Changes in Oligosaccharides and Sensory Quality of Soymilk During Germination

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Abstract

The effect of germination of soybeans on chemical and sensory qualities of soymilk were investigated. The soybeans were soaked and germinated at 25 °C for 5 days prior to grinding. The result showed that a rapid initial decrease in the contents of raffinose and stachyose and a slight increase in protein yield were found after 2 days of germination. Undesirable flavor such as beany and grassy was minimum and total acceptability was maximum for the soymilk prepared from soybeans germinated for two days. Changes in roasted nutty odor and taste were almost identical to the changes of total acceptability during-five day germination.

Introduction

Soymilk, one of the most promising soybean products as substitute for bovine milk, has been traditionally prepared in the Orient by soaking, grinding and filtration in cold water followed by boiling of the filtrate for about 30 minutes. (1) This simple preparation resulted in several undesirable characteristics, particularly in flavor. The major undesirable quality of soymilk are generally known as objectionable beany flavor, flatulence factors and some antinutritional components. (2) Among these factors, objectionable flavor and flatulence factors are regarded as principal cause of discouraging consumer's acceptance. In order to improve the quality of soymilk many workers have investigated various treatments.

For reduction or elimination of objectionable flavor described generally as beany, painty and rancid, heat treatment during soaking or grinding of soybeans or boiling of filtrate was found to be very effective for reduction of beany off-flavor. (3.4) Wilkens et al. (4) suggested that the flavor improvement by heating is mainly due to inactivation of lipoxygenase in soybeans. (4)

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Various methods investigated for heat treatment were hot water grinding,(4) steam-infusion cooking of ground soybean(5) and roasting of raw soybeans before grinding. Other methods studied for improvement of flavor and chemical characteristics were soaking soybeans in alkaline solution, (6,7) addition of salts to soymilk(8) and so-called Illinois process. (9,10) In the Illinois process, soybeans were soaked in NaHCO3 solution, blanched, ground and then heated to 93°C before neutralization and homogenization. This process which is principally different from other methods, because of no extraction involved, claimed near 90% recovery of solids and 99% of protein, producing a bland flavored soymilk. However this process requires many steps of processing and sophisticated equipment which result high cost for processing. Although some of the heat treatments and alkaline soaking improves soymilk flavor significantly, existance of beany off-flavor to some degree and a low protein yield due to protein denaturation by heating is still remained for further study.

Soybeans contained 1-2% of raffinose and 5-8% of stachyose in dry weight basis⁽¹⁾ and these oligosaccharides were proved to be the main cause of flatulence after injestion of soybeans.⁽¹¹⁾ In order to remove these oligosaccharides from soymilk, enzymatic hydrolysis with using α -galactosidase⁽¹²⁻¹⁴⁾ and ultrafiltration of soymilk⁽¹⁵⁾ were found to be very effective. However these methods need more study for economic feasibility. Germination of soybeans which is of our interest for this

study has been investigated by many workers for its effect on chemical characteristics of soybeans. East *et al.*(16) reported that oligosaccharides were almost completely removed by 4-5 days of germination and Kim *et al.*(17) achieved more than 2/3 reduction of raffinose and stachyose without root growth by resoaking the germinated soybeans for 24 hours. However there was no reports in literature concerning with the quality changes of soymilk by using germination method except the work on phytate changes in soymilk.(18)

In this study attempt was made to investigate the effect of germination of soybeans on some of soymilk qualities so that proper germination method can be proposed for reduction of flatulence factors and improvements in yields and organoleptic properties of soymilk.

