

# Changes in the Composition of Soybeans on Sprouting<sup>1</sup>

L. L. MCKINNEY, F. B. WEAKLEY, R. E. CAMPBELL, and J. C. COWAN, Northern Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture, Peoria, Illinois

SPROUTED SOYBEANS have long been used as human food in the Orient. Analytical determinations on the composition of sprouted soybeans and a literature search for comparable studies indicate that such beans could form the basis of a high-energy, high-protein broiler feed.

Block and Bolling (2) found that the amino acid composition of soybeans changed little, if any, on germinating for two to five days; Dunn and his students (5) observed a decrease of about 30% in the total amino acids after germinating for 12 days. The vitamin content of soybeans increases appreciably after germinating for two to six days. Burkholder and McVeigh (3) determined the changes in the B vitamin contents of seven varieties of soybeans after germinating for four days. Figure 1 is drawn from

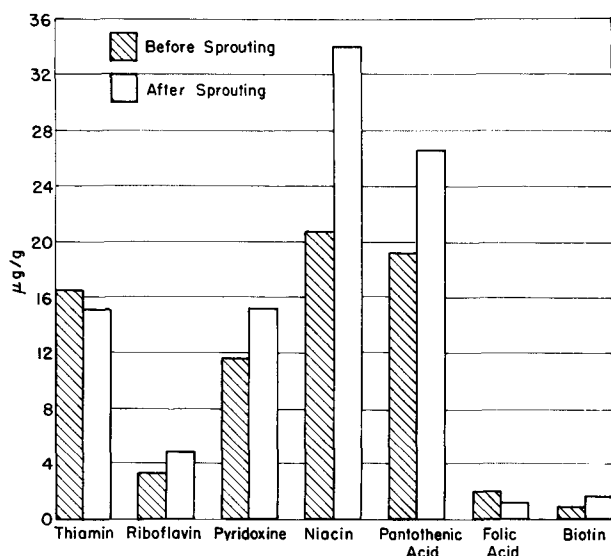


FIG. 1. Changes in the vitamin content of soybeans on sprouting for four days, from Burkholder and McVeigh (3).

their average values for the Lincoln, Dunfield, and Illini varieties, which are related to commercial varieties presently grown for use in animal feeds. With the exception of thiamin and folic acid, increases from 31 to 90% in the B vitamins were noted. Studies on vegetable varieties have shown large increases of carotene (13) and vitamin C (15) on germinating from two to six days. The optimum sprouting time of vegetable varieties, based on the coupled effects of vitamin content and taste, was reported to be 54 hrs. at 28°C., not including a 10-hr. soaking period (13).

Sasaki (11) investigated the compositional changes which occurred in Japanese and Korean varieties of soybeans during germination up to 12 days. There was no change in total nitrogen at the 5- and 10-day periods. However in five days a decrease of 21% in protein nitrogen and a corresponding increase in non-protein nitrogen was observed. After 12 days of germination 5% of the starting bean weight was isolated

as asparagine, which is in agreement with the results obtained by Dunn (5). The total carbohydrates increased from 22.6% in the seed to 25.8% by the fifth day, based on the starting weight of the seed, and then decreased slightly to 25.1% by the tenth day. Stachyose and sucrose were used up during the first five days, at which time reducing sugars reached a maximum of 4.7% and then decreased. Starch and dextrin increased steadily from 0.4 to 9.0%, based on the starting seed weight, during the first 10 days of germination. Petroleum ether extractables had decreased 17.0% by the fifth day and 37.0% by the eighth day of germination. From this study it appears that during the first five days of germination the loss in oil weight is equivalent to the gain in carbohydrate weight.

Feeding studies with rats (6) have shown increased protein efficiency after germinating soybeans for 60 hrs. although the percentage of nitrogen absorbed was not increased. Like mature beans, the germinated beans appear to contain antigrwth factors and must be cooked before feeding.

