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Abstract: Macronutrients and dietary fiber contents, digestibility (D%), biological value (BV), net protein utilization (NPU), dialyzability (%Da) and potential contribution of Fe and Zn were evaluated in pasta like product (spaghetti-type) made with corn flour and nutritionally improved (PNI) with 30% broad bean flour (C/BB) and 20% quinoa flour (C/Q). PNI showed a significant increase in NPU and BV and a slight decrease in D as compared with the control sample. One PNI portion supplies 10-20% of the daily dietary fiber recommended value. The C/Q PNI showed 14.84 of Fe %Da and the C/BB 52.34 of Zn %Da. EDTA addition quadruples the Fe %Da in C/BB and C/Q and increases the Zn %Da five-fold in C/BB PNI, but does so only 1.5 times in C/Q PNI. The EDTA addition may negatively affect the quality of pasta like product by decreasing firmness and increasing stickiness and solid losses during cooking.

Dear reviewers

We submit to FOOD CHEMISTRY our manuscript Entitled "Nutritional improvement of corn paste-like product with broad bean (Vicia faba) and quinoa (Chenopodium quinoa)", in which we evaluate the improvement in the nutritional characteristics resulting from combining corn flour with unconventional Andean whole grain flours for making gluten-free pasta like products.

We believe that the knowledge generated is important because there is very little information about extrusion-cooking of gluten free products and even less about the characterization of their nutritional properties.

We consider that it is an important issue due to the increasing of number of people suffering celiac disease and the frequent consumption of products made from refined flour, with low nutrient supply, causes low coverage of the recommended daily intake of various macro and micro nutrients.

On the other hand the use of corn, nutritional and economic support of Andean region, in the preparation of gluten free foods enhanced with other natural Andean crops or adapted to the region, adds value to the food chain.

Yours sincerely

Dra. Norma Sammán

Nutritional improvement of corn pasta-like product with broad bean (*Vicia faba*) and quinoa (*Chenopodium quinoa*)

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***Highlights (for review)**

- Broad bean and quinoa improved nutritional quality of corn pasta-like product
- The complementation increased protein biological value and decreased digestibility
- Broad bean and quinoa flours affect Fe and Zn bioavailability in opposite ways
- EDTA reduce sensorial and physicochemical qualities of free gluten pasta like

ABSTRACT

Macronutrients and dietary fiber contents, digestibility (D%), biological value (BV), net protein utilization (NPU), dialyzability (%Da) and potential contribution of Fe and Zn were evaluated in pasta like product (spaghetti-type) made with corn flour and nutritionally improved (PNI) with 30% broad bean flour (C/BB) and 20% quinoa flour (C/Q). PNI showed a significant increase in NPU and BV and a slight decrease in D as compared with the control sample. One PNI portion supplies 10-20% of the daily dietary fiber recommended value. The C/Q PNI showed 14.84 of Fe %Da and the C/BB 52.34 of Zn %Da. EDTA addition quadruples the Fe %Da in C/BB and C/Q and increases the Zn %Da five-fold in C/BB PNI, but does so only 1.5 times in C/Q PNI. The EDTA addition may negatively affect the quality of pasta like product by decreasing firmness and increasing stickiness and solid losses during cooking.

Key words: dialyzability, iron, zinc, protein digestibility, gluten free

1. INTRODUCCION

The celiac disease is an immunological disorder of the intestine caused by gluten intolerance. The only effective treatment for people who suffer from this disease is to omit any foods that contain wheat, oats, barley and rye from their diet. Due to the growing number of cases of gluten-intolerance, which affects 0.3-1% of the world's population (Schoenlechner, Drausinger, Ottenschlaeger, Jurackova & Bergehofer, 2010) it is necessary to guarantee the availability of a wide range of gluten-free products. According to Dermiskesen, Mert, Sumnu & Sahin, (2010) the gluten-free pasta and baked products that are currently available in the market are often made from refined flours and starches. For this reason, they contribute low amounts of protein, minerals and dietary fiber, which could increase the risk of nutritional deficiencies associated with the celiac disease. According to Stojceska, Ainsworth, Plunkett & Ibanoglu (2010), the iron and dietary fiber deficiencies are among the most frequent in people who suffer from this disease. Matos Segura & Rossell (2011) found great variation in the nutrient composition of the gluten-free products available in the market, and pointed out that they are low in protein content and high in fat and carbohydrate content, while they contain adequate levels of dietary fiber, in line with nutritional recommendations.

