- U.S. Department of Health and Human Services (1999). Toxicology Profile: Mercury. Retrieved from: <http://www.atsdr.cdc.gov/toxoprofiles/tp46.pdf>
- National Marine Fisheries Service (2010). Retrieved from: <http://www.nmfs.noaa.gov/fishwatch/species/dolphinfish.html>
- Department of Economic and Labour Affairs Bonaire Economy 2007. Retrieved from: http://www.bonaireeconomy.org/stats_pdfs/Economic%20Note%202007%20pdf.pdf
- Doan, T., Melvold, R., Vessili, S., Waltenbaugh, C. (2007). Lippincott's Illustrated Reviews: Immunology. Philadelphia, PA: Lippincott Williams & Wilkins
- United States Environmental Protection Agency (2001). Mercury Update: Impact on Fish Advisories. Retrieved from: <http://epa.gov/ost/fishadvice/mercpd.pdf>
- Lipske, M. (2006). On a Remote Canadian Lake, Scientists Track Mercury's Path Through the Food Chain. Smithsonian Institute Insider. Retrieved from http://www.si.edu/cpa/insiderresearch/articles/V14_Mercury.html
- World Resource Institute (2000). Coastal and Marine Ecosystems: Netherlands Antilles. Retrieved from: <http://earthtrends.wri.org/text/coastal-marine/country-profile-132.html>
- Hightower, J. (2009) A Call for Tougher Standards on Mercury Levels in Fish. Retrieved from: <http://e360.yale.edu/content/feature.msp?id=2113>
- Fish Base Website (2010). Retrieved from: www.fishbase.org
- Wener, K. (2009). Oceana: Mercury levels in Hair of Coastal Alabama Anglers and Residents. Retrieved from: http://na.oceana.org/sites/default/files/efileadmin/oceana/uploads/mercury/Rodeo_Hair_Report_Final.pdf
- Methylmercury (MeHg) (CASRN 22967-92-6) | IRIS | US EPA. US Environmental Protection Agency. <http://www.epa.gov/iris/subst/0073.htm>.

The Quantitation of Bacterial Contamination of Paper Currency

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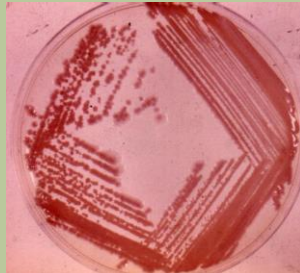
Introduction

Paper money is one of the items most exchanged between people, largely via the drawers of tills in shops. Inevitably there is transfer of skin surface bacteria onto the money and the reverse, thus in any locale most people will be exposed to most of these bacteria over time, and are likely to develop immunity to those bacteria. However, the bacteria present in geographically and/or climatically divergent locales are likely to be different, thus the money will carry different microflora populations adapted to the local conditions. The advent of rapid and frequent travel between such places exposes the traveller to the local microflora population, which may be different to those of their place of origin. This may help to explain the oft reported incidence of gastrointestinal distress amongst travellers but not the local population.

This study was aimed at assessing the degree and nature of the bacterial contamination of paper money from divergent locales in the Caribbean and North America.

Materials and Methods

Samples of low denomination, high turnover notes were obtained from 9 different locales. The samples were individually sealed into sterile bags on collection, then swabbed to sample the bacteria. The swabs were cultured and characterised by the local pathology laboratory on Bonaire, BonLab.



Site	1	2	3	4	5	6	7	8	9
Cultimara	+	++	+	-	+	-	-	-	+
	+	++	++	+	-	-	-	-	++
	+	++	+	-	-	-	-	-	+
SuperStore	+	++	+	-	-	-	-	-	++
	-	+	+	-	-	-	-	-	++
	+	++	+	-	-	-	-	-	+
Walmart1 -U.S	+	++	++	-	-	-	-	-	+
	+	++	+	-	+	-	-	-	+
	-	+	+	+	-	-	-	-	++
Walmart2- U.S	+	++	+	-	-	-	-	-	+
	+	++	+	-	-	-	-	-	++
	+	++	+	-	-	-	-	-	++
Centrum	-	+	+	+	-	-	-	-	+
	+	+	+	+	-	-	-	-	+
	+	+	+	-	+	-	-	-	++
La Curacao	+	+	++	-	-	-	-	-	+
	-	++	+	+	-	-	-	-	++
	-	+	+	-	-	-	-	-	+
Walmart1(Can)	++	+	+	++	-	-	-	+	++
	++	-	++	+	-	+	-	-	++
	++	+	+	-	-	-	-	-	++
Walmart2(Can)	+	+	+	+	-	-	-	-	+
	++	+	+	-	-	-	+	-	++
	++	+	+	+	-	+	-	+	++
Miami	++	+	+	++	+	+	-	-	++
	++	++	+	+	-	-	+	-	+
	++	+	-	+	-	-	-	-	++

