Pak. j. sci. ind. res., vol.39, nos. 9-12, September-December 1996

EFFECTS OF SOAKING AND COOKING ON PHYSICAL CHARACTERISTICS, TANNIN CONTENTS AND PROTEIN DIGESTIBILITY OF KIDNEY BEANS (*PHASEOLUS VULGARIS* L.)

ZIA-UR-REHMAN AND W.H. SHAH,

PCSIR Laboratories Complex, Shahrah-e-Jalaluddin Roomi Lahore-5600, Pakistan

(Received October 5, 1995)

This study reports the effects of soaking and cooking methods on some physical characteristics, tannin contents and in vitro protein digestibility of kidney beans. Soaking of kidney beans in sodium bicarbonate solution gave rise to lower hydration capacity, hydration index, swelling capacity and cooking time than soaking in simple water or neutral sodium chloride solution. Tannin contents of red kidney beans were reduced by soaking at 30° and 100°C for different periods. However, soaking in sodium bicarbonate solution with or without sodium chloride was more efficient in reducing tannin contents. Maximum improvement in protein digestibility was also observed by soaking kidney beans in mild alkaline solutions. Tannin contents were further reduced alongwith improvement in protein digestibility as a result of cooking.

Key words: Kidney beans, Soaking, Cooking, Tannin, In vitro protein digestibility.

Introduction

Kidney beans *(Phaseolus vulgaris L.)* are legume seeds and consumed widely in many areas of the world [1]. It is a good source of protein (25%) but the use of its protein has been limited by the presence of tannin and phytic acid [2]. Tannins are reported to be responsible for decreasing feed intake, growth rate, feed efficiency, net metabolizable energy and protein digestibility. Such effects are caused through the formation of complexes with protein, starch and digestive enzymes [3,4]. Attempts have been made to remove or inactivate tannins in food and feed. The technologies developed for this purpose involve the identification of low tannin cultivars in cereals and legumes, pearling or dehusking the grains, soaking of grains in water and various chemical solutions, cooking, sprouting and drying of grains [4-6]. Traditional methods such as soaking in water, salt solution, alkali solution, acidic solution and cooking improve the nutritive value of the legumes by inactivation of antinutritional factors [7-9]. The present paper reports the tannin contents and protein digestibility of raw kidney beans, after soaking in water, sodium chloride and sodium bicarbonate solutions and cooking at 15 lbs/inch^2 for 5 min. Tannin contents, protein digestibility and physical characteristics of red as well as white kidney beans after soaking have also been studied.

Materials and Methods

Red and white kidney beans were obtained from Ayub Agricultural Research Institute, Resalewala, Faisalabad. These cultivars were grown under similar soil and agro-climatic conditions. Beans were cleaned to remove broken seeds, dust and other foreign materials and then subjected to various soaking treatments prior to cooking.

Soaking treatments. The beans were soaked in simple water, 1% sodium chloride solution, 1% sodium bicarbonate solution and 1% sodium bicarbonate + 1% sodium chloride solution at 30° and 100°C for 30, 60 and 90 min. The beans to water ratio was 1: 5. The soaked beans were rinsed twice with distilled water and then dried in hot air oven at 55°C for 24 hr.

Cooking treatments. Presoaked beans (without drying) were placed in one litre beakers containing tap water (four times the weight of dry beans). Top of the beakers was covered with aluminum foil. After cooking in a pressure cooker (15 lbs/inch^2) for 5 min., the beans were mashed and dried in hot air oven at 55°C for 24 hr. Raw and processed kidney beans were ground in a willey mill to pass through a 40 mesh sieve.

Estimation of physical characteristics. Fifty beans (randomly selected) were weighed and transferred to a 250 ml Erlenmeyer flask. After adding 20 ml distilled water, the flask was tightly stoppered and left over night (16 hr.) at room temperature (23.5±1°C). The beans were then drained, blotted to remove excess water and reweighed. Hydration capacity per seed was recorded as weight after soaking minus weight prior to soaking [8].

Hydration index was determined as the ratio of the hydration capacity to the original seed weight. Some soaked seeds were reweighed and transferred to a 200 ml measuring cylinder containing 100 ml water. The increase in seed volume after hydration was recorded as swelling capacity, which was measured by water displacement as volume after soaking minus volume of the seeds before soaking [8].

