

Influence of soaking and cooking on the thiamin, riboflavin and niacin contents of legumes

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Abstract

The effects on thiamin, riboflavin and niacin contents of faba beans (*Vicia faba*, L.), chickpeas (*Cicer arietinum*, L.) and lentils (*Lens culinaris*, L.), of soaking in different solutions (citric acid solution pH 4.96 ± 0.02 , distilled water pH 7.00 ± 0.02 and sodium bicarbonate solution pH 7.85 ± 0.02) and cooking of the presoaked legumes in distilled water have been studied. The main factor determining the vitamin retention after each treatment was the legume genus. Soaking faba beans for 9 h produced losses of 0 and 15% thiamin, 0 and 11% riboflavin and no changes in niacin content ($P \leq 0.05$). In the case of chickpeas the effect of the treatment was more acute. Losses of 0–18% thiamin, 0–4% riboflavin and 0–46% available niacin was observed while in lentils, the thiamin and available niacin contents decreased by 5–10% and 26–42%, respectively and the riboflavin increased by up to 98%. In general, vitamin losses were greater when soaking was carried out in alkaline solution. In most of the studied legumes, cooking produced further decreases in vitamins. Faba beans lost up to 35 and 32%, respectively, of their thiamin and available niacin contents, while riboflavin was not affected. Chickpeas and lentils were more liable to lose their vitamins: up to 51% thiamin, 66% riboflavin and 78% available niacin in chickpeas and up to 61% thiamin and 61% available niacin in lentils. Cooking did not cause any additional loss of the riboflavin content of lentils. With few exceptions, cooking caused greater vitamin losses when a prior soaking was carried out in alkaline solution.

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1. Introduction

Food legumes form an important part of the human diet, providing a high proportion of proteins, fats, carbohydrates, dietary fibres, B-group vitamins (thiamin, riboflavin, niacin), and minerals. Worldwide, most grown legumes are soybean, peanut, beans, peas, chickpeas and lentils (Kadam & Salunkhe, 1989). In many developing countries they are the main sources for human and animal nutrition. In the developed countries, legumes have an increasing use in dietetic formulations in the treatment and prevention of diabetes, cardiovascular diseases, cancer of colon, and lowering of blood cholesterol levels (Brand, Snow, Nabhan, & Truswell, 1990), which indicates their possible therapeutic value in humans. Nowadays, legumes are

essential raw material for the modern food industry in the production of protein concentrates, fats and starches, and functional food ingredients (protein isolates, protein hydrolysates, dietary fibres, lecithin and isoflavones) (Salunkhe & Kadam, 1989; Linden & Lorient, 1994).

Dry grain legumes, however, contain several anti-nutritional factors, such as α -galactosides, trypsin and chymotrypsin inhibitors, phytates and lectins (Vidal-Valverde, Frias, & Valverde, 1992), which can impede the availability of nutrients and thus limit their consumption. α -Galactosides are known to be responsible for flatulence in humans, trypsin and chymotrypsin inhibitors are able to bind hydrolytic enzymes, such as trypsin and chymotrypsin, inhibiting their activity, whereas phytic acid is found to reduce the availability of some minerals. Removal of these factors is therefore essential for improving the nutritional quality of legumes and, subsequently, to increase their potential as human food. Some simple and inexpensive processing

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techniques, such as soaking and cooking, are highly efficient for the reduction of these antinutritional factors and for improving legume organoleptic quality.

Soaking is an integral part of a number of treatments, such as cooking, canning, germination, and fermentation. It consists of hydration of the seeds in water, usually until they reach maximum weight, with or without discarding of the soaking medium, and the results obtained depend of factors such as legume genus, species and variety, process duration, temperature, pH, salinity of the soaking media, and also the storage conditions undergone before processing. Numerous studies indicate that soaking can reduce the levels of total sugars, α -galactosides, minerals, phytic acid and proteolytic enzyme inhibitors (Frias, Vidal-Valverde, Sotomayor, Diaz-Pollan, & Urbano, 2000; Vidal-Valverde, Frias, Estrella, Gorospe, Ruiz, & Bacon, 1994; Vidal-Valverde et al., 2002), which can be partly or totally solubilised and eliminated with the discarded soaking solution. During soaking some metabolic processes can take place and usually affect the soluble carbohydrate and riboflavin contents (Frias, Prodanov, Sierra, & Vidal-Valverde, 1995; Vidal-Valverde et al., 1992, 2002).