1 = Escherischia coli

2 = Enterococcus

3 = Staphylococcus

4 = Streptococcus

5 = Lactobacillus

6 = Pseudomonas

7 = Klebsiella

8 = Haemophillus

9 = Corynebacter

++ = >3 colonies + = 1-3 colonies - = 0 colonies

Results and Discussion

The bacterial load comprised organisms commonly associated with human skin, and no other bacteria were found. However, this does not mean they were absent, they may have merely not multiplied under the culture conditions used.

The bacteria detected in the highest numbers did not vary in presence much between locales, but did vary in amount of colonies. In comparison, the minor components were more highly variable. For example, Pseudomonas, Klebsiella and Haemophillus were only detected in Ontario and Miami, while Lactobacillus was not detected in any of the colder locales. This suggests that it may be the minor components that play a role in the gastrointestinal distress, rather than the more generally distributed major components.

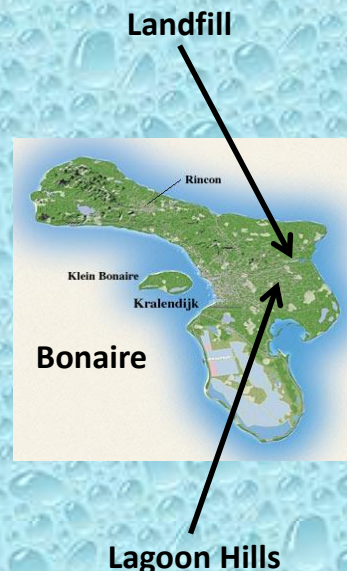
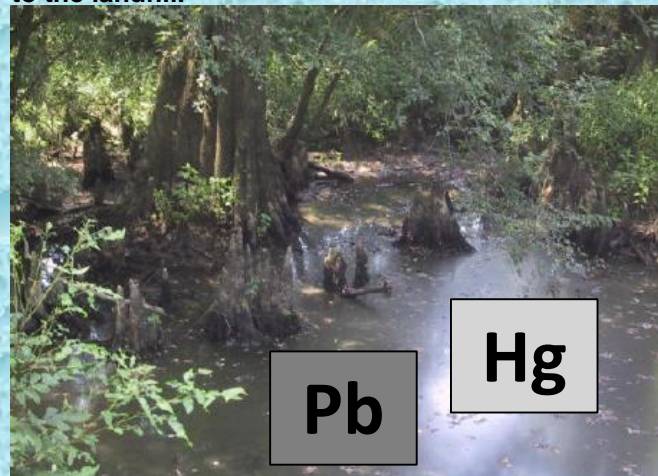
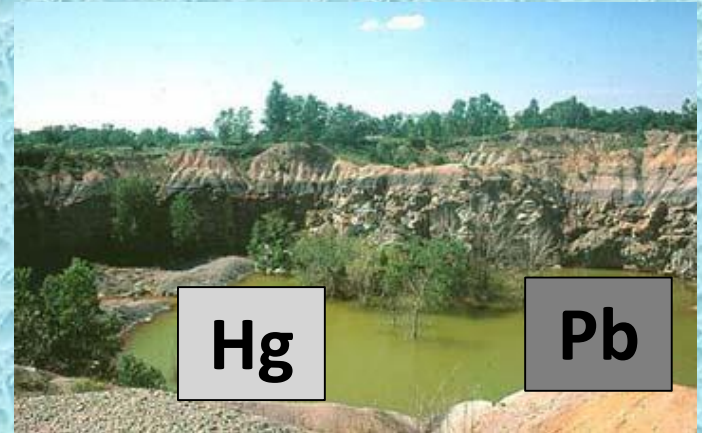
It will be interesting to repeat this work in the future to see whether the same pattern remains in place subsequent to the demise of the Netherlands Antilles guilder and its replacement with the US dollar.