Materials and Methods

Materials and soymilk preparation

Soybeans used in this study was seeed quality (wangyo variety which was harvested and supplied by 1stitute of Rural Development, Suwon, Korea. Soyeans were soaked for 3 hours and germinated at room imperature over a period of 5 days before soymilk

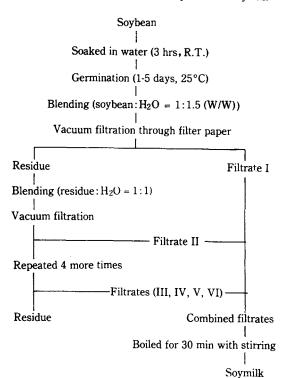


Fig. 1. Flow diagram for preparation of soymilk

preparation. Soymilk was prepared, as shown in Fig. 1, by the methods described by Kim *et al.*(18) All reagents used were reagent grade or equivalent and distilled water was used for soaking soybeans and preparation of soymilk.

Analysis

Proximate analyses of soymilk were carried out by the AOAC procedures. (19) Proteins were determined by micro-kjeldahl method(19) using a conversion factor of 6.25. Total sugars and crude lipids were determined by the phenol sulfuric acid method of Dubois *ct al.* (20) and Roese-Gottlieb method, (15) respectively. Moisture content was determined by evaporation of 20 *ml* soymilk in boiling water bath and dried at 105°C to a constant weight.

For analysis of sugars, soymilk was added ethyl alcohol to be 80% of final ethanol concentration and sugars were extracted at 80°C for 2 hours followed by centrifugation at $10,000 \times g$. After evaporation of ethyl alcohol from sugar extract under vacuum, lipids and proteins were removed by the method of ether extraction and lead acetate precipitation, respectively. The lipid and protein free extract was added sodium oxalate (2.5% of soymilk by weight basis), stirred and then centrifuged. The supernatant was cooled rapidly and stored in refrigerator overnight to remove any solutes precipitated by centrifugation at 10,000 x g for 30 minutes. The final sugar extract was clarified with Waters sample clarification kit (pore size of 0.45 um) before injection to HPLC. Sugars of glucose, fructose, galactose, sucrose, raffinose and stachyose were determined by Waters analytical high-pressure liquid chromatography (model HPLC/ALC-244, Waters Associates, Milford. MA) equiped with universal injector (U6K), Differential Refractometer detector (R401) and Carbohydrate Analysis Column. A mobile phase of acetonitrilewater in the ratios of 84:16 and 65:35 were used for separation of simple sugars and oligosaccharides, respectively. The flow rate was 20 ml/min and 20 ul of sample were injected. Areas of the peaks of the sample were calculated and expressed relative to the areas with standard solutions. Triplicate samples were analyzed and average was taken for the results of chemical data.

Sensory evaluation

A 40 graduate and undergraduate students were asked for descriptions of odor and taste of soymilk by using

flavor profile methods. (21) From the result, descriptions which were considered as the most representative for soymilk's odor and taste were selected. The descriptions used for sensory tests were beany, cooked beany, sour and roasted nutty for odor of soymilk and beany, cooked beany, grassy and roasted nutty for taste. Eleven panelist were selected from initial 40 persons and trained. Sensory test was carried out in sensory test room having separate booths at 10:30 am and 3:00 pm. Sensory quality of soymilks were compared by multiple comparison method (21) using 7 point scale where '1' represent extremely week, '4' for moderate and '7' for extreemly strong. Each panelist tested samples in triplicate and the results were statistically analyzed by analysis of variance. (21)

Results and Discussion

The proximate composition of soybeans used in this study was 9.3% of moisture, 35.5% of protein, 21.7% of lipids, 28.4% of carbohydrate and 4.7% of ash. The results obtained for germination effect on yields, oligosaccharides and organoleptic properties of soymilk are discussed as follow.

Yields

Soybeans were soaked for 4 hours and then germinated which was followed by soymilk preparation. The solids or protein yields, the ratio of their weights recovered in soymilk to those weight in dry soybeans, were shown in Table 1. The solids yield and protein yield were in the range of 70.9-81.8% and 77.1-87.9, respectively. These values are generally higher than those values of other data reported⁽³⁾ but comparable to those of steam-infusion cooking method.⁽⁵⁾ The high

yields obtained in this study is probably due to several repeatation of extraction which would recover most of extractible solids in soybeans. Eventhough the Illinois process yields higher solids and protein into soymilk than our results, it is not comparable because the process is based on nonextraction method while ours was prepared by extraction of soluble components.

