Available Online at http://journalijcar.org

International Journal of Current Advanced Research
Vol 4, Issue 8, pp 278-284, August 2015

ISSN: 2319-6505

International Journal of Current Advanced Research

ISSN: 2319 - 6475

RESEARCH ARTICLE

IN VITRO BIOAVAILABILITY OF IRON FROM SOME NON CONVENTIONAL LEAFY VEGETABLES CONSUMED IN CAMEROON

Djuikwo, NRV[1*], Ejoh AR[2], Mbofung CM[3], Tanya, NA[4] and Gouado I[5]

[1]Department of Biochemistry, Faculty of Science, University of Yaounde I, Cameroun
[2,3,4]Department of Food Science and Nutrition, University of Bamenda, Cameroon
[5]Department of Biochemistry, Faculty of Science, University of Douala, Cameroun

A R T I C L E I N F O

Article History:

Received 22th, July, 2015
Received in revised form 31th, July, 2015
Accepted 20th, August, 2015
Published online 28th, August, 2015

Key words:

Malnutrition, Leafy-vegetables, Dishes, Iron, Bioavailability.

A B S T R A C T

To determine the bioavailability of iron in leafy vegetable dishes made of *S. macrocarpon, H. sabdariffa thin morphotype, H. sabdariffa* white morphotype, *C. maxima* spotted morphotype, *C. maxima, C. sesamoides, C. olitorius, A. hybridus, V. calvoana M. oleifera* consumed in the northern part of Cameroon were processed using local processes techniques identified through a household survey. Total iron in the dishes was determined by atomic absorption and the bioavailability of iron was assessed *in vitro*. The results showed that iron contents varied with the species of vegetables and the cooking method (P <0.05). Iron contents of dishes are substantial and could contribute to the improvement of serum iron status of populations. The contents varied from 9.85mg/100gDW for the dish made of *V. calvoana* to 71.98mg/100gDW for that made using *C. sesamoïdes.* The addition of ingredients to the bleached vegetables with or without kanwa resulted to lower iron contents and the reduction percentage varies between 13% and 77% with the species of leafy vegetable. The results of this study show that, the dishes prepared with kanwa have higher iron levels than those prepared without kanwa. Unfortunately, kanwa reduces the bioavailability of iron of the leafy vegetables studied. The bioavailability of iron in the leafy vegetables studied is generally low and varies with the species and with the cooking method applied. *C. olithorus*had the highest content of ionizable iron (5mg/100gDM) while with *C. sesamoïdes*had the lowest (0.36mg/100gDW). The high iron content in a vegetable dish does not guarantee high iron bioavailability.

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INTRODUCTION

Malnutrition is still considered a public health problem worldwide (Bobby *et al*., 2002). FAO (2008) estimated that micronutrient deficiencies affect more than two billion people. Cameroon, despite its dietary diversity is not without these nutritional problems (Lowe *et al.*, 1993; Kollo *et al.*, 2001). Iron, classified as micronutrient is needed by our body in small amounts. Deficiency in iron, however, can have a major impact on health such as anemia (Norhaizan and Nor Faizadatul, 2009). Iron deficiency is quite prevalent, while insufficient intake and poor bioavailability are the major causes. Bioavailability is a general term that refers to how well a nutrient can be absorbed and used by the body.

It can be affected by many factors such as the presence of anti-nutrients (Lestienne *et al.,* 2005) as well as the quantity of food consumed (Borel, 2003). Iron deficiency among women and children are particularly related to low dietary intake of iron. This is explained by the fact that the iron-rich foods such as meat and fish are expensive (Dapi *et al*., 2011). It is established that the choice of food can have a positive or negative impact on the nutritional status of the consumer (Tanya, 2009). Vegetable is a vital component of human diet that is eaten all year round in Cameroon. Several authors have shown that raw leafy tropical regions are rich in minerals (Tchiégang and Aïssatou, 2004; Ejoh *et al*, 2005; Adepoju, 2008). These vegetables are eaten cooked usually after adding ingredients (salt, onion, peanuts.). The nutritional composition of leafy vegetables is linked not only with factors such as variety, season, stage of maturity, geographical location, but also with the cooking methods (Chang *et al.*, 2013).

In Cameroon, there have been a number of studies determining the chemical composition of dishes (Sharma *et al.*, 2007; Kana Sop *et al.*, 2008; Fokou *et al.*, 2009). However, because of the ethnic diversity at the national level, the food variety, the food processing and cooking methods, there is need for more elaborate studies on the nutritional value of the different dishes in Cameroon.

