A STUDY ON THE EFFECT OF GERMINATION ON THE PROTEIN OF SOME OILSEEDS Mohamed, Samira S. and Fakhriya S. Taha Fats & Oils Department, National Research Centre, Cairo, Egypt

ABSTRACT

Soybean, sunflower and cottonseed kernels were germinated in different aqueous solutions for 12, 24 and 48 hours. The germinating solutions included: sodium carbonate-sodium bicarbonate buffer (pH 9.4), 2% ammonium carbonate, 2% ammonium carbonate + yeast, tap water and acidified water. Germination for 48 hours with 2% ammonium carbonate gave the highest improvement in oil content, protein content and N-solubility of the investigated kernels and defatted meals. The meals prepared under these best conditions showed a decrease in phytate content, and a slight decrease in urease activity of soybean and gossypol of cottonseed. Nitrogen solubility and digestibility were improved. Protein isolates were prepared from germinated defatted kernels and the non germinated defatted kernels which showed improvement in their functional properties especially their nitrogen solubility index.

INTRODUCTION

Oilseed meal protein is a highly potential unconventional protein source, that might help in filling part of the world protein gap, especially in developing countries.

Treatments to improve the nutritional quality of oilseed proteins has been investigated. Among these treatments some are traditional while others are purely scientific achievements. Among the traditional treatments used by many cultures to produce new food products, improve the palatability or nutritional value of existing ones are fermentation, soaking and germination. It has been generally accepted that soaking and sprouting or germination of

seeds increase their nutritional value (Bau and Debry, 1979; Liener, 1978) and Smith and Circle, 1958).

In Egypt, three major seeds are eaten after being soaked, water leached or germinated. Faba beans (*Viciae faba*) are soaked in water for 24 hr to sprout and the resulting product is "Foul Nabet". Fanugreek (*Trigonella foeum* graceum) sprouts are eaten fresh and called "Helba", and lupines (*Lupinus termis*) are boiled water leached then salted and eaten as "Termis". Similar products are characteristic of other countries. Perhaps the most widely consumed or germinated and soaked to prepare traditional foods is soybeans especially in countries of south east Asia.

Seed germination is a biological process in which the tiny plant restores its activity to full potential. This involves vigorous metabolism, driven by mobilization of the food materials stored in the seed. Cotyledons act as reserves for these stored materials. Both biological and chemical changes take place during germination (KeShun, 1997).

The aim of this work was to study the effect of germination of some oilseeds in different solutions on the chemical and functional properties of their proteins.

MATERIALS AND METHODS

Soybean (*Glycine max*), sunflower seed (*Hebinthus annus*) and cottonseed (*Gossypium barbadence*) were supplied by the Ministry of Agriculture as a crop of 1998. The seed were cracked, dehulled by sieving and blowing and the cracked kernels were then subjected to germination.

1. Germination of Kernels

50 g lots of each of the three types of kernels studied were placed between layers of moistened paper towels (moistened with the germinating solution examined), placed in plastic pans which were wrapped in plastic and placed in a dark place at room temperature.

50 g lots of each type of kernel were taken after 12, 24 and 48 hrs, placed in a freezer to stop germination, then lyophilized, ground and stored in a dessicator for analysis. This will make a total of 15 samples (50 g lots) for each type of kernels.

Aqueous solutions used for germinating the kernels

- a. Sodium carbonate-sodium bicarbonate buffer (pH 9.4)
 - 1.5 g Na₂CO₃ + 4.5 g NaHCO₃ in 300 ml water.
- b. 2% Ammonium carbonate (pH 8.4) 6 g (NH₄)₂CO₃ in 300 ml water.
- c. 2% Ammonium carbonate + yeast (pH 8.0) 6 g (NH₄)₂CO₃ + 3 g yeast in 300 ml.
- d. Tap water (pH 5.6).
- e. Acidified water (pH 3.6)
- f. Tap water adjusted to pH 3.6 with dilute HCl.

2. Boiling of Kernels in Different Solutions

50 g lots were placed with 300 ml of the solutions and refluxed at 100°C for 30 minutes.

3. Defatting of Germinated Kernels

10 g dry ground germinated kernels were extracted with n-hexane in a soxhlet apparatus to remove the oil and determine the oil content of the germinated kernels, also prepare the meal for further analysis.