Various formulations and technological parameters have been widely investigated in recent studies. The purpose has been to improve both the quality and nutritional value of gluten-free pasta and baked products through the incorporation of nutritional flours or protein isolates of diverse origin, such as amaranth, quinoa, lupine, chickpea and other leguminous flours (Cabrera-Chávez et al., 2012; Demirkesen et al., 2010).

According to Zandonadi, Botelho, Gandolfi, Ginani, Montenegro & Pratesi (2012) pasta is one the most widely demanded products among people suffering from gluten-intolerance. However, information on the effect of different raw materials on protein biological utilization and on the availability of some nutritionally valuable minerals in this type of products is still limited. Extrusion-cooking is an alternative technology which is suitable to make gluten-free pasta like product, as it allows for the complementation of different flours or raw materials (Giménez, González, Wagner, Torres, Lobo &Sammán, 2013; Marti, Seetharaman & Pagani, 2010).

In the Andean region of Jujuy, the production of crops like quinoa and broad bean has seen continuous growth. Because of their nutritional properties, such as proteins that adequately complement the cereal proteins, high content of minerals like iron and zinc, vitamins, dietary fiber and functional components, added to the absence of gluten-forming protein, these foodstuffs prove ideal to enrich corn-based products and may be safely consumed by the celiac population (Hager, Wolter, Fritz, Zannini & Arendt, 2012).

However, their use in formulations also involves the addition of other anti-nutritional compounds that affect the actual bioavailability of protein and of some critical nutrients such as Fe and Zn. In order to counteract the negative effect of the inherent presence of these inhibitors, co-fortifiers such as EDTA are added. EDTA is generally used in flours to increase the bioavailability of Fe and Zn (Tripathi & Platel, 2011). The aim of this study is to evaluate the nutritional impact of gluten-free pasta like made with corn flour and nutritionally improved with 30% broad bean flour and 20% quinoa flour, obtained by extrusion-cooking using EDTA as co-fortifier.

2. MATERIALS AND METHODS

2.1. Raw materials

Corn flour was provided by Molinos Puerto Reconquista (Santa Fe, Argentina). size was reduced using a Buhler-Miag roller mill. Quinoa seeds and broad beans (Vicia faba) hulled manually and drying in solar dryer, were obtained from a

Cooperative of producers (CAUQUEVA- Tilcara, Jujuy, Argentina) and ground using a fixed hammer mill (Retschj, Germany), in order to obtain flour with a particle size between 0.191 and 0.490 mm.

2.2. Elaboration of spaghetti type pasta nutritionally enhanced (PNI)

PNIs were prepared in duplicate, as follows:

Blends flours were prepared by substituting corn flour with broad bean flour (C/BB) and quinoa flour (C/Q) as 30 and 20 g/100g (db) respectively; substitution levels suitable to obtain a product of acceptable quality (Gimenez et al., 2013). Homogenized blends were mixed with water up to a moisture content of 28% using a planetary mixer (Brabender).

