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EVALUATION OF TEMPE PRODUCED BY FERMENTATION OF SOYBEAN USING TWO SPECIES OF *Rhizopus*

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ABSTRACT

Soybean was dehulled, soaked, autoclaved and fermented by two species of *Rhizopus* namely, *R. oligosporus* and *R. arrhizus* ,to produce tempe. Chemical, nutritional, microbiological and organoleptical evaluation of soybean tempe products were investigated. The best products were obtained after 26 and 30 hrs at 31- 32°C by *R. oligosporus* and *R. arrhizus*, respectively. It was noticed that, soaking and autoclaving processes decreased significantly ($P \leq 0.05$) contents of ether extract, ash, total carbohydrate, total solids, soluble solids, insoluble solids, TP, minerals, number of yeasts and lactic acid bacteria. On the contrary, moisture, crude protein and NPN contents were increased significantly ($P \leq 0.05$) in soaked-autoclaved soybean grits, while crude fiber content and total bacterial spores count did not change in the same grits. However, fermentation process either by R. *oligosporus* or R. *arrhizus* elevated significantly ($P \leq 0.05$) contents of moisture, crude protein, NPN, crude fiber, soluble solids, counts of lactic acid bacteria and total bacterial spores. Conversely, ether extract, total carbohydrate, total solids, insoluble solids and TP contents were diminished significantly ($P \leq 0.05$) in tempe products. Fe and K contents increased slightly after fermentation process, but Ca, Cu and Na contents decreased slightly after this process. Contents of Mg, Mn, Zn and yeasts count did not alter appreciably after fermentation. Slight or no changes in the essential amino acids content were occurred as a result of tempe production. The most of essential amino acids were close to the amounts needed for protein balance according to the FAO / WHO (1973) reference protein. Sulfur-containing amino acids gave the lowest score, although they were increased by fermentation. Chemical scores of essential amino acids for tempe produced by *R. arrhizus* were higher than those for tempe fermented by *R.oligosporus* with the exception of leucine. Data also indicated that tempe production process improved significantly ($P \leq 0.05$) the protein digestibility, C-PER and biological value. Moreover, trypsin inhibitor and phytic acid contents were diminished significantly ($P \leq 0.05$) as a result of tempe production. Finally, sensory evaluation established that all fried tempe products were accepted.

Keywords: Soybean, *Rhizopus spp.*, tempe, chemical and nutritional evaluation, microbiological aspects, sensory properties.

INTRODUCTION

There is no doubt that the demand of animal protein products will be increased as a result of population growth. Therefore, current work is necessary to evaluate protein-rich meat substitutes. Legumes are an important source of proteins in the Egyptian diet and in many developing countries. Higher meat prices during recent years and the need for protein-rich foods have led people in most less developed countries to shift their consumption to certain legumes (Asker 1986).

Soybean plays a major role in agriculture, commerce, industry and nutrition and for centuries have been an important source of dietary protein for millions of people of the Orient (Sutardi and Buckle 1985).

Fermented foods may be defined as those foods that have been subjected to the action of microorganisms or enzymes so that desirable biochemical changes cause significant modification to the food. By fermentation, the food may be made, more nutritious, more digestible and have better flavor (Ashenafi and Busse 1991a).

Tempe is a popular fermented food in Indonesia. It is produced by fungal fermentation of soybean or other legumes (Ashenafi and Busse1991b). Tempe can be served as an excellent substitute for animal protein products and it therefore, holds promise to combat malnutrition in countries where proteins and calories are in short supply (Mital and Garg1990). Nout *et al.* (1987) reported that tempe technology meets an increasing interest in the developing countries as a small-scale method to derive nutritious food from locally available legumes and cereals.

The present study was designed to prepare tempe from soybean using two species of *Rhizopus, R. oligosporus* and *R. arrhizus*. Chemical, nutritional, microbiological and sensory evaluation of soybean tempe products were investigated. This work was extended to prepare fried tempe products for consumption through four recipes (with flavoring mixture, with blanched potatoes, with flavoring mixture of taamia and with flavoring mixture of kofta), which were also evaluated organolepically.

MATERIALS AND METHODS

Materials

Soybean (Glycin maxl-merr) Giza 82 variety was obtained from Agriculture Research Center, Giza, Egypt (season 2002). The beans were cleaned and ground into grits using a household blender (National, Japan). Hulls were separated out and bean grits were kept in polyethylene bags at 5 ± 1°C until used.

