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DETERMINATION OF SELENIUM LEVELS IN FOODS AND PLASMA IN A NORTHEAST REGION OF ALGERIA

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Abstract- Selenium (Se) is an essential nutrient of fundamental importance to human biology. Unfortunately until now very limited research has been conducted on selenium in Algeria. This study was conducted in order to quantify selenium levels in twenty foods and two hundred sixty six plasma collected in Souk Ahras (Northeast of Algeria). The samples had been analyzed using inductively coupled plasma-mass spectrometer (ICP-MS). The selenium concentration in food samples ranged from 22 to 1324 µg/kg. All plants alimentation have very low selenium levels than animals. The highest values were found in fish (sardine and tuna), then meats and eggs. However, cereals, fruits and vegetables contained the lowest levels. Whereas the selenium rate (mean ± SD) found in this population was (83.89 ± 21.61 µg/l). The mean value for 130 men was (85.75 ± 22.26 µg/l) and (82.08 ± 20.82 µg/l) for women but this difference was not statistically significant. Whereas, when samples were divided into age groups statistical analyses revealed a significant difference from thirty to fifty year and the difference is highly significant from ten to twenty year and very highly significant after fifty years. We also noted that the serum selenium rates increases from birth to 50 years and decrease after.

Keywords- Selenium (Se), plasma, human nutrition, inductively coupled plasma-mass spectrometer (ICP-MS), Keschan disease

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Introduction

Selenium (Se) is an essential trace element. The importance of selenium in animal nutrition has been known since 1957, but its implications for human biology and medicine have been clearly established in 1979, with the description of nutritional deficiency diseases. Its biochemical function is the formation of the active center of the enzyme glutathione peroxidase (GSH-Px) in mammals [1,2]. Besides this fundamental activity, selenium plays a role in the hepatic metabolism of xenobiotics and in the detoxification of heavy metals, and it contribute to maintain the integrity of skeletal and cardiac muscles and sperm. Several clinical and epidemiological studies show that the state of selenium deficiency, even marginal may cause muscular, hematological and neurological disorders and predisposes to cardiovascular disease and cancer [3,4].

Around the world, large variations exist in quantitative and qualitative selenium distribution in soils. These factors contribute to a variable selenium impregnation and therefore a variation of blood concentration in different countries or regions [5, 6]. Selenium levels in blood and tissues are very much influenced by dietary selenium intake [7-9]. Normal blood level varies from 0.05 to 0.34 µg/ml. In selenium deficient areas such us China, blood levels are as low as 0.009 µg/ml [10,11]. Therefore, it is necessary to monitor the selenium content in blood and representative consumed foods in each country [12] in order to determine if this region is seleniferous or not.

The aim of this investigation was the determination of selenium levels by ICP-MS in various samples from an agricultural region named Souk-Ahras situated in the north-east of Algeria. The latter is a North African country with a large area where each region has specific properties (climate, soil types and diet). Especially, that few data are available in the literature regarding the selenium status in general population, soil and foods in the northeast of Algeria except some preliminary research carried out [13-15].

Experimental Section

Study Area

The department of Souk-Ahras is situated in the northeast of Algeria located at 615 km from Algiers east, at 95 km south of Annaba and 165 km from the Constantine eas [16]. Its surface is 4.359,65 km², it is localized in a basin surrounded by of the Djbel Boussalah mountains [Fig-1] and it has a mediterranean climate. Its population is estimated at 438127 inhabitants [17].

