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Effects of Dehulling, Soaking and Germination on Chemical Composition, Mineral Elements and Protein Patterns of Faba Beans (*Vicia faba* L.)

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ABSTRACT

The effects of dehulling, soaking and germination on the changes in general chemical composition, nine mineral elements and the protein patterns of whole faba beans were studied. Such pretreatments had a significant effect on the changes in the chemical composition of whole beans. Dehulling of beans significantly increased the contents of copper, zinc and potassium, while it significantly decreased the amounts of iron, manganese, phosphorus, calcium, magnesium and sodium compared with whole beans. Whole beans soaked for 12 h, followed by dehulling, had higher amounts of iron, copper, zinc, calcium and sodium, and quite a lower level of potassium, than unsoaked dehulled beans. All of the mineral elements except zinc were significantly increased on germination. No obvious changes in the electrophoretic pattern of faba bean proteins were observed as the seeds were soaked for up to 12 h. Extending soaking time for a further 4 h led to the disappearance of one band at the region of MW 67 KD. Germination of seeds for 3 days initiated disappearance of some bands while other new bands were observed.

INTRODUCTION

Legumes in general are considered staple foods for many communities in different areas of the world. Faba beans are recognized as a good source of dietary protein and carbohydrate, since the protein content has been

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reported to range from 26% to 35% (Bhatty, 1974) and the carbohydrates from 51% to 68% (Cerning *et al.*, 1975). Furthermore, faba beans are characteristically a good source for some mineral elements (El-Shimi, 1980). Hegazi & Salem (1974) found that whole faba beans contained 3·5% ash.

The major obstacle in utilizing the dry beans is the tough nature of their seed coats and consequently the long hours of cooking needed to tenderize them (Cerny *et al.*, 1971).

In general, when faba beans are prepared for consumption they are usually subjected to some pretreatments, i.e. dehulling, soaking and/or germination. These primary processes are known to improve the nutritive value of dry beans by reducing or eliminating some of the antinutritional factors, such as tannins (Abd El-Aal *et al.*, 1985) and phytate content (El-Shimi, 1980).

Numerous studies have suggested that phytate reduces the biological availability of dietary copper and manganese (Davies & Nightingale, 1975), iron (Welch *et al.*, 1974), magnesium (Guenter & Sell, 1974) and zinc (Oberleas, 1973).

The purpose of this work was to study the changes in chemical composition, contents of nine mineral elements (iron, copper, zinc, manganese, magnesium, phosphorus, calcium, potassium and sodium) and the changes in the protein patterns of faba beans as a result of dehulling, soaking and germination.

MATERIALS AND METHODS

Whole faba beans

Faba bean seeds of the Giza 3 variety were obtained from the Agricultural Experimental Station, belonging to the Ministry of Agriculture in Sakha location, Egypt, during the season of 1985. The seeds were divided (using a sample divider), packed in sealed polyethylene bags and stored in a deep freezer at $-20°C$ until used.

Dehulling of seeds

The whole beans were mechanically dehulled using a PRL 'Mini' Dehuller, National Research Council, Canada.

Soaking of seeds

The whole faba beans were soaked in distilled water (1:5 w/v) for 12 h at room temperature ($\sim 25°C$). At the end of the soaking period, the beans

were removed and dehulled manually. The cotyledons were then disintegrated using a Waring blender, then dried at 50°C in a cabinet drier for 6 h.

Germination of seeds

Three lot samples of the whole beans were first steeped for 12 h in distilled water (1:5 w/v) at room temperature, then transferred to moistened cotton layers and allowed to germinate in the dark at room temperature (~25°C) for 3 days. During germination the cotton layers were kept always moist by rinsing with distilled water. After the germination period (the degree of germination was 98·0 ± 1·0%), the seeds were dehulled manually and the separated cotyledons were disintegrated and dried as before.

Preparation of samples

The whole faba beans and their dried processed products were ground to pass through a 40-mesh sieve.

Analytical methods

Chemical composition analysis
Moisture, crude protein (N × 5·85), crude fat, crude fibre and total ash were determined by the methods of the AOAC (1980). Non-protein nitrogen was determined as described by Bhatty (1973). Phytic acid was determined according to the method of Wheeler & Ferrel (1971).

Mineral element analysis
Total phosphorus was determined colorimetrically. Calcium, copper, iron, magnesium, manganese, zinc and potassium were determined by atomic absorption (Bye Unicam SP 1900), while sodium was determined by flame photometer (Gallenkamp) as outlined by the AOAC (1980).

Statistical analyses for analysis of variance and Duncan's Multiple Range test were carried out according to Nie *et al.* (1970).

