

## Fish consumption frequency and lipid peroxidation in the riverside population of Lower Tocantins, Pará

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### ABSTRACT

**Introduction:** Several studies report the benefits of fish consumption to the prevention of cardiovascular diseases such as atherosclerosis, thrombosis and arrhythmia. On the other hand, regular consumption of fish can induce the accumulation of methylmercury in the body.

**Objective:** To evaluate the relationship between frequency of fish consumption, mercury concentrations and intensity of lipid peroxidation.

**Methods:** A cross-sectional, observational study. It was evaluated riverside in Limoeiro do Ajuru, Pará. Variables were sociodemographic variables, frequency of weekly fish consumption, concentration of total mercury in hair and the dose of malondialdehyde in plasma. The concentrations of total mercury ( $\mu\text{g/g}$ ) and malondialdehyde ( $\text{nmol/ml}$ ) were respectively  $0.63 \mu\text{g/g}$  and  $0.54 \text{ nmol/ml}$  for weekly fish consumption less than two meals,  $0.51 \mu\text{g/g}$  and  $0.42 \text{ nmol/ml}$  for consumption in two to four meals and  $0.88 \mu\text{g/g}$  and  $0.31 \text{ nmol/ml}$  for consumption major than four fish meals. There was a significant difference between groups of two to four and major than four meals, only for the total mercury variable ( $p = 0.008$ ).

**Discussion:** In this study, low levels exposure and high fish consumption can influencing the bioaccumulation of mercury in this population. Santos et al, found the total mercury concentration ( $0.09$  to  $3.79 \mu\text{g/g}$ ) in the rivers and compared the levels according to the intake categories of fish, did not obtain significant statistical difference. This divergent result suggests that deforestation in the Amazon has been increased and is an important vehicle for mercury exposure which affects local survival and subsistence.

**Conclusion:** Riparians in the studied region have high fish consumption and low concentrations of mercury. Although the group with low fish consumption had higher levels of MDA there was no significant difference when compared with other groups.

### KEY WORDS

Fishes, Lipid Peroxidation, Mercury, Malondialdehyde.

### INTRODUCTION

The Food and Agriculture Organization of the United Nations<sup>1</sup> recommends that fish intake should be at least  $12 \text{ kg/inhabitant/year}$ , however, the riverine populations fish corresponds to about 60% of protein intake. Several studies show the high consumption and the adequate energy-protein contribution that the fish possesses to the populations bordering the Amazon<sup>2,3</sup>.

Several studies report the benefits of fish consumption or fish oil supplementation to human health, among which the

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beneficial effects in the prevention of cardiovascular diseases such as atherosclerosis, thrombosis and arrhythmia<sup>4</sup>. The omega-3 ( $\omega$ -3) exerts a protective effect on lipid metabolism, triglyceride level, systemic arterial pressure, and has anti-inflammatory effect, in diabetes, hypertension, obesity and certain types of cancer<sup>5-8</sup>.

The families of polyunsaturated fatty acids (PUFAs)  $\omega$ -3 integrate into the membrane phospholipids more easily in place of arachidonic acid, and are able to produce eicosanoids or docosanoids, with anti-inflammatory and antithrombotic properties, since they compete for cyclooxygenase (COX) receptor and are preferred substrates in the lipoxygenase (LOX) pathway<sup>4</sup>. However, PUFAs present on the plasma membrane are more susceptible to lipid peroxidation, a process in which the reactive oxygen species (ROS) attack the lipid part of the cell membrane, and malondialdehyde (MDA) is a toxic byproduct released during lipoperoxidation<sup>5,9</sup>. Thus, an increase in MDA concentration in blood would be expected due to this  $\omega$ -3 susceptibility<sup>4</sup>.

On the other hand, regular consumption of fish can induce the accumulation of methylmercury in the body, causing neurotoxicity<sup>10</sup>. In addition, methylmercury causes an imbalance in antioxidant status due to increased reactive oxygen species and depletion of antioxidant molecules and enzymes, as demonstrated in some studies<sup>11,12</sup>.

However, there are still few studies on the regular consumption of fish in riverine populations, with no history of other forms of exposure to mercury, and its influence on serum levels of lipoperoxides. The aim of this work is to evaluate the relationship between frequency of fish consumption, levels of exposure to MeHg and lipid peroxidation marker in rivers.

## MATERIAL AND METHOD

A cross-sectional, retrospective cross-sectional study was carried out based on the review of the research forms of the resident riverside communities in the municipality of Limoeiro do Ajuru participating in the Project "Oxidative stress and antioxidant defenses related to mercury exposure due to fish consumption (approved financing through Process: 479624/2012-7 MCT/CNPq-n. 14/2012), attended by the research team of the Nucleus of Tropical Medicine (NMT) in on-site visits conducted in the 2014.