Two species of Rhizopus: *Rhizopus oligosporus NRRL 2710* and *Rhizopus arrhizus NRRL 1526*, were supplied by Northern Regional Research Laboratory, Peoria, Illinois, USA. The two strains were maintained on slants of potato – dextrose-agar (PDA) at 5 ± 1°C and subcultured at intervals of 2 months.

Methods

1.Technological methods

1.1.Tempe production

Figure (1) shows flow sheet of tempe production (Berghofer and Werzer 1986) from soybean in the laboratory. The inoculum was prepared by shaking each slant with 2ml sterile distilled water for 1 minute and 0.4 ml of such suspension was used to inoculate each flask contained 40 gems dry bean grits (Moeljopawiro *et al.*, 1987). The fermentation process was stopped when bean grits were tightly bound together into a compact cake by white mycelia and could be removed easily from the Petri dish (El-Bagoury 1996).

Whole, clean beans

Dry dehulling

Soaking
17-18hrs at room temperature in
acidified soak water (1%of 85%lactic acid was added)
and the ratio of beans to water was 1: 3 (w/v)

Draining

Autoclaving
121°C for 20 min.

Cooling to 37°C

Inoculation with spores suspension
of *Rhizopus spp.*

Mixing and packing
In Petri dishes

Incubation at 31-32°C

Fresh tempe

Figure (1): Flow sheet of tempe production from soybean in the laboratory

1.2. Preparation the tempe for the consumption

Tempe product was prepared for consumption by four methods. In the first method, raw tempe was mixed with flavoring mixture containing cumin, hot pepper, coriander and sodium chloride as reported by El-Bagoury (1996). In the second method, raw tempe was mixed with blanched potatoes as outlined by Yueh *et al.* (1979). In the third recipe, taamia was prepared by replacing broad bean with 10, 30 and 50% of raw tempe. While in the fourth recipe, kofta (meat) was manufactured by replacing minced meat with 10, 30 and 50% of raw tempe. In the four methods, the mixed pastes were divided and shaped into small discs, 4 cm in diameter and 1 cm thick, and then deep-fried in sunflower oil. The controls in the first and the second methods were unfermented soybean grits, while in the third and the fourth recipes were broad bean and minced meat, respectively.

Analytical methods

Sample of soybean grits was ground in a laboratory mill to pass through 40 mesh sieve, packed in polyethylene bags and kept at 5±1°C. While, samples of fermented and unfermented soybean (soaked and autoclaved only) were homogenized and divided into two portions. The first one was taken directly for moisture and antitryptic activity determinations, while the second part was dried at 55 – 60 °C for 48 hrs, ground, packed in polyethylene bags and kept at 5±1°C until analysis.

Proximate chemical composition including moisture, crude protein (TN X 6.25), ether extract, crude fiber and ash contents was determined *according to the methods given in* A.O.A.C. (1990). Carbohydrate content was calculated by difference.

Water-soluble solids were measured according to the procedure of Agosin *et al.* (1989).

The **Na and k** contents were determined using Flamephotometer, while the **Fe, Mn, Cu, Zn and Ca** contents were estimated using a Perkin- Elmer atomic absorption spectrophotometer as reported in A.O.A.C. (1990).

Non-protein nitrogen (NPN) was estimated as described by Paredes-Lopez and Harry (1989). Protein nitrogen (PN) was calculated by difference between total nitrogen (TN) and non-protein nitrogen. True protein was obtained by multiplying PN with conversion factor of 6.25.

Amino acids content *was determined using Beckman amino acid* analyzer (Model 119 CL) as described by Moore and Stein (1963). Tryptophan was estimated colorimetrically in the alkaline hydrolyzate following the method of Miller (1967). Chemical scores of essential amino acids were estimated according to the method of Pellet and Young (1980).

In-vitro protein digestibility was performed according to the method mentioned by Salgo *et al.* (1984). The true and apparent digestibilities were calculated form the following equations:

% True digestibility = 425.78 – 47.64 pH_{10}

% Apparent digestibility = 392.51 – 44.84 pH_{10}

Where: pH_{10} = pH value of the sample suspension after 10 minutes digestion with enzymes

Computed protein efficiency ratio (C–PER) was accounted according to the computation procedure of Hsu *et al.* (1978).