Electrophoretic analysis
Raw faba beans, soaked beans (4, 8, 12 and 16 h) and germinated beans (24, 48 and 72 h) were manually dehulled and homogenized in a mortar. Then they were defatted with cold acetone (1:3 w/v, three successive times). Samples of the defatted powder were each mixed with 10 ml of distilled water and shaken for 1 h, then centrifuged at 3000 *g* for 15 min. The supernatants were loaded with sodium dodecyl sulphate (SDS) and

mercapto-ethanol (ME) according to the method described by Hamza (1983). Sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS–PAGE) was performed in 3-mm slabs in the PANTA-PHOR apparatus according to the method of Stegemann *et al.* (1985). The molecular weight marker proteins in KiloDaltons (KD), were: phosphorylase, 97; albumin, 67; alcohol dehydrogenase, 37; chymotrypsinogen, 25; and lysozyme, 14.

RESULTS AND DISCUSSION

Chemical composition

The proximate chemical composition of the whole, dehulled, soaked and germinated faba beans is presented in Table 1. The true protein content of whole beans was 28·6%. Dehulling of the seeds reduced the protein content by 3·2% of that originally present. The values obtained in this respect were quite comparable with those obtained by Barratt (1982). When faba beans were soaked for 12 h and/or germinated for 3 days, a further significant reduction in the protein content of the cotyledons was observed.

The crude fat content of whole beans was about 1·3% and the value for dehulled beans was quite similar. Soaking for 12 h or germination for 3 days, followed by dehulling, had no effect on crude fat content. Fordham *et al.* (1975) reported a slightly lower lipid content in bean sprouts than in dry beans. Kylen & McCready (1975) stated that, during germination, the lipids are broken down by the action of lipases which are not accumulated in the seeds.

The whole beans had 9·5% crude fibre. The dehulling process led to a significant decrease in the crude fibre content of the cotyledons by about 89·9% of that originally present in the whole beans. On the other hand, the soaking or germination processes followed by dehulling had no effect on the crude fibre content.

The total ash contents of whole and dehulled beans were 3·3% and 3·2%, respectively. Soaked beans had less ash content, while germinated seeds exhibited highly significant amounts of ash (3·7%) compared with ungerminated cotyledons.

Table 1 shows that dehulling of faba beans led to a significant increase in phytic acid content. This increase can be attributed to localization of phytic acid in cotyledons (Deshpande *et al.*, 1982). In contrast, soaking of whole beans for 12 h, followed by dehulling, resulted in a significant decrease in phytic acid compared with dehulled beans. It has been reported that there was a slight decrease in phytate content in soaking of dry beans (Tabekhia & Luh, 1980).

TABLE 1
Chemical Composition of Whole, Dehulled, Soaked and Germinated Faba Beans (on Dry Weight Basis)*

Material	*Per cent crude protein* ($N \times 5{\cdot}85$)	*Per cent non-protein nitrogen*	*Per cent true protein* ($N \times 5{\cdot}85$)	*Per cent crude ether extract*	*Per cent crude fibre*	*Per cent ash*	*Per cent N-free† extract*	*Phytic acid (mg/100 g)*
Whole beans	$31{\cdot}6^{a}$	$0{\cdot}5^{c}$	$28{\cdot}6^{a}$	$1{\cdot}27^{ab}$	$9{\cdot}5^{a}$	$3{\cdot}3^{ab}$	$54{\cdot}3^{b}$	425^{b}
Dehulled beans	$31{\cdot}1^{ab}$	$0{\cdot}6^{b}$	$27{\cdot}7^{b}$	$1{\cdot}33^{ab}$	$1{\cdot}0^{b}$	$3{\cdot}2^{b}$	$63{\cdot}4^{a}$	485^{a}
Soaked beans	$30{\cdot}9^{bc}$	$0{\cdot}7^{a}$	$27{\cdot}1^{c}$	$1{\cdot}35^{a}$	$1{\cdot}0^{b}$	$3{\cdot}0^{b}$	$63{\cdot}7^{a}$	427^{b}
Germinated beans	$30{\cdot}4^{c}$	$0{\cdot}6^{b}$	$26{\cdot}9^{c}$	$1{\cdot}26^{b}$	$1{\cdot}0^{b}$	$3{\cdot}7^{a}$	$63{\cdot}7^{a}$	411^{b}

* Means of three replicates.
† By difference.
Mean values in a column not sharing the same superscript are significantly different at $P < 0{\cdot}05$.

TABLE 2
Mineral Element Content of Whole, Dehulled, Soaked and Germinated Faba Beans (mg/100 g, Dry Weight Basis)*

Material	*Fe*	*Mn*	*Cu*	*Zn*	*Na*	*Ca*	*Mg*	*K*	*P*
Whole beans	$7{\cdot}7^{b}$	$1{\cdot}4^{b}$	$2{\cdot}8^{c}$	$2{\cdot}7^{b}$	254^{a}	$98{\cdot}6^{a}$	188^{a}	$1\,731^{b}$	383^{ab}
Dehulled beans	$6{\cdot}1^{c}$	$1{\cdot}2^{c}$	$3{\cdot}2^{b}$	$2{\cdot}9^{a}$	137^{d}	$30{\cdot}6^{d}$	173^{b}	$1\,800^{a}$	369^{b}
Soaked beans	$7{\cdot}7^{b}$	$1{\cdot}1^{d}$	$3{\cdot}3^{ab}$	$2{\cdot}9^{a}$	226^{c}	$40{\cdot}9^{c}$	143^{c}	$1\,161^{c}$	341^{c}
Germinated beans	$9{\cdot}2^{a}$	$1{\cdot}5^{a}$	$3{\cdot}4^{a}$	$1{\cdot}6^{c}$	243^{b}	$63{\cdot}4^{b}$	172^{b}	$1\,848^{a}$	400^{a}

* Means of two replicates.
Mean values in a column not sharing the same superscript are significantly different at $P < 0{\cdot}05$.

