Pure Appl. Chem., Vol. 84, No. 2, pp. 181–190, 2012.
http://dx.doi.org/10.1351/PAC-CON-11-07-22

Concentrations of toxic and essential elements in Lebanese bread*

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Abstract: Concentrations of 20 minor, trace, and ultratrace elements relevant to human health (Ag, Al, As, Cd, Co, Cr, Cu, Fe, Hg, Li, Mn, Mo, Ni, Pb, Sb, Se, Si, Sn, V, and Zn) were determined in three varieties of Lebanese bread (white and brown double loaf bread, Saj bread) sampled at five geographical regions (Grand Beirut, South of Lebanon, North of Lebanon, Mount of Lebanon, and Beka'a) during the wet and dry seasons. The analyses were carried out by double-focusing sector-field inductively coupled plasma-mass spectrometry (DF-SF-ICP-MS). The data were discussed in terms of nutritional elements supplied by type of bread, the risk of contamination by toxic elements (As, Cd, Pb) owing to the manufacturing process, and variations related to geographic distribution and seasonal sampling.

Keywords: analysis; bread; Lebanon; sector-field inductively coupled plasma-mass spectrometry (SF-ICP-MS); MS trace elements.

INTRODUCTION

For ages, bread has been a popular staple food. Its nearly ubiquitous consumption makes it of paramount importance in international nutrition. Bread and cereals contribute considerably to the daily diet of the Lebanese population. According to the total diet study (TDS) for the Lebanese population made in 2010, an average quantity of 136.8 g of bread was consumed daily [1].

Ideally, bread should have a low glycemic index, be an important source of proteins and contain tolerable dietary fibers, vitamins, trace elements and antioxidants [2]. The elemental composition of bread is reflected by the composition of wheat flours and different substances added during its processing, such as, e.g., water, yeast, and salt, and is affected by the baking conditions or containers [3,4].

Pure Appl. Chem.* **84, 169–333 (2012). A collection of invited papers based on presentations at the 4th International IUPAC Symposium on Trace Elements in Food (TEF-4), Aberdeen, UK, 19–22 June 2011.

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Wheat, rye, barley, and oats are classified as a moderate source of Ca and Mg (100–200 mg/100 g), Fe and Zn (1–5 mg/100 g), or Cu (0.1–1 mg/100 g) in bread [4]. Cereals and wheat bread can also be rich in phytic acids knowm for their antinutritional effects in human diet owing to chelating of bivalent cations, such as Ca, Fe, Mg, and Zn [4–7]. Multi-element analysis of bread and other food products is of a particular interest when concerning nutritional value of these products, evaluation of nutrients which bioavailability might increase with their increased concentrations as is in case of Fe, Mn, Se, and Zn [3] and determination of any possible contaminating toxic elements. Even though some minerals are known as nutrients, they could induce intoxication in the case of an overexposure [8].

The elemental content of bread and cereal products and contribution of these products to total daily intakes of different elements were extensively studied in Spain [8–12]. Information on diets of other Mediterranean countries where bread is a staple food is missing. The objective of this study was to determine the element composition of three types of the most popular Lebanese bread, including normal white Arabic bread, brown or whole wheat bread, and Saj (typical Mediterranean type of bread), and to discuss the data in terms of bread geographical origin and seasonal variations.

EXPERIMENTAL

Sampling

Three types of bread popular in the Middle East were selected for this study considering their market share and popularity. They included "normal white Arabic bread" (Br1) made of white flour (usually a loaf of two layers), "brown or whole wheat bread" (Br2) made of whole wheat grain flour and labeled with different constituents (loaf with two layers), and "Saj bread" (Br3) mostly made of brown and whole wheat flour. The types of bread were selected on the basis of its high consumption by the population of Lebanon while available brands were collected arbitrarily at sampling sites. Samples were collected at the most frequented markets of 10 cities with the highest population densities unequivocally distributed to 5 geographical Lebanese departments, i.e., Grand Beirut (GB1, GB2), North of Lebanon (NL1, NL2), Mount of Lebanon (ML1, ML2), Beka'a region (Bk1, Bk2), and South of Lebanon (SL1, SL2). Samples were collected during two seasons, i.e., dry (on August 2010) and wet on (January 2011). Breads were bought as packed in nylon bags with 10 portions of each kind. A part from each portion was cut and oven-dried at 50 °C overnight in polyethylene containers. Dried samples were ground to a fine homogenous powder using an acid-washed mortar and pestle.

