

Changes in the Quality of Soybean During Storage. Part 1—Effect of Storage on some Physico-chemical Properties of Soybean

Ranjana Narayan, G. S. Chauhan & N. S. Verma

Department of Food Science and Technology,
G. B. Pant University of Agriculture and Technology,
Pantnagar-263 145, India

(Received 30 October 1986; accepted after revision 6 May 1987)

ABSTRACT

Soybean (Glycine max) seeds of the Bragg variety stored in Jute bags under ambient conditions for 1, 2, 3 and 9 years were analysed for some physico-chemical characteristics to investigate the effect of storage period. The kernel weight and density decreased whereas hardness increased. The colour changed from creamy yellow to brown and grains stored for three years were slightly infested whereas those stored for nine years were heavily infested with insects. Among chemical characteristics, moisture content, fat, water-soluble nitrogen (WSN), nitrogen solubility index (NSI), sugars, trypsin inhibitor activity, available lysine, pigment and lipoxygenase activity of seeds decreased during storage whereas non-protein nitrogen (NPN), extent of browning, free fatty acid (FFA) content and peroxide value increased. Ash content and phytic phosphorus first increased and thereafter decreased with increasing storage period.

INTRODUCTION

Although soybean cultivation has been known for a long time, commercialisation is in its infancy in most parts of the world (Frank, 1974). However, due to its high content of good quality protein, the cultivation of soybean is now under consideration in some countries. With the increasing production of soybeans, a proper storage technology is important to maintain quality and to reduce losses. Maintenance of quality in stored seeds of all kinds is

essential if economic losses are to be avoided and availability is to be enhanced (Christensen & Kaufmann, 1969).

Various physical, chemical and biological changes take place in grains during storage depending on the storage conditions, storage time and condition of grains to be stored. If these physical and chemical changes reach beyond a certain limit, the grains are considered deteriorated. Thus the acceptability and suitability of grains are directly related to the various physico-chemical changes which occur during storage. Since the information on the effect of storage under ambient conditions (on physico-chemical properties of soybean seeds) is scanty, the present investigation was undertaken.

MATERIALS AND METHODS

Soybean seeds (Bragg variety) harvested in different years—1976, 1982, 1983 and 1984—and stored under ambient conditions (temperature ranged from 16 to 40°C and relative humidity from 50% to 90%) were analysed for various physico-chemical characteristics.

Physical characteristics

The observations regarding the changes in colour and insect infestation were made visually. Soybean grains of different samples were subjected to physical tests such as thousand kernel weight (Pomeranz, 1971), density and kernel hardness (Obuchowski & Buschek, 1980).

Proximate composition

This was determined by using standard AOAC (1966) methods.

Proteinaceous components

Water-soluble nitrogen was estimated by the AACC (1976) method, non-protein nitrogen by the Bhatti & Finglayson (1973) method and available lysine by Carpenter's (1960) method.

Fat characteristics

Oil samples extracted from different soybean samples were analysed for peroxide value using the AOCS (1960) method and free fatty acids by the AOCS (1954) method. The methyl esters of fatty acids were prepared by

the sodium methoxide trans-esterification procedure. Esters were separated by a GLC dual column gas chromatograph equipped with flame ionisation detector for free fatty acid determination. The conditions used for operating GLC were: stainless steel column (8 ft \times $\frac{1}{8}$ in) packed with 15% DEGS on chromosol (80–100) mesh; column temperature, 180°C isothermal; injector and detector adjusted to 200°C; carrier gas, N₂, 25 ml min⁻¹; sensitivity, 0.75×10^{-9} and chart speed, 1 cm min⁻¹. The peaks thus obtained were subjected to quantitative analysis.

Pigment, browning and sugars

Pigments and sugars were analysed by the AACC (1976) method and the extent of browning by the procedure as described by Ranganna (1979).

Water absorption and extraction of total solids and proteins

Soybean grains were soaked in water overnight and blanched in 0.5% sodium bicarbonate solution for 30 min. Per cent water absorption and losses of total solids in soaked and blanched water were determined using the standard methods of the AOAC (1966) and proteins by the AACC (1976) method.

Anti-nutritional factors

Trypsin inhibitor activity was determined by the Kakede *et al.* (1974) method. Lipoxygenase activity was determined by using the thiocyanate method of Sumner (1943) as modified by Koch *et al.* (1958). Phytic phosphorus was determined by using the method described by Tara *et al.* (1971).

