EFFECT OF DURUM WHEAT SEMOLINA SUBSTITUTION WITH BROAD BEAN FLOUR (*Vicia faba*) ON THE MACCHERONCCINI PASTA QUALITY

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ABSTRACT

This study investigated the effect of *Maccheronccini* fortification with different levels (10, 30 and 50%) of broad bean flour (*Vicia faba*) on its nutritional and technological quality.

Incorporation of the legume flour significantly increased protein, dietary fibre, and mineral contents of produced pasta ($P<0.05$). The mineral and protein dietary reference intake contributions (DRIs) were higher in enriched pasta considering an intake of 200g/day/person (cooked pasta). Cooking losses were relatively low regardless of the substitution level. Colour parameters of produced pasta indicated comparable brightness and higher redness values for enriched pasta. Higher levels of phytates were also found which could compromise iron bioavailability as was predicted through phytate/mineral molar ratios which remained higher the inhibitory threshold values for calcium (Ca) and iron (Fe) intestinal absorption. Enriched pasta showed significantly lower glycaemic index and slightly greater percent protein digestibility as regard to the control.

Produced *Maccheronccini* pasta had good technological properties with regard to colour and cooking behaviour. Moreover, contribution to DRIs and nutritional value were enhanced upon broad bean flour addition.

KEYWORDS: broad beans, *Maccheronccini* pasta, dietary reference intake, nutritional quality, cooking properties, colour parameters

INTRODUCTION

Pasta is a very popular food in several countries around the world. It is a cheap product, easy to prepare and well accepted by all age groups. Furthermore, it has a long shelf life and its consumption has largely increased over the last years. Pasta is considered as a healthy food since it presents low sodium content and produces a low glycaemic response and provides good amounts of complex carbohydrates, protein and vitamin B [1, 2]. Because of its characteristics, pasta was among the first foods to be authorized for enrichment by the Food and Drug Administration.

Broad beans consumption is popular in the Middle East, North Africa and South America. They represent a source of energy, protein, folic acid, niacin, vitamin C, magnesium, potassium, iron and dietary fibre [3, 4, 5]. Legume proteins are known to contain high levels of lysine and threonine, two essential amino acids that lack in cereal products [6]. Hence, they represent an adequate complement to cereal proteins [1].

Epidemiologic studies have indicated that legume consumption is inversely associated with the risk of coronary heart disease, type II diabetes mellitus and obesity [7, 8]. Furthermore, it promotes a slow post-prandial blood glucose increase due to the presence of a high amount of non-digestible carbohydrates including resistant starch and fibre [9].

Many authors have investigated the technological and nutritional quality of legume-fortified pasta [10, 11]. Moreover, high levels of substitution can decrease the quality of pasta products [12, 13]. Fortification of pasta sometimes produces particle aggregation which renders the feeding of the extrusion screw difficult during mixing [10]. Consequently, the choice of pasta fortification level is a compromise between improving its nutritional quality, and getting a product with acceptable technological properties. Despite the nutritive value of broad beans, there have been reports of the presence of anti-nutritional factors, such as phytic acid, tannins, trypsin and chymotrypsin inhibitors and α -galactosides [14]. Phytic acid is strongly negatively charged and thus has a great potential for complexing positively charged multivalent cations such as calcium, zinc and iron. This has adverse effects on mineral bioavailability, owing to the formation at physiological pH values of insoluble complexes which are not absorbable in the human gastrointestinal tract [15]. The antinutritional effect of phytic acid depends on both the concentration of phytates and minerals as well as the strength of the binding [16].

The aim of this study was to investigate the utility of *Vicia faba* produced in Algeria as ingredient to produce *maccheronccini* pasta with high amount of broad bean flour by semolina substitution (up to 50%) and to evaluate their technological and nutritional quality. The *Maccheronccini* shape was chosen because it is thicker and less fragile than the commonly used spaghetti and is therefore more appropriate for enrichment.

MATERIALS AND METHODS

Materials

The durum wheat semolina was of the type commonly used for pasta production and was kindly supplied by *Belladauna* factory (Foggia, Italy). Broad beans were cultivated and harvested in a mountainous region (Feraoun township, Bejaia-Algeria), dehulled and ground with a traditional mill situated in the same area. The obtained flour was then sieved to pass through a 500 μm mesh sieve.