The Quantitation of Heavy Metals in Ground Water

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Introduction

Waste disposal on Bonaire is a major problem. Waste water, solutes and water-borne solids are disposed of via water trenches without any form of treatment. Solid waste is disposed of, unsorted and uncontrolled, via a landfill. This landfill was not properly installed and does not have a barrier membrane between it and the subsoil, which means any leachate as a result of rainfall is not retained within the landfill and may enter the groundwater system. At the same time the leachate also passes into the streams that run from the landfill and contaminate the surrounding areas, as well as the Lagoon and adjacent marine environment. The failure to control the disposal of the solid waste also predisposes to mutually reactive chemicals coming into contact with each other, thus the exact nature of the contamination cannot be deduced. Heavy metal-containing wastes are also disposed of this way, thus such metals (Hg, Pb, Ni, Zn, Mn, Cd, Cu) may be present in the leachate. To assess the degree of contamination, samples from the runoff were collected, as well as from wells adjacent to the landfill.



Materials and Methods

Samples were collected in October 2010 during a period of increased rainfall. Approximately 50ml of water was collected from each of 15 sites. 9 samples were taken from wells on 3 separate properties close to the landfill (Lagoon Hills), while 6 samples were collected from sites around the landfill itself. These samples were stored at -10C until analysed for heavy metal content using Osumex heavy metal screening kits (Osumex, Toronto, Canada). Distilled water was used as negative control.

Results and Discussion

All of the samples collected from the wells were positive for the presence of lead, as were 3 of the landfill samples. 2 of the landfill samples were positive for mercury, while 1 showed traces of manganese and nickel.

The apparent contamination of the wells is a concern as some people in the locale use them as sources of water for all their general activities, including cooking and drinking. However, exact levels of contamination could not be ascertained with the screening kits, and more accurate specific kits will be used to further research.

on management. Some species are merely especially in certain regions of their (e et al., 2011).

the Indian Ocean coastline of South Africa, bears the KwaZulu-Natal Sharks Board shore of the more popular water-based s lead to the accumulation of an enormous brates that inhabit, or pass through, the e scientists at the KZNSB have engaged in itutions, both internal to, and external to,

gy is extensive, that relating to turtle istry, is relatively limited. However, the om several species, tissues and) published data on the fatty acid ipose and liver, in loggerhead turtles 1) examined changes in lipid derivatives, d Lin and Huang (2006) reported on the man et al., (1971, 1972) described the freshwater and marine, including et al., (1985) and Seaborn et al., (2005) while Lawniczak and Teece (2009) turtle egg development in the common e et al., (1995) also assessed lipid changes species, painted (*Chrysemys picta*), *leia blandingii*).

art of the bycatch in the nets off South on. Samples from adipose tissue under the fatty acid analysis to ascertain how closely bled those of animals from other regions.

Loggerhead turtle.



Figure 1. Global distribution of loggerhead turtles.

Materials and methods

Unless otherwise stated all reagents were obtained from Merck Pty. Ltd., Randburg, South Africa. The loggerhead liver and adipose samples were obtained from the KwaZulu-Natal Sharks Board, Umhlanga, South Africa. Samples collected from the five turtles in October 2008 were from one adult male, one juvenile female and three adult females. Samples were taken as soon post mortem as possible and frozen at -20°C.

Aliquots of the samples were thawed, blended and extracted at 4°C overnight using 20 volumes per weight of chloroform:methanol (2:1, v/v) (Folch et al, 1957). The extracts were purified by washing with 20% of their volume of 0.9% saline at 4°C overnight. The chloroform layer was then removed, reduced to dryness, and the dry samples made to 20ml with chloroform, transferred to glass vials and stored at -20°C.