The extrusion process was carried out in a Brabender 10 DN single screw extruder, at 100°C, using a 3:1 compression ratio screw, a 1.5 mm x 3 (diameter x nº of holes) die and a screw speed of 60 rpm. The products were dried at low temperature 40°C and 40 % relative humidity during 16 h. Na_2EDTA ($C_{10}H_{14}N_2O_8Na_2.2H_2O$; PM: 372,24) was added to the mixture before extruding in a molar ratio 1:1 EDTA:Fe, to investigate its effect on availability of Fe and Zn

2.3. Cooking loss (CL): Ten grams of the PNI samples, of 10 cm of long approximately, were placed into 500 ml beaker with 200 ml of boiling distilled water. After required cooking time, the cooked product was drained 3 min. The cooking water was then collected in aluminum vessel, placed into an air oven at

105 °C and evaporated to dryness. The residue was weighed and reported as percentage of the starting material.

2.4. Global Sensorial score (GSS)

Sensory evaluation was carried out with a trained panel of three persons to evaluate firmness and stickiness. The global score was obtained by consensus in two replications.

A 0-5 scale was used. The 0 value was assigned for the firm and not sticky noodle and 5 for the most soft and very sticky one. A global score (firmness+stickiness) less than or equal to 5 was considered acceptable.

2.5. Compositional analysis

Raw materials and PNI were analyzed for protein, fat, ash, and moisture using the standard procedures of AOAC (1995). Moisture was determined in a vacuum oven (SHE- LAB 1410), AOAC 925.09 method. Lipid content was determined according to the acid hydrolysis method, AOAC 922.06. Total protein content was determined using Kjeldahl (BUCHI DIGESTIÓN UNIT K-435) procedure with a nitrogen-to-protein conversion factor of 6.25, AOAC 984.13 method. Ash analysis used a carbonization at 550°C (Mufle furnace), AOAC 923.03 method. Total iron and zinc content was analyzed using atomic absorption spectrometer. Calibration of the measurements was performed using commercial standards. Dietary Fiber was determined by enzymatic-gravimetric method, AOAC 985.19. The fatty acid profile was determined using gas chromatography following the methylation acid process.

2.6. Protein quality

2.6.1. Chemical Score

Protein quality of the different flour blends (C/B y C/Q) were analyzed according to the chemical score (ChS), and was calculated using the amino acid composition of the corn flour (Sourci, Fachman, & Kraut 1994), quinoa (Reppo-Carrasco, 1992) and broad bean (Lattanzio, Bianco, Criveli & Miccolis, 1983) flours. A preschooler reference patron FAO/WHO/ONU (1985) was utilized. The chemical score was calculated from the following equation:

$$\text{Chemical Score} = \frac{\text{aae}_i \text{ (flour blend)}}{\text{aae}_i \text{ (reference protein)}} * 100$$

aae_i: limiting amino acid

2.6.2. NPU and Protein Digestibility of PNIs

Sprague-Dawley rats of both sex and aged 25-35 days, with average of 60±1 g, were used. Animals were housed individually in screen-bottom cages in a temperature-controlled room at 21±1°C with a 12 h light-dark cycle.

The tests were carried out with isocaloric diets formulated so as to cover all the nutrient requirements whose only variable was the protein type.

Test diets and water were offered ad libitum for 24 h, starting at midday. At the end of the feeding period, diets were withdrawn and the animals reweighed. The consumed diet was established by weight difference.

The classical method of Bender & Miller (1953) was performed to determine NPU using body water as an index of body nitrogen. Three groups of four rats each were used for each sample of protein during 10 days, and biological values

(BV=NPU/digestibility) were calculated. Diet composition was 4 g of salt mixture (Phillips and Hart salt mixture, ICN Nutritional Biochemical, Cleveland, OH), 2.2 g of vitamin mixture (Vitamin Diet Fortification, ICN Nutrition Biochemical), and 5 g of lipid (corn oil). The amount of PNI added was adjusted so as to obtain 10 g of protein in the diet. A mixture of starch and sucrose (1:1) was added so as to complete 100 g of the diet. Casein as protein reference was used.