4. Isoelectric Precipitation of Protein

The meal resulting from germinated, defatted kernels were used to prepare a protein extract for the determination of the isoelectric precipitation curve of the meal protein. 50 g of the meal were dissolved in 1250 ml of 0.02 N NaOH. 40 ml aliquots of the protein extract were placed in 60 ml centrifuge tubes and the pH values were adjusted to values between pH 3.0-7.0 using 6 N HCl. This was followed by centrifugation and % protein precipitated at each pH was calculated according to Taha *et al.* (1981).

5. Preparation of Protein Isolate

A protein extract is prepared as described in experiment (4). The pH was adjusted to the isoelectric point (IEP) of the protein determined in experiment (4). The supernatant was separated by decantation, then the precipitated protein washed with water and lyophilized EI-Nockrashy *et al.* (1977).

Analysis

Moisture, oil, protein, ash, fiber and urease activity were determined according to AOCS Method of analysis (1998). Nitrogen solubility according to Lyman, *et al.* (1953). Phytates were determined according to Wheeler and Ferrel, (1971). Free and total gossypol according to Pons, *et al.* (1958). Digestibility (Pedersen and Eggum (1983), water absorption capacity (Sosulski, 1962), emulsifying capacity (Swift, *et al.*, 1961). Nitrogen solubility index and protein despersibility index (AOCS, 1998).

RESULTS AND DISCUSSION

I: Effect of Germination on the Oilseed Kernels and their Defatted Meals

In an attempt to improve the nutritional quality of soybean, sunflower seed and cottonseed meal protein, mainly by improving the solubility and digestibility of the protein. Soybean, sunflower and cottonseed kernels were germinated in several aqueous solutions for three different time intervals. Boiling for 30 minutes in the difference aqueous solutions was also investigated.

Dehulled soybean, sunflower seed and cottonseed kernels were germinated in carbonate-bicarbonate buffer, 2% (NH₄)₂CO₃, 2% (NH₄)₂CO₃ + yeast, tap water and acidified water for 12, 24 and 48 hrs. These aqueous solutions had different pH values, ranging from 3.6 to 9.2.

I.1. Soybeans

Table 1. shows the effect of germination of soybean kernels in the different solutions for different periods of time on the oil and protein contents of the kernels. Generally, oil content of all germinated soybean kernels increased to varying degrees ranging from 4.8% - 20.2% increase over the non germinated kernels. The highest increase (25.03%) was achieved by germinating the kernels in 2% (NH₄)₂CO₃ for 48 hrs. compared to an increase of (20.82%) in non germinated soybean kernels (control).

Table 1. also indicate that germination of soybean kernels in all solutions resulted in a considerable increase in protein content which 4-12% more than protein content of the non-germinated meal. This highest increase was achieved by germinating soybean kernels in 2% (NH₄)₂CO₃ solution for 48 hrs. at room temperature. Increase in time of germination is concomitant with increase in protein content of germinated kernels.

Defatting of germinated kernels followed by grinding resulted in soybean meals with improved nutritional quality when compared to defatted soybean meal (control) prepared from non-germinated kernels.

Table 2. shows that, highest protein content and nitrogen solubility of defatted meal was achieved by using 2% (NH₄)₂CO₃ solution for 48 hours as in the kernels. As would be expected the protein content and nitrogen solubility are directly proportional to time of germination, increase in time causes increase in the protein and nitrogen solubility.

Highest increase in nitrogen solubility of meal protein of germinated soybean kernels in the different solutions for 48 hrs was in the following order 2% (NH₄)₂CO₃ (13.29%) > Na₂CO₃/NaHCO₃ (12.95%) > acidified water (10.3%) > water (9.73%) > 2% (NH₄)₂CO₃/yeast (8.03%), while highest increase in meal protein was in the following order (NH₄)₂CO₃ > Na₂CO₃/NaHCO₃ buffer > (NH₄)₂CO₃/yeast > water > acidified water with values 39.0% > 35.48% > 34.67% > 30.0% > 26.96%, respectively.

Results in both Tables 1. and 2. show that boiling of soybean for 30 min. in the different solutions is not recommended although it results in increase in the protein content due to the solubilization of some of the oil and carbohydrates during boiling, yet the nitrogen solubility of the protein is drastically decreased.

I.2. Sunflower

Table 3. gives values for oil content and protein content of sunflower germinated kernels in the different solutions investigated and Table 4. gives the protein values and nitrogen solubility % of the meal protein resulting from the germinated kernels after being defatted.