The following equation was used to calculate the biological value as given by Mitchell and Block (1946):

Biological value = 49.9 + 10.53 C-PER

Trypsin inhibitors were extracted as described by Wang *et al.* (1975). Trypsin inhibitor activity was estimated by the caseinolytic procedures described by Kakade *et al.* (1969). Trypsin inhibitor unit (TIU) was expressed in terms of the Tryptic units inhibited per gram dry sample.

Phytate phosphorus was extracted and determined following the procedure of Mohamed *et al.* (1986). Phytic acid content was calculated by multiplying mg phytate phosphorus with a factor of 3.553.

Microbiological methods

Lactic acid bacteria, yeasts count and total aerobic bacterial spores count *of soybean tempe samples were counted* according to the method of Ashenafi and Busse (1991b).

Organoleptic evaluation

Fresh tempe samples were evaluated organoleptically as reported by Nout *et al.* (1987). The following criteria were used: color = white, surface = covered entirely by mold mycelium, physical characteristic = compact, texture = elastic and rubbery, and flavor = acceptable.

Fried tempe samples were evaluated organoleptically according to the method of Gorczyca and Zabik (1979). The panel was composed of 10 judges, using a fully structured 9 points rating scale, to evaluate color, texture, odor, taste, appearance and overall acceptability.

tatistical analysis

Data were statistically analyzed according to procedures outlined by Gomez and Gomez (1984)

RESULTS AND DICUSSION

Organoleptic properties of fresh tempe:

Table (1) shows the organoleptic properties of fresh soybean tempe products. It is clear that two species of *Rhizopus* , *R. oligosporus* and *R. arrhizus* , grew very well on soybean grits. They gave highly compact cakes with white cotton- like color after 26 and 30 hrs at 31- 32°C, respectively.

Table (1): Organoleptic properties of fresh tempe* produced by two species of Rhizopus

Properties	Tempe products by	
	R.oligosporus	*R. arrhizus*
Color	White cotton like	White cotton like
Surface covered by mold mycelium	Entire	Dense
Physical characteristic	Highly compact	Highly compact
Texture	Elastic and rubbery	Elastic and rubbery
Flavor	A yeast like aroma and ester flavored	
Production of spores	Non	Non

*The best products were obtained after 26 and 30 hrs of fermentation at 31 – 32 °C by *R.oligosporus* and *R. arrhizus*, respectively.

Proximate chemical composition of tempe :

The obtained results In Table (2) indicated that moisture and crude protein contents of dehulled soybean increased significantly ($P \le .05$) after soaking and autoclaving processes. On the contrary, ether extract, ash and total carbohydrate contents decreased significantly ($P \le .05$). While, crude fiber content of dehulled soybean did not change significantly ($P \le 0.05$) after these processes. These changes may be due to leaching out some components during soaking. After fermentation, either by *R. oligosporus* or by *R. arrhizus*, moisture, crude protein and crude fiber contents of tempe products were elevated significantly ($P \le 0.05$). In contrast, ether extract and total carbohydrate contents were decremented significantly ($P \le 0.05$).

Table (2) showed that ash content of tempe fermented by *R. oligosporus* was not significantly affected, but ash contents of tempe produced by *R. arrhizus* were increased significantly ($P \le 0.05$). These changes may be due to fungi growth where they have consumed carbohydrate and ether extract as a source of energy and the development of a fiber –rich fungous mycelium (Shurtleff and Aoyagi 1979).

The results obtained in Table (2) also illustrate the influence of Rhizopus species on chemical constituents of tempe products. It could be noticed that *the moisture, crude protein, ether extract and ash* contents of tempe produced by *R. arrhizus* (64.02, 44.13, 23.33 and 4.85 %, respectively) were *higher significantly* ($P \leq 0.05$) than those fermented by *R. oligosporus* (61.01, 42.61, 23.25 and 4.76 %, respectively). In contrary, crude fiber and total *carbohydrate contents of tempe* produced by *R. arrhizus* (5.84 and 21.83%, respectively) were lower significantly ($P \leq 0.05$) than those fermented by *R. oligosporus* . (5.92 and 23.46%, respectively). These results are in agreement with those findings of Shurtleff and Aoyagi (1979); Agosin *et al.* (1989); Paredes-Lopez *et al.*(1990); Ashenafi and Busse (1991a); Njoku *et al.* (1991); De- Reu *et al.* (1995) and El-Bagoury (1996).