2.7. Mineral dialysability determination (%DaFe) y Zn (%DaZn)

The samples were prepared to 10% (w/w) of solids concentration using deionised water. Aliquots (25 g) of homogenized samples were adjusted to pH 2.0 with 6 N HCl and after the addition of 0.8 mL pepsin digestion mixture 16% pepsin (Sigma P-7000) solutions in 0.1 HCl, were incubated at 37 °C for 2 h in a shaking water bath. At the end of the pepsin digestion, dialysis bags containing 20 mL 0.19 M PIPES (piperazine-N,N0-bis[2-ethanesulfonic acid] disodium salt) buffer (Sigma P-3768) were placed in each flask and were incubated for 50 min in a shaking water bath at 37°C. Pancreatin–bile mixture (6.25 mL of 2.5% bile (Sigma B-8631), 0.4% pancreatin (Sigma P-1750) solution in 0.1 NNaHCO3) was then added to each flask and the incubation continued for another 2 h. The bag contents were then weighed and analyzed for mineral content by flame atomic absorption spectroscopy (AAS). Assessment of minerals in samples was made by AAS after dry ashing (AOAC, 1985). Mineral dialysability was calculated from the amount of each dialyzed mineral expressed as a percentage of the total amount present in each sample (Wolfgor, Drago, Rodríguez, Pellegrino, & Valencia, 2002).

Dialyzable Mineral (%)= %Da=[D/(W A)]x 100

Where: %Da is the total amount of dialyzed mineral (mg); W is the weight of sample (g) and A is the concentration of each mineral in the sample (mg/g).

The recommended daily intake (RDI) of Fe 18 mg/day and Zn 15 mg/day were used to calculate the percentage of RDI which is supplied by a cooked PNI ration (120 g).

2.8 Statistical Analysis

The data were analyzed and presented as average value ± standard deviation. Statistical differences between the average values were analyzed by using Tukey's HSD comparison test. A level of confidence of 95% ($p<0.05$) was estimated. The program STATISTICA 5.0 for Windows of StatSoft Inc, Tulsa (USA) was used

3. Results and discussion

3.1. Macronutrient content and assessment

Table 1 shows the results of composition analysis corresponding to PNIs

The addition of 30% broad bean flour to corn flour significantly increases the protein content of PNI, reaching values close to the 15% recommended by FAO/WHO/ONU (1985). This content is higher than that of gluten-free commercial spaghetti made from rice and corn flour or starches (Mariotti, Iametti, Cappa, Rasmussen & Lucisano; Marti et al. 2010) and similar to those obtained in wheat spaghetti fortified with broad bean and other leguminous flours (Gallegos-Infante, Bello-Pérez, Rocha-Guzmán, González-Laredo & Avila-Ontiveros, 2010). The protein increase in the PNI is lower when quinoa flour is used, due to its lower protein tenor and the lower substitution percentage.

The addition of 30% broad bean flour to formulate the compound flour increases the lysine content in the C/BB PNI up to 51.51 mg/g protein and the chemical score increases 50% with respect to pure corn pasta like product. In this case, tryptophan becomes the limiting amino acid. Drago, González, Chel-Guerreo & Valencia, (2007) informed a similar increase in corn-cowpea blends with only 15% substitution. Even when the complementation of corn flour with quinoa increases the lysine content by 44%, the limiting amino-acid in the C/Q blend is still the lysine.

PNI showed an increase in NPU and BV values by over 50% in relation with the values of corn spaghetti type pasta (Table 2); the blend with quinoa flour is more efficient, which shows that there is adequate complementation in the studied proteins. Their digestibility values decrease 15% approximately in relation with the control sample. This may be due to factors such as greater compaction in the broad bean and quinoa protein structure, presence of other components like fiber and minerals, the formation of protein-starch compounds, the crosslinks between proteins (Drago et al., 2007) and the presence of anti-nutritional compounds such as polyphenols and phytates whose activity has failed to be suppressed by means of the mechanical and thermal efforts administered during the extrusion-cooking process. Values found are similar to those yielded by macaroni enriched with 20% chickpea (Herken, Ibanoglu, Oner & Ibanoglu, 2006), and lower than those informed for spaghetti enriched with lupine flour (Rayas-Duarte, Mock & Satterleei,

1996) and spaghetti enriched with mustard protein concentrates (Alireza Sadeghi & Bhagya 2008).