A 1ml aliquot of each extract was used to determine total lipid dry weight, and a further aliquot approximating to 20mg of total lipid was transmethylated using 10% acetyl chloride in methanol to prepare the fatty acid methyl esters (FAME) (Christie, 2003). These were then extracted into hexane, dried under a stream of nitrogen, redissolved in a minimum volume of hexane and the methyl esters separated using a Varian 3400 gas chromatograph run isothermally at 195°C, with a 10% SP2330 on Chromosorb WAW 100/120 6'x1/8" packed column (Supelco Pty. Ltd., Randburg, South Africa) and with flame ionisation (FID) detection. The peaks were quantitated using a Varian 4270 integrator and identified by comparison with authentic FAME standards (Sigma-Aldrich Pty. Ltd., Sandton, South Africa).

Statistical analyses were carried out using a standard package (SPSS). Comparisons between liver and adipose tissue fatty acids were done using the 't' test.

Table 1. The liver and adipose tissue lipid and FAME profiles of the 5 turtles.

Tissue	Liver						Adipose					
Constituent	1	2	3	4	5	$\bar{x} \pm SD$	1	2	3	4	5	$\bar{x} \pm SD$
Lipid (g/g)	0.237	0.316	0.402	0.272	0.351	$0.316 \pm 0.06^*$	0.716	0.652	0.703	0.725	0.675	$0.695 \pm 0.03^*$
12:0	0.34	0.61	0.75	0.49	0.43	0.52 ± 0.16	0.92	0.37	0.67	0.76	0.88	0.72 ± 0.22
14:0	11.22	12.35	9.08	10.76	12.71	$11.22 \pm 1.44^*$	8.55	6.73	7.37	8.20	10.50	$8.27 \pm 1.44^*$
16:0	15.52	12.36	14.34	16.01	14.73	14.59 ± 1.41	14.08	15.77	16.38	14.05	14.76	15.01 ± 1.04
18:0	6.45	7.65	7.04	5.36	6.44	6.59 ± 0.85	7.06	5.89	7.11	6.42	5.81	6.46 ± 0.62
18:1n7	22.53	22.07	24.24	22.62	24.24	22.82 ± 1.15	22.64	22.76	24.55	22.42	24.05	22.46 ± 1.25

Conservation Union) states in the 2004 Red Data Book (turtles and tortoises) are relatively well conserved. Of the 305 described species evaluated, 128 (42%) are considered threatened (IUCN, 2004). Within the IUCN, the Species Survival Commission published a review of the status of marine turtles in 2001, and identified several regional populations of different species of marine turtles - the north east Indian Ocean, the western Indian Ocean (Wallace et al., 2011). However, conservation of marine turtles is difficult, as loggerhead turtles are migratory, so even in the relatively safe waters of South Africa, many are not protected (Figure 1.). Thus any work which aims to understand the history of the species may be advantaged by studying multiple species.

In this study we assessed the lipid and fatty acid profiles of loggerhead turtles caught in a single period of their cycle. All turtles were juvenile, there were no gender or age related differences in fatty acid profiles. While there were significant differences in fatty acid saturates, given their largely energy metabolism, these were not significant on a holistic level. The differences in fatty acid profiles were metabolically important difference. The degree of conformity between the adipose and liver fatty acid profiles are of limited availability and hence conservation of polyunsaturates, with significantly greater dietary availability and hence not conserved. When comparing our data with those of Christie and Ackman et al., (1971) on loggerhead turtles, we found significant differences. The only apparently major difference was in the polyunsaturates. The author groups demonstrated much lower levels of polyunsaturates, which also caused a reduced total polyunsaturates in the liver. The fatty acid profiles for liver, with much greater dietary availability, were similar to others (Ackman et al., 1992, Joseph et al., 1993). The fatty acid profiles from the central Pacific showed adipose tissue fatty acid profiles similar to loggerheads. These may reflect true inter-population differences as the Chinese soft shelled turtles were wild caught. Lin and Huang (2006) also found differences in fatty acid profiles of soft shelled turtles, which may be due to captive alligator (*Alligator mississippiensis*) and crocodile (*Crocodilus*) effect in wild versus captive caiman (caiman) and crocodile (crocodile) et al., (1993) in wild versus captive Nile crocodile (*Crocodilus*) acid metabolism is adaptable to the fatty acid profiles of a species there appears to be a predominance of the n3 polyunsaturates in loggerhead turtles, which reflects differences between oceanic populations and those of availability from dietary sources, or both,