Cooking is probably the oldest treatment for making legumes edible. Usually it includes a previous soaking of the seeds and a subsequent cooking in boiling water until they become completely soft. Addition of mineral salts to the soaking and/or cooking media can produce a reduction of the cooking time (Lu, Hsu, & Wilson, 1984; Van Buren, 1986). In general, cooking produces denaturalisation of proteins and their diffusion to the liquid phase (Haytowitz & Matthews, 1983), inactivation of heat-sensitive factors, such as trypsin inhibitors (Frias et al., 2000), decrease of phytic acid (Iyer, Kadam, & Salunkhe, 1989; Khalil & Mansour, 1995; Vidal-Valverde et al., 1994) and α -galactoside contents (El-Adawy, 2002).

Soaking and/or cooking can cause considerable losses in some essential nutrients, such as hydrosoluble vitamins, due to their high solubility and thermal instability (Edijala, 1980; Abdel-Hamid, 1983; El-Adawy, 2002).

Although some research has been carried out on the chemical composition of legumes, not much work has been carried out on the effect of simple processes, such as soaking and/or cooking on their vitamin contents. Literature reports are mainly related to soy, peanuts and different species of *Phaseolus* beans, whereas information concerning chickpeas, faba beans and lentils is scarce and unsystematic. It is important to note that these three legumes are among the most cultivated in the world (Kadam & Salunkhe, 1989) and the most consumed in the Mediterranean countries. In addition, it is noteworthy that, most reports refer to both processes together, without distinction of the influence of each of them, which introduces additional confusion.

The present work is part of a complex study on the global improvement of faba bean, chickpea and lentil nutritional value by processing and comprises research on a wide range of antinutrient and nutrient substances, and their biological availability. Here, we offer a detailed study on the individual effects of soaking and cooking processes on the contents of some of the most significant hydrosoluble vitamins in chickpeas, faba beans and lentils—thiamin, riboflavin and niacin and the influence of some additives in the soaking medium, such as citric acid and sodium bicarbonate, on the vitamin retention after soaking and/or soaking and cooking. The results contribute to a better understanding of the effects of these processes on legume composition and allow selection of the best conditions for improving the quality of these legumes when they have to be processed for human or animal nutrition.

2. Materials and methods

2.1. Legumes

Lentils (*Lens culinaris* L., var. *vulgaris*), faba beans (*Vicia faba* L., var. *major*) and chickpeas (*Cicer arietinum* L., var. *kabuli*) were randomly obtained from the local market in Madrid (Spain). The whole raw seeds were ground and passed through an 0.18 mm sieve before analysis.

2.2. Soaking

Lentil, faba bean or chick pea seeds were mixed with 0.1% (w/v) citric acid solution (pH 4.96 ± 0.02), distilled water (pH 7.00 ± 0.02) or 0.07% (w/v) sodium bicarbonate solution (pH 7.85 ± 0.02) in a proportion of 1:3 (w/v) seeds:soaking medium. The legumes were allowed to imbibe water at room temperature for 9 h. The legume seeds were strained to drain off the soaking medium, and the soaked seeds were freeze-dried, ground, and passed through an 0.18 mm sieve before analysis.

2.3. Conventional cooking

After soaking and draining of the soaking medium, as described above, the legume seeds were cooked for 35 min in distilled water [seed:water ratio 1:6.7 (w/v)]. The cooking water was drained off and the seeds were freeze-dried, ground, and passed through an 0.18 mm sieve before analysis.

2.4. Vitamin determination

A single extraction procedure for thiamin, riboflavin and available niacin was carried out using acid (0.3 M HCl, 121 °C, 15 min) and enzymatic (20% Taka-Diastase

solution) digestion of the legumes (raw and treated) according to a previous paper (Vidal-Valverde et al., 1993). Thiamin, riboflavin and available niacin were determined by HPLC, as described by Prodanov, Sierra, and Vidal-Valverde (1997).

2.5. Statistical methods

Data were subjected to multifactor ANOVA using the least-squared difference test with the Statgraphic 5.0 Programme (Statistical Graphics Corporation, Rockville, MD, USA) for Windows using a PC-Pentium.

3. Results and discussion

The contents of thiamin, riboflavin and available niacin of raw, soaked and soaked and cooked faba beans, chick peas and lentils are collected in Table 1.