The solids yields were decreased relatively slowly during 5 day-germination, while protein yield was rather increased in initial germination period reaching maximum of 92.5% after 2 days. The initial increase in protein yield may indicate that the rate of increase in soluble protein by enzymatic hydrolysis of insoluble protein was faster than decreasing rate of protein which was used for metabolism for germination. This result is agreeable with the data of Suberbie et al. (22) They found an increase in dispersibility of full-fat soy flour when soybeans were germinated. However Yang and Kim (23) who studied the changes in nitrogen compounds during germination did not show such an increase in soluble nitrogen but showed a rapid increase in soluble non-protein nitrogen.

Flatulance factors

Fig. 2 and 3 show the changes in simple sugars and oligosaccharides, expressed as glucose, in soymilk prepared from germinated soybeans. The oligosaccharides such as raffinose and stachyose known as flatulence factors, and sucrose were reduced rapidly in general as the germination proceeds. However the decrease appears somewhat different between sucrose and oligosaccharides. The raffinose and stachyose showed a fast initial decrease while sucrose slightly increased at first day before rapid reduction thereafter. It is probably due to the mode of α -galactosidase action

Table 1. Effect of germination on yields of solids and protein in soymilk

	Soaked	Germination (days)				
		1	2	3	4	5
Solids yield(%)a	81.77	81.75	79.18	75.57	72.55	70.94
Protein yield(%)b	87.90	91.50	92.50	89.21	85.79	77.09

 $^{^{}a}$ % solids yield = $\frac{\text{wt. of total solids recovered in soymilk}}{\text{wt. of total solids in dry soybean}} \times 100$

^b% protein yield = $\frac{\text{wt. of protein recovered in soymilk}}{\text{wt. of protein in dry soybean}} \times 100$

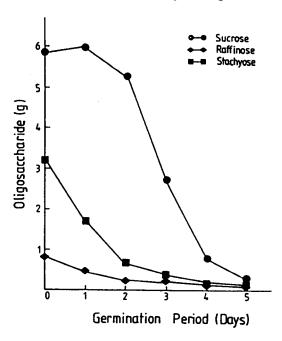


Fig. 2. Changes in oligosaccharides content of soymilk prepared from germinated soybeans. The values of sugars are expressed as glucose recovered from 100g of dry soybeans

which breakdown the glycosidic linkage from galactose unit of the oligosaccharides. Therefore the stachyose would produce raffinose, sucrose, and monosaccharide,

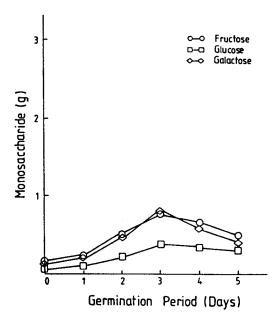


Fig. 3. Changes in monosaccharides content of soymilk prepared from germinated soybeans. The values of sugars are expressed as glucose recovered from 100 g of dry soybeans

in order, as the enzyme hydrolysis proceeds. After 2 days of germination, raffinose and stachyose reduced by 71.2% and 82.3%, respectively.

On the other hand, simple sugars in soymilk increased steadly until initial 3 day-germination which was followed by a slow decrease (Fig. 3). The decrease of simple sugars at later stage of germination would be caused by a fast utilization of monosaccharides for growth metabolism and a decrease in rate of monosaccharide production from oligosaccharides. Our data obtained are agreed with other reports(16,17) and 2-3 days of germination was found to be effective for removal of most of oligosaccharids from soymilk.

Sensory quality

To determine the proper boiling time for evaluation of sensory qualities of soymilk as they are affected by germination of soybeans, the soymilk prepared from ungerminated soybeans were boiled over a period of 40 minutes. The descriptions of odor and taste which were selected from flavor profile method were scored by using 7 point hedonic scale test. Fig. 4 showed that odor intensity of beany and sour was decreased and cooked beany and roasted nutty was increased as boiling time increased. The reduction of beany off-flavor were evidient by boiling. All of the changes were slowed down after 20 minutes of boiling.

