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Toxic Elements in Marine Products and Human Hair Samples in Mersin, Turkey

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Metallic elements are found in all living organisms, where they play variety of roles. Some metals are essential elements and their deficiency results in impairment of biological function, but when present in excess, essential metals may also become toxic (Rivai 2001). Fishes have the ability to accumulate heavy metals in their tissues by the absorption along the gill surface and gut tract wall to higher levels which are several hundred times more than the concentration of metals in their surrounding water medium. Accumulation of heavy metals is reported to be influenced by many factors such as the organ, species, age, sex and the environmental factors like temperature, salinity and pH (Suresh et al. 1993).

The impact of environmental exposure to toxic metals on human health has been discussed in numerous publications over the past 20 years (Badsha and Golspink 1988; Renzoni et al. 1998; Raghunath et al. 1999; Nowak and Chmielnicka 2000). Hair is a metabolic end product that incorporates metals into its structure during the growth process. Heavy metal content of human hair has been used for some time as an index to assess environmental and occupational exposure to toxic metals. The previous studies have reported when that if compared to other biological material, hair offers a number of advantages; (1) Easy to obtain without injuring the donor, (2) Can be stored for long periods of time before being analyzed without deterioration since the elements in the sample are assumed to be stable, and (3) The concentration of most elements are relatively high in hair as compared to the rest of the body (Rivai 2001). Unlike other biological specimens (e.g. the blood and urine) that indicate recent exposure, trace metal content of hair correlates with body stores (Ahmed and Elmubarak 1990). The elemental analysis of hair is, therefore, becoming a popular method for determining systematic toxication, diseased states and also as a method in forensic work (Kunç 1987; Yoshinaga et al. 1997; Mickeley et al. 1998). The amount of Hg in hair is 250 times more than those in blood (Ünlü 1993). On the other hand, Pb is nearly 100 times more concentrated in hair than those in body liquids (Kunç 1987). In this study, Hg, Pb, Cu, and Zn levels in marine product samples consumed in Mersin and in human scalp hair samples taken from people living in this region were investigated. The distributions and relationships of above elements between human scalp hair samples taken from people who eat fish regularly and who never eat fish as test persons were compared by using Mann-Whitney U statistics.

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MATERIALS AND METHODS

Human scalp hair samples were snipped with clean stainless steel scissor to 0.5 cm from the scalp. 50 of which were taken from people who eat fish regularly and 16 of which were taken from people who never eat fish. The samples were put in polyethylene bags and kept in dark until the time of experiment. The samples were washed with a non-ionic detergent. For mercury analysis the digestion procedure was as follows: dried sample (0.1 g), nitric and perchloric acid mixture (a ratio of 1:5 v/v) were placed in a Digestion Bomb apparatus, and then the temperature was gradually heated to 100 ^{0}C and maintained for 2 h. Cooling and diluting resulted in a volume of 25 mL (Kunç 1987).

Marine product samples were collected from Mersin Fish Market and brought to laboratory and then frozen in a -22 ^{0}C freezer. Frozen samples were thawed prior to use. 0.85 g muscle tissue of the samples and 3 mL nitric acid were placed in the Digestion Bomb. A temperature-controlled and time-controlled heated digestion unit was used as follows: the solutions were gently heated to 140-150 ^{0}C (2.5 h) until digestion was complete. After cooling, the solutions were quantitatively transferred to a standard flask and diluted to 25 mL with deionized water of which the trace element content was carefully checked (AOAC 1990).

The amount of Hg in 25 mL samples was determined by using UNICAM 939 Hidrid Generation VP 90 Continuous Flow Vapour System with 253.7 nm wavelength.

Ammonium pyrrolidine dithiocarbamate (APDC) extraction in methyl iso buthyl ketone (MIBK) solution was carried out using the digestion solution with following pre-treatment for Pb, Cu and Zn analysis (Rand 1985). Samples were concentrated to 5 mL and kept in refrigerator at 4 ^{0}C. A flame Atomic Absorption Spectrometer (Perkin-Elmer 3100) was used for the determination of the above metals. MIBK was used as a blank.

RESULTS AND DISCUSSION

The concentration ranges of Hg were found varied from 0.010 to 0.345 μg/g, 0.085 to 0.708 μg/g, 0.033 to 1.857 μg/g, 0.049 to 2.662 μg/g and 0.049 to 0.820 μg/g in grey mullet (n=30), hake (n=30), red mullet (n=30), common sole (n=30) and shrimp (n=40) samples, respectively. Pb concentrations were not determined in the samples due to the detection limit of the AAS, except for hake (Pb concentration of which varied from 0.228 to 2.159 μg/g, 7 samples were above the permitted value) and red mullet samples (Pb concentration of which varied from 0.809 to 1.863 μg/g, 11 samples were above the permitted value). Cu was determined only in shrimp samples, which belong to the crustacea family and varied from 0.196 to 10.676 μg/g.