The thiamin content of faba beans did not change when the soaking was carried out in acid solution and a decrease (15%) was observed when it was carried out in water or basic solution ($P < 0.05$) (Table 1, Fig. 1). Riboflavin decreased (11%) only after soaking the faba beans in sodium bicarbonate while available niacin did

not change after soaking in any solution ($P \leq 0.05$). When faba beans were soaked in citric acid, water or sodium bicarbonate solution and cooked, the thiamin content decreased (19, 24 and 35%, respectively) while the riboflavin content did not change ($P \leq 0.05$). The available niacin contents decreased by 32 and 23% ($P \leq 0.05$), in the cases when faba beans were soaked in water or sodium bicarbonate, respectively, and cooked (Table 1, Fig. 1).

The thiamin contents of chickpeas decreased by 11, 7 and 18% after soaking in citric acid, water or sodium bicarbonate solution, respectively ($P \leq 0.05$). The riboflavin content of chickpeas did not change after soaking in water, but decreased 14 and 13% when the process was carried out in acidic or basic medium ($P \leq 0.05$), while the available niacin decreased after soaking in water or in sodium bicarbonate solution by 26 and 46%, respectively. Cooking of the pre-soaked chickpeas in acid, water or basic medium produced decreases of the thiamin, riboflavin and available niacin contents (45, 47, 51%), (53, 52, 66%) and (79, 72, 78%), respectively, (Table 1, Fig. 1).

When lentils were soaked in citric acid, water or sodium bicarbonate, decreases of the thiamin and available niacin contents were observed (5, 6, 10% and

Table 1
Influence of processing on the vitamin content of legumes^a

Legumes	Thiamin (mg/g d.m.)	Riboflavin (mg/g d.m.)	Available Niacin (mg/g d.m.)
Faba Beans	0.253±0.004d	0.123±0.009bc	1.52±0.068c
<i>Soaking</i>			
Citric acid soaking	0.253±0.004d	0.119±0.001ab	1.48±0.034c
Water soaking	0.216±0.009c	0.123±0.005bc	1.49±0.011c
Sodium bicarbonate soaking	0.215±0.002c	0.109±0.002a	1.44±0.094c
<i>Soaking and cooking</i>			
Citric acid soaking + cooking	0.205±0.007c	0.134±0.000 c	1.43±0.094c
Water soaking + cooking	0.192±0.003b	0.122±0.013 bc	1.03±0.026a
Sodium bicarbonate soaking + cooking	0.164±0.004a	0.113±0.003 ab	1.17±0.042b
Chickpeas	0.241±0.005f	0.118±0.001d	0.612±0.031d
<i>Soaking</i>	0.24±0.00f	0.12±0.00e	0.62±0.03d
Citric acid soaking	0.215±0.002d	0.102±0.005c	0.591±0.027d
Water soaking	0.224±0.001e	0.115±0.003d	0.452±0.022 c
Sodium bicarbonate soaking	0.198±0.005c	0.102±0.007c	0.332±0.039b
<i>Soaking and cooking</i>			
Citric acid soaking + cooking	0.132±0.003b	0.056±0.006b	0.130±0.012a
Water soaking + cooking	0.127±0.003b	0.057±0.003b	0.173±0.034a
Sodium bicarbonate soaking + cooking	0.117±0.002a	0.040±0.004a	0.135±0.017a
Lentils	0.433±0.006e	0.061±0.005a	1.16±0.142d
<i>Soaking</i>			
Citric acid soaking	0.409±0.003d	0.118±0.002d	0.671±0.042b
Water soaking	0.406±0.007d	0.120±0.001d	0.450±0.023a
Sodium bicarbonate soaking	0.389±0.009c	0.106±0.004c	0.863±0.061c
<i>Soaking and cooking</i>			
Citric acid soaking + cooking	0.200±0.012b	0.071±0.001b	0.594±0.045b
Water soaking + cooking	0.166±0.004a	0.069±0.002b	0.451±0.046a
Sodium bicarbonate soaking + cooking	0.169±0.002a	0.057±0.001a	0.872±0.057c

For each legume, the same letters in the same column means no significant differences ($P \leq 0.05$).

^a Mean value of at least four determinations±standard deviation.