Pasta production

Pasta produced was of the type *Maccheronccini*, fabricated in a pilot scale made of an extruder and a dryer (Italpast, Parma - Italy) by mixing durum wheat semolina with water at 41°C, in order to obtain a final water content of 44–45% in the dough (control sample). Fortified formulations were prepared by replacing durum wheat semolina with broad bean flour in 3 levels: 10, 30 and 50%. The final water content was obtained by adding different amounts of water as a function of the percentage of semolina replaced by broad bean flour. Increasing bean flour levels resulted in a slight increase of dough firmness and a decrease of stickiness but these changes did not affect the pasta-making procedure that was performed under the same conditions (temperature 50°C, kneading time 15 min; vacuum degree 700 mm Hg) independently of formulation with the exception of the pressure that ranged between 60 and 125 atm as a function of the specific dough composition. Pasta was dried at 55°C for 16 h. This temperature was applied from the beginning to the end of the drying cycle and then pasta was equilibrated to room conditions. The relative humidity of the hot air was ~50%. After drying, part of the samples was stored in polyethylene bags for cooking experiments. The rest was milled (Moulinex grinder, France) and sieved to pass through a 500 μm mesh sieve for chemical analysis. Obtained powders were stocked at 4°C in polyethylene tubes.

Chemical composition

Raw materials and milled pasta samples were analyzed for moisture (Method 925.09, AOAC [17]), ash (Method 08-03, AACC [18]), total lipids (Method 945.16, AOAC 1996 [17]), proteins (Kjeldahl, Method 46-13, AACC [18]) and starch (Method 996.11., AOAC 1996 [17]). Total, soluble and insoluble dietary fibre were determined according to the Official Method 991.43 [18]. The resistant starch of cooked pasta, considered as the starch fraction not hydrolysed *in vitro* by pancreatic α -amylase, was determined according to the methodology described by McCleary et al [19]. All measurements were done at least in triplicate.

Determination of minerals

The total Fe, Ca and Zn concentrations in raw material, uncooked and cooked pasta samples were determined in triplicate using a flame atomic absorption spectrometer. Previously, samples were placed in a Teflon perfluoroalkoxy (PFA) vessel and treated with HNO₃ (14 M) and H₂O₂ (30% v/v). The Teflon PFA vessel was irradiated at 800 W (15 min at 180°C) in a Microwave Accelerated Reaction System (MARS) from CEM (Vertex, Spain). At the end of the digestion program, the digest was placed in a polypropylene tube and made up to final volume with 5% HCl.

Phytate determination

Phytate content of raw material, uncooked and cooked pasta was measured by using the enzymatic kit (K-Phyt 07/11 Megazyme, Ireland 2011). The method is based on phytic acid (myo-inositol hexakisphosphate) hydrolysis by phytase (myo-inositol (phosphate) + inorganic phosphate (Pi)) and further myo-inositol (phosphate) hydrolysis by alkaline phosphatase. The resulting Pi reacts with ammonium molybdate and lead to the formation of molybdenum blue. The amount of molybdenum blue formed is proportional to the amount of Pi present in the sample and is measured by the increase of absorbance at 655 nm. Pi is quantified as phosphorus from a calibration curve generated using standards of known phosphorus concentration.

***In vitro* starch digestion and GI**

In vitro starch digestion and glycaemic index estimation was employed according to the modified method reported by Sanz-Penella et al. [20]. Total starch content was determined by the AOAC Official Method 996.11 [17] and the area under the curve (AUC) from 0 to 120 min and total digestible starch was used to calculate the *in vitro* glycaemic index value normalized against white bread (SigmaPlot software, Version 12.0) expressed as a percentage. Measurements were done in quadruplicate.

Pasta cooking procedure

The cooking procedure of Gelencsér et al. [21] was followed with slight modifications. Briefly, 25g of pasta were cooked (triplicate) in 250 mL of boiling distilled water to the optimal cooking time which corresponds to the time required to get complete gelatinisation of starch. This latter was determined by removing a piece of

pasta from the water at 30 s intervals and pressing it between fingers. After cooking, the samples were rinsed with distilled water and allowed to drain for 2 min.

Cooking properties

Cooking loss was evaluated after combining the cooking and rinse waters in a baker. Aliquots of 1 mL were transferred to a pre-weighed microfuge tubes and concentrated in a concentrator (Eppendorf Concentrator 5301 AG 22331, Germany) to evaporate the water. Microfuge tubes were then dried overnight at 105°C to constant weight. The resulting weighed residue is reported as a percentage of the original pasta sample.