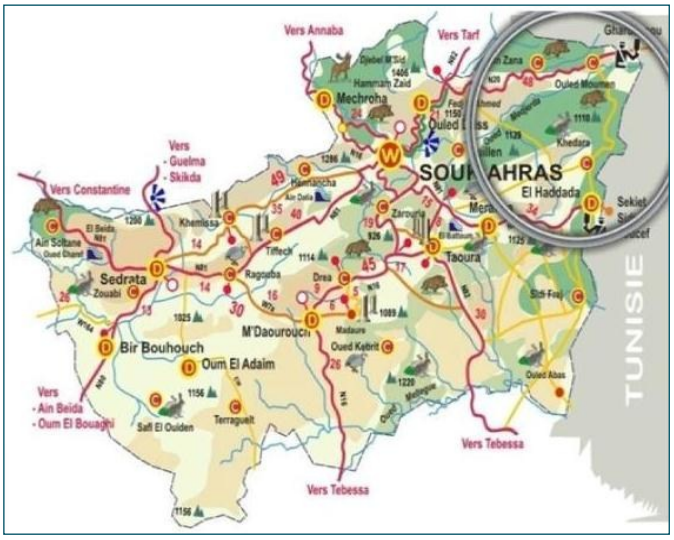

Fig. 1- Geographical map of Souk Ahras

Food Samples

Sampling

Food Samples

Foods were selected including those frequently consumed and those consumed in larger amounts in the region residents diet. Vegetables, cereals, were harvested in Souk-Ahras land while the rest of foods were purchased at shops. Three samples for each food item were purchased in 2010-2011. The twenty food samples were cleaned and dried at 65°C until constant weight than they were ground into a homogenous matter, aliquoted in 150 g amounts and stored in air-tight polyethylene bottles at -18°C until analysis.

Serum Samples

Serum samples were collected from 266 people in Souk Ahras (130 males and 136 females), ranging in age from few days to 70 years and they were in good general health at a regular medical examination. The collection of blood samples was carried out from diferent hospitals since December 2010 to November 2011, plasma was separated using the centrifugation without any addition of anticoagulants.

Digestion and Analysis

The plant samples were digested by acid (1g tissue and 10ml HNO_3 65% and 4 ml $HClO_4$ 72%), soaked for 48 hours and then digested for 30 min on a hotplate at temperature 80°C. Then the temperature is gradually increased for 20 minutes. After evaporation solutions are allowed to cool for 5 minutes and then 10 ml of HNO_3 were added to all tubes including witnesses. We puts the flask heating until complete evaporation of HNO_3 and white fumes appear. All samples were double-filtered using Whatman N°.42 filter paper and syringe regenerated cellulose filters the treatment ends when the medium became clear. After cooling, 2 ml of 10% HCl were added to reduce the Se^{-6} en Se^{+4}. Mixed and boiled for 5 minutes. Removed from the plate 3 ml of distilled water was added and after cooling we added also 10 ml of distilled water to each flask and the Erlenmeyer flask. Finally we added 10 ml of EDTA and a few milliliters NH_4OH so that the pH of the solution was 1-2. The solution can be kept in this state until analysis.

While animal samples were digested by acid (0.5 g of meat, 8 ml HNO_3, 5 ml HCL and 2 ml HF). Then we put the samples in the microwave at temperature 180°C for 45 min; after the solution must be filtered and than diluted with distilled deionized water to 50 ml.

Serum was separated into acid-washed tubes and, until analysis, stored at -20°C. A working standard solution (10 µg/ml) was prepared from a 1 mg/ml Se stock solution (SPEX CertiPrep®, Metuchen, NJ, USA). During analysis, all serum samples and working solutions were prepared by dilution (1:19, v/v) in an acidic solution (1% HNO_3, 0.01mM EDTA, 0.07% Triton X-100, 1.5% 1-butanol).

Selenium Determination

The selenium concentration in food digests were measured by the inductively coupled plasma-mass spectrometer (ICP-MS model 7500 from Agilent Technologies, Japan). Also, Yttrium (89Y) was used as the internal standard. The recovery for spiked samples with 5 and 10 µg Se/kg was more than 80% and the detection limit for Se was 0.2 µg/kg.

Statistical Analysis

The data were expressed as mean ± SD (standard deviation) and were subjected to statistical evaluation, using students - test. Statically significance was set on $P < 0.05$ (Minitab 13.0).

Results and Discussion

Serum Samples

The mean of the individual selenium levels in the studied population was 83.89 ± 21.61 µg/L. The range for all samples was 38.95-122.52 µg/L. As indicated in the [Table-1] the plasma selenium levels tended to be somewhat lower in females than in males but at a statistically non significant level ($P < 0.01$).

Table 1- Plasma selenium levels (µg/l) in healthy population of Souk Ahras (8d-70 y)

Sex	Number (n)	Mean ± SD	Range
M	130	85.75 ± 22.6	41.01-122.52
F	136	82.08 ± 20.82	38.95-121.83
All	266	83.89 ± 21.61	122.52-38.95

M : males; F : females; d : days; y : years; SD : standard deviation.