RESULTS AND DISCUSSION

Table 1 summarises the results showing the changes in physical characteristics of soybeans during storage. Colour of stored soybeans changed from creamy yellow to brown with the increase in storage period and the intensity of the colour change increased at ambient temperature during storage. This was in accordance with the earlier findings of Friedlander & Navarro (1972), Yoshino *et al.* (1977), Saio & Baba (1980) and Saio *et al.* (1982). The thousand kernel weight and density of soybeans decreased whereas hardness increased during storage (Table 1). The decrease in thousand kernel weight and density and increase in hardness of soybean

TABLE 1
Effect of Storage Period on some Physical Characteristics of Soybeans

Storage period (years)	Colour	Insect infestation	Thousand kernel weight (g) ^a	SD	Density (g/ml) ^a	SD	Kernel hardness (kg per grain) ^a	SD
1	Creamy yellow	—	151.8	0.08	0.7474	3.3×10^{-4}	11.9	0.06
2	Creamy yellow	—	148.9	0.04	0.7455	4.1×10^{-4}	12.9	0.03
3	Slight brownish yellow	+	143.3	0.08	0.7425	2.6×10^{-4}	13.4	0.05
9	Light brown	++	127.8	0.06	0.7150	2.9×10^{-4}	14.6	0.06

—, no infestation; +, slightly infested; ++, heavily infested; ^a Average of 25 replications; SD, Standard deviation.

TABLE 2
Effect of Storage Period on Proximate Composition (per cent, dry basis) of Soybeans

Storage period (years)	Moisture	SD	Protein	SD	Fat	SD	Ash	SD	Carbohydrate (by difference)	SD
1	9.00	0.07	36.8	0.04	20.7	0.03	5.34	0.02	37.2	0.08
2	8.4	0.06	36.6	0.08	19.3	0.04	5.35	0.03	38.8	0.04
3	8.00	0.07	35.5	0.12	18.8	0.06	5.30	0.03	40.4	0.06
9	7.70	0.06	27.4	0.04	18.1	0.06	5.00	0.1	49.5	0.07

SD, Standard deviation.

grains was probably due to certain biochemical changes resulting in the evolution of CO₂ and due to evaporation of moisture. The possibility of insect infestation for these changes can also not be ruled out. These findings compared favourably with those obtained by the studies of Matsui *et al.* (1980), Saio & Baba (1980) and Iwata *et al.* (1982).

Proximate composition

Moisture and fat contents decreased constantly with the increase in storage period whereas protein content decreased considerably in the samples stored for 9 years as compared to others (Table 2). Ash content of the samples stored for 1, 2 and 3 years remained almost the same whereas it decreased in the samples stored for nine years. This was attributed to the loss of cotyledons due to insect infestation during storage of soybeans. These results were in accordance with those reported by Lakshminarayana *et al.* (1973), Gavrechenkov & Sinha (1980) and Ochiai *et al.* (1985).

Proteinaceous components

From Table 3 it can be seen that the changes in WSN, NSI and NPN and available lysine contents were not so conspicuous in the samples of soybeans stored for 1, 2 and 3 years as in the samples stored for 9 years. The decreases in WSN, NSI and available lysine were probably due to the occurrence of the Maillard reaction in which some insoluble compounds are formed, thereby reducing the solubility of nitrogen (Saio *et al.*, 1980, 1982) and availability of lysine (Zimmerman *et al.*, 1969). The observed increase in NPN content of soybeans may be due to enzymic hydrolysis during storage (Saroj, 1979; Tomar, 1985).

Fat characteristics

Table 4 shows the effect of storage period on peroxide value and some fatty acid contents of soybeans. The peroxide value and FFA content increased remarkably after two years' and nine years' storage of soybeans. The increase in PV was attributed to the formation of peroxides from unsaturated fatty acids whereas the increase in FFA content was due to hydrolytic changes in fat components. From Table 4 it can be seen that the trienoic acids underwent the maximum reduction as compared to dienoic and monoenoic acids. The palmitic, stearic and oleic acid contents increased during storage whereas linoleic and linolenic acid contents decreased considerably. The decrease in polyunsaturated fatty acids during storage is due to oxidative deterioration. These results compared favourably with

TABLE 3
Effect of Storage Period on some Proteinaceous Components of Soybean

Storage period (years)	W/SN ^a (%)	SD ±	NSI ^b (%)	SD ±	NPN ^c (%)	SD ±	Available lysine (g/100 g of protein)	SD ±
1	3.62	0.05	61.5	0.62	0.04	2.8×10^{-3}	5.9	0.02
2	3.37	0.08	57.6	0.58	0.05	3.6×10^{-3}	5.4	0.03
3	2.80	0.06	50.0	0.85	0.06	2.4×10^{-3}	5.0	0.02
9	1.90	0.08	42.0	0.49	0.11	3.6×10^{-3}	3.8	0.03

^a Water-soluble nitrogen.