Sunflower kernels which contain 36.0% oils, after being germinated in Na₂CO₃/NaHCO₃ buffer, 2% (NH₄)₂CO₃, 2% (NH₄)₂CO₃/yeast, water and acidified water for 48 hrs at room temperature contained 37.5, 39.2, 37.9, 37.3 and 37.0% oils, respectively, as well as 38.8, 40.5, 37.2, 37.9 and 38.9% protein, respectively, compared to 34.6% in control kernel. Highest % increase in both oil and protein of sunflower kernels was achieved with germination in 2% (NH₄)₂CO₃ reaching 8.9% and 17.0% increase, respectively, after 48 hrs.

Defatting of the sunflower germinated kernels in the 5 examined solutions resulted in sunflower seed meals (Table 4), with increased protein content and improved nitrogen solubility of the meal protein.

This improvement was true for all germinating solutions and for all germinating periods. In accordance with the results of germination of soybean, highest improvement in meal protein and its nitrogen solubility was reached after 48 hrs germination of sunflower kernels in 2% (NH₄)₂CO₃ solution. Protein content reached 64.3% compared to 57.2% for sunflower meal (control) and nitrogen solubility of protein reached up to 85.6% compared to 72.8% for control. Tables 3. and 4. also show that increasing time of germination has a positive effect on all the investigated criteria. While boiling for 30 min. has a negative effect on the nutritional quality of the protein.

I.3. Cottonseed

Table 5. shows the effect of using different solutions and different periods of time for the germination of cottonseed kernels on their oils and protein contents. Table 6. gives the values for the protein content and

nitrogen solubility of the meals resulting from the germinated kernels after being defatted.

1+2

3+4

5+6

Results of Tables 5. and 6. are in accordance with the results of Tables 1., 2., 3. and 4. in that they confirm that both oil and protein content increase as a result of germination of cottonseed kernels in the different solutions. Also that of the germinating time increases the protein and oil contents and nitrogen solubility increases.

Highest oil and protein content of kernels were 35.3% oil for cottonseed germinated in 2% (NH₄)₂CO₃/yeast for 48 hs. Compared to 32.9% oil in control kernels and 39.2% protein for germinated cottonseed in 2% (NH₄)₂CO₃ for 48 hrs. compared to 33.6% protein in control kernels.

Increase in meal protein (Table 6) was 15.18% protein increase over the control meal for sample resulting from germination for 48 hrs. with 2% (NH₄)₂CO₃. This condition also resulted in highest improvement in the nitrogen solubility of the protein reaching 82.2% compared to 73.0% for meal control, achieving 12.6% improvement in nitrogen solubility. Boiling cottonseed kernels resulted in decrease in oil content, increase in protein content and a drastic decrease in nitrogen solubility of protein.

The energy needed during the germination of seeds is supplied first by the carbohydrates which when depleted the seeds start the utilization of their oil reserves to supply the energy. It was reported that carbohydrates from soybean cotyledons are quickly depleted in early phases of germination within three days KeShun (1997). Yoshida *et al.* (1974) found that as soybeans germinated, the lipid content in hulls and cotyledons decreased while the oil in the hypocotyl increased.

Cunningham *et al.* (1978) germinating cottonseed in water for 3 days, and found that the protein and oil content both increased slightly with progressing time of germination. Protein solubility data indicated increase with increasing time of germination. Ultra-centrifuge data revealed no difference in relative concentrations of 2S, 7S or 12S peaks at different times of germination. Amino acid analysis also showed hardly any change in essential amino acids content due to germination.

In general, the results of germination study showed that germination of soybean, sunflower and cottonseed kernels for 48 hrs in the different solutions is a good treatment to improve the nutritional quality of both the germinated kernels and the meal protein prepared therefrom.

This conclusion is in agreement with the recommendations of Bau and Debry (1979), Liener (1978) and Smith and Circle (1958) that soaking and germination of seeds increase their nutritive value. Ibrahim (1988) gave results indicating that soaking soybean meals and flours in the aqueous solutions used in this study resulted in improvement of their nutritional value. Abdel-Moniem (1998) and Riztey and Sistruik (1979) reported good nutritional results by soaking soybean and cow peas in sodium carbonate.