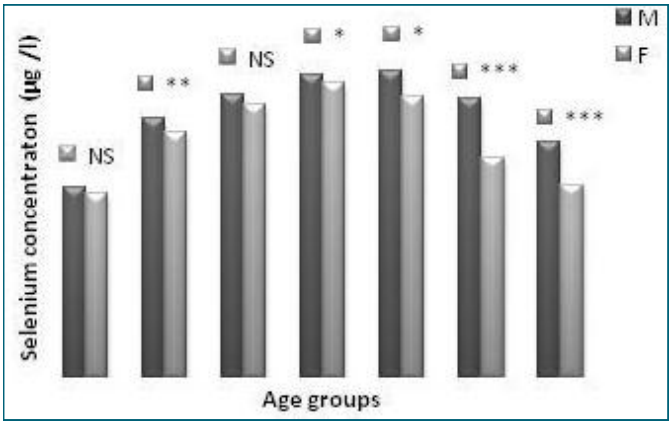

Fig. 2- Plasma selenium levels and age groups

NS: P > 0.05 statistically no significant difference;
** P < 0.05 statistically significant difference;*
*** P < 0.01 statistically highly significant difference;*
**** P < 0.001 statistically very highly significant difference*

As shown in [Fig-2], we can notice that the selenium rate increases from birth until fifty years old and decrease after. The statistical analysis showed that there is no significant difference ($P > 0.05$)

between males and females concerning the first and the third age groups (< 10 and [20-30]). However, there is a significant difference ($P < 0.05$ and $P > 0.01$) for the fourth and the fifth age ones ([30-40] and [40-50]). On the other hand, the difference is highly significant ($P < 0.001$) for the second group ([10-20]) and very highly significant ($P < 0.001$) within the sixth and seventh age groups ([50-60] and < 80).

Food Samples

The mean values of selenium concentration in different foods samples analyzed in this study are reported in the [Table-2].

Table 2- Selenium levels in analyzed food samples in Souk Ahras-Algeria (µg / kg).

Food	Mean Selenium Content (µg/kg)
onion	33 ± 2
garlic	53 ± 1
carrot	29 ± 1
zucchini	33.7 ± 1.1
Potato	43.7 ± 1.5
Haricot bean	35 ± 1
lentils	61.3 ± 0.6
Tomato	23 ± 1
Peach	28.3 ± 0.6
Orange	23 ± 1
Fig	37 ± 1
Wheat	63.3 ± 1.5
Barley	53.3 ± 1.5
Cow milk	73.4 ± 1.6
sheep	179.3 ± 0.6
beef	293 ± 1
chicken	352.7 ± 1.5
Chicken eggs	282.4 ± 1.6
Sardine	1079.5 ± 0.6
Touna	1321.6 ± 2.1

The selenium concentration ranged from 22 to 1324 µg/kg. All plants alimentation have very low selenium levels than animals. The highest values were found in fish (sardine and tuna), then meats and eggs. However, cereals, fruits and vegetables contained low levels of selenium.

The highest level of selenium in legumes was obtained in lentils followed respectively by garlic, potato, haricot bean, onion and zucchini. However the highest one in fruits was obtained in fig, peach, orange and also tomato.

Therefore, in cereals samples the highest selenium level was obtained in wheat than barely. However, cow's milk has a higher value compared to the plants alimentation but lower than the animal one.

The mean serum selenium levels obtained in this study was 83.89 ± 21.6 µg/l. This rate [Table-3] was similar to the results obtained in the northwest of Algeria [14] and Turkey [16] but lower than those observed in some population from seleniferous regions such as Saudi Arabia [17], Nigeria [18], India [20] and Belgium [21]. However, it is higher than others data observed in people from seleno-deficient areas such as Czech Republic [26], Poland [25], Austria [26], Egypt [27] and China [28].

No person had plasma selenium values lower than 45µg/L, the level considered to reflect inadequate selenium status [28] and this confirmed that the Algerian populations don't risk a deficit of selenium.

The selenium content in plasma of males does not differ significantly ($P > 0.05$) from females and this was also observed in the blood of Tlemcenian city [14], Austrian [29], Spanish [30], French [31] and Greek populations [32]. While Kafai and Ganji [33] have observed that selenium concentrations were significantly lower in women than in men.

Table 3- Reported serum selenium levels in different countries

Country	Overall mean (µg/L)	References
This study	83.89	
Algeria(northwest)	85.65	[14]
Saudi Arabi(Riyadh)	100	[17]
Spain(granada)	74.9	[21-23]
Czech Republic	67.5	[24]
Nigeria	119	[18]
Poland	54.8	[25]
Turkey (various regions)	81.5	[16]
Austria	75	[26]
India (Mumbai)	100	[19]
Belgium (antwerp region)	90	[20]
Egypt	68	[25]
China (Keschan disease areas)	21-24	[27]

The sex difference noticed with age groups is in agreement with the results of various studies carried out in some countries such as Italy [34]. This difference can be due to hormonal status, food habits, economic class and activities [35].