Toxic metal concentrations of human hair samples taken from people who eat fish regularly and who never eat fish are given in Table 1 a, b and Table 2.

Table 1a. Hg, Pb and Zn concentrations in the human hair samples taken from people who eat fish regularly (M: Male, +: Smoker, -: Non-smoker)

No	Sex	Age	Profession	Smoking Habit	Hg (μg/g) Mean ± SD	Pb (μg/g) Mean ± SD	Zn (μg/g) Mean ± SD
1	M	10	Fisherman	-	0.452 ± 0.018	22.631 ± 0.578	193.125 ± 0.019
2	M	13	Fisherman	-	0.650 ± 0.030	26.852 ± 0.030	215.741 ± 0.029
3	M	15	Fisherman	-	1.897 ± 0.537	96.774 ± 0.700	366.129 ± 0.540
4	M	15	Fisherman	-	0.974 ± 0.177	41.667 ± 0.170	259.722 ± 0.176
5	M	16	Fisherman	+	1.519 ± 0.313	27.152 ± 0.314	177.152 ± 0.313
6	M	17	Fisherman	+	0.679 ± 0.220	67.248 ± 1.006	422.401 ± 0.230
7	M	17	Fisherman	-	0.777 ± 0.052	33.810 ± 0.061	188.095 ± 0.053
8	M	18	Fisherman	-	1.249 ± 0.330	47.368 ± 0.600	411.842 ± 0.329
9	M	18	Fisherman	-	0.840 ± 0.080	38.631 ± 3.114	408.571 ± 0.070
10	M	18	Fisherman	+	0.729 ± 0.031	64.216 ± 3.466	157.682 ± 0.030
11	M	22	Fisherman	-	0.403 ± 0.059	29.534 ± 7.119	257.238 ± 0.060
12	M	23	Fisherman	-	0.461 ± 0.020	29.612 ± 0.028	231.068 ± 0.020
13	M	23	Fisherman	+	0.878 ± 0.095	61.194 ± 0.090	194.776 ± 0.096
14	M	23	Fisherman	+	1.337 ± 0.431	11.161 ± 0.332	272.768 ± 0.433
15	M	23	Fisherman	+	0.905 ± 0.150	40.000 ± 0.049	438.462 ± 0.149
16	M	24	Fisherman	+	3.284 ± 0.803	17.327 ± 0.903	255.446 ± 0.802
17	M	24	Fisherman	-	0.621 ± 0.025	43.363 ± 0.026	226.991 ± 0.027
18	M	25	Fisherman	+	1.153 ± 0.235	107.843 ± 0.231	225.714 ± 0.234
19	M	25	Fisherman	-	0.819 ± 0.078	37.500 ± 0.088	556.818 ± 0.077
20	M	25	Fisherman	+	2.138 ± 0.605	29.651 ± 0.507	202.907 ± 0.607
21	M	26	Fisherman	+	1.186 ± 0.283	86.250 ± 0.254	362.500 ± 0.284
22	M	27	Fisherman	+	1.078 ± 0.000	65.909 ± 0.001	305.682 ± 0.000
23	M	27	Fisherman	-	1.542 ± 0.345	44.000 ± 0.348	329.333 ± 0.347
24	M	28	Fisherman	+	0.866 ± 0.085	57.407 ± 0.079	244.444 ± 0.082
25	M	29	Fisherman	-	0.580 ± 0.025	24.380 ± 0.029	269.421 ± 0.026

Permitted limit for total mercury in marine products, which is also adopted by various countries, is 0.5 μg/g fresh weight as reported by FAO (1983). The WHO (1987) recommends a permitted value of 0.31 μg/g. Accepted limits for human consumption (150.0, 10.0 and 1.5 μg/g wet weight, for zinc, copper and lead, respectively) were set by the Australian National Health and Medical Research Council (Sharif et al. 1991).

There were 13 hake, 9 red mullet, 11 common sole and 5 shrimp samples above the permitted value for Hg concentrations. Only grey mullet samples were found to below the permitted value.

Relationships between metal concentrations of the marine product samples and parameters (weight and length) are shown in summary in Table 3.

The values that below the Pb= 0.02 mg/L, Zn= 0.002 mg/L and Cu= 0.012 mg/L could not be analyzed due to the detection limit of the AAS.