Moreover studies on the bioavailability of iron from these foods consumed in Cameroon are limited (Kana Sop *et al.*, 2004) and therefore needs to be elaborated. The purpose of this study is to determine the iron content and assess their bioavailability in some leafy vegetables dishes consumed in the northern part of Cameroon.

MATERIALS AND METHODS

Sample collection and preparation

After a survey to determine the different leafy vegetables consumed and their methods of preparation in the northern part of Cameroon, the leafy vegetables having a high consumption frequency or/and high iron content were selected for the preparation of dishes. The dishes were those made principally of *S. macrocarpon (kuitadje), H. sabdariffa morphotype mince (foléré Ngaoundéré), H. sabdariffa morphotypeblanc (foléréGaroua), C.maxima morphotypetacheté (hakomborho). C. maxima (hakomborho), C. sesamoïdes (gouboudo), C. olitorius (lalo), A. hybridus (hakondiam), V. calvoana (souhaka)* and *M. oleifera (giligandja).*

About 2000 g of each of these leafy vegetables were randomly obtained from 3 different farms early in the morning, when temperatures were below 15˚C, within the region and immediately (within an hour) transported in open baskets to the Biochemistry laboratory at the University of Ngaoundéré for preparation prior to analysis. The leaves were sliced into small particles, rinsed under tap water and left 10 min to drain until subjected to different processing conditions. The ten species were each divided into 3 lots. One was left unprocessed to dry at 45˚C in an electric oven for 12 hours while the others were subjected to either squeeze-washing and boiling, or boiling in kanwa (a local alkaline salt obtained from the Ngaoundéré market) and squeeze-washing. For the latter, 500g portions of fresh samples were boiled in 1L water containing 10 g kanwa for 10 minutes before squeeze-washing and rinsing. Squeeze-washing and rinsing is a traditional process which involves crushing the vegetable with the hands and squeezing out the water and foam. This is followed by rinsing with fresh clean tap water and squeezing out the water. Data from the survey mentioned earlier were used to determine the traditional concentration of kanwa used in the preparation.

Preparation of the dishes

The blanched vegetable samples were prepared with the addition of the ingredients indicated in Table 1. In a dry pot placed on an electric heater (Indesit), 50ml of cotton oil (diamaor) were introduced and 15g of sliced onions introduced when the oil was already hot (100°C for 3 minutes) and left for a minute to be cooked brown. Fresh tomatoes (25g), water (300ml), and salt (5g) were added and left to boil before peanut paste (50g) were added and left to cook for 15 minutes (Peanut was not added in the preparation of *C. Sesamoïdes* and *C. olitorius*dishes). The squeeze-washed leafy vegetables blanched with or without kanwa were then added and the whole lot left on mild fire for 20 minutes. The sauces obtained are generally consumed with corn, millet and cassava fufu. The dishes were prepared in the kitchen facilities of the Department of Science and Food Nutrition, National School of agro-Industrial Sciences (ENSAI), of University of Ngaoundéré using the ingredients and methods that were traditionally used in the study area as described in the household survey.

After cooking, the samples were left to cool and dried at 45°C in 24h then ground with a Culatti (polymix, France) , sieved through a mesh screen size of 500 microns, packed in air tight plastic bags, labelled and stored at-18°C in a freezer. Water contents of the samples were determined immediately after cooking.

Moisture and total iron content determination

Moisture content was determined by drying samples to constant weight in an electric oven at 105°C (AOAC, 1980). Iron contents in all the samples were determined after wet digestion with mixture of nitric, sulphuric and hydrochloric acid using Atomic Absorption Spectrophotometer (AAS aalae spectrophotometer S11) (Kawashima and Soares, 2003).

Bioavailability and ionizable iron estimation

The *in vitro* bioavailability of iron from the leafy-vegetables samples were determined by the method described by Rao and Prabhavati (1983) and the Estimation of ionizable iron in the filtrates estimated by adaptation of the α-dipyridyl method (AOAC, 1965). Duplicate aliquots (5 ml) of the filtrates were taken and the volume in one set was made up to 15 ml and served as the extract blank. The other aliquot was treated with 1 ml of 10% hydroxylamine hydrochloride, 5 ml of acetate buffer pH 4.2, 2 ml of 0.1% of α, α-dipyridyl reagent, and made up to 15 ml with water. The colour developed was measured at 510 nm in the Spectronic S 11 against a reagent blank. Iron standards (1-15 mg) were simultaneously run in the assay. The ionizable iron content in the sample was derived after subtracting the optical density of the extract blank from that of the sample.