Water absorption of drained pasta is evaluated by weighing the cooked pasta and reported as $(W_1 - W_2)/W_2$, being W_1 : weight of cooked product, g, and W_2 : weight of raw pasta, g.

For ash determination 10 mL of cooking water were transferred to a previously heated and pre-weighed porcelain vessels and left to dehydrate in a sand bath overnight at 40°C. The vessels were then ashed in a muffle furnace at 600°C for 2 hours and weighed after cooling. The difference is reported as percentage of the initial dry pasta. Samples were analysed in quadruplicate, unless cooking loss which was performed in hexaplicate.

Colour of pasta

Colour of cooked and uncooked pasta was determined by a Chromameter (Konika Minolta, Sensing INC, Japan) using the Hunter L^* , a^* and b^* . L^* values measure black to white (0–100); a^* values measure redness when positive and greenness when negative; b^* values measure yellowness when positive and blueness when negative. Each data colour represents the mean of three measurements on different pasta samples.

Statistical analysis

Multiple sample comparison of the means (ANOVA) and Fisher's least significant differences (LSD) were applied to establish statistical significant differences between samples. All statistical analyses were carried out with the software Statgraphics Plus 7.1 (Bitstream, Cambridge, MN) and differences were considered significant at $P < 0.05$.

RESULTS AND DISCUSSION

Chemical composition of raw materials

The nutrient content of durum wheat semolina and broad bean flour are shown in Table 1. While starch content is the main compound in semolina, the amount of proteins, lipids, fibre and ash is found to be significantly ($P<0.05$) higher in bean flour. As expected from its higher ash content, the amount of calcium, iron and zinc is significantly more important in broad bean flour. Legumes are known to contain relatively higher amounts of antinutrient factors than cereals [22]. This is supported by our study that shows an increase ($P<0.05$) of phytate content in bean flour.

Effect of replacement durum wheat semolina by broad bean flour on chemical composition of uncooked and cooked pasta

The greater level of proteins and ash registered in broad bean flour with regard to semolina (Table 1) directly affected the increase ($P<0.05$) of these parameters in fortified pasta as expected (from 14.6 to 20.3 g 100g dm^{-1}) in traditional and 50% broad bean enriched pasta respectively). In the same tendency, Torres et al. [23] developed pasta products with lupine and found an enhancement of their nutritional quality. Cooking of pasta did not have any effect on protein content. This result shows that protein loss in cooking water is not significantly important. Similar results were found by Sissons et al. [24], who described no relationship of cooking loss with protein content in pasta. Fibre (total and insoluble) and resistant starch contents also increased significantly in enriched pasta, whereas, soluble dietary fibre remained almost constant. This is in accordance with the findings of Sanz-Penella et al. [25] when studying amaranth enriched bread. Fortification of pasta with broad bean flour induced an increase in the mineral content. This result is predictable since the amount of minerals is significantly higher in beans (Table 1). Thus, the highest mineral content is found in 50% enriched pasta (1.69 g 100 g dm^{-1} instead of 0.89 g 100 g dm^{-1} in traditional *Maccheronccini*). Cooking of pasta had a negative effect on ash content and subsequently on the tested minerals (Ca, Fe and Zn) due to leaching of soluble material into cooking water. While 10% enriched pasta didn't show significant loss in calcium content, the decrease of this mineral was significantly important in 30 and 50% enriched pasta ($P<0.05$). An opposite trend was observed for the cooking effect on iron content, since this latter remained constant in all the formulations. Zinc content decreased only in cooked pasta with high broad bean substitution levels (30 and 50%) taking in account that cooking loss is significantly higher in samples with beans (Figure 2). As seen in Table 2, increasing broad bean substitution level resulted in high phytate values in dry pasta (0.17, 0.33, 0.46 and 0.83 g 100 g dm^{-1} in the control, 10, 30 and 50% enriched pasta respectively). High phytate content indicates that pasta preparation

steps including drying were not effective on phytate destruction due to heat-stable properties of phytic acid. [25] Cooking process was able to decrease phytate content only in 50% fortified pasta.