None of the above studies report the loss in dry matter on germination. Since it is known that cereal grains rapidly lose dry matter during germination (4), it might be suspected that soybeans would do likewise. The data reported here were obtained to determine the loss in dry matter and to follow the changes in oil, nitrogen, thiamin, and ascorbic acid during a practicable germination period for a typical American variety in order to evaluate the possibility of germinating soybeans to produce a high-energy, high-protein broiler feed.

## Experimental

For each germination period, except those involving the thiamin and ascorbic acid studies, 100 g. (605-611 seeds) of Hawkeye variety soybeans from the 1955 crop were washed with chloride-of-lime solution (350 p.p.m.  $Cl_2$ ) to inhibit mold growth, then the beans were soaked in 275 ml. of tap water over-night (17 hrs.). The drained beans (about 240 g.) were spread out on moist cotton, covered with three layers of moist cheesecloth, and allowed to germinate in the dark at room temperature (24°-27°C.). The soaking time is included in the germination time.

The germinated beans were dried over-night at 70°C., ground, and dried to constant weight at 100°C. Corrections were made for solubles in the soaking liquor (av. 0.7%) and for sterile or spoiled beans (av. 1.4%). Nonprotein nitrogen was measured by the amount of nitrogen soluble in 0.8N trichloroacetic acid (1), and other analyses were made by the methods of the Association of Official Agricultural Chemists. Analyses are reported on a dry weight of the starting beans as determined by drying to constant weight at 100°C. at atmospheric pressure. Higher drying temperatures or vacuum drying were avoided for fear of removing volatiles from the germinated beans since the reported (11) increase of Reichert-Meissel value for oil from germinated beans indicates the possibility that low-molecular-weight fatty acids were formed.

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Ungerminated soybeans destined for use in the studies involving changes in thiamin and ascorbic acid on germination were also treated for inhibition of mold growth, but the subsequent soaking time in water was limited to approximately 2 hrs. The beans, whether soaked for 2 hrs. or 17 hrs., appeared to attain the same wet weight within the first day of germination. The germinated beans were taken as is and analyzed for thiamin and ascorbic acid. The vitamin values were calculated to the dry weight of the starting beans. Thiamin was measured by the thiochrome method of Johnson (9). A modified indophenol-xylene extraction method (14) was used to measure ascorbic acid.

**Results and Discussion**

The growth rate of the sprouts and the loss in dry matter on germinating from two to six days are shown in Table I. Since corn has been reported (4)

TABLE I  
Growth of Sprouts and Weight Loss During Germination

Germination time (hours)	Length of sprouts (inches)	Weight loss of dry matter (%)
48.....	1/4	0.8
72.....	1/4-1/2	1.5
96.....	1/2-3/4	2.0
120.....	1 1/2-2	2.5
144.....	2 1/4-2 1/2	2.6

to lose 10.8% of its dry weight during a two-day germination period, it is noteworthy that less than 3% of the dry matter of soybeans was lost during a six-day germination period.

Figure 2 shows the changes which occur in nitrogen values. Protein nitrogen was determined by sub-

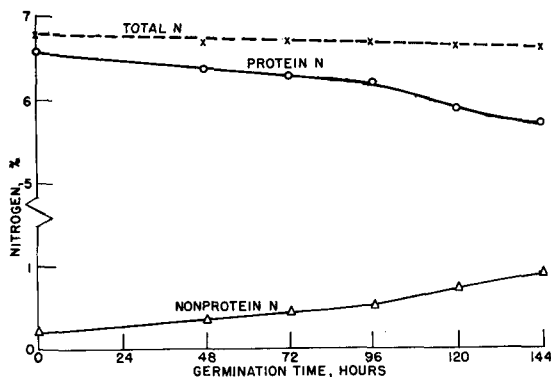


Fig. 2. Changes in nitrogen on germinating soybeans. Percentages of nitrogen are based on the dry weight of the starting beans.

tracting nonprotein nitrogen from total nitrogen. After a six-day germination period the loss in total nitrogen was 2.6%, and the decrease in protein nitrogen was 15% with a corresponding increase in nonprotein nitrogen. The apparent loss in protein does not necessarily reflect a loss in protein feeding value because the breakdown products, such as amino acids and peptides, should appear in the nonprotein fraction and be available to the animal.