Statistical analysis

The experiments were conducted in threes. Statistical Analysis System software (SAS Institute Inc. Version 8.2, Cary, NC, USA; 2001) was used for data analyses. One-way analysis of variance was used to find out the mean difference between the groups. Multiple comparisons with the least significant difference (LSD) method were used to assess

Table 1 Name of leafy-vegetables and ingredients used for the preparation of dishes

Local names (fufulde)	Scientific names	Main ingrédients
Foléré Ngaoundéré	*H. sabdariffa M.*	Onions, tomatoes, oil, water, salt peanut paste
Hakokuitadje	*S. macrocarpon*	Onions, tomatoes, oil, water, salt peanut paste
Hakomborho	*C. maxima T.*	Onions, tomatoes, oil, water, salt peanut paste
Goubdo	*C. sesamoïdes*	Onions, tomatoes, oil, water, salt
Lalo	*C. olitorius*	Onions, tomatoes, oil, water, salt
Folere Garoua	*H. sabdariffa B.*	Onions, tomatoes, oil, water, salt peanut paste
Hakomborho	*C. maxima*	Onions, tomatoes, oil, water, salt peanut paste
Hakondiam	*A.hybridus*	Onions, tomatoes, oil, water, salt peanut paste
Souaka	*V. calvoana*	Onions, tomatoes, oil, water, salt peanut paste
Giligandja	*M. oleifera*	Onions, tomatoes, oil, water, salt peanut paste

which combination was different from the rest. $P < 0.05$ was considered significant.

RESULTS

Total iron contents

The total iron contents of dishes from leafy vegetables were presented in Figure 1. These levels vary with the species and the cooking method ($P < 0.05$). The levels of total iron range from 9.85mg/ 100g DW for *V. calvoana* to 71.98/ 100g DW mg for *C. sesamoides* and 9.95mg/ 100g DW 81.63mg/ 100g DW respectively for the same vegetables prepared with or without kanwa. The dishes made with kanwa showed higher iron contents regardless of the vegetable considered. Thus the *H. sabdariffa* dishes without kanwa has a content of 15.52 ± 0.72 mg/ 100gDW while with kanwa we have in the same vegetable a value worth 45.23 ± 2.01 mg/ 100gDW, that is three times the previous value. In the *M. oleifera* dish without kanwa, there is a value of 12.22 ± 1.02 mg/ 100gDW and a double value in the dish of the same vegetable with kanwa that is 24.49 ± 2.05 mg/ 100 g DW.

The addition of ingredients to the blanched vegetables with or without kanwa causes the loss of iron and the percentage of reduction varies with the species. The *S. macrocarpon* dish without kanwa experienced a reduction of 20.78% (Table 2) and with kanwa the reduction was 27.71%. In the *H. sabdariffa* dish without kanwa, the reduction is 22.3% and 16.75% in the same dish with kanwa. Yet the losses were more pronounced in the dishes of *C. maxima*. Indeed, there has been an iron content move in the blanched vegetables from 89.74mg/100g DW to 38,5mg/100g DW after addition of ingredients thus a 57.09% discount. In the *C. maxima* dish with kanwa, the percentage loss was 69.34 (Table 2). In the ten vegetables studied, the dishes prepared with kanwa have higher iron levels than those dishes prepared without kanwa. Is the iron found in these vegetables bioavailable?

In vitro iron bioavailability of leafy vegetables

The results of the bioavailability of iron from leafy vegetables are presented in Tables 3a to 3j. These results show a significant difference between the dishes of the same species of vegetable and also between dishes made from different species ($P < 0.05$).