Contribution of proteins and microelements to the relevant dietary reference intakes (DRI s) for consumption of a daily average portion of 200g of cooked pasta

Table 3 shows the contribution of mineral intake from traditional and enriched pasta to the DRIs given by the Food and Nutrition Board of the Institute of Medicine, National Academy Science [26, 27]. When expressed in terms of DRIs, traditional pasta contributes 54.0 to 65.7% of the protein recommendation for male and female adults, respectively (Table 3). Whereas, enriched pasta could supply significantly higher amounts of protein, from 61.2 to 74.7% (10% enriched pasta and 50% enriched pasta, respectively) in the case of adult males and from 74.5 to 91.0% in the case of adult females, which is close to the complete supply of protein. Regarding minerals, consumption of pasta provides only 1.9-2.47 to 2.22-2.88% (traditional and 50% enriched pasta, respectively) of the daily requirements of this macro-element for adult male and female, whatever are their ages. Thus, a pasta based diet should be accompanied by a product that is rich in Ca to make a balanced diet. While consumption of traditional pasta provides 22.65% of iron recommendation for young, adult and over 70 aged males, and 10.06% for adult females, broad bean fortified pasta increases this contribution from 39.23 to 54.55% (10% to 50% enriched pasta, respectively) for males and from 17.44 to 24.19% (10% to 50% enriched pasta, respectively) for adult females, who have greater needs of iron (Table 3). The same tendency was observed with zinc, where increasing bean flour substitution levels in pasta resulted in higher zinc contribution to dietary requirements, from 19.74 to 28.5% (control and 50% enriched pasta, respectively) for young males and young, adult and more than 70 years old aged females. Phytate (*myo*-inositol hexakisphosphate) is a form of phosphorus storage in cereals and legumes. Its content in cooked pasta varied from 0.22 to 0.56 g 100 g dm⁻¹ (control and 50% fortified pasta respectively). It is known that phytic acid can reduce the bioavailability of divalent minerals by formation of insoluble complexes in the gastrointestinal tract [15]. Hence, the predicted intakes that are derived from DRIs for minerals in this study are overestimated. In order to evaluate the effect of the resulted higher phytate presence in enriched pasta on the bioavailability of the tested minerals (Ca, Fe and Zn), the phytate/mineral molar ratios has been investigated [28]. Phytate/calcium molar ratio could induce less calcium bioavailability in humans at values higher than 0.24. Regarding iron, the molar ration is more than 1; whereas if the phytate/Zn molar ratio is higher than 5, the bioavailability of Zn could be less than 50% [28]. The phytate /Ca molar ratios varied from 0.08 to 0.34 (control and 50% enriched pasta, respectively). Thus, 50% substituted

pasta will adversely affect the availability of this element. The same tendency is observed for zinc. Indeed, phytate/Zn ratio is superior to 5 only in the case of 50% enriched pasta (1.9, 2.8 and 3.9 in the case of pasta substituted with 0, 10 and 30% bean flour respectively). Consequently, the amount of phytic acid present in traditional and fortified pasta can reduce the bioavailability of zinc only at the highest substitution level. On the other hand, molar ratios of iron were 1.4, 1.6, 1.9 and 2.9 (in the control, 10, 30 and 50% enriched pasta, respectively). Therefore, iron intake could be limited by the presence of phytates in both traditional and fortified pasta.

Enzymatic digestion

The kinetic curves of traditional and enriched pasta products with comparison to white bread taken as a reference are shown in (Figure 1). White bread showed the highest levels of starch hydrolysis compared to traditional and enriched pasta. Increasing broad bean substitution levels resulted in significantly ($P<0.05$) lower values of GI (95.7, 89.8, 84.8 and 83.3 for control, 10, 30 and 50% enriched pasta, respectively). In general, GI depends on the food texture and particle size, type of starch, degree of starch gelatinization, physical entrapment of starch molecules within food, food processing and other ingredients [20]. Legume seeds, due to their poor digestibility related to the inherent physical and structural properties of starch, generally exhibit higher resistant starch content (Table 2) and lower GI (Figure 1) when compared to cereal grains [29]. Petitot et al. [11] also reported that high levels of legume flour addition induce minor structural changes in pasta, keeping low its glycaemic index.

Cooking properties

Fortification of pasta with broad bean flour decreased significantly the optimal cooking time (Figure 2). This reduction was accompanied by a lower water uptake and a higher cooking loss. A physical disruption of the gluten network due to the presence of fibre may have facilitated the penetration of water to the core of pasta [1]. The higher water uptake in the control is possibly due to its higher starch content. Indeed, Dexter et al. [30] found cooked weight to be directly related to degree of swelling of starch granules. Sozer et al. [31] also reported that longer optimal cooking time induced higher water uptake since more water can diffuse and interact with both starch and protein matrices.