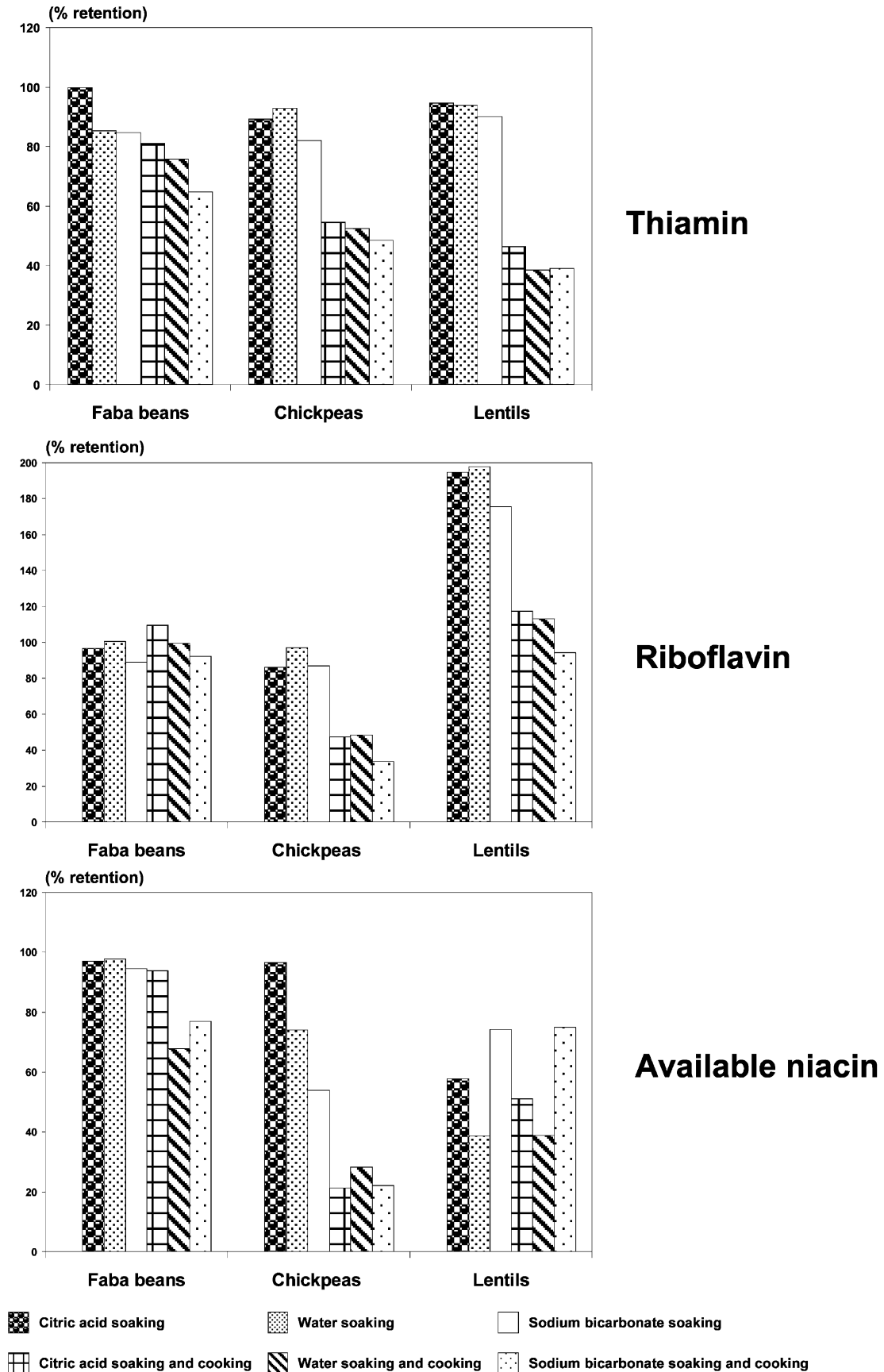


Fig. 1. Effect of processing on the thiamine, riboflavin and available niacin content of legumes.

42, 61, 26%, respectively) ($P \leq 0.05$), while the riboflavin content always increased (95, 98 and 76%, respectively). Cooking lentils in water (previous soaking in citric acid solution, water or sodium bicarbonate solution) produced a reduction in the thiamin and available niacin contents (54, 62, 61% and 49, 61, 25%, respectively), while the riboflavin content remained higher than in the raw seeds in the cases when a previous soaking was carried out in citric acid (17%) or in water (13%) (Table 1, Fig. 1).