The percentage of bioavailability of Iron varies according to the treatment applied to the vegetable. Indeed, the iron in the dishes of vegetables prepared without kanwa is more bioavailable than that of those prepared with kanwa. Similarly, the iron contained in the bleached leafy vegetables without kanwa is more bioavailable than iron in the bleached vegetables with kanwa. This is the case of *M. oleifera* whose bleached leaves without kanwa has 1.89% bioavailable iron and 4.76% respectively at pH 1.35 and pH 7.5 against 1.29% and 0.57% of bioavailable iron bleached vegetables with kanwa. In terms of dishes of this same vegetable, the one made without kanwa gives 7.28% bioavailable iron at pH 1.35 and 2.70% at pH 7.5 while the *Moringa* dish with kanwa offers only 3.80% and 0.61% respectively at pH 1.35 and pH 7.5. The *H. sabdariffa dish* without kanwa has bioavailable iron percentages of 3.67% and 0.45% respectively to 1.35 and pH 7.5 while with kanwa and these pH these percentages are reduced to 0.99 % and 0.24%. The iron contained in kanwa is not bioavailable.

The addition of ingredients increases the bioavailability of iron of certain leafy vegetables. Thus in the case of *V. calvoana* with kanwa at pH 1.35, we'll move from a percentage of bioavailable iron of 5.80 to 10.75 respectively for the bleached vegetables and the dishes prepared from it. At pH 7.5, we move from 0.17 to 1.91. The results of Table 3a shows that at pH 1.35, folere dish has a higher percentage of ionizable iron of 8.47 compared to simply bleached folere and at duodenal pH 1.15, the folere dish is given a percentage of 8.07 while it is only 0.34 for bleached folere without kanwa.

At pH 1.35 the vegetables dishes prepared with or without kanwa have higher percentages of ionizable iron than at pH 7.5. In the *M. oleifera* dish without kanwa, at acidic pH, the percentage of ionizable iron is of 7.98 and 2.70 at pH duodenal. With kanwa, the same trend is observed: 3.80% and 0.61% respectively in the acidic pH and duodenal pH. The iron content is not proportional to its bioavailability.

Indeed, with a total iron content of 71.98mg/ 100gDW, *C. sesamoïdes* contains in his dishes without kanwa 0.24 ± 0.05 mg/ 100gDW ionizable iron at pH 1.35 and 0.12 mg/ 100gDW in pH 7.5 or 0.33% and 0.17% respectively bioavailability. On the contrary *S. macrocarpon* which contains in its dishes without kanwa a total iron content of 11.09 mg/ 100gDW at 2.01 mg/ 100gDW ionizable iron at pH 1.35 and 1.36 mg/ 100gDW at pH 7.5 thus 18.12% and 12.27% respectively.

Generally, the bioavailability of iron from the leafy vegetables studied is small and is less than 5%. That of *A .hybridus* varies from 0.96 to 1.98% at pH 1.35 and from 0.21 to 1.01% at pH 7.5. The bioavailability of iron in *C. sesamoïdes* ranges from 0.33 to 1.75 at pH 1.35 and 0.17 to 0.84%.

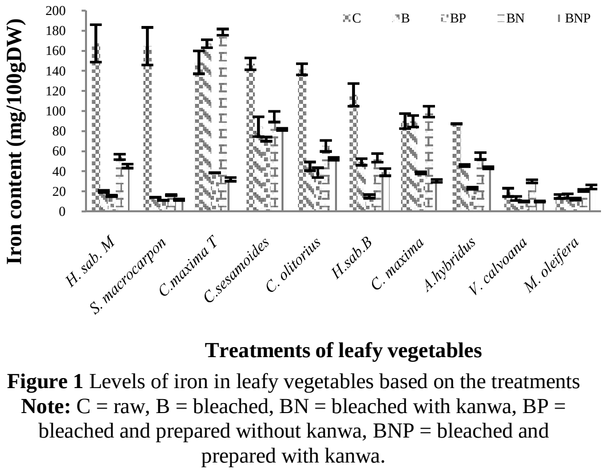

Figure 1 Levels of iron in leafy vegetables based on the treatments
Note: C = raw, B = bleached, BN = bleached with kanwa, BP = bleached and prepared without kanwa, BNP = bleached and prepared with kanwa.

Digestion in the body is done in two steps. The first stage takes place at acidic pH with pepsin and the second continues to pH 7 with pancreatin. The bioavailability of iron in this summary table is given by the average of the two pH.