I.4. Relation Between pH of Germination Solutions and Nutritional Quality

Tables 1. to 6. show that 2% (NH₄)₂CO₃ is the choice germinating solutions investigated as it resulted generally in an increase in oil and protein content as well as nitrogen solubility of soybean, sunflower and cottonseed kernels.

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Figure 1. shows the relation between pH of germinating solutions and the % improvement in nitrogen solubility. It is clear that there was an improvement in nitrogen solubility with germinating solutions with different pH values investigated. Least % improvement was achieved with tap water pH 5.6, nitrogen solubility improved slightly by lowering pH of water to 3.6, while the highest improvement in nitrogen solubility was achieved with 2% (NH₄)₂CO₃ (pH 8.4).

I.5. Chemical Composition of Soybean, Sunflower and Cottonseed Meals Resulting after Germination

Since 2% (NH₄)₂CO₃ proved to be the choice solution for germination and the time of germination 48 hrs, the kernels resulting from this treatment were defatted and subjected to complete analysis. Table 7. gives the chemical composition of the resulting soybean, sunflower and cottonseed meals resulting from germinated and non-germinated (control) kernels for comparison. It can be clearly seen that the three meals resulting from germination with 2% (NH₄)₂CO₃ for 48 hrs showed increase in their protein content as well as improvement in the N-solubility and digestibility over the non-germinated meals. Ash, fiber and phytate content of the three germinated meals were reduced in comparison to the control meals.

Also this treatment resulted in a reduction in the urease activity of germinated soybean and a slight reduction in gossypol content of cottonseed meal.

The improvement in nitrogen solubility, digestibility, phytates and urease activity may be due to the activation of endogenous enzymes breaking the protein to peptides, degrading the phytates and inhibiting the urease activity.

Ologhobo and Fetuga, (1984), Chang *et al.* (1977) and Tabekhia and Luh, (1980) reported that germination reduced phytic acid in seeds and legumes.

El-Mahdy, (1985) found that germination of kernels decreased values of phytic acid, trypsin inhibitor and tannins.

Results in Table 7. clearly recommends germination of seeds to improve their nutritional quality. They can be eaten as sprouts or used for preparing other food products from the meal protein.

I.6. Effect of Germination on the Iso-Electric Point of the Proteins

Figures 2, 3 and 4 are the isoelectric precipitation curves of the meal proteins resulting from the germinated and non-germinated soybean, sunflower and cottonseed kernels, respectively. It can be seen that germination of seeds in 2% (NH₄)₂CO₃ for 48 hrs, resulted in a slight shift in the isoelectric points of the meals, but were still in the range of pH 4-5 characteristic for oilseed proteins. Figure 1. indicates that IEP of soybean meal (control) is 4.6 shifted to pH 4.4 on germination. Figure 2. shows that sunflower seed meal protein had an IEP pH 4.2 that was shifted by germination to pH 4.6. Cottonseed protein which has an IEP pH 4.0 shifted to pH 4.6 on germination. Figure 3. also shows that the amount of protein precipitated was reduced by germination in all cases.

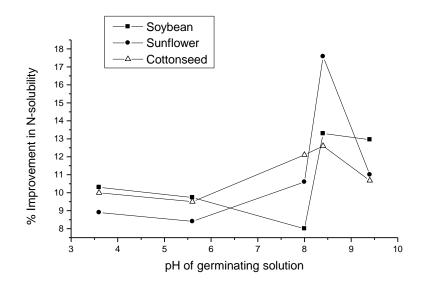


Fig. (1): Relation between pH of germinating solution and % improvement in nitrogen solubility of meal protein after 48 hrs germination at room temperature.

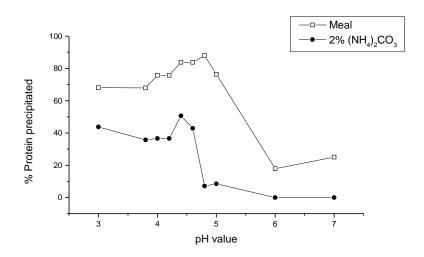


Fig. (2): IEP curves of soybean protein (untreated soybean meal and (NH₄)₂CO₃ germinated soybean meal).

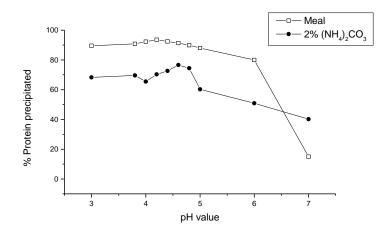


Fig. (3): IEP curves of sunflower protein (untreated sunflower meal and (NH₄)₂CO₃ germinated sunflower meal).