Chemical analysis. Moisture content of kidney beans was estimated by placing a ground sample (3-5g) at 100 ±

5°C in an oven for 20-24 hr. till a constant weight was attained, whereas ash content was determined after ignition at 550 ± 5°C in a muffle furnace for a period of 4hr. [10]. Crude protein contents of the kidney beans were estimated after digestion with sulphuric (conc) acid using $CuSO_4$: K_2SO_4: SeO_2 (1: 9: 0.02) as catalyst according to Micro Kjeldahl Method. Fat contents were determined after extracting with n.hexane, while crude fibber was measured after digestion with 1.25% sulphuric acid and 1.25% sodium hydroxide solutions as described in A.O.A.C. [10]. Neutral detergent fiber and acid detergent fiber was estimated by the method of Van Soest and Wine [11]. Tannin content of the samples was estimated on spectrophotometer at 760 nm using Folin Denis Reagent accoding to A.O.A.C. method after extraction with 1% hydrochloric acid in methanol [11].In vitro protein digestibility was determined after digestion with pepsin-HCI solution at 37.5°C for 24 hr. [12]. Data of soaked and cooked beans were recorded and statistically analyzed according to Steel and Torrie [13]. Differences between treatments at 5% level were considered significant.

TABLE 1. EFFECT OF STORAGE ON PROXIMATE COMPOSITION, TANNIN CONTENTS AND PROTEIN DIGESTIBILITY OF RAW KIDNEY BEANS.

Parameters* (%)	Red kidney beans		White kidney beans	
	Freshly harvested	After 6 months storage	Freshly harvested	After 6 months storage
Moisture	3.69±1.1	3.43±0.3	4.01±0.6	3.71±0.8
Crude protein	24.77±3.6	24.44±4.4	23.96±3.9	23.86±1.2
Ash (minerals)	4.26±0.7	4.31±0.3	3.98±0.6	3.86±0.5
Crude fat	2.11±0.8	2.73±0.2	2.76±0.2	2.90±0.2
Crude fibre	4.68±0.3	4.93±0.2	4.73±0.3	5.1±0.2
Neutral detergent Fibre	18.32±0.5	19.73±0.6	17.86±0.7	18.32±1.8
Acid detergent Fibre	7.09±0.4	8.1±0.4	7.12±0.8	7.93±0.9
Tannin	1.13±0.2	1.16±0.3	0.77±0.2	0.73±0.1
Protein digestibility before cooking**	41.20±0.3	39.92±2.3	47.36±2.9	45.62±2.8
Protein digestibility after cooking***	58.61±1.0	54.00±1.6	62.40±1.7	59.09±2.0

* Average of three determinations alongwith standard deviation.
** Unsoaked and uncooked beans.
*** Unsoaked cooked beans.

Results and Discussion

It would be noted from Table 1 that the chemical composition of the freshly harvested red and white kidney beans was almost the same. However, the amount of tannin in red beans was higher (1.132) than white kidney beans (0.772), whereas in vitro protein digestibility before cooking was 41.20 and 47.36% respectively. These results indicated that in vitro protein digestibility (IVPD) was inversely proportional to tannin contents of kidney beans. Therefore, low tannin cultivar (white kidney beans) showed higher IVPD than high tannin cultivar (red kidney beans). Similarly, other workers also observed a higher protein digestibility for white compared with pigmented cultivars of *Phaseolus vulgaris* [3,14].

It would be evident from the data presented in Table 1 that chemical composition of both the cultivars remained unchanged during 6 months storage at ambient temperature. However, the decline in the protein digestibility observed during this storage might be due to the formation of insoluble complex compounds between tannins and proteins which probably reduced the availability of amino acids to proteolytic enzymes and ultimately reduced the digestibility of proteins. Reduction in protein digestibility after storage was comparatively less in red kidney beans, which might be due to the presence of larger amount of tannin. Statistical analysis showed that reduction in protein digestibility during storage was significant ($p<0.05$).

Effect of soaking on physical characteristics. It would be seen from Table 2 that physical characteristics of kidney beans were changed to some extent on soaking in different salt solutions. Hydration capacity, hydration index and swelling capacity values of kidney beans soaked in simple water and sodium chloride solution were significantly ($P<0.05$) higher than the kidney beens soaked in sodium bicarbonate solution with or without sodium chloride.

On the other hand, cooking time of the kidney beans soaked in sodium bicarbonate solution with or without sodium chloride was remarkably reduced as compared with beans soaked in other salt solutions. This could be attributed to restriction of inter and intra cellular membranes of the beans in alkaline solutions. The results indicated that soaking of kidney beans in sodium bicarbonate solution gave rise to lower hydration capacity, hydration index, swelling capacity and cooking time than soaking in water or sodium chloride solution alone.