With the addition of broad bean and quinoa, the lipid content increases by more than 100% in relation with the control sample. The determined values are higher than those informed for gluten-free pasta made from green banana flour (Zandonadi et al., 2012) and lower than those presented by other pasta made from quinoa, amaranth and rice (Marti et al., 2010; Schoenlechner et al., 2010). Even though pasta is not a relevant source of lipids, the increase in polyunsaturated fatty acids attained with this complementation of flours could improve its contribution of essential fatty acids (5-10%). Brar et al. (2006) determined that the strict diet maintained by celiacs may reduce cardiovascular risk due to the ensuing improvement in the lipoprotein profile. However, Matos Segura and Rosell (2011) found a high content of lipids (7-26%) in commercial gluten-free baked products with formulations presenting a predominance of saturated fats. Therefore, Brar et al.'s considerations could be put into question.

Ash content in PNI doubles that of the control samples, which improves the contribution of minerals to the diet.

The energetic value of the PNI C/Q and control sample is similar to that found by Torres, Frias, Granito & Vidal-Valverde (2007) in pasta that has been nutritionally improved with different raw materials. In the case of PNI C/BB, the energetic value decreases by 14%.

3.2. Dietary fiber

The content of dietary fiber in compound flours increases with the addition of quinoa and broad bean flour, and is higher when broad bean flour is added. Values found are higher than those informed for rice gluten-free pasta that has been nutritionally improved with amaranth flour (Cabrera-Chavez et al., 2012); for parboil-rice pasta and for green banana pasta (Marti et al., 2010; Zandonadi et al., 2012).

There is growing interest in increasing the intake of dietary fiber in celiac patients, since their diets, in general, contribute less than 20g/day of dietary fiber (Stojceska et al., 2010). However, adequate dietary fiber contents were found (>3 g/100g) in commercial gluten-free baked products as a result of the addition of hydrocolloids (guar, xanthan and pectin gums) (Matos Segura & Rosell, 2011). In the studied pasta types, the fiber is contributed by the raw materials used or by the extrusion-cooking process applied (Gimenez et al., 2013). This characteristic increases the value of these products as natural foodstuffs. No reports on dietary fiber content in commercial gluten-free pasta were found. One portion of C/Q or C/BB PNI dried (50-70g) contributes approximately 10-20% of the recommended daily intake.

3.4. Iron and zinc contents and potential availability

As shown in Table 2, the addition of quinoa and broad bean flours significantly increases the Fe and Zn content in PNI. The positive effect of using leguminous flours and pseudocereals as a mineral-rich source has been shown by different authors (Alvarez-Jubete, Arendt & Gallagher, 2010; Drago et al., 2007; Pastor-Cavada et al., 2011). PNI mineral content are higher than that found by Torres et al. (2007) and Dyner et al. (2007) in wheat pasta enriched with different raw materials. However, no reports are available on the mineral content of gluten-free pasta.

Fe %Da values of PNI are similar to those reported by Drago et al. (2007) and Pastor-Cavada et al. (2011) for extruded corn and rice flour blends nutritionally improved with leguminous flours, and higher than those found by Dyner et al. (2007) in wheat spaghetti enriched with amaranth.

The effect of the complementation of the flours studied on Fe dialyzability depended on the raw material used. Fe %Da increases by 40% with the addition of quinoa, and decreases by the same percentage with the addition of broad bean. This behavior was similar to that observed by Vitali, Dragojevic, Sebecic & Vujic (2007) in wheat crackers enriched with different whole-grain raw materials.