Table 1b. Hg, Pb and Zn concentrations in the human hair samples taken from people who eat fish regularly (M: Male, +: Smoker, -: Non-smoker)

No	Sex	Age	Profession	Smoking Habit	Hg (μg/g) Mean ± SD	Pb (μg/g) Mean ± SD	Zn (μg/g) Mean ± SD
26	M	29	Fisherman	+	1.525 ± 0.320	50.000 ± 0.322	389.130 ± 0.320
27	M	29	Fisherman	+	1.103 ± 0.275	71.581 ± 1.200	645.349 ± 0.280
28	M	30	Fisherman	+	0.438 ± 0.135	21.875 ± 0.130	204.375 ± 0.138
29	M	30	Fisherman	+	0.899 ± 0.141	41.026 ± 0.145	239.744 ± 0.143
30	M	30	Fisherman	+	1.242 ± 0.335	33.929 ± 0.330	209.524 ± 0.334
31	M	30	Fisherman	-	0.443 ± 0.013	28.972 ± 0.017	242.056 ± 0.015
32	M	31	Fisherman	-	0.934 ± 0.164	48.014 ± 6.698	254.285 ± 0.162
33	M	31	Fisherman	+	0.470 ± 0.022	44.400 ± 0.021	266.400 ± 0.024
34	M	32	Fisherman	+	0.927 ± 0.147	36.932 ± 0.149	259.659 ± 0.148
35	M	35	Fisherman	+	1.245 ± 0.337	21.278 ± 0.402	166.023 ± 0.335
36	M	35	Fisherman	+	0.818 ± 0.077	81.034 ± 0.077	400.000 ± 0.077
37	M	40	Fisherman	+	1.689 ± 0.412	58.182 ± 0.371	255.455 ± 0.415
38	M	42	Fisherman	+	0.709 ± 0.049	36.869 ± 0.046	271.212 ± 0.050
39	M	43	Fisherman	+	0.622 ± 0.025	88.793 ± 0.023	299.138 ± 0.025
40	M	44	Fisherman	+	1.050 ± 0.000	158.036 ± 0.002	403.571 ± 0.000
41	M	45	Fisherman	+	1.278 ± 0.312	94.565 ± 0.318	410.870 ± 0.312
42	M	47	Fisherman	+	0.920 ± 0.144	34.158 ± 0.141	279.208 ± 0.145
43	M	50	Fisherman	+	0.863 ± 0.084	53.636 ± 0.081	373.636 ± 0.085
44	M	50	Fisherman	+	0.988 ± 0.158	51.408 ± 0.160	211.972 ± 0.159
45	M	50	Fisherman	+	0.594 ± 0.020	26.263 ± 0.020	168.182 ± 0.020
46	M	51	Fisherman	-	0.456 ± 0.019	28.879 ± 0.020	233.621 ± 0.018
47	M	53	Fisherman	-	0.905 ± 0.140	147.692 ± 0.137	535.385 ± 0.141
48	M	53	Fisherman	-	1.153 ± 0.234	43.137 ± 0.236	294.118 ± 0.232
49	M	57	Fisherman	+	3.065 ± 0.755	98.750 ± 0.791	151.667 ± 0.756
50	M	63	Fisherman	-	1.729 ± 0.521	67.647 ± 0.525	269.118 ± 0.520

Mann-Whitney U statistics were used for comparisons of each groups in terms of concentration, depth, feeding, etc. The correlation (Kendall Test) was used to investigate the relationship between toxic metal concentrations and parameters (such as length, weight and age).

There were no significant differences in Hg and Pb concentrations between *Mullus barbatus* and *Upeneus moluccensis* ($p>0.05$). Hg concentrations in *Upeneus moluccensis* were higher than those in *Mullus barbatus*. This should be caused by *Upeneus moluccensis* live on coastal area, whereas *Mullus barbatus* live in depth. It is suggested that living and feeding places are important to heavy metal accumulation. Pb concentrations in *Mullus barbatus* are higher than those in *Upeneus moluccensis* samples.

The level of Hg concentrations was found high for common sole (*Solea solea*) samples. This should be caused by feeding on fish and invertebrates that live on the bottom sediments in the sea.