Table 2 Percentage of reduction of iron levels in leafy vegetables dishes

	H. sab. M	*S. macrocarpon*	*C.maxima T*	*C.sesamoides*	*C. olitorius*	*H.sab.B*	*C. maxima*	*A.hybridus*	*V. calvoana*	*M. oleifera*
C-B	88,08	91,49	-12,50	42,57	68,27	57,53	0,27	47,49	31,61	-2,20
C- BN	67,57	90,16	-20,32	35,75	53,91	54,00	-10,41	36,77	-57,81	-38,89
B-BP	22,21	20,79	76,96	14,71	13,89	69,57	57,10	49,44	24,23	20,23
BN-BNP	16,70	27,72	82,13	13,54	19,42	26,71	69,34	20,96	66,83	-17,63

CB = reduction between the raw and blanched samples; C-BN = reduction between raw and blanched samples with kanwa; B-BP =reduction between the bleached samples and the prepared samples; BN-BNP = reduction between samples bleached with kanwa and samples prepared with kanwa

Table 3 *In vitro* bioavailability of iron in cooked *H. sabdariffa 'white'*

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionizable iron (mg/100g)	% of total iron	Ionizable iron (mg/100g)	% of total iron
H.sab 'white' B	44.29	0.51±0.05^{a}	1.15	0.15^{a}	0.34
*H.sab 'white'*BN	55.38	0.06±0.02^{b}	0.11	1.26±0.01^{d}	2.28
H.sab 'white' BP	15	1.27±0.01^{c}	8.47	1.21±0.01^{c}	8.07
H.sab 'white' BNP	39.14	0.02^{a}	0.05	0.8±0.02^{b}	2.04

Table 4 *In vitro* bioavailability of iron in cooked *S. macrocarpon*

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionizable iron (mg/100g)	% of total iron	Ionizable iron (mg/100g)	% of total iron
S.macrocarpon B	14	3.56±0.14^{d}	25.43	3.46^{d}	24.74
S.macrocarpon BN	16.20	1.84^{b}	11.36	1.62^{c}	10
S.macrocarpon BP	11.09	2.01	18.12	1.36^{b}	12.27
S.macrocarpon BNP	11.71	1.66±0.03^{b}	14.18	0.94^{a}	8.03

Table 5 *In vitro* bioavailability of iron in cooked *C. maxima T.*

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionizableiron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
C.maxima T. B	167	20.08±0.48^{d}	12.02	0.45^{b}	0.27
C.maximaT. BN	178.6	1.88^{c}	1.05	0.22±0.05^{a}	0.12
*C.maximaT.*BP	31.91	1.04±0.01^{a}	2.70	0.38±0.04^{b}	0.99
*C.maximaT.*BNP	38.48	1.22±0.01^{ab}	3.82	0.41±0.07^{b}	1.23

Table 6 *In vitro* bioavailability of iron in cooked *C. sesamoïdes*.

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionizableiron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
C.sesamoides B	84.39	1±0.06^{B}	1.18	0.71^{d}	0.84
C.sesamoides BN	94.41	0.34±0.06^{a}	0.36	0.56±0.02^{c}	0.59
C.sesamoides BP	71.98	0.24±0.05^{a}	0.33	0.12^{a}	0.17
C.sesamoides BNP	81.63	1.43±0.01^{c}	1.75	0.2±0.01^{b}	0.25

Table 7 *In vitro* bioavailability of iron in cooked *C. olitorius*

Vegetables	Total iron (mg/100g)	pH 1,35		pH 7,5	
		Ionizableiron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
C.olitorius B	44,91	0,28±0,01^{a}	0,62	0,63±0,03^{b}	1,40
C.olitorius BN	65,23	1,47±0,02^{c}	2,25	0,54±0,02^{a}	0,82
C.olitorius BP	38,67	0,87±0,01^{b}	2,25	4,15±0,09^{c}	10,73
C.olitorius BNP	52,56	0,3±0,08^{a}	0,57	0,75±0,02^{b}	1,43

Table 8 *In vitro* bioavailability of iron in cooked *H. sabdariffa*'thin'

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionizableiron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
H.sab 'thin' B	19.95	0.5±0.12^{a}	2.51	1.75±0.01^{d}	8.77
H.sab 'thin' BN	54.30	0.95±0.41^{b}	1.75	0.47^{c}	0.87
H.sab 'thin' BP	15.52	0.57±0.07^{a}	3.67	0.07^{a}	0.45
H.sab 'thin' BNP	45.23	0.45±0.02^{a}	0.99	0.11^{b}	0.24

Table 9 *In vitro* bioavailability of iron in cooked *C. maxima*

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionisable iron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
C.maxima B	89.74	1.05^{c}	1.17	0.31±0.01^{b}	0.35
C.maxima BN	99.35	1.2±0.02^{d}	1.20	0.32^{c}	032
C.maxima BP	30.46	0.73	2.39	0.07	0.3
C.maxima BNP	38.05	0.86±0.01^{b}	2.26	0.01^{a}	0.03