According to these results, soaking produced no (or slight) changes in the thiamin contents of faba beans, chickpeas or lentils. The same effect was observed for the riboflavin contents of faba beans and chickpeas but, in lentils, a marked increase was found after soaking. The available niacin content did not change significantly after soaking faba beans in any solution or chickpeas in acid solution, but a marked decrease was observed when chickpeas were soaked in water or sodium bicarbonate solution and when lentils were soaked in any solution. Soaking and cooking of the legumes produced marked decreases in thiamin and available niacin contents of faba beans, chickpeas and lentils. The riboflavin content did not change after cooking the pre-soaked faba beans, decreased very considerably in chickpeas and increased in lentils when they were pre-soaked in acid or water medium. In general, cooking had a marked reduction effect on the vitamin content of the soaked legumes, except for the riboflavin content of soaked faba beans and available niacin of soaked lentils, where no change was observed ($P \leq 0.05$).

4. Discussion

From the above results (Table 1, Fig. 1), we can conclude that the behaviour of each legume in the soaking process was very specific and depended on the legume genus. Faba beans were the legumes least affected, with small losses in their vitamin contents; chickpeas were more affected and lentils showed the greatest changes: small decrease of the thiamin content, considerable loss of the niacin content and an important increment of the riboflavin content.

Vitamin losses after soaking can be explained by the multiple effect of several factors. First, it is necessary to consider the vitamin removal with the drained liquid after soaking. The faster the legume hydration, the faster is the mass transfer from the seeds to the liquid phase, which shows very well that the higher vitamin losses obtained in lentils corresponds to their faster hydration rate and the higher vitamin retention in faba beans corresponds to their slower hydration rate.

Second, it is known that, in vegetables, vitamins exist in different forms: thiamine as thiamin, mono-, di- and tri-phosphates; riboflavin as, flavin-mononucleotide

(FMN) and flavin-adenine dinucleotide (FAD) and niacin as nicotinic acid, nicotinamide, nicotinamide-adenine dinucleotide (NAD) and nicotinamide-adenine dinucleotide phosphate (NADP), which are bound to the base cellular structure by covalent or other less stronger links (Machlin, 1984). The relative solubility of each of these forms in water, and/or the relative strength of their corresponding bonds to the base cellular structure, can define the corresponding vitamin diffusion to the soaking medium and its subsequent loss. This relates very well the higher water solubility of niacin and its highest loss in chickpeas and lentils and the smallest water solubility of riboflavin and its smaller loss in all studied samples (Budavari, 1996) and indicates that, in chickpeas and lentils, the predominant forms of niacin, probably, are its simplest forms, nicotinic acid and/or nicotinamide, whereas thiamine, and especially riboflavin, probably exist in more complex forms.

As can be seen from Table 1 (Fig. 1), greater vitamin loss occurred when the soaking process was carried out in sodium bicarbonate solution (alkaline pH) and water (neutral pH). This effect can be explained by the higher rates of decomposition of thiamin and, at certain levels, riboflavin, due to their chemical instability in neutral and alkaline solutions (Machlin, 1984). On the other hand, it is known that, the water solubility of the studied vitamins increases with increase of the pH (Machlin, 1984), which can lead to further losses of these vitamins. Varriano-Marston and Omana (1979) also found that the introduction of sodium salts into the soaking medium alters the integrity of the seed cellular walls, thus favouring the diffusion of intracellular matter to the outer liquid. These mechanisms explain the greater decrease of thiamin, riboflavin and niacin after soaking in sodium bicarbonate solution in most of the studied samples. The higher loss of niacin found after soaking of lentils in citric acid solution should be considered as an exception and related to the specific behaviour of the lentils in this process.

Riboflavin increase, during soaking of lentils, can be explained by microbial activity. This phenomenon was observed in our previous investigations on germination (Urbano et al., 1995; Prodanov et al., 1997) and fermentation (Vidal-Valverde et al., 1993; Vidal-Valverde, Prodanov, & Sierra, 1997) of lentils, where soaking is an initial part of these treatments and was attributed to the existence of some microorganisms (mainly *Lactobacillus*) in the lentil cover, which are able to synthesize riboflavin immediately after humidification of the lentil seeds (Vidal-Valverde et al., 1997, 2002).