Table 2. Hg, Pb and Zn concentrations in the human hair samples taken from people who never eat fish as test persons (M: Male, F: Female +: Smoker, -: Non-smoker)

No	Sex	Age	Profession	Smoking Habit	Hg (μg/g) Mean ± SD	Pb (μg/g) Mean ± SD	Zn (μg/g) Mean ± SD
1	M	14	Student	-	0.863 ± 0.081	10.167 ± 0.878	144.921 ± 0.080
2	F	18	Student	-	0.869 ± 0.134	21.458 ± 0.884	265.795 ± 0.133
3	F	19	Student	-	0.290 ± 0.006	20.500 ± 4.243	181.671 ± 0.005
4	M	19	Student	+	0.654 ± 0.033	8.250 ± 0.250	166.556 ± 0.031
5	F	20	Student	+	0.427 ± 0.131	10.375 ± 0.530	343.498 ± 0.130
6	F	20	Student	-	0.408 ± 0.000	14.667 ± 1.233	274.750 ± 0.000
7	F	20	Teacher	+	1.019 ± 0.000	15.000 ± 0.000	229.375 ± 0.000
8	F	20	Secretary	+	0.237 ± 0.000	9.583 ± 1.627	170.750 ± 0.000
9	F	20	Student	-	0.240 ± 0.005	17.000 ± 0.000	229.333 ± 0.006
10	M	20	Student	+	0.180 ± 0.000	11.833 ± 0.382	191.625 ± 4.066
11	F	21	Nurse	-	0.237 ± 0.000	8.000 ± 0.148	228.750 ± 0.010
12	F	23	Technician	-	0.237 ± 0.000	13.833 ± 0.577	236.250 ± 0.000
13	F	23	Student	+	0.394 ± 0.055	22.375 ± 4.369	232.250 ± 3.536
14	F	25	Student	-	0.143 ± 0.000	13.500 ± 0.000	198.500 ± 0.031
15	F	63	Housewife	-	0.180 ± 0.000	15.125 ± 0.530	178.500 ± 0.001
16	F	73	Housewife	-	11.836 ± 2.091	9.750 ± 0.250	190.881 ± 2.092

There were no differences in Hg concentrations between *Lisa aurata* and *Lisa saliens* ($p>0.05$). This may be due to the fact that they were living in the same places. The level of Hg concentrations was found low for grey mullet samples, as grey mullet are pelagic.

There were highly significant differences in Hg and Cu concentrations between *Metapenaeus monoceros* and *Parapenaeus longirostris* ($p<0.001$). This should be a result of living area and feeding; since *Metapenaeus monoceros* are living in coastal area, while *Parapenaeus longirostris* are living in depth.

Table 3. Correlations between mercury and lead concentrations and weight and length of the marine product samples

Species	Significance of correlation				
	Hg & Wt	Hg & Lt	Pb & Wt	Pb & Lt	Hg & Pb
Lisa aurata *Lisa saliens*	ns	ns	-	-	-
Merluccius merluccius	ns	ns	p<0.05	p<0.05	p<0.001
Mullus barbatus *Upeneus moluccensis*	ns	ns	ns	p<0.05	ns
Solea solea	ns	ns	-	-	-
Metapenaeus monoceros *Parapenaeus longirostris*	ns	ns	-	-	-

Hg and Zn concentrations were negatively correlated with age variability ($p>0.05$), whereas Pb concentrations were positively correlated with age variability ($p<0.05$) for 50 human scalp hair samples taken from people who eat fish regularly. Correlation between Hg and Zn was found to be not significant ($p>0.05$), whereas correlations between Hg and Pb, Zn and Pb were found to be highly significant ($p<0.01$).

Hg concentrations were positively correlated with age variability ($p<0.01$), while Pb and Zn concentrations were negatively correlated with age variable ($p>0.05$) for 16 human scalp hair samples taken from people who never eat fish as control group. Correlations among the Hg, Pb and Zn were found to be not significant ($p>0.05$).

Hg concentration of 16th sample of the control group was 11.836 ± 2.091 μg/g. This value was higher than the human scalp hair samples taken from people who eat fish regularly. This may be due to the fact that all teeth of the person are amalgamated. The person shows Minamata Diseas (Hg poisoning) semptoms such as lost of equilibrium, sight and hearing problems (Gonzales 1985). These problems started after the amalgamation. Therefore, the sample was kept out of statistical analysis (Doğan, 1997).

Statistical analysis of the data indicated that concentrations of Hg, Pb and Zn in human scalp hair samples taken from people who eat fish regularly and who never eat fish were significantly different ($p<0.001$). This shows that, the toxic elements are introduced human body through the food chain.

Kyle and Ghani (1982) found that mean Hg concentration in the scalp hair of 27 person eating basically fish in Papua New Ginea was 15.5 μg/g. Kayakırılmaz and Oral (1989) found that there was a positive relationship between diet and Zn concentration in human scalp hair.

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