^b Nitrogen solubility index.

^c Non-protein nitrogen.

SD, Standard deviation.

TABLE 4
Effect of Storage Period on Peroxide Value and some Fatty Acid Contents of Soybeans

Storage period (years)	PV ^a (meq/kg of oil)	SD ±	FFA ^b (%)	SD ±	Palmitic acid (%)	Stearic acid (%)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)
1	18	0.82	0.69	0.04	14.9	1.97	26.40	48.7	5.53
2	40	0.78	4.31	0.03	15.2	2.00	27.21	47.0	4.41
3	65	0.54	5.37	0.04	17.0	2.86	27.34	45.5	3.22
9	98	0.65	9.85	0.03	20.0	3.41	28.45	45.1	2.10

^a Peroxide value.

^b Free fatty acid.

SD, Standard deviation.

those obtained by Yanichek & Iraskova (1971), Maga & Johnson (1972), Robertson *et al.* (1973), Harman & Mattick (1976), Gupta & Rost (1976) and Stewart & Bewley (1980).

Extent of browning, pigment and sugars

The results showing the effects of storage period on extent of browning, pigment and sugar content of different samples are summarised in Table 5. The increase in extent of browning of samples with increase in storage period was attributed to the occurrence of enzymatic and non-enzymatic browning reactions. Friedlander & Navarro (1972) reported that the Maillard reaction was the main contributing factor to this. Pigment content (carotenoids) decreased as a function of storage time. This decrease was due to degradation of the carotenoid pigment during storage. The reduction in reducing sugars was attributed to their participation in Maillard reactions whereas the reduction in non-reducing sugars may be attributed to their enzymatic hydrolysis (Iwata & Shirahata, 1979; Matsui *et al.*, 1980).

Water absorption and extraction of total solids and protein during soaking and blanching of stored soybeans

Table 6 shows the results of water absorption and extraction of total solids and protein during soaking and blanching of soybeans as affected by storage period. The water absorption decreased whereas the extraction of total solids and protein increased during soaking and blanching of soybeans with increase in storage period. The reduction in water absorption capacity of seeds with increase in storage period may be due to the losses in hygroscopicity of proteins and carbohydrates during storage (Yoshino *et al.*, 1977). However, the increase in extractability of total solids and protein during soaking and blanching may be due to the breakdown of cellular membrane comprised of phospholipids (Saio *et al.*, 1980).

Anti-nutritional factors

Table 7 summarises the changes in phytic phosphorus content, trypsin inhibitor and lipoxygenase activity of soybeans during storage. Phytic phosphorus content decreased during storage but the result also depended upon the amount of total phosphorus present in the sample at the time of determination. The observed decrease in phytic phosphorus content of soybeans stored for nine years may be due to breakdown of phytic acid by the enzyme phytase during storage (Ebine *et al.*, 1983). Trypsin inhibitor and lipoxygenase activity decreased during storage, possibly because trypsin

TABLE 5
Effect of Storage Period on the Extent of Browning, Pigments and Sugars of Soybean

Storage period (years)	Extent of browning (OD)	SD ±	Pigment (carotenoids) (ppm)	SD ±	Sugars			
					Reducing (mg maltose 10 g^{-1} of soy flour)	SD	Non-reducing (mg sucrose 10 g^{-1} of soy flour)	SD
1	0.06	0.008	22.5	0.49	176.9	0.41	214	0.074
2	0.16	0.008	18.2	0.45	164.8	0.34	192	0.09
3	0.22	0.04	12.3	0.54	141.4	0.37	175	0.12
9	0.29	0.04	8.9	0.49	125.7	0.42	139	0.31

SD, Standard deviation.