Wet acid digestion and analysis

Concentrations of 20 elements in analyzed bread samples, i.e., Ag, Al, As, Cd, Co, Cr, Cu, Fe, Hg, Li, Mo, Mn, Ni, Pb, Se, Si, Sb, Sn, V, and Zn, were determined according to the analytical procedure described in detail elsewhere [13]. In brief, a 0.25-sample was hot-plate digested at 85 °C with a mixture of conc. HNO_3 and H_2O_2 (10:3) until a transparent solution was obtained. The solution was analyzed by double-focusing sector-field inductively coupled plasma-mass spectrometry (DF-SF-ICP-MS). Isotopes of 20 elements were measured by DF-SF-ICP-MS in low-resolution (300) mode: ^{7}Li, ^{95}Mo, ^{107}Ag, ^{111}Cd, ^{118}Sn, ^{121}Sb, ^{202}Hg, ^{208}Pb; medium-resolution (4000) mode: ^{27}Al, ^{28}Si, ^{51}V, ^{52}Cr, ^{55}Mn, ^{56}Fe, ^{59}Co, ^{60}Ni, ^{63}Cu, ^{66}Zn; and high-resolution (10 000) mode: ^{75}As, ^{78}Se. De-ionized water (18.3 MΩ cm) and Instra-analyzed 69–70 % (m/m) HNO_3 and 30 % (m/m) H_2O_2 solutions (Baker, Deventer, The Netherlands) were used throughout to minimize the blanks.

All polyethylene tubes for sample preparation were thoroughly washed and soaked overnight in a 1 % (*m*/*v*) HNO_3 solution. Then, they were thoroughly rinsed with de-ionized water. Quantification of elements in sample solutions was made using matrix-matched standard solutions with 10 calibration points. Two independent replicates were made for each type of sample, and respective procedural blanks, carried out in the same way as samples, were run and considered in final results. Wheat flour SRM 1567a from NIST and brown bread BCR 191 from IRMM were analyzed in duplicate at each sequence of samples for the purpose of quality control and assurance. The data for the analysis of reference materials proving the method's accuracy are summarized in Table S1 (Supplementary Information).

Statistical evaluation of data

Data were analyzed by the Statistical Package for Social Sciences (SPSS) for Windows (version 17). Student *t*-test and one-way analysis of variance (ANOVA) were used to determine statistically significant levels for differences in the mean values of measured elements among subjects grouped by region and season. A probability level of 0.05 was considered to be statistically significant.

RESULTS

The determined concentrations of trace elements in 60 bread samples are shown in Fig. S1 (Supplementary Information). Figure 1 shows, for each element, the mean, median, and concentration distribution in the samples sampled during the wet and dry seasons. The data were compared with those reported in the literature. Note that for many elements our data are the first reported in bread.

DISCUSSION

For the purpose of further discussion, two groups of the determined elements were considered: one referred to as toxic: Ag, Al, As, Cd, Hg, Pb, Sb, and Sn, and the other including components of enzymes and considered as essential: Co, Cr, Cu, Fe, Li, Mn, Mo, Ni, Se, Si, V, and Zn. Note that at high concentrations or when present at particular oxidation states, e.g., Cr(VI) or Se(VI), these elements can show toxic effects. First, the total element concentrations found will be discussed in the context of the literature for bread samples from other countries, and then the variations observed between the different bread varieties. Seasonal variations and the effect of sampling location in Lebanon will also be briefly discussed.