Changes which occur in the oil are shown in Figure 3. There was no loss in oil during the first three days of germination, during which time the oil maintained

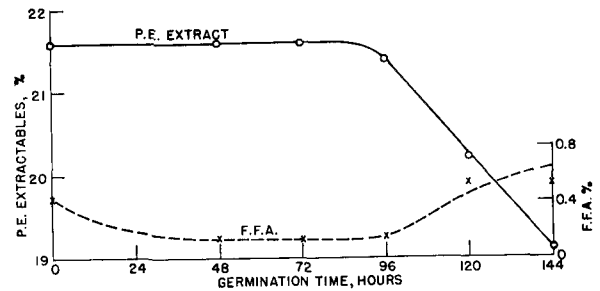


Fig. 3. Changes in the oil (petroleum ether extractables, P.E.) content of soybeans on germination, and of the free fatty acids (F.F.A.) in the oil. Percentages are based on the dry weight of the starting beans.

its natural yellow color. The oil from the five- and six-day germinated beans was dark, which indicated the formation of a petroleum ether soluble chromogenic substance. At the end of six days of germination 12% of the oil had been lost. The decrease in free fatty acids from 0.44% in the bean oil to 0.14% at the two- and three-day germination period was unexpected. A repetition of the analyses gave the same values. According to Hill *et al.* (8), the hydrolysis of fats by lipase in the germinating seed proceeds slowly at first, but as the percentage of acid formed increases, the rate increases. In other words, a little acid accelerates the hydrolytic action of lipase. The decrease in free fatty acid at the beginning of the germination therefore probably results from its being metabolized at a greater rate than it is being liberated.

In general, the nitrogen and oil values reported for Hawkeye soybeans are in agreement with those reported by Sasaki (11) for Japanese and Korean varieties. He did not observe any decrease in total nitrogen during the first 10 days of germination; Dunn (5) reported a loss of only 2.3% during a 12-day period. Sasaki reported a greater decrease in both protein nitrogen and oil for a five-day germination period. These differences may result from a different technique used in sprouting the beans; for example, he maintained his beans at 40°C. during the first two days.

Changes which occur in the thiamin and ascorbic acid content of the soybeans on germination are shown in Figure 4. No appreciable change was observed in the thiamin content during the first four days of germination, which is in agreement with the observations of Burkholder (3) and Wai (13). Sugi-

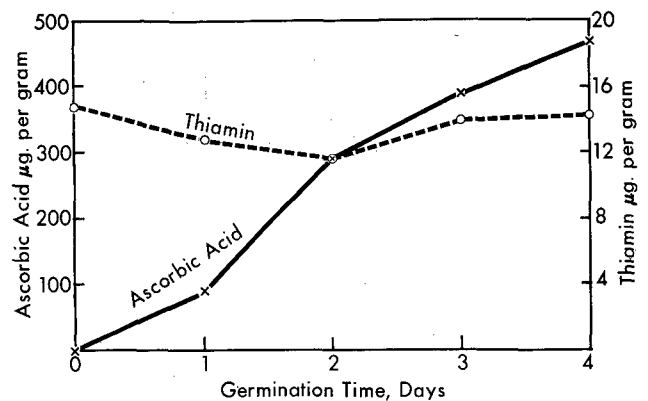


Fig. 4. Changes in ascorbic acid and thiamin content of germinating soybeans, calculated to dry weight of starting beans.

