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Effect of germination on the carbohydrate, protein and amino acid contents of broad beans

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With 2 tables

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Broad bean is considered to be the most important and abundant source of dietary protein for the majority of people in Egypt. In recent years some evidence has been gathered that germination improves the nutritive value of beans (1, 2). During germination the diastitic and proteolytic break down fairly elaborate which results in the formation of readily assimilable diet when eaten as human food. In addition, germination eliminates toxic factors which may be present in the edible beans (1, 2).

Studies on germinated cereals and legumes pointed to marked changes in their nutrients (3, 4, 5, 6). These changes depend upon the duration of soaking and the time of germination. Proteins showed an extensive breakdown accompanied by inter-conversion and utilization of amino acids in the production of new nitrogen compounds (7).

Some workers have demonstrated that a complicated series of changes occurred in the carbohydrate contents of pulses during germination (4), some of the starch being converted to sucrose, glucose and fructose in the earlier stages, while later increasing quantities of maltose appear by apparent increase in the reducing sugars and dextrins.

The experimental work in this study aimed to determine the changes in the carbohydrate, protein and amino acid contents of broad bean seeds due to germination.

Experimental

Sampling: 1 kg of the broad bean seeds (Vicia *faba*), Giza 1 variety was obtained from the Plant Breeding Department, Ministry of Agriculture.

Germination procedure: Germination was carried out by soaking the beans in tap water (1 kg beans in 4 l of water) at room temperature (25° C), for 4 days. Water was changed every 24 hours. At the end of the germination process, the beans were dried in an oven provided with fan at 50° C, then ground into a fine powder for analysis.

Methods of analysis: Samples were analysed for moisture and nitrogen contents following the standard methods (A.O.A.C., 1960) (8). Reducing sugars were determined by the *Somogi* method (9).

Determination of total sugars and starch contents: Total sugars were extracted with alcohol 80% (10). Extraction was repeated three times and the combined alcoholic extracts were evaporated under vacuum and the remaining

cloudy aqueous fraction was diluted with distilled water to a known volume. The remaining material which was extracted with alcohol, was treated by the addition of perchloric acid solution 52 %, and the supernatant liquid was diluted with water then adjusted to known volume. Determination of sugars and starch was carried of out according to the colorimetric method of *Cleeg* (10).

Determination of amino acids: Samples were hydrolysed with 6 N HCl for 22 hours at 110° C, according to *Khan* and *Baker* method (11). The HCl hydrolysate was evaporated off under vacuum to dryness and the residue was dissolved in 10 % isopropyl alcohol (V/V) to a certain volume. Portions were spotted on Whatman No. 1 filter paper and two dimensional ascending chromatograms were prepared using the buffered method of *Levy* and *Chung* (12). After development with ninhydrine reagent, the located amino acids were eluted and the concentration of each individual amino acid was determined colorimetrically at 510 mμ. Cystine and methionine were determined in the hydrolysate after performic oxidation (13).

Determination of total free amino acids: Twenty grams of the germinated beans were extracted with successive amounts of 5 % (W/V) potassium sulphate solution, 0.02 N sodium hydroxide and hot 70 % (V/V) ethanol (7). The free amino acids in the extract were determined by the method of *Lee* and *Takahashi* (14).

Results and discussion

The effect of germination on carbohydrate contents of the broad beans is presented in table 1.

Table 1. Effect of germination of carbohydrate components of the Egyptian broad beans (on dry wt. basis)

Carbohydrate %	Raw beans	Germinated beans
Reducing sugars	6.10	6.75
Non-reducing sugars	1.76	2.90
Total sugars	7.86	9.65
Starch	37.80	30.95
Total carbohydrate	45.66	41.80

A comparison between the carbohydrate contents of the raw beans and the germinated beans indicates that total sugars, reducing sugars and the non-reducing sugars were increased by germination. This increase can be explained by the action of α- and β-amylases which developed during germination. Some of the starch being converted into sugars (table 1), its value was markedly decreased from 37.8 % in the raw beans to 30.9 % in the germinated ones. This decrease was partly due to respiration and partly through the formation of soluble carbohydrates. In favour of such explanation is that given by *Jorgenson* (15) who showed that α-glucosidase activity was higher in the germinated barley than in the ungerminated one.

Protein content of the raw beans was slightly decreased from 28.1 % to 26.7 % in the germinated beans (table 2). This decrease is the result arising from the breakdown of proteins by proteolytic action of enzymes which leads to an apparent increase in soluble amino acids.

Table 2. Effect of germination process on proteins, amino acids and α-free amino acids of the Egyptian broad beans (on dry wt. basis)

Amino acid	Raw beans mg/g nitrogen	Germinated beans mg/g nitrogen
Cystine	50	54
Methionine	41	45
Tryptophan	65	66
Arginine	347	374
Lysine	351	436
Phenylalanine	273	290
Leucine + isoleucine	779	775
Valine	336	328
Tyrosine	245	300
Alanine	221	289
Threonine	237	289
Glycine	326	415
Serine	331	397
Glutamic acid	1026	1125
Aspartic acid	530	545
% of total protein	28.1	26.7
Total free amino acids (mg/100 g)	72	290

Acid hydrolysates of protein revealed the presence of the same amino acids in both of raw and germinated beans, table 2. It is evident that germination leads to a noticeable increase in the concentration of amino acids. This increase is mainly due to the proteolytic action of enzymes which break down the complex protein molecules into simpler units of amino acids. Differences were observed in the amounts of glutamic acid, lysine, threonine, tyrosine, alanine, serine, glycine and phenylalanine which were highly increased by germination. They were present in large proportion of total amino acid contents. The sulphur containing amino acids, cystine and methionine, were slightly increased. Other amino acids did not change considerably in their amounts during germination. This may reflect a balance between synthetic and degradation processes. The simplest explanation is that they are liberated from reserve proteins and utilized more or less unchanged (7). It is believed that during germination the newly formed proteins differ in their amino acid contents.

In general, the interesting feature of the germination process is the formation of readily assimilable diet rich in most of the essential amino acids required for the human nutrition.

Summary

Broad bean seeds were germinated for 4 days and the changes occurred in carbohydrate, protein and amino acids of germinated seeds were studied. Proteins showed slight reduction in their content, while variations were observed in the amounts of amino acids. The germination process leads to an increase in most of the essential amino acids needed for human consumption. Starch content was decreased accompanied by an apparent increase in both of reducing and non-reducing sugars.

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