Effect of soaking on tannin contents. Data presented in Table 3 indicated that considerable amount of tannin was removed from kidney beans by soaking in different salt solutions at 30° and 100°C for different periods. However, removal of tannins was found to be minimum in case of simple water soaking process. Soaking of kidney beans in simple water at 30° and 100°C for 90 min. removed about 11.5 and 36.2% tannins respectively, whereas about 17.69% at 30°C and 43.13% at 100°C, tannins were removed as a result of soaking in 1% neutral sodium chloride solution for 90 min. Removal of tannins during these soaking processes indicated easy penetration of water molecules into the beans which leached out tannins from the material. Rao and Doesthal

[15] also found that substantial amount of tannin was reduced from various leguminous materials during water soaking process.

Soaking in sodium bicarbonate solution alone or in combination with sodium chloride resulted in better removal of tannins from kidney beans than simple water or sodium chloride solution. Soaking of kidney beans in 1% sodium bicarbonate solution at 30°C and 100°C for 90 min. resulted in a reduction of tannins by 51.32 and 60.17% respectively. Almost similar results were obtained when kidney beans were soaked in sodium bicarbonate solution containing 1% sodium chloride. Statistical analysis of this data revealed that the reduction in tannin content was significantly different from each other ($P<0.05$) under the two conditions of soaking. The results identified that soaking temperature significantly affected the rate of extraction of tannins from kidney beans. They also suggested that rate of extraction of tannins was remarkably increased under alkaline conditions. Most likely some soluble sodium salts of tannic acid were formed in the presence of sodium bicarbonate which might be responsible for maximum reduction of tannins. However,exact mechanism of leaching of tannins is still unknown. It has already been reported earlier [16] that significant amount of tannins was removed from winged beans on soaking in sodium bicabonate solution.

Effect of soaking on protein digestibility. In vitro protein digestibility (IVPD) of kidney beans was significantly ($P<0.05$) improved due to soaking in different salt solutions

TABLE 2. EFFECT OF SOAKING ON PHYSICAL CHARACTERISTICS OF RAW KIDNEY BEANS*.

Soaking solution	Red kidney beans				White kidney beans			
	Hydration capacity. (mg/seed)	Hydration index	Swelling capacity. (ml.)	Cooking time (minutes)	Hydration capacity (mg/seed)	Hydration index	Swelling capacity (ml.)	Cooking time (minutes)
Simple water	8.6±1.9	60.2±1.2	78±1.0	10	11.9±1.5	77.4±2.2	90±1.2	9
1% NaCl solution	8.9±1.1	56.9±1.6	73±0.9	7	12.3±1.2	69.5±1.9	87±0.9	6
1% $NaHCO_3$ solution	8.4±2.3	55.5±1.3	70±2.1	4	11.4±1.4	66.9±1.7	84±1.6	4
1% $NaHCO_3$ + 1% NaCl	8.1±2.1	52.6±2.9	69±1.1	4	10.8±2.0	65.0±1.3	80±2.1	4

* Average of three determinations alongwith standard deviation.

TABLE 3. EFFECT OF SOAKING ON TANNINS AND PROTEIN DIGESTIBILITY OF RED KIDNEY BEANS*.

Soaking conditions		Simple water			Sodium chloride			Sodium bicarbonate			Sodium bicarbonate + Sodium chloride		
Temp °C	Time Minutes	Tannin %	Removed	IVPD %	Tannin %	Removed	IVPD %	Tannin %	Removed	IVPD %	Tannin %	Removed	IVPD %
30	30	1.09±0.09	3.53	43.61±1.21	1.03±0.07	8.84	44.04±1.45	0.61±0.09	46.01	54.38±1.71	0.59±0.07	47.78	55.72±1.73
	60	0.98±0.80	8.84	45.26±1.36	0.98±0.06	13.27	46.0±1.50	0.57±0.07	49.55	61.60±1.63	0.58±0.05	48.67	62.81±1.80
	90	1.00±0.09	11.50	49.00±1.26	0.93±0.05	17.69	51.26±1.33	0.55±0.05	51.32	63.36±1.40	0.52±0.08	53.98	64.00±1.43
100	30	0.81±0.07	28.31	47.27±1.11	0.77±0.08	31.85	49.36±1.27	0.50±0.07	55.75	64.00±1.44	0.48±0.08	57.52	65.73±1.06
	60	0.74±0.06	34.51	56.77±1.36	0.61±0.07	46.01	59.53±1.37	0.46±0.03	59.29	67.89±1.63	0.45±0.06	60.17	67.00±1.36
	90	0.72±0.08	36.28	58.60±1.25	0.62±0.09	45.32	60.32±1.50	0.45±0.09	60.17	68.47±1.38	0.44±0.07	61.06	68.30±1.55

*Average of three replicates along with standard deviation. (a) Significant at $P<0.05$ within the coloumn.