In another study, Lu et al. (1984) observed that, after soaking of faba beans in water for 12 h (previous bleaching for 3 min at 80 °C), the riboflavin content did not change and thiamin decreased by 50%. Addition of sodium bicarbonate to the soaking medium, at

concentrations of 0.5, 1.0 and 1.5%, did not produce additional changes in the retention of either vitamins. Our results for the thiamin content are not in agreement with the above authors, probably due to the different conditions of performing the experimental processes.

Cooking, in general, produced a further decrease in the vitamin content of all studied legumes (Table 1, Fig. 1). These losses, with some exceptions, were higher when a previous soaking was carried out in alkaline and neutral solutions and can be explained by the increasing leaching rates, promoted by the high cooking temperature and the increased levels of sodium ions (retained after the draining of the soaking medium). According to Varriano-Marston and Omana (1979) and Iyer et al. (1989), these are the main two factors responsible for the intensification of the cellular wall lyses and the diffusion of ions from the intracellular media to the outer liquid. Another important factor, which explains some of the riboflavin, and especially thiamine, losses, is their chemical instability in neutral and alkaline solutions at elevated temperatures (Machlin, 1984).

Haytowitz and Matthews (1983) found that 15 min cooking of lentils (*Lens culinaris* L.) had no effect on their riboflavin contents and produced 15 and 25% losses, respectively, in the thiamin and total niacin contents, whereas 150 min cooking produced 44, 78 and 28% losses, respectively, in the thiamin, riboflavin and total niacin contents of chickpeas (*Cicer arietinum* L.) and 53, 79 and 36% losses, respectively, in thiamin, riboflavin and total niacin contents of faba beans (*Vicia faba* L.). Khalil and Mansour (1995) found 75, 42 and 94% losses, respectively, in the thiamin, riboflavin and available niacin contents of faba beans (*Vicia faba* L.) after 12 h of soaking and 1 h of cooking in water and El-Adawy (2002) found 66, 52 and 96% losses of thiamin, riboflavin and available niacin contents of chickpeas (*Cicer arietinum* L.) after 12 h of soaking and 90 min of cooking. The differences between these results and our own can be explained by the longer cooking process carried out by these authors, which can favour leaching of the studied vitamins to the cooking media and the higher heat instability of thiamin and riboflavin at neutral or higher pH values of the cooking media.

Abdel-Hamid (1983) found that 1 h direct cooking (without soaking) of chickpeas produced up to 29 and 16% losses, respectively, in their thiamin and total niacin contents, which is considerably less, than that obtained in our experiment. These differences can be attributed to the additional soaking process carried out in our study.

Reports on the influence of salt/acid addition to the cooking medium in faba beans, chick peas and lentils and their effect on vitamin retention were not found. In cowpeas (*Vigna unguiculata* L.), Uzoraga, Morton, and Daniel (1991) observed that the addition of 0.1% sodium bicarbonate to the cooking liquid reduced the

thiamin content by a further 57%, than when the process was carried out in distilled water; riboflavin was maintained without substantial changes and total niacin content increased slightly.

Thus, when legumes have to be processed for human and/or animal nutrition by soaking alone, the following two considerations have to be taken in account: (a) acidifying the soaking media improves the vitamin retention in most cases and the addition of sodium bicarbonate increases vitamin loss; (b) discarding the soaking liquids leads to certain loss of thiamin and a considerable loss of niacin, especially in chickpeas and lentils. The best way to reduce these losses is to carry out the soaking process in 0.1% citric acid solution or in water with $\text{pH} \leq 7$. It was found that the same conditions produced the best retentions in other nutrients, such as starch, glucose, fructose, sucrose and a greater destruction of antinutrients, such as α -galactosides and trypsin inhibitors in faba beans (Vidal-Valverde, Frias, Sotomayor, Diaz-Pollan, Fernández, & Urbano, 1998) and in chickpeas (Frias et al., 2000). It is also noteworthy that soaking is an important tool for increasing the riboflavin content of lentils.

With respect to the whole soaking and cooking process, the best conditions, which caused minimum vitamin loss, were 9 h soaking in 0.1% citric acid solution or in water with $\text{pH} \leq 7$ and subsequent boiling of the seeds for 35 min. These conditions agree very well with those found as optimal for of best retentions of other nutrients such as starch, glucose, fructose and sucrose, and maximal destruction of antinutrients, such as α -galactosides, trypsin inhibitors and phytic acid (Vidal-Valverde et al., 1998; Frias et al., 2000).

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