In order to discuss differences in concentrations as a function of element, season, and geographical site, statistical analysis of the obtained data was carried out using *t*-test for two independent samples at the 95 % confidence limit ($p < 0.05$) for the set of results from two studied seasons [14,15]. Kolmogorov–Smirnov and Mann–Witney tests by rank for independent samples were used for elements which do not fulfill the normal distribution criteria [16]. The ANOVA test at the 95 % confidence limit was used to indicate any significant differences in geographical sites within the same season as all elements showed normal distribution in each geographical site for each studied season [14,15]. The normality of elements was examined by the Kolmogorov–Smirnov test.

Concentrations of toxic elements

The mean concentrations of toxic elements for all the analyzed samples follow the sequence: Al > Sb > Pb > As > Cd > Ag > Sn > Hg.

Aluminum

The mean concentration level of Al in bread originated from the wet season was found to be 11.58 ± 0.66 mg/kg (the range of 6.32–32.25 mg/kg). In case of the dry season, a lower mean value by about 50 % was determined in all bread samples, i.e., 5.88 ± 0.27 mg/kg (the range within 2.37–11.59 mg/kg). Breads Br2 and Br3 from region SL2 were established to have the highest concentrations of Al; they

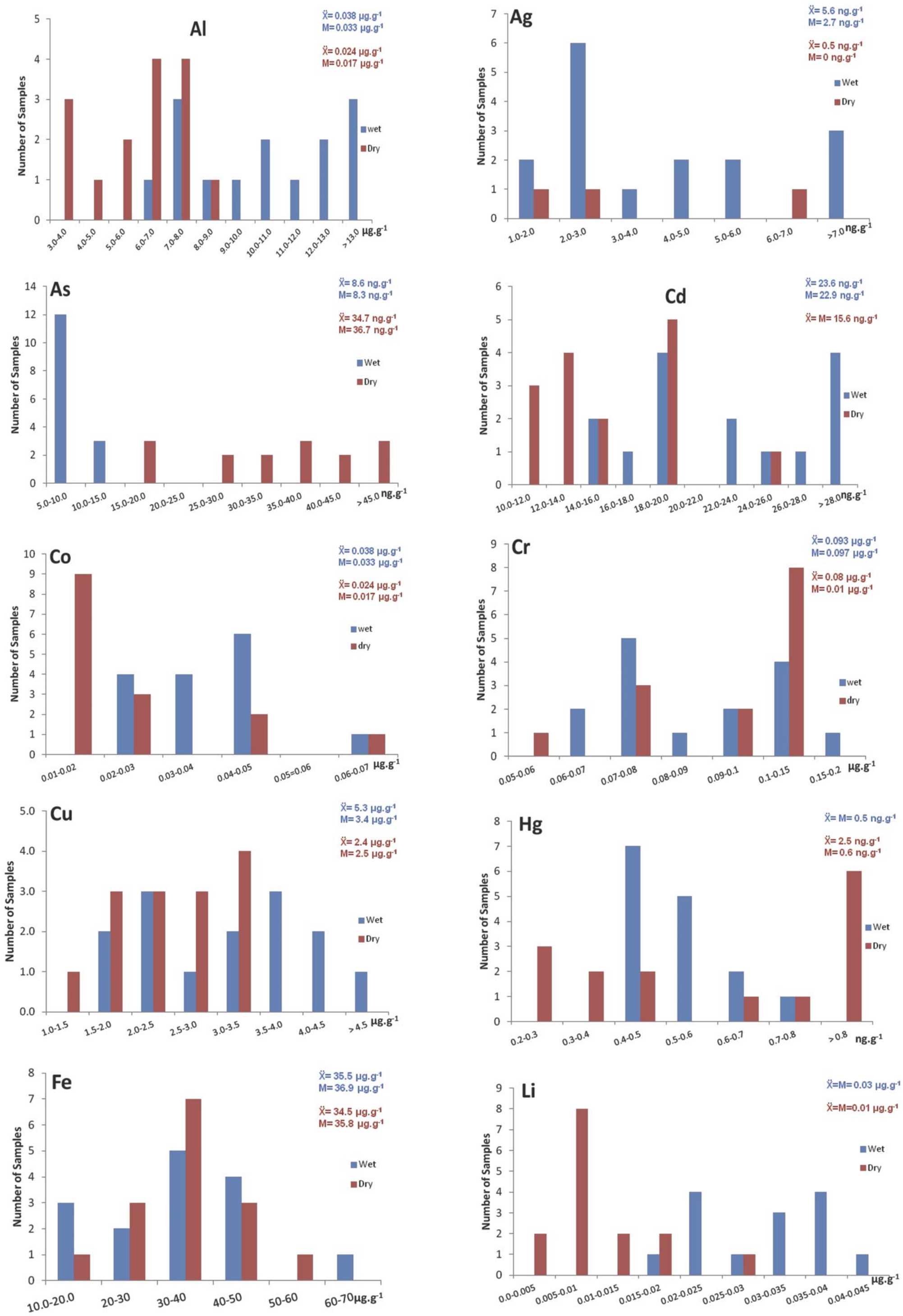

Fig. 1 Distribution of sample concentrations comparing to mean value (X) and median value (M) from each season (wet and dry) with a reference to the interval of results previously reported in literature.

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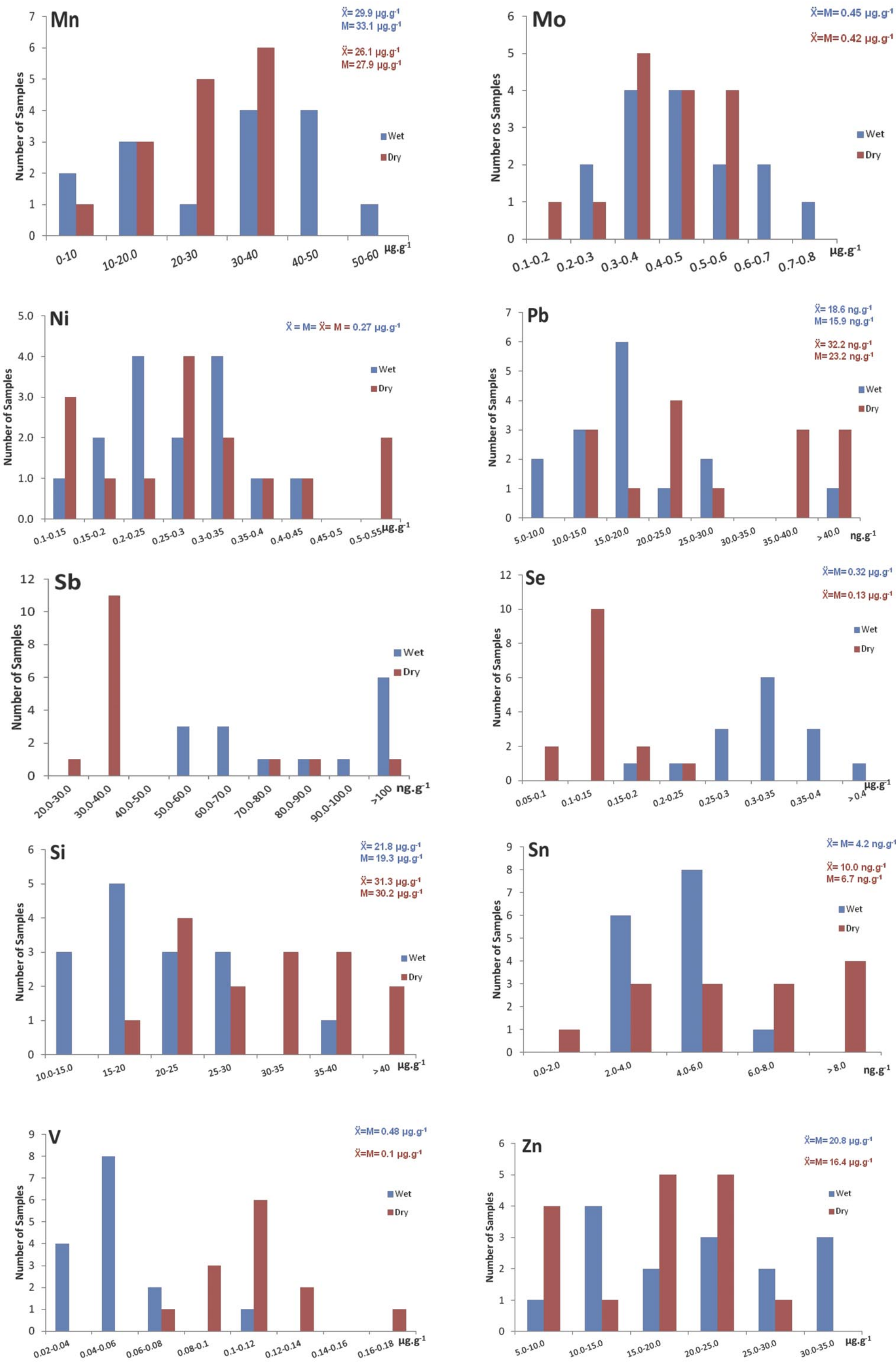