TABLE 4. EFFECT OF COOKING ON TANNINS AND PROTEIN DIGESTIBILITY OF RED KIDNEY BEANS.*

Treatment		Simple water			Sodium chloride			Sodium bicarbonate			Sodium chloride + Sodium bicarbonate		
Temp °C	Time Minutes	Tannin %	Removed %	IVPD %	Tannin %	Removed %	IVPD %	Tannin %	Removed %	IVPD %	Tannin %	Removed %	IVPD %
											(a)		(a)
30	30	0.98±0.09	13.27	67.06±4.1	0.92±0.06	18.58	69.0±1.9	0.49±0.05	56.63	77.23±2.1	0.47±0.06	58.40	76.01±1.8
	60	0.81±0.07	28.31	69.11±4.0	0.77±0.02	31.85	72.56±2.6	0.41±0.07	63.71	78.11±2.6	0.38±0.07	66.37	78.44±2.6
	90	0.79±0.06	30.08	73.08±5.0	0.70±0.01	38.05	74.00±3.0	0.37±.0.06	67.25	80.26±3.3	0.35±0.03	69.02	79.13±2.1
100	30	0.65±0.02	42.47	71.78±3.1	0.68±0.02	39.82	74.11±3.5	0.38±0.01	66.37	80.00±3.0	0.33±0.02	70.79	79.42.±4.4
	60	0.53±0/06	53.09	74.61±2.6	0.55±0.01	51.32	76.67±3.4	0.31±0.03	72.56	82.61±2.6	0.28±0.03	75.22	83.23±4.1
	90	0.49±0.05	56.63	75.66±2.0	0.50±0.03	55.75	77.71±4.1	0.29±0.07	74.33	84.00±2.7	0.27±0.03	76.10	85.00±3.7

* Average of three determinations alongwith standard deviation. (a) Significant at $P<0.05$ within the column. IVPD = In vitro protein digestibility.

at 30° and 100°C for different periods (Table 3). These results indicated that soaking temperature and time also affected protein digestibility of kidney beans to various extent. IVPD of unsoaked red kidney beans was 41.20% without cooking. However, IVPD of kidney beans became 49 and 58.60% on soaking in simple water at 30° and 100°C for 90 min. respectively. Similar results were obtained on soaking kidney beans in 1% sodium chloride solution at 30° and 100°C which indicated that sodium chloride did not play any significant role in the improvement of digestibility of proteins. As reported elsewhere [17], improvement in protein digestibility was probably due to partial removal of tannins by soaking in simple water.

Soaking in 1% sodium bicarbonate solution with or without sodium chloride improved IVPD of kidney beans to almost same extent. IVPD of kidney beans was 54.38 and 63.36% on soaking in 1% sodium bicarbonate solution at 30°C for 30 and 90 min. respectively. However, IVPD become 64.00 and 68.37% as a result soaking in sodium bicarbonate solution at 100°C for 30 and 90 min. respectively. Improvement in protein digestibility was about 40% more due to soaking in alkaline solutions than soaking in simple water or neutral sodium chloride solution. It has already been observed that protein digestibility of winged beans was considerably improved on soaking in sodium bicarbonate solution [17]. Significant improvement in protein digestibility due to soaking in alkaline solution may be attributed to the removal of polyphenolic compounds particulary tannins from red kidney beans. Partial removal of tannins from the kidney beans probably created large spaces within the matrix, which allowed the proteolytic enzymes to attack easily on protein profile available in soaked kidney beans and consequently increased its digestibility.

Effect of cooking on tannin contents. Tannin contents of red kidney beans were further reduced ($P<0.05$) as a result of cooking (Table 4). However, reduction of tannin was almost the same when kidney beans, soaked in simple water or in 1% sodium chloride solution, were cooked under pressure for 5 min. About 56% reduction in tannin contents was observed on cooking the presoaked red kidney beans in simple water or sodium chloride solution. Maximum reduction in tannins (about 75%) was achieved on cooking the kidney beans after soaking in sodium bicarbonate solution alone or in combination with sodium chloride. Previously it was observed [18,19] that tannin from winged and field beans were reduced during cooking process.