Table 10 *In vitro* bioavailability of iron in cooked *A .hybridus*

Vetgetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionizableiron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
A.hybridus B	45.81	0.7±0.06^{b}	1.53	0.1^{a}	0.21
A.hybridus BN	55.16	0.83±0.07^{b}	1.50	0.47^{c}	0.85
A.hybridus BP	23.16	0.46±0.04^{a}	1.98	0.34±0.04^{b}	1.46
*A.hybridus*BNP	43.6	0.42±0.08^{a}	0.96	0.44±0.03^{c}	1.01

Table 11 *In vitro* bioavailability of iron in cooked *V. calvoana*

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionisable iron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
V.calvoana B	13	1.83±0.01[d]	14.08	0.9±0.01[c]	6.92
V.calvoana BN	30	1.74±0.04[c]	5.80	0.05[a]	0.17
V.calvoana BP	9.85	0.98±0.02[a]	9.94	0.16[b]	1.62
V.calvoana BNP	12.5	1.07±0.03[b]	10.75	0.19±0.02[b]	1.91

Table 12 *In vitro* bioavailability of iron in cooked *M. oleifera*

Vegetables	Total iron (mg/100g)	pH 1.35		pH 7.5	
		Ionisable iron (mg/100g)	% of total iron	Ionizableiron (mg/100g)	% of total iron
M. oleifera B	15. 32	0.29±0.03[a]	1.89	0.73±0.02[c]	4.76
M. oleifera BN	20.82	0.27±0.04[a]	1.29	0.12±0.02[a]	0.57
M. oleifera BP	12.22	0.89[b]	7.28	0.33±0.05[b]	2.70
M. oleifera BNP	24.49	0.93[b.]	3.80	0.15[a]	0.61

The mean ± standard deviation of the same letters followed by exposing the same column are not significantly different (P <0.05) (N = 3)
B = bleached, BN = bleached with kanwa, BP = bleached and prepared without kanwa, BNP = bleached and prepared with kanwa.

Table 13 Summary of the bioavailability of iron in leafy vegetable dishes

Vegetables	Total iron in raw samples (mg/100gDW)	Total iron in BP (mg/100gDW)	Ionizable iron in BP (mg/100gDW)	%	Total iron(BNP) (mg/100gDW)	Ionizable iron In BNP (mg/100gDW)	%
C. sesamoides	146.95±5.58[a]	71.98	0.36±0.05[g]	0.5	81.63	1.45±0.02[c]	1.77
C. olithorius	141.52±11.26[a]	38.67	5.02±0.1[a]	12.98	52.56	5.75±0.1[a]	10.93
C. maxima	89.98±7.47[b]	30.46	0.80[f]	2.62	38.05	0.87±0.02[e]	2.26
C. maxima T	148.44±5.97[a]	31.91	1.42±0.05[d]	4.45	38.48	1.63±0.09[c]	4.23
A hybridus	87.24±0.27[b]	23.16	0.80±0.08[f]	3.45	43.6	0.86±0.11[e]	1.97
M. oleifera	15±2[c]	12.22	1.22±0.05[e]	9.98	24.49	1.08[d]	4.43
H. sab. M	167.42±18.63[a]	15.52	0.64±0.07[f]	4.12	45.23	0.56±0.02	1.23
S.macrocarpon	164.58±18.7[a]	11.09	2.37[c]	21.4	11.71	2.6[b]	22.2
H .sab.B	116.05±114	15	2.48±0.02[b]	16.5	39.14	0.82±0.02[e]	2.09
V. calvoana	19.01±4.12[c]	9.85	1.14±0.02	11.57	12.5	1.26±0.05	10.1

The mean ± standard deviation followed by the same letters inexponenton the same column are not significantly different (P <0.05) (N = 3)
BP = dishes prepared without kanwa; BNP = dishes prepared with kanwa

DISCUSSION

The iron contents of dishes are interesting and could contribute to the improvement of serum iron status of populations depending on their bioavailability. Iron contents were very higher compared to those reported by Kana Sop *et al.*, (2008, 2004) in Cameroon, Oguntona*et al.,* (1987) and Onabanjo and Oguntona (2003) in Nigeria. This can be explained by the fact that the dishes in this study contain vegetable sauces without supplements such as couscous, bananas, and yams. The significant difference (P <0.05) between the dishes with or without kanwa in the same vegetable species is due to the addition of kanwa in the blanched vegetables. Indeed, the analysis of the mineral composition of kanwa used in this study shows a total iron value of (173.46 ± 1.88mg/ 100gDW).