Fig. 1 (*Continued*).

contained 14.5 mg/kg of Al in the wet season and 11.50 mg/kg of Al in the dry season. Only three samples showed much higher concentrations in the wet season, i.e., Br3 from NL2 region (32.25 mg/kg) and Br2 (25.25 mg/kg) and Br3 (15.26 mg/kg) from ML2 region. A high level of Al could be attributed to additives of bread or to contamination originating from contact with materials of containers at the production and distribution stage. Reported results for Al in this study are similar to those reported in Pakistan bread (14.6 ± 3.9 mg/kg for nonbrand bread and 4.8 ± 2.0 mg/kg for brand bread) [17].

Arsenic

As had a mean concentration in bread from the wet season of 9 ± 2 µg/kg while the respective value for the dry season was much higher, that is, 33 ± 4 µg/kg. Less than 18 µg/kg was reported in Chilean bread [18].

Antimony

The mean concentration of Sb in bread from the wet season was 136 µg/kg and 63.64 µg/kg for the dry season. Samples Br2 from GB2 region in addition to Br2 and Br3 from NL2 region showed values higher than 100 µg/kg in both seasons. High concentrations of Sb were also found in different types of bread from ML1 and ML2 regions.

Cadmium

The mean concentrations of Cd in bread samples collected in the wet and dry seasons were 24 and 15 µg/kg, respectively. Results for Cd concentrations in bread samples obtained in this study are in agreement with mean concentrations of this element (15 µg/kg) reported for bread and cereals in the TDS report from 2010 [1] and with results reported by Cuadrado [10]. Concentrations of Cd determined in bakery products from Turkey were also similar (7–33 µg/kg, dry weight) [8]. A lower concentration of Cd (<2 µg/kg) was reported in Chilean bread [18].

Lead

Pb mean concentrations were 18 and 28 µg/kg for bread collected in the wet and dry seasons, respectively. The results of Pb in three samples were exceptionally higher than in other samples, i.e., samples Br2 from ML1, Br3 from GB2 and Br3 from Bk from the dry season, concentrations of Pb of 100, 74, and 62 µg/kg, respectively, were found; in the wet season in sample Br3 from NL(2) the concentration of 66 µg/kg was measured. The most probable source of contamination with Pb in this case could be wheat flour and tap water used in bread production [8]. A study on the contribution of cereals to the total dietary intake of heavy metals made by Cuadrado [10], indicates that mean concentrations of Pb in white bread were 36 µg/kg (dry weight) and 59 µg/kg (dry weight) in whole wheat bread. Much higher concentrations of Pb in bread from Turkey were also found and reported, i.e., 23–1819 µg/kg (dry weight) [8]. Similar high concentration of Pb (136 ng/g) in Chilean bread was reported [18].