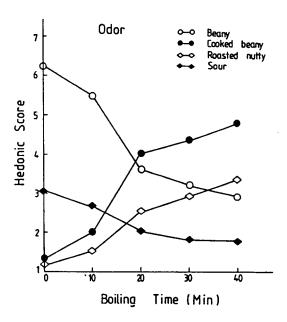


Fig. 4. Effect of boiling time on odor scores of soymilk at 8% solid level

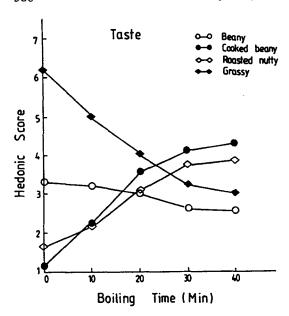


Fig. 5. Effect of boiling time on taste score of soymilk at 8% solid level

Fig. 5 shows the effect of boiling time on taste of soymilk. It was found that grassy taste reduced significantly and both roasted nutty and cooked beany increased, in similar tendency, from very little intensity to moderate, while beany taste decreased very slowly. The F-values calculated by analysis of varience for each flavor description showed that all of them were signifi-

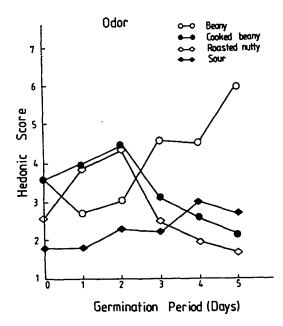


Fig. 6. Effect of germination on odor score of soymilk at 8% solid level

cant at 5% level for treatment of boiling.

Since the objectionable flavor decreased and desirable flavor such as roasted nutty increased rapidly until 20-30 minutes of boiling, boiling of soymilk for 30 minutes was accepted for further study of germination effect on soymilk's sensory quality.

Fig. 6 and 7 show the changes in odor and taste of soymilk as affected by germination of soybeans. The beany odor and taste reduced their intensity to minimum at first and second day of germination, respectively, which were followed by a rapid increase. Grassy taste which has an unfavorable effect on consumers preference showed a similar change as beany taste. Therefore it is conclusive that germination of soybeans has a significant effect for reduction of undesirable flavor of soymilk. On the other hand roasted nutty flavor, a generally accepted as desirable flavor for soymilk, increased to maximum after two days of germination for both odor and taste, and then decreased rapidly to the intensity score of between the range of 1-2. The total acceptability of soymilk was also found to be changed in same manner as roasted nutty flavor.

From the results of our study it is concluded that germination of soybeans for two days at room temperature before grinding was found to be benificial for reduction of flatulence factors and improvement of protein yield and sensory properties of soymilk.

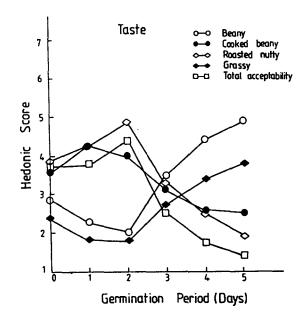


Fig. 7. Effect of germination on taste score of soymilk at 8% solid level

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대두발아에 의한 콩우유의 과당과 관능적품질의 변화

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25°C 에서 5일간 발아시킨 대두로 콩우유를 제조한후 발아가 콩우유의 당과 향미에 미치는 영향을 조사하였다. 2일간 발아시켰을때 단백질의 수율이 약간 중가함과 동 시에 stachyose와 raffinose는 급격한 감소를 보여 콩우 유의 화학적 품질 향상을 보여 주었다. 또한 2일 발아시 불쾌한 콩비린맛과 풀맛 그리고 콩비린 냄새가 최소치에 도달하였으며 전체적인 기호도는 가장 높았다. 구수한 냄새와 맛은 5일간의 발아 과정에서 전체적인 기호도의 변화와 유사한 변화 경향을 보여 주었다.