Durum wheat protein are mainly composed of glutenins and gliadins that are insoluble and have the propriety to form intra and inter-molecular disulphide bonds during processing of pasta or bread, which leads to the

formation of a strong tridimensional gluten network having the ability to entrap starch granules. On the other hand, legumes proteins are essentially composed of salt-soluble globulins and water-soluble albumins [32]. Thus, the increase of cooking loss in enriched pasta may be due to the introduction of non-gluten proteins that diluted and weakened the gluten network strength. The weakening of pasta structure results in leaching of more dry material in cooking water as shown in Figure 2. These results are consistent with those reported by Torres et al. [13] and Petitot et al. [11]. It is described that the main components that leach into the cooking water are amylose and some salt-soluble proteins [11]. In the current work the cooking loss ranged between 4.1 and 5.5% for control pasta (100% semolina) and 50% enriched pasta, respectively (Figure 2). No loss of starch or proteins seems to be occurred (Table 2). It was reported that during cooking, insoluble fibre fractions may get suspended in viscous gelatinized starch and prevented amylose from leaching into the cooking water as well as the formation of lipid-amylose-complexes that probably contributed to a decrease in the amylose loss [33,34]. In the present investigation the increment of bean flour in formulations had a slight effect on this parameter, affected basically by the mineral loss after cooking (Table 2). All the samples lost approximately 4% of solids (not minerals) during cooking which could be represented by soluble compounds occurring in pasta like soluble carbohydrates and/or fibre. Our results show cooking losses inferior to 10% in all formulation, indicating good cooking quality products [35]. The mineral content increased when the enrichment level increase (Table 2), mineral loss in cooking water also increase with the inclusion of bean (from 0.0087% in the control to 0.01-0.013 % in fortified pasta respectively).

Colour parameters

Pasta colour is an essential quality parameter. Generally consumers prefer pasta with a bright yellow colour [36]. Fortification of pasta with broad bean flour did not affect their brightness (L^* values) at all substitution levels. Cooking process did not have influence on control pasta lightness (Figure 3), but it significantly decreased ($P<0.05$) in enriched pasta. These results don't agree with those reported by Wood [10] and Zhao et al. [37] who found a decrease in brightness of legumes fortified pasta. A significant increase in redness (a^* values) was also observed in enriched cooked and uncooked pasta compared to the control. This increase may be due to the development of Maillard reaction during pasta drying cycle. b^* values, indicating yellowness decrease significantly in 10 and 30% enriched pasta, but were comparable to the control in 50% enriched pasta. In cooked pasta, yellowness decreased at all the substitution levels ($P<0.05$). The comparison between uncooked and cooked pasta indicates differences in yellowness in the case of the control and 10% pasta. However, 30% and

50% pasta yellowness remained constant when cooked. The total colour difference between control sample and pasta with bean flour, ΔE , were lower or equal than five units, indicating that not significant differences are perceptible to consumers by visual observation.

CONCLUSION

Our study indicated that it was possible to produce *Maccheronccini* pasta with good technological properties with regard to aspect, colour and cooking properties even with high legume substitution levels (50%). The addition of broad bean flour to semolina in pasta-making significantly improved its nutritional quality, translated into higher amounts of protein, dietary fibre and minerals (especially iron and zinc). The mineral DRIs contribution was higher in pasta formulations with bean flour although higher levels of phytates were also found which could compromise the mineral availability. Pasta cooking process did not affect protein content at all the substitution levels getting higher value of DRI contribution after consuming 200 g/day/person. Consumption of bean enriched pasta change matrix structure and composition which could also have health benefits by reducing the glycaemic index.

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FIGURE CAPTIONS

Figure 1. **A.** Kinetic curves of different pasta formulations and white bread as a reference; **B.** Decrease in pH of pasta during in vitro protein digestibility measurement.

Figure 2. Cooking properties of pasta.

Figure 3. Aspect of dry raw pasta. Pasta formulation: A. control, B. 10% broad bean enriched pasta, C. 30% broad bean enriched pasta, D. 50% broad bean enriched pasta.