The municipality of Limoeiro do Ajuru is located in the Lower Tocantins Microregion, is one of the newest municipalities in the State of Pará. It has a total population of 25,021 inhabitants, of which 11,906 are women. The majority of the population inhabit in the rural area in near the river. In general, the population survives from catch and açai's extraction.

Were included the research forms of men and women riverside, permanent residence in the municipality for more than a

year and age between 13 and 86 years. Were excluded patients with acute illness and illicit drug use. The social and demographic variables included in this study were: age, sex, length of residence in the community, occupation, schooling, infectious and chronic degenerative diseases, exposure to environmental and occupational toxics. From this, 118 riparians were included in the study.

The frequency of weekly fish consumption was evaluated according to the classification adopted by Brune, Nordberg et al<sup>13</sup>: Category I, no fish consumption; Category II, <2 fish meals/week; Category III, 2-4 fish meals/week; Category IV, > 4 fish meals/week and Category V, unknown consumption.

The total mercury concentration (HgT) in each participant's hair sample was analyzed in the NMT Human and Environmental Toxicology Laboratory, where all the samples were processed for quantification of HgT by atomic absorption spectrophotometry with gold leaf amalgamation by hot steam technique using the Japanese-made Mercury Analyzer SP3D model<sup>14</sup>.

The MDA concentrations measured by blood plasma—and the methodology used for the quantification of Thiobarbituric Acid Reactive Substances (TBARS) in cell membranes was by Yagi<sup>15</sup>, with modifications made by Percario et al<sup>16</sup>.

The numerical data were recorded in spreadsheet in Microsoft Excel® 2010 software. For the comparison of numerical variables, the Kruskal Wallis test was used. The significant difference was considered,  $p < 0.05$ .

This study was approved by the Ethics Committee of the Nucleus of Tropical Medicine no. 334.523.

## RESULTS

The sociodemographic characterization of the population participating in the research, resident in Limoeiro do Ajuru, showed that 69.5% were represented by women. As to age, 78.8% are in the age group of 13 to 52 years, 72.1% self-declared brown, more than 50% presented low schooling, 43.3% were married, the predominant occupation was fishing and / or agriculture with 42.4%, and 50% presented monthly income less than one minimum wage (Table 1).

As for the frequency of weekly consumption of fish, 15.8% reported consumption less than two fish's meal per week, 36.6% consume between two and four fish's meal / week and about 47.7% consume more than four meals of fish / week.

The concentrations of HgTotal in category II are below 2  $\mu$ g/g, however in the categories where fish consumption is higher, mercury levels are above 2  $\mu$ g/g (6.6%), maximum values of 2, 46 and 6.27  $\mu$ g/g respectively in categories III and IV. There was a significant difference be-

**Table 1.** Sociodemographic characterization of study population, Limoeiro do Ajuru, Pará, Brazil.

	n	(%)
<b>Genre</b>		
Female	82	69.5
Male	36	30.5
<b>Age (years)</b>		
13  - 32	40	33.9
33  - 52	53	44.9
≥ 53	25	21.2
<b>Race</b>		
White	23	19.5
Brown	85	72.1
Black	9	7.6
No response	1	0.8
<b>Schooling*</b>		
No literate	4	3.4
≤ 9 years	70	59.3
> 9 years	44	37.3
<b>Civil status</b>		
Single	28	23.7
Married	51	43.3
Stable union	32	27.1
Divorced	1	0.8
Widower	6	5.1
<b>Occupation</b>		
Housewife	26	22.1
Fisherman/Farmer	50	42.4
Public server	14	11.8
Retired	11	9.3
Others	17	14.4
<b>Monthly income**</b>		
No income	8	6.8
< 1	59	50
1 to 3	50	42.4
3 to 5	1	0.8

Source: Research Protocol, Nucleus of Tropical Medicine.

\* ≤ 9 years: elemental school; high school and > 9 years: higher education. \*\*Minimum wage (R\$ 724.00).

tween categories II and IV, with higher fish intake/week ( $p=0.008$ ) (Tables 2 y 3).

The median levels of MDA according to the category of fish consumption were below the reference value for both men and women<sup>17</sup>. Although there was a tendency for MDA concentration to decrease as fish consumption increased, there was no statistically significant difference between the categories ( $p=0.1245$ ).

## DISCUSSION

The high consumption of fish in the region shows that this food contributes to the survival, subsistence and maintenance of the food culture, also to contribute to the energetic diet of the riverside inhabitants of the Amazon region, as demonstrated by several studies in the region, the importance of this food for the survival of many riverine families according to the eating habits and seasonal period<sup>2,18</sup>.

In addition, high fish intake may make these fish-dependent populations more susceptible to exposure to mercury, since this metal is not excreted from the organism<sup>19</sup>. Thus, carnivorous fish, with higher trophic level, have higher Hg concentrations in their tissue due to the capacity of biomagnification<sup>20,21</sup>. The Ordinance nº 685, of August 27, 1998, of the National Agency of Sanitary Surveillance (ANVISA)<sup>22</sup>, recommends the maximum limit of mercury in fish and fishery products (excluding predators) of 0,5 mg/kg and predator fish of 1,0 mg/kg.