Mercury, silver, and tin

Mean concentrations of Hg in bread from the wet season were 0.5 µg/kg. Concentrations in samples from the dry season were below the limit of detection. Ag mean concentrations were between 2 and 6 µg/kg. Similar concentrations were also measured for Sn. These are, to our knowledge, the first data reported for these elements in bread.

Concentrations of essential elements

The mean concentrations of essential elements for all the analyzed samples follow the sequence: Li < Cr < Co < V < Ni < Se < Mo < Cu < Zn < Si < Mn < Fe.

Copper

The concentration of Cu in all analyzed bread samples was within 1.7–6.0 mg/kg. The mean value of Cu in whole wheat (Br2) and Saj (Br3) breads showed higher concentrations (2.7–4.0 mg/kg) than white bread (Br1) (1.6–2.2 mg/kg). The concentration of Cu in white bread was close to the mean value of Cu for bread and cereals category from the TDS made in Lebanon in 2010 [1] and to those reported in Spain [10] and Turkey [19]. However, another study from Turkey reported a mean concentration of Cu in bakeries of 22.1 ± 1.0 mg/kg of dry weight [8].

Zinc

The mean concentration of Zn in bread from the wet season was assessed to be 20.7 ± 0.76 mg/kg (ranged between 8.2 and 40.9 mg/kg, respectively). A slightly lower mean concentration of 15.35 mg/kg was found in samples originated from the dry season ranged between 7.5 to 29.9 mg/kg. The analysis of breads demonstrates an important nutritional value especially of whole wheat bread (Br2), which can contain on average 25.5 and 19 mg/kg, respectively, for the wet and dry seasons. The corresponding concentrations found for white bread (Br1) were 11.4 and 8.58 mg/kg. The concentration of Zn reported in breads and cereals from the TDS made in Lebanon in 2010 was 9.48 mg/kg [1]. Our results are in agreement with those reported for cereal products in Spain: 9.2 mg/kg (dried bread) and 18.4 mg/kg (the whole bread) [10]. The Zn level in bakeries from Turkey was reported to be 10.0 ± 3.0 mg/kg (dry weight) [8].

Selenium

Wheat has a capacity to accumulate high amounts of Se and is considered a good source of this element [20]. A mean concentration of 0.32 ± 0.08 mg/kg of Se was assessed in bread samples collected during the wet season and was three times lower in the dry season: 0.13 ± 0.03 mg/kg. Because the content of Se in foodstuffs is related to their protein percentage, wheat contains more Se than cereals, legumes, or dried fruits [9]. Interestingly, our values are about one [10] to two [9] orders of magnitude higher than those reported elsewhere.

Cobalt

No difference was observed in Co content between the types of bread and seasons (0.02–0.04 mg/kg). These concentrations are ca. five times higher than those reported in the TDS [1].

Iron

Concentrations of Fe in bread samples ranged from 15.7 to 59.4 mg/kg (a mean value of 33.9 mg/kg). Whole wheat and Saj (Br2 and Br3) breads were established to have a higher mean concentration (40.4 mg/kg) than white bread (20.4 mg/kg) for both seasons. Only one sample, Br3 from SL2 region, showed a higher concentration of 88 ± 7.7 mg/kg, likely due to contamination from containers during bread processing. The reported value of Fe from TDS was 15.04 mg/kg [1]. Tokalioglu and Gurbuz [19] reported mean concentration values of Fe in some Turkish cereals to be 19.5 ± 0.25 mg/kg in wheat and 12.5 ± 0.8 mg/kg in flour. Another study from Turkey reported a mean concentration of Fe as 18.2 ± 8.1 mg/kg (dry weight) in different bakery products [8].