Effect of cooking on protein digestibility. In vitro protein digestibility was significantly ($P<0.05$) improved as a result of cooking the soaked kidney beans. It was observed that protein digestibility became 75.66% on cooking the water soaked kidney beans. Almost similar results were obtained when sodium chloride soaked kidney beans were subjected to cooking process. Significantly higher ($P<0.05$) protein digestibility (about 85%) was observed on cooking the kidney beans after soaking in sodium bicarbonate solution alone or in combination with sodium chloride. Improvement in protein digestibility of the red kidney beans may be due to the partial removal of tannins during soaking and cooking processes. Laurena *et al.* [19] and Poel [20] observed a significant improvement in protein digestibility due to removal of tannin from different legumes during cooking process.

It may be concluded from these findings that different soaking methods significantly affect the extraction of tannin from red kidney beans. Sodium bicarbonate soaking method is the most effective way for reducing tannin from kidney beans. Soaking temperature also plays significant role in the extraction of tannin from red kidney beans. Rate of tannin extraction from red kidney beans is about 2-3 times higher at 100° than 30°C. Cooking also brought about a greater reduction in the levels of tannin and significantly improved the protein digestibility. Removal of tannin may be attributed to leaching out of the antinutrient into the soaking solution under the influence of concentration gradient. Such losses may be taken as a function of changed permeability of seed coat. Soaking is an integral traditional method of processing legumenous grains in this part of the world and offers the dual advantage of reduction in energy cost by shortening cooking time as well as rendering the grains nutritionally superior.

References

1. V. C. Sgarbieri, World Rev. Nutr. Diet, **60,** 132 (1989).
2. D. K. Salunkhe, Curr. Sci., **51,** 387 (1982).
3. J. H. Hulse, Polyphenols in cereals and legumes (IDRC, Ottawa, Canada (1980).
4. S. S. Deshpande, S. K. Sathe and D. K. Salunkhe, Chemistry and Safety of plant Polyphenols, Adv. Exp. Med. Biol., **177,** 457 (1984).
5. J.K. Chavan, S.S. Kaelam, C.P. Ghonsikhe and D.K. Salunkhe, J. Fd. Sci., **44,** 1319 (1979).
6. S.S. Deshpande, S.K. Sathe, D.K. Salunkhe and D.D. Coruforth, J. Fd. Sci., **47,** 1846 (1982).
7. L.B. Rockland, R.J. Hayes, E. Metzler and L.J. Binder, U.S. Patent No. 3318708, May 9 (1967).
8. O. Onayemi, O.A. Osibogun and O. Obembe, J. Fd. Sci., **51,** 153 (1986).
9. R. A. Tentonico and D. Knorr, Fd. Tech., **39,** 127 (1985).
10. A. O. A. C., *Official Methods of Analysis* (Association of Official Analytical Chemists, Washington D.C.,

USA, 1980), 13th ed.
11. P. J. Van Soest and R.H. Wine, J. Assoc. Off. Anal. Chem., **50,** 50 (1967).
12. M.L. Price, L.G. Butler, J.C. Rogler and W.R. Featherston, J. Agric. Fd. Chem., **27,** 441 (1979).
13. R. G. D. Steel and J.H. Torrie, Principles and Procedures of Statistics (McGraw Hill, London, 1980), pp.345.
14. Z.Glick and P. Joslyn, J. Nutr., **100,** 516 (1970).
15. P. V. Rao and Y.J. Doesthal, J. Sci. Fd. Agric., **33,** 89 (1982).
16. B. O. Delumen and L.A. Salamat, J. Agric. Fd. Chem., **28,** 533 (1980).
17. A. C. Laurena, V.V. Garcia and E.T. Mendoza, Qual. Plant. Plant Foods Hum. Nutr., **36,** 107 (1986).
18. L.Elias, D.C. Fernandez and D.G. Bresani, J. Fd. Sci., **44,** 524 (1979).
19. A. C. Laurena, T. Van, Den and M. A. T. Mendoza, J. Agric. Fd. Chem., **32,** 1045 (1984).
20. A. F. B. Van der Poel, Animal Feed Sci. Tech., **29,** 179 (1990).