TABLE 6
Effect of Storage Period on Water Absorption and Extraction of Total Solids and Protein during Soaking and Blanching of Soybeans

Storage period (years)	Soaked water				Blanched water					
	WA ^a (%)	SD ±	Total solids (%)	SD ±	WA ^a (%)	SD ±	Total solids (%)	SD ±	Protein (%)	SD ±
1	48.9	0.26	6.76	0.05	35.0	0.37	6.35	0.05	1.30	0.01
2	45.5	0.17	7.10	0.11	31.7	0.21	7.64	0.04	1.54	0.01
3	43.3	0.25	7.26	0.06	29.2	0.21	8.00	0.07	1.90	0.02
9	42.2	0.26	8.35	0.05	25.0	0.21	8.90	0.05	2.24	0.02

^a Water absorption.
SD, Standard deviation.

TABLE 7
Effect of Storage Period on some Antinutritional Factors of Soybeans

Storage period (years)	Total phosphorus (%)	SD \pm	Phytic phosphorus		TIA ^a (%)	SD \pm	Lipoxygenase activity (units g ⁻¹)	SD \pm
			On sample basis (%)	SD \pm				
1	0.62	0.005	0.44	0.006	56.9	0.36	26 528	5.1
2	0.62	0.006	0.48	0.006	53.6	0.33	17 784	3.4
3	0.67	0.004	0.60	0.008	49.4	0.17	8 514	3.9
9	0.58	0.006	0.30	0.007	42.4	0.29	—	—

^a Trypsin inhibitor activity (% per millilitre of extract).

SD—Standard deviation.

inhibitor, being proteinaceous in nature, might be denatured on exposure to varying temperature and humidity during storage (Shlosberg, 1977; Yao *et al.*, 1983). The reduction in lipoxygenase activity may be attributed to the denaturation of lipoxygenase enzyme and also to the reduction in the substrate concentration, i.e. linoleic acid, during the storage period (Shlosberg, 1977; Melton *et al.*, 1981; Yao *et al.*, 1983).

REFERENCES

- AACC American Association of Cereal Chemists (1976). *Cereal Laboratory Methods*, St. Paul, Minn., USA.
- AOAC Association of Official Analytical Chemists (1966). *Official methods of analysis* (10th edn), Washington, DC, USA.
- AOCS American Oil Chemist's Society (1954). *Official and tentative methods* (1st edn), (Hopper, T. H. (Ed.)).
- AOCS American Oil Chemist's Society (1960). *Official and tentative methods* (2nd edn), Chicago, USA. AOCS.
- Bhatty, R. S. & Finglayson, A. J. (1973). Extraction of non-protein nitrogen from oil seed meals with different solvents. *Cereal Chem.*, **50**(3), 329–39.
- Carpenter, K. J. (1960). Corrected straight acids. Procedure for determination of available lysine in foods. *Biochem. J.*, **77**, 604.
- Christensen, C. M. & Kaufmann, H. H. (1969). *Grain storage: The role of fungi in quality loss*, Minneapolis, Univ. Minn. Press.
- Ebine, H. (1983). Phytin in foods and its action—Miso brewing gets rid of the phytin problem. *Miso no kagaku to Gijutsu*, (353), 248–53.
- Frank, E. H. (1974). Soy protein products and their production. *JAACS*, **61**, 67–73.
- Friedlander, A. & Navarro, S. (1972). The role of phenolic acids in the browning, spontaneous heating and deterioration of stored soybeans. *Experientia*, **28**(7), 761–3.
- Gavrenchenkov, Y. D. & Sinha, R. N. (1980). Keeping quality of soybeans stored under aerobic and anaerobic conditions. *Cand. J. Pl. Sci.*, **40**(4), 1087–99.
- Gupta, A. K. S. & Rost, H. E. (1976). Changes in the lipids of soybeans during storage and germination. *Fette Seife Arisrichmittel.*, **78**(2), 59–61.
- Harman, G. E. & Mattick, L. R. (1976). Association of lipid oxidation with seed aging and death. *Nature*, **260**, 323–4.
- Iwata, T. & Shirahata, K. (1979). Keeping quality of green soybeans. I. The factors concerning post harvest quality deterioration and the effects of atmosphere and whole plant packing. *J. Jap. Soc. Hort. Sci.*, **48**(1), 106–13.
- Iwata, T., Sugiura, H. & Shirahata, K. (1982). Keeping quality of green soybeans by whole plant packaging. *J. Jap. Soc. Hort. Sci.*, **51**(2), 224–30.
- Kakede, M. L., Rackis, J. J., McGhee, J. C. & Pusking, G. A. (1974). Determination of trypsin inhibitor activity of soy products. *Cereal Chem.*, **51**, 376.
- Koch, R. B., Stern, B. & Ferrari, C. G. (1958). Linoleic acid and trilinolein as substrate for soybean lipoxydases. *Arch. Biochem. Biophys.*, **78**, 165.
- Lakshminarayana, T., Surendranath, M. R., Rao, S. D. T. & Reddy, B. R. (1973). Indian soybean: Composition and storage. *Oils and Oilseeds J.*, **26**(2), 7–12.