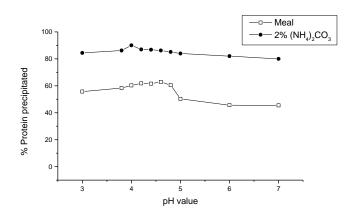


Fig. (4): IEP curves of cottonseed protein (untreated cottonseed meal and (NH₄)₂CO₃ germinated cottonseed meal).

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II. Protein Isolate from Germinated and non-Germinated Seed Meal

Table 8. gives the composition of protein isolates prepared by the solubilization of the proteins then isoelectric precipitation at their elucidated isoelectric points.

Protein isolates were prepared from soybean, sunflower and cottonseed meal protein and the proteins resulting from their germinated kernels. All proteins isolates have protein content above 90%. Protein content of isolates prepared from untreated meals are slightly higher than the ones prepared from germinated meals.

Components	Soyl	bean	Sunfl	lower	Cottonseed			
Components	Isolate I	Isolate II	Isolate I	Isolate II	Isolate I	Isolate II		
Protein	98.3	95.3	98.8	96.0	95.0	93.5		
Ash	0.05	0.0	0.07	0.0	1.59	0.0		
Fiber	0.0	0.0	0.1	0.0	0.0	0.0		
NEF	1.65	2.1	2.03		3.41			

Table (8): Chemical a	alysis of	protein	isolates.
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* Values on moisture free basis.

* Isolate I: Isolate prepared from non germinated meal,.

* Isolate II: Isolate prepared from 2% NH4CO3 germinated defatted meal.

III. Functional Properties of Protein Isolates

Some functional properties of the prepared isolates as well as the untreated meal were investigated. These properties included water absorption capacity, emulsifying capacity, protein dispersibility index and nitrogen solubility index. Table 9. indicate that the isolates prepared from the 2% (NH₄)₂CO₃ germinated seeds showed some improvement in all the functional properties especially in the protein dispersibility index and nitrogen solubility index.

Table (9): Functional properties of protein isolates.

Functional		Soybeaı	Su	unflow	er	Cottonseed			
properties	Meal	P _l I	P _l II	Meal	P _I I	P _l II	Meal	P _i I	P _I II
Water absorption Capacity WAC (%)	300	320	380	260	300	270	160	180	160
Emulsifying Capacity (ml oil / 100 g sample)	20.8	20.9	20.3	26.8	20.8	20.9	20.8	20.6	21.5
Dispersibility Index (DPI)	16.25	18.32	22.62	14.32	14.32	16.28	13.39	15.60	19.89
Nitrogen solubility Index (NSI)	15.48	16.50	19.98	11.54	12.83	15.83	10.33	13.28	18.26

* Values on moisture free basis.

* Meal: Untreated meal

* P_I I: Isolate prepared from non germinated meal,.

* P₁II: Isolate prepared from 2% NH₄CO₃ germinated defatted meal.

CONCLUSION

The art of germination of seeds prior to eating or processing generally improves the nutritional quality. Among the several aqueous solutions investigated for germination $(NH_4)_2CO_3$ solution resulted in highest improvement of the nutritional quality. The protein isolates prepared from the 2% $(NH_4)_2CO_3$ germinated kernels defatted meals also possessed improved protein dispersibility index and nitrogen solubility index.

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دراسة على تأثير الإنبات على بروتينات بعض البذور الزيتية سميرة سعيد محمد، فخرية سيد طه قسم الزيوت والدهون - المركز القومى للبحوث - القاهرة - مصر

كلاً من لب بذور الصويا وعباد الشمس والقطن قد تم إنباتها في محاليل مختلفة لمدة 12، 24، و48 ساعة. والمحاليل التي تم إنباتها فيها هي محلول صوديوم كربونات-صوديوم بيكربونات، 2% كربونات الأمونيوم، 2% كربونات الأمونيوم وخميرة، ماء عادى، وماء حمضى. وقد وجد أن الإنبات في محلول 2% كربونات أمونيوم لمدة 48 ساعة يعطى أعلى نسبة تحسين لكلٍ من نسبة الزيت والبروتين ونسبة إذابة البروتين للب.