moto (12) reported no remarkable change in thiamin up to 13 days of germination. At most it appears that alternating minor gains and losses of thiamin occur during germination. Ascorbic acid was absent in ungerminated beans but appeared within the first 24 hrs. of germination and reached a value of 390 micrograms per gram at the end of three days. It was found in the reduced form only (14). Previous investigators (3, 13, 15) have reported that ungerminated soybeans contained ascorbic acid and that the original value increased several-fold during the first three to five days of germination. Previous reports (3, 13) also show varying amounts of ascorbic acid present in the dehydro form. A report (14) of the modified indophenol-xylene extraction method used in the present study to estimate ascorbic acid showed that the analytical methods used by previous investigators gave high values for ascorbic acid because of interference by sulfhydryl groups. It was also shown that the ascorbic acid values reported for ungerminated soybeans and the values reported for dehydro-ascorbic acid resulted from interference associated with sulfhydryl group activity.

From several excellent studies (3, 7, 13, 15) on the increase in the vitamin content of soybeans on germination it appears that a substantial increase can be expected in two to three days. Since the over-all composition of soybeans does not change appreciably during the first 72 hrs. of germination, it appears that sprouted beans could be used as a basis for producing a high-energy, high-protein broiler feed. Hulls are easily removed from sprouted soybeans because they become loose and detached on drying. This property would facilitate removal of undesired fiber. However grinding followed by screening would have to be used because aspiration would probably remove the dried sprouts.

### Summary

Changes in nitrogen and oil contents and loss in dry weight have been followed during the first six days of germination of the Hawkeye variety of soy-

beans; thiamin and ascorbic acid content have been followed for four days. During the first three days about 1.5% of the dry matter was lost; the nonprotein nitrogen increased from 3 to 6% of the total nitrogen, with no change in the petroleum ether extractables and with a decrease in free fatty acids. At the end of six days, with sprouts about 2½ in. long, only 2.6% of the dry matter was lost, the nonprotein nitrogen had increased to 13% of the total nitrogen with 2.6% loss of total nitrogen, and a 12% loss was observed in petroleum ether extractables; the free fatty acids did not increase appreciably. No change in the thiamin content occurred during the first four days of germination. Ascorbic acid was found to be absent in mature beans but appeared after the start of germination and increased rapidly during the four-day period.

An analysis of these results and of those from the literature indicates that soybeans sprouted for two to three days have possibilities for use in high-energy, high-protein broiler feed.

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## Solubilities of Fatty Acids and Derivatives in Acetone<sup>1</sup>

O. S. PRIVETT, E. BREAU, J. B. COVELL, L. N. NORCIA,<sup>3</sup> and W. O. LUNDBERG,  
The Hormel Institute, University of Minnesota, Austin, Minnesota

KNOWLEDGE of the solubilities of fatty acids and derivatives in organic solvents at various temperatures is useful in the research laboratory and in industry in connection with low-temperature fractional crystallization and liquid-liquid distribution processes. The presently available information has been covered quite completely in two reviews (1, 2). It is characterized by many gaps, largely for two reasons: a) many of the compounds are difficult to prepare in sufficiently pure form, and b) technical problems introduced by such phenomena as polymorphism and supercooling sometimes make reliable measurements difficult.

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<sup>2</sup> Present address: U. S. Food and Drug Administration, Minneapolis, Minn.

<sup>3</sup> Present address: Oklahoma Medical Research Foundation, Oklahoma City.

In previous measurements, two methods have been used to determine solubilities. Brown *et al.* (2, 3) and Bailey *et al.* (1) have used a so-called analytical method, which involves withdrawal and analysis of a portion of a solution in which equilibrium has been established between the solvent phase and an excess of solute. Others have used a "synthetic" method, which is somewhat simpler and has the added advantage of using appreciably less material without sacrifice of accuracy. This method consists of observing the temperature at which the last crystal disappears when raising the temperature of a solution of known gross composition or, alternatively, of observing the temperature at which the first crystal appears when the temperature is lowered. Hoerr and Harwood (5) employed the rising temperature technique. Ward and Singleton (11), using the falling temperature method, introduced refinements involving an examination of the cooling curves at different rates of