Despite the high consumption of fish in the region, total mercury concentrations in hair samples from the population of Limoeiro do Ajuru are low, where only 6.6% of the studied population presented levels of mercury above 2 µg/g, considered a reference by the WHO to fish-consuming populations<sup>23</sup>. Of these, 57.1% are men, 50% are fishermen and consume more than two fish meals per week, suggesting that gender and occupation may also influence the concentration of mercury. The highest level (6.27 µg/g) was found in a female housewife who consumes more than four fish meals per week.

When comparing the concentrations of HgTotal according to the frequency of fish consumption/week, there was a significant difference between categories III (two to four meals) and IV (more than four meals), thus low levels exposure, and high fish consumption is influencing the bioaccumulation of mercury in this population. In the year 2013, in the Lima study, Santos et al<sup>2</sup>, the concentration of HgTotal in the rivers of Limoeiro do Ajuru ranged from 0.09 to 3.79 µg/g, and compared the levels according to the intake categories of fish, did not obtain significant statistical difference. This divergent result in a short period of time suggests that, deforestation in the Amazon has been increased and is an important vehicle for mercury exposure—which affects local survival and subsistence.

**Table 2.** Levels of HgTotal ( $\mu\text{g/g}$ ) in hair samples according to fish consumption frequency/week, Limoeiro do Ajuru, Pará, Brazil.

Fish consumption frequency/week	n (%)	HgTotal ( $\mu\text{g/g}$ )	p value
Category II: < 2 meals	16 (15.8)	0.63 (0.49 – 1)	0.008**
Category III: 2-4 meals	37 (36.6)	0.51 (0.49 – 0.87)	
Category IV: > 4 meals	48 (47.7)	0.88 (0.84 – 1.40)	

Source: Laboratory of Human and Environmental Toxicology, Nucleus of Tropical Medicine, UFPA. Data are represented in median and confidence interval. \*\*Kruskal-Wallis test.

**Table 3.** Levels of malondialdehyde plasma samples according to fish consumption frequency/week, Limoeiro do Ajuru, Pará, Brazil.

Fish consumption frequency/week	n (%)	MDA (nmol/mL)	p value
Category II: < 2 meals	11 (17.7)	0.54 (0.47 – 1.31)	0,1245
Category III: 2-4 meals	18 (29)	0.42 (0.32 – 0.84)	
Category IV: > 4 meals	33 (53.2)	0.31 (0.37 – 0.79)	

Source: Laboratory of Human and Environmental Toxicology, Nucleus of Tropical Medicine, UFPA. Data are represented in median and confidence interval. Kruskal-Wallis test.

In the case of Imperatriz, Maranhão<sup>3</sup> and Furo do Maracujá, Pará<sup>14</sup>, with respective mean values of 0.87 and 0.73  $\mu\text{g/g}$  of total mercury in hair. These results resemble this study, suggesting that these populations are considered control for studies involving mercury. However, other localities, such as the Tapajós River Basin region, high levels of mercury, are influenced by gold mines. Several studies in this region showed the high exposure to Hg<sup>24,20</sup>.

When comparing the levels of MDA in different categories of fish consumption, there was no statistically significant difference between the groups, and it was observed that the concentration of MDA decreased and fish consumption increased. In a study with animal models<sup>25</sup>, supplementation with 1: 1 and 1: 2 (EPA: DHA) fish oil evidenced a less oxidative environment and modulation in the production of pro-inflammatory biomarkers. A study conducted by Jamilian, Samini and cols<sup>26</sup>, observed that the group of pregnant women with gestational diabetes supplemented with omega-3s for 6 weeks had beneficial effects on maternal serum high-sensitivity CRP and MDA plasma levels, the incidence of hyperbilirubinemia in the newborn, as well as the hospitalization rate, when compared to placebo. Another study by Ateya, Sabri and cols<sup>27</sup>, found that in supplementing pediatric patients with end-stage renal disease on regular hemodialysis for 16 weeks, levels of endogenous antioxidants increased significantly, such as glutathione peroxidase and superoxide dismutase, however, there was no modulation of serum MDA concentration.

The sample size may be a limiting factor of the study, since studies conducted with a considerable sample size

have clearer and more accurate results. However, our study reported different from those already carried out with fish oil supplementation, the fish consumption. This fact may be relevant, since the population of this study high fish frequency consumption that among several nutrients is the  $\omega$ -3, with antioxidant capacity that modulates and prevents lipid peroxidation.

## CONCLUSION

The riverine population of the present study shows high weekly fish intake, which may further bioaccumulation of mercury in organism. However, the fish not appear to influence the serum levels of MDA, since is observed in the decrease concentrations of this marker of oxidative stress with the increase of fish consumption. Thus, more studies are needed to understand the role of polyunsaturated fatty acids as of regular fish consumption in riparian populations from Amazon.

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