The negative effect of the addition of broad bean on iron availability could be due to an adverse balance between compounds that decrease Fe solubility, such as phenolic compounds, phytates, flavonoids and fibers in general (Chaieb, Gonzalez, López-Mesa, Bouslama & Valiente, 2011) and to the lower content of the histidine and cystine, which favor bioavailability (Cumming, Edmond & Magee, 2004). On the other hand, the increase in Fe %Da by the addition of quinoa is due to the higher content of the amino-acids mentioned above (Bhargava, Shuka & Ohri, 2006) and to the lower content of chelating compounds (Alvarez-Jubete et al; 2010).

Zn %Da was higher than Fe %Da. Unlike Fe, Zinc is only found in vegetables in divalent state, which could explain its greater bioaccessibility (Garrow, James & Ralph, 2000). The samples that were nutritionally improved with broad bean flour presented the highest dialyzability percentages, and increased Da by 33-58% in relation with the control sample. According to Hemalatha, Platel & Srinivasa (2007) the protein content and quality of leguminous flours could improve Zn dialyzability in the samples enriched with broad bean flour. Also Lucarini, Di Lullo, Cappelloni & Lombardi-Boccia (2009) found that the content of tannins, phytates and dietary fiber in leguminous flours may not affect Zn bioavailability in the same way they would affect that of Fe. Else, Ramirez-Cárdenas, Leonel, Costa & Reis (2010) found that polyphenols affect Fe bioavailability but do not have an important effect on zinc bioavailability.

Although it remains uncertain how Zn bioavailability can be affected by minerals with similar chemical configuration, the opposing effects on the dialyzability of Zn and Fe caused by the addition of quinoa and broad bean suggest that they compete for absorption sites (Hemalatha et al., 2007).

EDTA addition remarkably improves the dialyzability of both minerals with respect to the control sample. Greater efficiency in the increase of Fe %Da is observed in the C/BB blend and of Zn %Da in the C/Q blend. Other studies have demonstrated that the addition of EDTA increases two- to three-fold the absorption of Fe and Zn in flours (Tripathi, Platel & Srinivasan, 2012).

The complementation of the flours studied, C/BB and C/Q, significantly increases the Fe and Zn potential contribution. Moreover, the addition of EDTA quadruples the RDI% of Fe per portion in relation with the control sample.

For Zn, the addition of EDTA increases the RDI% five-fold in C/BB PNI, and 1.5 times in C/Q PNI as compared with the control sample.

Figure 1 shows the increase in the potential Fe and Zn contribution of a serving of pasta like product when corn flour is enriched with quinoa and broad bean, and the beneficial effect of adding EDTA as a promoter of bioavailability. However, the presence of this promoter or co-fortifier weakens the structure of the starch net created, negatively affecting some important characteristics relative to the sensory quality of the spaghetti type pasta, such as loss of soluble solids during cooking (CL%) (Table 3) and stickiness (Figure 2).

4. CONCLUSIONS

The use of quinoa and broad bean flours may contribute to improve the nutritional value of gluten-free spaghetti type pasta based on corn flour. The substitution percentages studied remarkably improve the contribution of protein, dietary fiber, unsaturated fatty acids, iron and zinc.

The efficiency of amino-acid complementation increased the biological value of protein in pastas like product (spaghetti-type) by over 50% in spite of the decrease in digestibility.

Although broad bean and quinoa flours had different effects on the dialyzability of Fe and Zn, both improved the potential contribution of these minerals by increasing the RDI percentage covered per portion.

EDTA addition favors the biological use of iron and zinc. Nevertheless, its negative effect on the physico-chemical and sensory quality of these pasta like products makes it necessary to expand studies to the use of other co-fortifiers.