Table 1. Chemical composition of raw material

	Durum wheat semolina	Broad bean flour
Main components (g 100g d.m⁻¹)		
Moisture	12.0±0.1 ^b	10.7±0.2 ^a
Starch	78.6±0.1 ^b	46.6±0.1 ^a
Proteins	15.9±0.1 ^a	30.9±0.5 ^b
Lipids	2.02±0.04 ^a	2.72±0.16 ^b
Ash	0.95±0.01 ^a	2.97±0.01 ^b
Dietary fibre (g 100g d.m⁻¹)		
Insoluble	2.39±0.07 ^a	14.06±0.36 ^b
Soluble	2.40±0.45 ^a	2.91±0.98 ^a
Total	5.96±0.6 ^a	17.1±1.5 ^b
Mineral content (µg g d.m⁻¹)		
Ca	113±16 ^a	321±29 ^b
Fe	11.3±0.6 ^a	41.3±1.6 ^b
Zn	8.0±0.6 ^a	27.1±1.5 ^b
Phytates (g 100g d.m⁻¹)	0.35±0.02 ^a	1.17±0.02 ^b

^{a-b} Mean±SD, n≥3. Values followed by the same letter in the same line are not significantly different at 95%

confidence level.

d.m. dry matter

Table 2. Effect on durum wheat semolina after addition of broad bean flour on chemical composition of uncooked and cooked pasta

Component	Units	Cooking	% of bean flour in pasta formulation			
			Control	10	30	50
Main components						
Moisture	g 100g	Uncooked	9.68±0.02 ^d	8.96±0.04 ^{bc}	8.85±0.06 ^b	8.67±0.03 ^a
Starch	g 100g d.m ⁻¹	Uncooked	76.9±0.2 ^d	68.5±0.3 ^c	66.3±0.1 ^b	63.2±0.1 ^a
		Cooked	75.9±0.5 ^d	68.2±0.4 ^c	66.0±0.3 ^b	63.0±0.4 ^a
Proteins	g 100g d.m ⁻¹	Uncooked	14.6±0.4 ^a	17.1±0.2 ^b	18.3±0.1 ^c	20.3±0.5 ^d
		Cooked	15.1±0.2 ^a	17.13±0.02 ^b	18.0±0.02 ^c	20.9±0.2 ^d
Lipids	g 100g d.m ⁻¹	Uncooked	2.02±0.04 ^b	2.09±0.01 ^b	1.95±0.01 ^a	1.89±0.01 ^a
		Cooked	n.d	n.d	n.d	n.d
Ash	g 100g d.m ⁻¹	Uncooked	0.89±0.01 ^c	1.19±0.01 ^d	1.35±0.01 ^e	1.69±0.03 ^f
		Cooked	0.53±0.01 ^a	0.72±0.08 ^b	0.82±0.02 ^c	0.97±0.05 ^{cd}
Dietary Fibre						
Insoluble	g 100g d.m ⁻¹	Uncooked	2.9±1.0 ^a	3.3±0.2 ^a	3.7±0.6 ^{ab}	5.5±0.4 ^b
Soluble	g 100g d.m ⁻¹	Uncooked	1.7±0.3 ^{ab}	1.1±0.1 ^a	1.2±0.3 ^a	2.4±0.3 ^b
Total	g 100g d.m ⁻¹	Uncooked	5.2±0.1 ^a	5.6±0.5 ^{ab}	5.8±1.0 ^{ab}	7.1±0.2 ^b
Resistant Starch	g 100g d.m ⁻¹	Cooked	1.44±0.02 ^a	1.59±0.03 ^b	1.72±0.01 ^c	1.99±0.01 ^d
Minerals						
Ca	µg g d.m ⁻¹	Uncooked	148.2±0.1 ^b	149.31±0.3 ^b	186.0±0.6 ^c	190.0±0.5 ^c
		Cooked	129.8±2.6 ^a	150.1±3.6 ^b	147.6±0.4 ^a	144.3±0.01 ^a
Fe	µg g d.m ⁻¹	Uncooked	9.58±0.07 ^a	15.11±0.06 ^b	19.31±0.14 ^c	24.78±0.86 ^d
		Cooked	9.98±0.34 ^a	17.33±0.04 ^e	20.0±1.0 ^c	23.9±1.0 ^d
Zn	µg g d.m ⁻¹	Uncooked	8.38±0.17 ^a	10.74±0.3 ^b	12.9±0.65 ^c	15±0.54 ^d
		Cooked	8.7±0.4 ^a	11.5±0.1 ^{bd}	11.7±0.3 ^{de}	12.5±0.1 ^{ce}
Phytic Acid						
	g 100g d.m ⁻¹	Uncooked	0.17±0.01 ^a	0.33±0.01 ^b	0.46±0.01 ^c	0.83±0.02 ^d
		Cooked	0.22±0.01 ^e	0.39±0.01 ^f	0.49±0.01 ^g	0.56±0.01 ^h