- Maga, J. A. & Johnson, J. A. (1972). Effect of processing and storage conditions on lipid composition of soy products. *Cereal Chem.*, **49**(1), 79–85.
- Matsui, S., Hayashi, K. & Nakaymura, M. (1980). Influence of low temperature storage and treatment of some chemicals on changes in the quality of green soybeans. *Res. Bull. Faculty Ag., Gifu Univ.* (43), 223–31.
- Melton, S. L., Moyers, R. E. & Jaynes, J. T. (1981). Storage effect on selected characteristics and lipids of defatted soy flours. *JAOCS*, **58**(11), 959–66.
- Obuchowski, W. & Bushcek, W. (1980). Wheat hardness: Comparison of methods of its evaluation. *Cereal Chem.*, **58**, 421.
- Ochiai, Yanagi, S., Galcazzi, M. A. M. & Saio, K. (1985). Properties of soybean in model storage studies. *Agri. and Biol. Chem.*, **49**(2), 525–9.
- Pomeranz, Y. (1971). *Wheat chemistry and technology* (2nd edn), American Association of Cereal Chemists, St. Paul, Minn., USA.
- Ranganna, S. (1979). *Manual of analysis of fruit and vegetable products*, New Delhi, Tata McGraw-Hill, 634 pp.
- Robertson, J. A., Morrison, W. H. & Burdict, D. (1973). Chemical evaluation of oil from field and storage damaged soybeans. *JAOCS*, **50**(11), 443–5.
- Saio, K. & Baba, K. (1980). Microscopic observation on soybean structural changes during storage. *J. Jap. Soc. Fd. Sci. Tech.*, **27**(7), 343–7.
- Saio, K., Nikkuni, I., Ando, Y., Otsuru, M., Terauchi, Y. & Kito, M. (1980). Soybean quality changes during model storage studies. *Cereal Chem.*, **57**(2), 77–82.
- Saio, K., Kobayakawa, K. & Kito, M. (1982). Protein denaturation during model storage studies of soybeans and meals. *Cereal Chem.*, **59**(5), 408–12.
- Saroj, S. (1979). *Storage studies on proteinsnack*. MSc Thesis submitted to G. B. Pant University of Agriculture and Technology, Pantnagar.
- Shlosberg, S. S. (1977). *The effect of soybean storage conditions on trypsin inhibitor, lipooxygenase and quality*. MSc thesis, University of Illinois, Urbana, Ill., USA.
- Stewart, R. R. C. & Bewley, J. D. (1980). Lipid peroxidation associated with accelerated aging of soybean, *Axe's Pl. Physiol.*, **65**(2), 245–8.
- Sumner, R. J. (1943). Lipid oxidase studies. Method for the determination of lipoxidase activity. *Ind. Engg. Chem. Anal. ed.*, **15**, 14.
- Tara, K. A., Usha, M. S., Prabhavati, C. & Bains, G. S. (1971). Vitamins, minerals, colour grade and limiting amino acid contents of Indian commercial flours. *Res. Ind.*, **16**(3), 196.
- Tomar, N. S. (1985). *Bottle processing of soy milk*. MSc thesis submitted to G. B. Pant University of Agriculture and Technology, Pantnagar.
- Yanichek, G. & Iraskova, L. (1971). Resistance of soybean flour to oxidative rancidity. *Voprosypitaniya*, **30**(5), 63–5.
- Yao, J. J., Wei, L. S. & Steinberg, M. P. (1983). Effect of maturity on chemical composition and storage stability of soybeans. *JAOCS*, **60**(7), 1245–9.
- Yoshino, U., Iwasaki, Y., Okubo, M. & Okuyama, T. (1977). Effect of storage conditions on soybean protein. *J. Jap. Soc. Fd. Sci. Tech.*, **24**(10), 526–9.
- Zimmerman, G., Ben-gara, I., Weissmann, S. & Yanai, S. (1969). Storage under controlled conditions of dry staple foods (defatted milk powder, wheat, soybeans and defatted soybean meal) and its influence on their protein nutritive value. *Deut. Ges. Chem. Apparateswesen. Monograph.*, **63**, 347–80.