Manganese

The mean concentration of Mn in bread samples was 26.6 ± 0.8 mg/kg (the range within 7.5–58.7 mg/kg) and showed no seasonal variation. A remarkable difference was established to be between bread samples made from whole grain or whole wheat (Br2 and Br3) for (31–39 mg/kg) and those in white bread (Br1) (10.75 mg/kg). The mean concentration of Mn for bread and cereals reported in the TDS made in Lebanon in 2010 was 10 times lower (3 mg/kg) [1]. A similar study of Spanish cereals showed a mean concentration of 6.7 mg/kg of Mn in white bread and 17.1 mg/kg in the whole wheat bread (dry weight) [10].

Chromium

Dietary intake constitutes the main contribution of Cr to the human organism. Concentrations of Cr determined in bread samples were at the 0.1 mg/kg level. Diets containing food products made from whole wheat and brown sugar contribute to relatively high Cr intakes. In addition, technology used in food processing can increase natural levels of Cr in raw products [21]. The content of Cr in several types of breakfast cereals in Spain was found to contribute on average about 0.23 mg/kg of Cr (the range within 0.09–0.55 mg/kg) with a large variability of concentrations between different samples, likely due to highly variable sample composition, the origin of raw materials, and methods of processing [11]. Knowing that the estimated safe and required dietary intake (ESADI) of Cr is about 50–200 μg [11], bread analyzed here contributes 9.8 % to the total daily intake.

Nickel

Concentrations of Ni in bread samples collected in both seasons were similar and ranged from 0.1 to 0.51 mg/kg with mean of 0.27 ± 0.01 mg/kg. Only in the case of brown or whole wheat bread (Br2) from NL2 region collected in the dry season, the concentration of Ni had a higher value: 0.77 mg/kg. Nasreddine [1] reported the mean concentration of Ni in bread and cereals of 0.17 mg/kg. Mean concentrations of Ni in white bread samples from Spain were 0.15 mg/kg (dry weight) and 0.13 mg/kg (dry weight) in the whole wheat bread [10].

Molybdenum

Mo, being an essential element, was found to be at almost the same level of 0.44 ± 0.02 mg/kg in all bread samples analyzed here. Concentrations of this element in breads taken from 2 seasons were homogeneously distributed between 0.13–0.84 mg/kg. The average concentrations of Mo in white bread from Spanish were 0.21 mg/kg (dry weight) and 0.26 mg/kg (the whole wheat bread) [10].

Vanadium, lithium, and silicium

Mean concentrations of V and Li in bread samples showed a difference between two seasons. Li had a higher concentration during the wet season, while V showed the opposite trend. In fact, the mean concentration of V in bread from the wet season was 0.05 ± 0.004 mg/kg, while in the dry season it was 0.1 ± 0.005 mg/kg. No difference due to the type of bread was observed. Regarding Li, respective mean concentrations of this element in bread collected in the wet and dry seasons were 0.03 ± 0.005 and 0.01 ± 0.001 mg/kg. In the case of Si, it was found that higher concentrations of this element were observed in bread collected in dry season. Accordingly, mean concentrations of Si in bread samples were 21.83 ± 1.36 and 29.97 ± 1.58 mg/kg, in the wet and dry seasons, respectively. In addition, it was found that concentrations of Si showed a higher variability between samples of bread from the dry season; concentrations of Si in samples of bread from the wet season were more consistent. No similar study for Li, V, and Si was reported before in literature.

Effect bread type, seasonal and regional variations

Bread type

This study shows that nutrients, such as Cu, Zn, Mn, Mo, Fe, Co, and Se are present in brown and whole wheat grain flour-based bread (Br2 and Br3) in much higher concentrations than in white flour-based bread (Br1). Indeed, brown and whole wheat flour are products of whole grains, which contain higher levels of trace elements in comparison with white flour owing to the presence of the outer kernel layers where minerals are concentrated [20,22]. This was confirmed by a study on buckwheat, the bran of which has a higher concentration of Se, Zn, Co, Ni, and Ag than the flour part [23].