In general, the results obtained in the present work are in good agreement with those published for the whole beans (Hegazi & Salem, 1974), the effect of dehulling (Youssef, 1978) and soaking and germination (El-Shimi, 1980).

Mineral elements composition

The mineral element content of the dry whole, dehulled, soaked and germinated faba beans is given in Table 2. Whole beans were relatively high in iron, copper, zinc, phosphorus, magnesium, potassium and sodium, and relatively low in manganese and calcium. However, considerable variations were found in this respect among the whole beans and their processed products for the content of several of the determined minerals. Furthermore, the contents of each particular mineral element of the whole, dehulled, soaked and germinated faba beans were comparable with other published data (Youssef, 1978; El-Shimi, 1980; El-Tabey Shehata *et al.*, 1984). The same trend was obtained for other legumes (Meiners *et al.*, 1976).

The whole faba beans contained 7·7, 1·4, 2·8, 2·7, 383, 98·6, 187·9, 1731 and 254 mg per 100 g of iron, manganese, copper, zinc, phosphorus, calcium, magnesium, potassium and sodium, respectively. Dehulling of beans significantly reduced their contents of iron, manganese, phosphorus, calcium, magnesium and sodium. In contrast, copper, zinc and potassium were increased.

Soaked seeds showed a wide variation in their mineral element contents compared with unsoaked dehulled beans. The soaked cotyledons were more or less high in iron, copper, zinc, calcium and sodium, and quite low in potassium content. The high level of sodium in soaked beans compared with unsoaked beans may be due to leaching out of sodium from the seed coats in the soaking medium, which is thereby re-absorbed by the imbibed cotyledons. El-Tabey Shehata *et al.* (1984) reported that the sodium content of whole faba beans ranged from 169 to 294 mg per 100 g.

Germination of faba beans for 3 days resulted in an increase in all of the tested mineral elements (with the exception of zinc) compared with ungerminated cotyledons. Data concerning some mineral element contents for germinated beans are in accordance with a report by El-Shimi (1980). However, germination of faba beans can be considered as a best and simple pretreatment for increasing nutrients.

Electrophoretic patterns

The electrophoretic patterns of dry, soaked and germinated faba beans are illustrated in Fig. 1. Compared with the protein banding pattern of raw

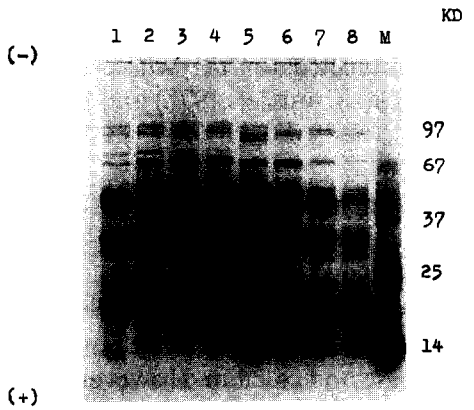

Fig. 1. SDS–PAGE of dehulled, soaked and germinated faba bean seed proteins. 1 = Dry and dehulled beans. 2 = Soaked beans (4 h). 3 = Soaked beans (8 h). 4 = Soaked beans (12 h). 5 = Soaked beans (16 h). 6 = Germinated beans (24 h). 7 = Germinated beans (48 h). 8 = Germinated beans (72 h). M = Marker proteins (from 14 to 97 KD) for MW. Water extracts from dry, soaked and germinated beans were first loaded with SDS/ME, then applied to 3-mm thick gel in Tris-borate buffer (pH 7·1). Electrophoresis was performed for 2 h at 300 V, 60 mA with cooling (2°C).

beans, a slight shift in the intensity of the resolved bands was observed due to the soaking treatment, but no obvious changes in the number of the separated bands were noticed after soaking for 4, 8 and 12 h. Soaking for 16 h led to the disappearance of one band having a molecular weight of 67 KD. There was also a slight change in the migration rate of the smaller MW subunits.

Germination of soaked beans reduced the number and changed the intensity of high and medium MW subunits. At the same time, a new band appeared in the lower MW region (14 KD). This may be due to the hydrolysis of higher MW proteins into smaller subunits by the action of hydrolytic enzymes during germination (Hamza, 1983).

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