بتحليل الكسب الناتج من الإنبات في 2% كربونات الأمونيوم لمدة 48 ساعة للبذور الثلاثة المختبرة وجد أنه قد نتج إنخفاض في نسبة الفيتات، وإنخفاض طفيف في نشاط اليوريز بالنسبة لكسب الصويا، وكذلك في نسبة الجوسيبول بالنسبة لبذرة القطن، كما أظهرت التحاليل تحسن أيضاً في كل من نسبة الإذابة ونسبة الهضم للبروتين.

وَبناءً على النتائج السابقة تم تحضير مستخلصات بروتين من أنواع الكسب الثلاثة المعاملة بالإنبات. وبتحليل المستخلصات وجد تحسن في بعض خواص البروتين الوظيفية مثل معامل الإذابة ومعامل الإنتشار.

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Solutions	۳Ц	12	hrs.	24	hrs.	48	hrs.	Boiling 30 min.		
3010110115	рН	% Oil	% Protein	% Oil	% Protein	% Oil	% Protein	% Oil	% Protein	
Na ₂ CO ₃ /NaHCO ₃ buffer	9.4	21.85	38.21	22.90	40.82	23.52	42.31	14.62	45.2	
2% (NH ₄) ₂ CO ₃	8.4	22.29	39.69	23.68	42.13	25.03	44.03	19.83	48.6	
2% (NH ₄) ₂ CO ₃ /yeast	8.0	21.67	38.21	22.86	4013	23.98	42.56	20.93	46.9	
Tap water	5.6	21.82	39.0	23.01	39.9	24.36	40.26	19.52	45.3	
Acidified water	3.0	23.38	40.26	23.58	40.58	24.93	41.81	20.36	42.3	

Table (1): Effect of germination of soybean kernels in different solutions.

* Control (non germinated soybean kernels) contain 20.82% oil and 35.9% protein.

* All values are given on moisture-free basis.

Table (2): Effect of germination of soybean kernels in different solutions on the protein content of the defatted meal and its nitrogen solubility.

	0 /											
Solutions	рН	12 h	rs.	24	hrs.	48 hr	S.	Boiling 30 min.				
		Protein	N-Sol	Protein	N-Sol	Protein	N-Sol	Protein	N-Sol			
Na ₂ CO ₃ /NaHCO ₃ Buffer	9.4	46.70	89.26	55.32	92.63	58.69	94.34	60.89	13.57			
2% (NH ₄) ₂ CO ₃	8.4	52.82	88.53	58.78	89.91	60.21	94.62	61.30	19.69			
2% (NH ₄) ₂ CO ₃ /yeast	8.0	49.73	79.32	58.61	86.62	58.34	90.23	60.0	19.92			
Tap water	5.6	50.53	82.69	52.88	85.29	56.32	91.65	59.31	32.36			
Acidified water	3.6	52.69	79.32	52.36	82.63	55.0	92.1	60.28	39.02			

* Control (hexane defatted non germinated soybean kernels) contain 43.32% protein and 83.52% nitrogen solubility.

* All values are given on moisture-free basis.

Solutions	Hq	12	hrs.	24	hrs.	48	hrs.	Boiling 30 min.	
Solutions	рп	% Oil	% Protein	% Oil	% Protein	% Oil	% Protein	% Oil	% Protein
Na ₂ CO ₃ /NaHCO ₃ buffer	9.4	35.6	35.2	36.6	36.6	37.5	38.8	28.9	41.3
2% (NH ₄) ₂ CO ₃	8.4	36.9	35.9	38.2	37.9	39.2	40.5	28.6	42.0
2% (NH ₄) ₂ CO ₃ /yeast	8.0	36.8	34.9	38.0	35.8	37.9	37.2	29.9	40.5
Tap water	5.6	35.09	35.2	36.5	36.9	37.3	37.9	25.7	41.3
Acidified water	3.6	36.5	34.6	36.9	37.0	37.0	38.9	24.3	43.8

Table (3): Effect of germination of sunflower kernels in different solutions.

* Control (non germinated sunflower kernels) contain 36.0% oil and 34.6% protein.

* All values are given on moisture-free basis.

Table (4): Effect of germination of sunflower kernels in different solutions on the

protein content of the defatted meal and its nitrogen solubility.