The decrease in iron content in dishes with or without kanwa compared to bleached samples is justified by the fact that the use of ingredients such as oil, onions, tomatoes or peanuts has diluted the iron contents. Indeed, Shashi and Salil (1999) showed that the addition of oil, seasoning and spices during the cooking of *Chenopodium album* and *Trigonellafoemun* leaves may be responsible for the increase of the total mass which may lead to the decrease concentration of minerals.

The dishes without kanwa have high percentages of bioavailable iron than those with kanwa. The iron contained in kanwa is not bioavailable. If the addition of kanwa in the preparations increases the pH and protects chlorophyll, basic seasoning such as sodium bicarbonate, affects the ascorbic acid (Shashi and Salil (1999). It is known that vitamin C promotes the absorption of iron (Stevels 1990). Kana Sop *et al.,* 2004 showed that dyalisable iron was increased with the lime juice and significantly decreased with legumes (beans, soybeans and peanuts), eggs and egg yolk. Narasimga and Prabhavathi (2013) showed after a study on the *in vitro* method for predicting the bioavailability of iron from food that at pH 7.5, the ionizable iron content increased in the presence of ascorbic acid and meat extract while it decreased in the presence of phytates and tannins. The extent of bioavailability of iron from such sources must be considered when evaluating the adequacy of dietary iron intake from the diet. All the dishes analyzed in this study are of plant origin. Animal products are good sources of bioavailable minerals (Gibson and Fergusson, 1999).

This is the reason why iron is more available from animal products than diets of plant origin certainly because of the phytate and fiber content of the plant-based diets, which inhibit the intestinal absorption of iron by forming insoluble chelates (Gibson and Ferguson, 1999; Oberleas, 2003). The iron content is not proportional to its bioavailability. Anti-nutrients such as phytates, tannins and dietary fiber would be strongly present in some vegetable like *C. sesamoïdes*and might have contributed to reduce the bioavailability of iron (Minihane and Rimbach, 2002). Generally, the bioavailability of iron from the leafy vegetables studied is small and is less than 5%. These results confirm those of Minihane and Rimbach (2002) who have shown that the absorption of non-hemenic iron is less than 5%. However *S. macrocarpon* made the difference in having an iron bioavailability ranging from 11.36 to 25.43% at pH 1.35 and 8.03 to 24.74% in pH 7.5. Bioavailability of iron is very influenced with the meal content and mostly when it is from plant source (Stolzfus, 2003). Kana Sop (2000) reported very low *in vitro* iron bioavailability from corn meal with okra (0.89% total iron) that had high content of total iron (31 mg/100 g DW).

The addition of ingredients increases the bioavailability of iron of certain leafy vegetables. This could be explained by the presence of ascorbic acid, citric acid, lactic acid content in the onions and tomatoes.

At pH 1.35 the vegetables dishes prepared with or without kanwa have higher percentages of ionizable iron than at pH 7.5. We know that at pH 1.35, most of the iron is in its ionized form. During transit of the iron to the duodenum (pH 7.5), most of the iron is insoluble, unless it is chelated by dietary compounds that enhance its absorption such as ascorbic acid, citric acid, lactic acid. This is what would explain the exceptions made by the dishes *C. olitorius*and *H. sabdariffa* thin observed in Tables 3e and 3f. Indeed, for the dish without kanwa*C. olitorius* for example, the percentage of ionizable iron has improved from 2.25% at acidic pH to 10.73% in duodenal pH. Similarly, with the flat kanwa, it is from 0.57% at pH 1.35 to 1.43% at pH 7.5. Dietary iron which is ionizable at duodenal pH is susceptible to be available for absorption.

CONCLUSION

The aim of this study was to assess iron content and the bioavailability in ten most frequently consumed vegetables dishes in the northern part of Cameroon selected after the interview of household. From their content, it was possible to conclude that all the ten leafy vegetables are good sources of iron. However, from the comparison of their bioavailability only *C. olitorius* contributed the largest part of iron intake. This study also shows that the addition of kanwa while cooking helps increase iron however kanwa reduces the bioavailability of iron.

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