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Table 1 Proximate composition of corn-based spaghetti-type pasta enhanced with 30% broad bean (C/BB) and 20 % quinoa (C/Q) flours

	C	**C/BB 70:30**	**C/Q 80:20**
Calories (Kcal/100g)	348.94	299.87	347.76
Moisture	11.10±0.04^{a}	11.18±0.06^{a}	11.21±0.07^{a}
Protein	6.07±0,13^{a}	14.51±0,20^{b}	7.90±0,18^{c}
Lípids	0.58±0,09^{a}	1.27±0,04^{b}	2.0±0,06^{c}
Ash	0.32±0,04^{a}	1.48±0,06^{b}	0.78±0,08^{c}
Dietary fiber	2.07±0,17^{a}	7.35±0,62^{b}	3.57±0,36^{c}
*HC	79.86	64.21	74.54
Zn (mg/100g)	0.54±0,03^{a}	1.73±0,11^{b}	1.54±0,06^{b}
Fe (mg/100g)	3.90±0,24^{a}	8.48±0,94^{b}	5.80±0,26^{c}
Fatty acids			
C16:0 Palmitic	18.84±0.51^{a}	14.78±0.05^{b}	10.82±0.24^{c}
C18:0 Estearic	4.59±0.08^{a}	2.4±0.01^{b}	1.42±0.00^{c}
C20:0. Arachidic	n.d	1.07±0.01^{a}	0.43±0.06^{b}
C 18: 1 Oleic	27.74±0.06^{a}	20.71±0.02^{b}	28.69±0.27^{c}
C20: 1 cis-11-eicosenoic	n.d.	0.49±0.01^{a}	1.22±0.08^{b}
C 18:2 Linoleic w6	46.04±0.54^{a}	55.77±0.00^{b}	51.92±0.19^{c}
C 18:3. Linolenic w3	2.77±0.07^{a}	3.74±0.03^{b}	5.49±0.08^{c}

C: control sample (corn 100%); nd: undetermined

Each point corresponds to the average value of four independent determinations.

Differents letters for each row represent statistical differences between samples ($p<0.05$)

*HC: digestible carbohydrates, estimated by differences

Table 2 Values of nutritional quality of proteins of corn-based spaghetti-type pasta enhanced nutritionally

Samples	NPU	D%	BV	ChS
C	34.81±1.9	90.93±2.6	38.28	41,38
C/BB 70:30	55.72±2.1	80.81±2.1	68.95	61.76
C/Q 80:20	58.65±1.4	78.06±3.2	75.19	65.37

Values corresponds to the mean of the groups of four rats each±SD

NPU: Net protein utilization; D: Digestibility; BV: Biological Value; ChS: Chemical Score

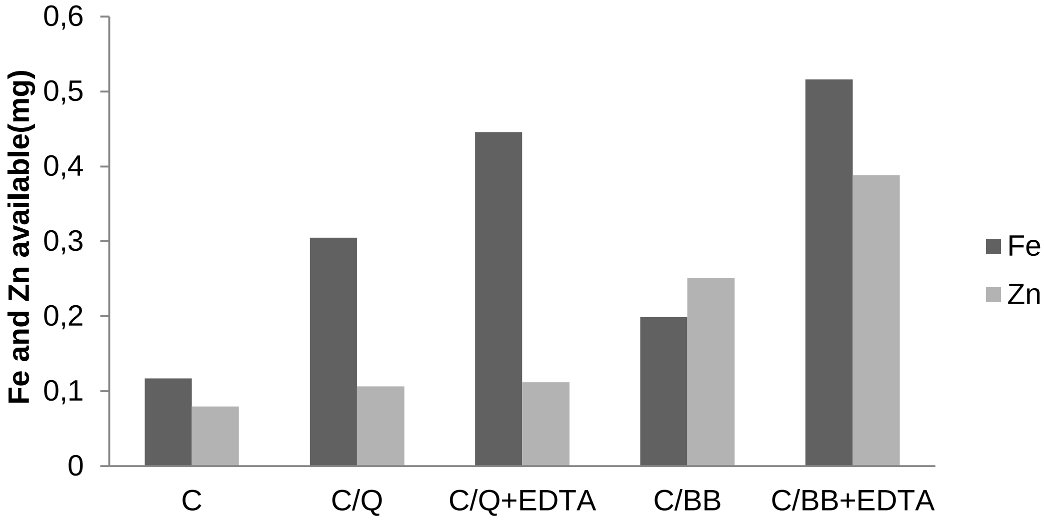
Table 3 Cooking loss (%), dialyzability (%) and recommended daily intake which is supplied by a120-g ration, corresponding to PNI samples.