Seasonal variations

Statistical data analysis for detection of any potential difference in elemental composition due to seasonal variation showed that results for most elements during both seasons are normally distributed except for Ag and Sb. The mean concentration of Al, Co, Cu, Li, Si, Se, V, Zn, As, Cd, Ag, and Sb showed a significant difference between the two seasons. Al, Co, Cu, Li, Se, Zn, and Cd showed higher mean concentration values during the wet season. Si, V, and As had a lower mean concentration values during the wet season. Mn, Fe, Cr, Mo, and Ni showed a similar mean concentration between samples collected from both seasons.

The seasonal variation of elemental concentration in bread is mainly a factor of the seasonal variation of the composition of wheat grain and water as main ingredients of bread. Cd, As, Hg, Pb, and Cr are considered as the most toxic contaminants having their source from the agriculture soil. Thus, a variation in soil composition between different periods of year is reflected in the crops. The seasonal variability was discussed by Matos-Reyes [12], who observed a wide variation in results for As, Sb, Se, Te, and Bi in vegetables and cereals without a common trend.

Regional variations

The data of each element and each geographical region within one season showed a normal distribution. The ANOVA test showed that no significant changes between mean concentrations of almost all the elements as a function of the region. The notable exception was the high concentrations of As and Hg observed in the Beirut region. Also, bread from North Lebanon showed much higher concentrations of Al, Fe, Cu, Cr, Ni, Pb, Si, and Zn as compared to bread from other regions, possibly due to some kind of contamination. This contamination of bread may have been derived from production, e.g., old metal containers used during bread processing. In addition, other samples show rather nonsystematic high concentration values that cannot easily be correlated to a specific geographical zone.

The regional variation on elemental composition of bread is due not only to the impact of manufacturing process (contamination by and/or loss of certain elements), but it is also reflected by the composition of the main ingredients of bread and notably the wheat grains. A previous study on the variation of elemental concentration (Na, K, Mn, Cu, Fe, Zn, and Se) from different cultivars in the Canary Islands showed a significant difference in the concentration of these elements, which might be associated with the genotype species and the environmental effects (type of soil and water composition in this area) [20]. This was emphasized by Oury [22] about the environmental effect on Mg, Zn, and Fe concentration but not for genotype factors on Fe concentration. Demirozut [8] reported a difference in element concentration in flour for bakery products related to the source of flour (mainly for Pb, Zn, and Fe but not for Cd and Cu).

CONCLUSIONS

The study allowed us to state that the concentrations of the most notorious essential and toxic elements in Lebanese bread were similar to those reported elsewhere, and allowed, for the first time, concentrations of ultratrace elements such as Ag, Co, Hg, Li, Sn, Sb, and V to be determined. The daily intake of elements from bread in Lebanon was: 0.32 mg/day for Cu, 4.4 mg/day for Fe, 3.3 mg/day for Mn, 2.1 mg/day for Zn, and 0.18 mg/day for Se, which accounts for 36, 25, 30, 22, and 32 % of respective RDA values given for these elements. The contribution of these nutrients from wheat flour was similar to that reported in the Canary Islands [20]. The daily intake of toxic elements was determined to be 4.5 μg/day in the case of As, 2.1 μg/day in the case of Cd, and 3.8 μg/day in the case of Pb comparing to their corresponding PMTI value (0.002 mg/day for ingested As, 260 μg/day for Pb, 72.8 μg/day for Cd) [18]. All of the toxic element values are below the threshold limits set by regulatory agencies.

SUPPLEMENTARY INFORMATION

Supplementary Information is available online (http://dx.doi.org/10.1351/PAC-CON-11-07-22).

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