Solutions	PH	12 hrs.		24	4 hrs.	48 I	nrs.	Boiling 30 min.				
	гп	Protein	N-Sol	Protein	N-Sol	Protein	N-Sol	Protein	N-Sol			
Na ₂ CO ₃ /NaHCO ₃ Buffer	9.4	57.2	71.9	58.5	74.3	63.5	80.8	62.6	18.2			
2% (NH ₄) ₂ CO ₃	8.4	58.0	73.6	58.9	78.9	64.3	85.6	66.7	14.8			
2% (NH ₄) ₂ CO ₃ /yeast	8.0	57.9	72.8	57.2	75.6	63.8	80.5	62.9	16.9			
Tap water	5.6	58.3	71.5	58.8	72.9	61.6	78.9	68.5	22.6			
Acidified water	3.6	57.0	70.6	59.3	73.3	62.8	79.3	66.3	34.3			

* Control (hexane defatted non germinated sunflower kernels) contain 57.2% protein and 72.8% nitrogen solubility.

* All values are given on moisture-free basis.

Solutions	рН	12	2 hrs.	24	4 hrs.	48	3 hrs.	Boiling 30 min.		
Solutions		% Oil	% Protein	% Oil	% Protein	% Oil	% Protein	% Oil	% Protein	
Na ₂ CO ₃ /NaHCO ₃ buffer	9.4	31.9	33.8	32.8	34.2	33.6	36.8	28.6	42.3	
2% (NH ₄) ₂ CO ₃	8.4	32.5	34.0	33.5	35.4	34.8	39.2	293	44.8	
2% (NH ₄) ₂ CO ₃ /yeast	8.0	32.6	33.5	33.3	34.8	35.3	36.7	30.1	43.6	
Tap water	5.6	31.8	33.6	32.9	35.6	33.9	37.9	26.0	42.1	
Acidified water	3.6	30.9	33.8	32.8	34.6	32.9	37.0	28.3	40.5	

Table (5): Effect of germination of cottonseed kernels in different solutions.

* Control (non germinated cottonseed kernels) contain 32.9% oil and 33.6% protein.

* All values are given on moisture-free basis.

Table (6): Effect of germination of cottonseed kernels in different solutions on the defatted meal and its nitrogen solubility.

Solutions	pН	12 hr	S.	24	nrs.	48	hrs.	Boiling 30 min.		
Solutions		Protein	N-Sol	Protein	N-Sol	Protein	N-Sol	Protein	N-Sol	
Na ₂ CO ₃ /NaHCO ₃ buffer	9.4	51.3	72.8	53.9	75.8	56.8	80.8	65.9	26.0	
2% (NH ₄) ₂ CO ₃	8.4	52.1	74.4	54.8	78.9	58.4	82.2	68.4	29.5	
2% (NH ₄) ₂ CO ₃ /yeast	8.0	50.8	73.0	52.6	77.6	55.5	81.9	64.3	20.8	
Tap water	5.6	50.5	74.8	52.3	75.0	56.3	79.9	67.6	19.9	
Acidified water	3.6	51.0	72.9	53.5	76.7	55.9	80.3	64.6	23.6	

* Control (hexane defatted non germinated cottonseed kernels) contain 50.7% protein and 73.0% nitrogen solubility.

* All values are given on moisture-free basis.

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Sample	Protein	Oil	Ash	Fiber	NEF	Phytate			Free Gossypol	N-Sol	Digestibility
Soybean meal (control)	43.22	0.8	7.2	4.8	43.88	5.2	1.8	-	-	83.52	84.0
Soybean germinated in2% (NH ₄) ₂ CO ₃ (48 hrs)	60.21	0.6	6.8	4.5	27.89	4.5	0.2	-	-	94.62	92.4
Sunflower meal (control)	57.2	0.5	8.8	9.2	24.3	10.6	-	-	-	72.8	75.6
Sunflower germinated in 2% (NH ₄) ₂ CO ₃ (48 hrs)	64.3	0.2	7.3	8.9	19.3	8.5	-	-	-	85.6	89.0
Cottonseed meal (control)	50.7	0.9	8.0	1.0	39.4	12.9	-	1.24	0.66	73.0	79.8
Cottonseed germinated in 2% (NH ₄) ₂ CO ₃ (48 hrs)	58.7	0.5	7.6	0.9	32.6	10.3	-	1.02	0.58	82.2	86.8

Table (7): Analysis of germinated and non-germinated (control) defatted soybean, sunflower and cottonseed kernels (meals).

All values are calculated on moisture free basis.