Samples	CL (%)	DaFe (%)	DaZn(%)	DRI $_{Fe}$(%)	DRI $_{Zn}$(%)
C	7,08±0,34[a]	10,04±1.68[a]	39,36±1.50[a]	0.78±0.04[a]	0.53±0.03[a]
C/BB 70:30	9.07±0.23[b]	6,10±0.47[b]	52.34±4.00[b]	1.10±0.06[b]	1.39±0.07[b]
C/Q 80:20	7,43±0,56[a]	14,84±0.68[c]	23.22±0.68[c]	1.62±0.02[c]	0.71±0.03[c]
C/BB 70:30+EDTA	11,80±0.15[c]	18,22±0.71[d]	62,20±2.48[d]	2.87±0.11[d]	2.59±0.09[d]
C/Q 80:20+EDTA	9,41±0.20[d]	22.80±0.21[e]	47.63±0.56[e]	2.48±0.05[e]	0.75±0.01[c]

DaFe: dialyzability of Fe; DaZn: dialyzability of Zn; CL: Cooking loss; DRI $_{Fe}$, DRI $_{Zn}$: % recommended daily intake of Fe and Zn. Each value represents the mean±S.D. of three determinations. Differents letters for each column represent statistical differences between samples ($p<0.05$)

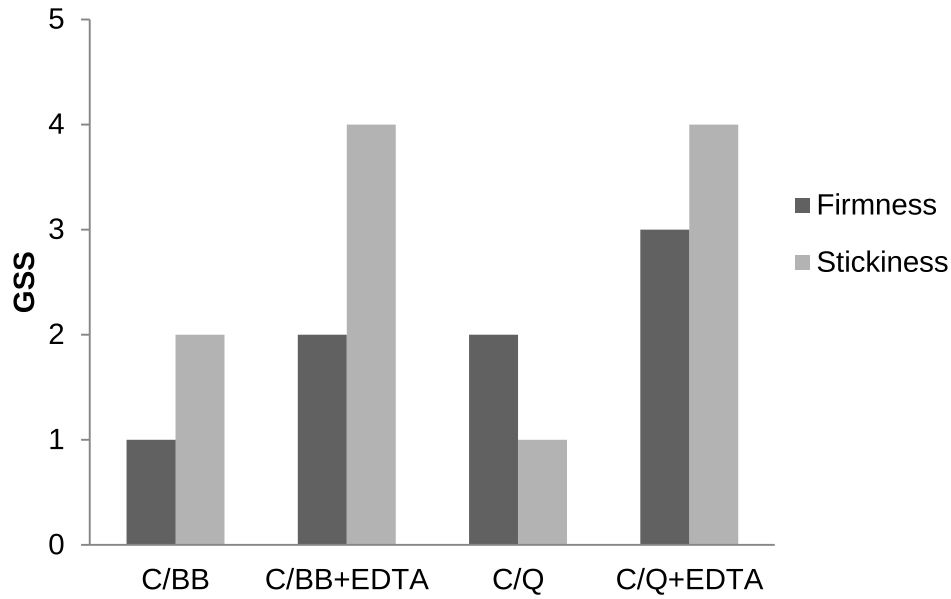
Figure(s)

Figure 1 Effect of EDTA on the potential availability of Fe and Zn corresponds to a 120-g ration of PNI



C: control sample, C/Q: PNI with 20 % of quinoa , C/BB: PNI with 30 % of broad bean.

Figure 2 Effect of EDTA on Global Sensory Score of PNI



C/Q: PNI with 20 % of quinoa, C/BB: PNI with 30 % of broad bean.