^{a-h} Mean±SD, n=3. Values followed by the same letter in the same line and the same column for a common parameter are not significantly different at 95% confidence level.

d.m. dry matter.

n.d. not determined.

Table 3. Contribution of proteins and microelements to the relevant dietary reference intakes (DRIs) for consumption of a daily average portion of 200g of cooked pasta

Nutrient	Gender	DRIs	% of bean flour in pasta formulation			
			Control	10	30	50
Proteins, g/day	Adult male					
	14-18	52	58.1	65.9	69.4	80.5
	31-50	56	54.0	61.2	64.4	74.7
	≥70	56	54.0	61.2	64.4	74.7
	Adult female					
	14-18	46	65.7	74.5	78.4	91.0
	31-50	46	65.7	74.5	78.4	91.0
	≥70	46	65.7	74.5	78.4	91.0
	Minerals, mg/day	Male and female				
Ca						
	14-18	1300	1.9	2.34	2.27	2.22
	19-70	1000	2.47	3.05	2.95	2.88
	≥70	1200	2.06	2.54	2.46	2.4
Fe	Male					
	14-18	11	16.47	28.53	33.09	39.59
	9-13, 31-70, ≥70	8	22.65	39.23	45.5	54.44
	Female					
	14-18	15	12.08	20.92	24.27	29.03
	19-50	18	10.06	17.44	20.22	24.19
Zn	9-13, 31-70, ≥70	8	22.65	39.23	45.5	54.44
	Male					
	9-13	8	19.74	26.11	26.53	28.5
	31-50	11	14.36	18.99	19.29	20.73
	Female					
	14-18	9	17.54	23.21	23.58	25.34
	9-13, 31-70, ≥70	8	19.74	26.11	26.53	28.5

DRI (Dietary Reference Intakes) contribution (%) for a daily average intake of 200 g of pasta if the protein mineral absorption inhibitors are absent, NAS (2005, 2015).

Table 4. Colour parameters of uncooked and cooked pasta

Parameter	Cooking	% of bean flour in pasta formulation			
		Control	10	30	50
L^*	Uncooked	85.5±1.8 ^{de}	84.6±0.7 ^{cd}	85.7±0.6 ^d	84.9±1.2 ^{cd}
	Cooked	84.8±1.1 ^{cd}	81.5±0.2 ^a	83.6±0.1 ^{bc}	82.2±0.3 ^{ab}
a^*	Uncooked	-1.94±0.08 ^b	-1.19±0.01 ^e	-1.49±0.05 ^c	-1.84±0.07 ^b
	Cooked	-2.16±0.09 ^a	-1.19±0.04 ^e	-1.3±0.01 ^{de}	-1.42±0.14 ^{cd}
b^*	Uncooked	19.8±0.8 ^d	14.2±0.2 ^a	15.3±0.5 ^b	17.7±0.4 ^d
	Cooked	20.4±0.5 ^e	16.5±0.1 ^c	15.7±0.1 ^b	17.5±0.3 ^d
C	Uncooked	17.9±0.8 ^d	14.2±0.2 ^a	15.4±0.5 ^b	17.8±0.4 ^d
	Cooked	20.5±0.5 ^e	16.7±0.4 ^c	15.7±0.1 ^b	17.5±0.3 ^d
h	Uncooked	96.2±0.2 ^d	94.8±0.0 ^b	95.6±0.4 ^a	95.9±0.1 ^{cd}
	Cooked	96.1±0.4 ^d	92.8±0.0 ^a	94.8±0.1 ^b	94.6±0.4 ^b
ΔE^*	Uncooked	--	3.8	2.5	0.7
	Cooked	--	5.1	4.8	4.0

^{a-c} Mean±SD, n=3. Values followed by the same letter in the same line and the same column for a common parameter are not significantly different at 95% confidence level.

Figure1.

Figure2.

Figure 3.