Selenium levels in most fresh fruits are rather low [Table-4]. This findings were in accordance with other reports in Table 4 such as the west-north of Algeria and lower than those obtained in Saoudi Arabia but higher than those carried in China (suzhou). The selenium values were anticipated because of the high water fraction and low protein fraction in these products. Food selenium is mainly present in the form of selenoamino-acids [32]. Similar to fruits, most vegetables are poor sources of selenium and this is in agreement with those finding by other workers [Table-4]. However, animal foods which are the richer protein products have higher selenium levels.

Table 4- Selenium levels in foods from various countries

Foods	This study	Algeria (northwest)	Saudi Arabia	Saudi Arabia	China
onions	33 ± 2	16 ± 0.4	24 ± 0.04	43 ± 0.0022	1.86 ± 1.17
garlic	53 ± 1.02	/	9 ± 0.02	/	/
carrot	29 ± 1.04	/	190 ± 1.07	2 ± 0.0004	/
zucchini	33.7 ± 1.1	/	21 ± 0.37		/
Potatoes	43.7 ± 1.5	5 ± 0.2	51 ± 0.09	1 ± 0.0003	6.18 ± 2.26
Haricot bean	35 ± 1.01	27 ± 2	/	/	/
lentils	61.3 ± 0.6	672 ± 26	/-	76 ± 0.0043	/
Tomato	23 ± 1	/	1.8 ± 3.07	/	1.54 ± 1.06
Peach	28.3 ± 0.6	/	/	/	/
Orange	23 ± 1.03	/	/	28 ± 0.0017	/
Figs	37 ± 1.08	/	/	32 ± 0.0009	/
Wheat	63.3 ± 1.5	65 ± 3	/	165 ± 0.0012	/
Barley	53.3 ± 1.5	/	/	69 ± 0.0029	/
Cow milk	73.4 ± 1.6	/	/	/	15 ± 2.78
sheep	179.3 ± 0.6	/	/	/	/
beef	293 ± 1.2	/	/	390 ± 0.0081	89.6 ± 14.9
chicken	352.7 ± 1.5	/	250 ± 0.33	353 ± 0.0074	111.7 ± 32.0
Chicken eggs	282.4 ± 1.6	402 ± 21	260 ± 0.30	226 ± 0.0035	151.7 ± 48.7
Sardine	1079.5 ± 0.6	1033 ± 25	/	658 ± 0.0063	/
Touna	1321.6 ± 2.1	/	290 ± 0.72	/	/

The average of selenium level in cereals was 58.3µg / kg and this findings were in accordance with other reports such us Khemis and Oum el bouaki in Algeria [15] but lower than those obtained in Tiaret, Saida, Guelma and Setif in north- west and of Algeria and lower than Khroub one. We note also that our study value was lower than those obtained in other countries such as Saoudi Arabia [17 -36] but higher than those carried in China [37].

The highest selenium levels were found in sardine's samples, this rate is very important compared to the other foods. Generally, sardine contains an amount five times greater than beef [38].

Conclusions

The results of our study indicate that the selenium levels of healthy eastern-Algerian population tended to be lower than a lot of word population but similar to the north-west Algerian population level. We also noted that selenium rates increase from childhood until 50 years and decrease after. The difference between males and females was statistically significant in particular age groups. Therefore the Algerian population in Souk-Ahras city doesn't risk a deficit of selenium. Also this study showed that fish, meat and eggs contained the highest selenium concentration; cereals and milk were on average levels whereas fruits and vegetables had the lowest concentration. Therefore, meat, fish and eggs contribute the major part of daily dietary selenium. However, cereals, milk, fruits and vegetables are not negligible sources because high amounts of them are consumed daily. Moreover, it should be noted that good selenium sources cannot be identified only from high selenium concentration, but bioavailability of selenium in foods must also be considered.

Owing to the limitation of resources, the data reported here derive from the analysis of samples purchased in the northwest part of Algeria (Souk-Ahras). To establish more representative data, samples from different parts of Algeria should be collected. Furthermore, the selenium content of infant milk, wide variety of foods and readily cooked dishes that are widely consumed in different regions should be determined so that we can calculate the selenium daily intake.

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