Table (2): Proximate chemical composition of dehulled soybean and tempe products (on dry weight basis)

Constituents (%)	Dehulled soybean grits	Soaked and autoclaved (before fermentation)	Tempe produced by *R. oligosporus*	Tempe produced by *R. arrhizus*
Moisture	8.70 d	57.10 c	61.01 b	64.02 a
Crude protein	37.95 d	38.53 c	42.61 b	44.13 a
Ether extract	23.56 a	23.38 b	23.25 d	23.33 c
Ash	4.73 b	4.49 c	4.76 b	4.85 a
Crude fiber	4.77 c	4.73 c	5.92 a	5.84 b
Total carbohydrate	29.00a	28.87 b	23.46 c	21.83 d

In a row, means followed by the same letter are not significantly different at P ? 0.05.

Solids contents of tempe products:

Table (3) shows an alteration in total solids, soluble solids and insoluble solids contents of tempe products. It has been observed that total solids, soluble solids and insoluble contents of dehulled soybean decreased (by 53.01, 60.27 and 51.52%, respectively) after soaking and autoclaving processes. This reduction may be due to draining the soak water after soaking process as stated by El-Bagoury (1996).

Table (3): Solids contents of dehulled soybean and tempe products

Sample	Total solids (%)	Soluble solids(%)	Insoluble solids (%)
Dehulled soybean	91.30	15.53 (17.01%)	75.77(82.99 %)
Soaked and autoclaved (before fermentation)	42.90	6.17 (14.39%)	36.73 (85.61%)
Tempe produced by *R. oligosporus*	38.99	13.77 (35.31%)	25.22 (64.69%)
Tempe produced by *R. arrhizus*	35.99	14.22 (39.50%)	21.73 (60.50%)

However, fermentation of soaked and autoclaved soybean either by *R.oligosporus* or by *R. arrhizus* caused a marked declination in total solids and insoluble solids. These decreases were higher in tempe produced by *R.*

arrhizus (16.11 and 40.73%, respectively) than those in tempe fermented by *R. oligosporus* (9.11 and 31.34%, respectively). It could be attributed to fungi metabolic processes during tempe production (El- Bagoury1996). In contrary, soluble solids content of soaked autoclaved soybean increased after fermentation by two species of *Rhizopus*. These increases were 55.19% and 56.61% in tempe produced by *R. oligosporus* and *R. arrhizus*, respectively. The increases in soluble solids of tempe reflected the hydrolyzing capacity of fungi. The obtained results are coincided with the results of Agosin *et al.* (1989), Ashenafi and Busse (1991a) and El-Bagoury (1996).

Minerals content

The changes in minerals content of dehulled soybean and tempe products are illustrated in Table (4). It was noticed that soaking and autoclaving processes lowered the minerals content of dehulled soybeans. The reduction percent of Ca, Cu, Fe, Mg, Mn, K, Na, and Zn contents in soaked-autoclaved soybean reached to 26.15, 16.28, 19.47, 6.25, 10.71, 11.48, 19.05, and 39.29%, respectively. Theses results are in agreement with those published for other legumes by Youssef (1978) and Khalil (1996). On the other side, fermentation of soaked-autoclaved soybean either by *R. oligosporus* or by *R. arrhizus* induced a slight increase in Fe and k contents.

Table (4): Mineral content of dehulled soybean and tempe products (mg/ 100gm dry weight basis)

Minerals	Dehulled soybean	Soaked and autoclaved	Fermented by	
			R.oligosporus	*R.arrhizus*
Ca	218	161	130	135
Cu	2.15	1.80	1.70	1.71
Fe	11.30	9.10	10.70	11.00
Mg	256	240	245	240
Mn	2.80	2.50	2.50	2.60
K	1864	1650	1733	1753
Na	2.10	1.70	1.55	1.60
Zn	5.60	3.40	3.40	3.40
Total	2361.95	2069.50	2127.85	2148.31

The increment of Fe and K in tempe produced by *R. arrhizus* (about 17% and 6%, respectively) was more than that occurred by *R. oligosporus* (about 15% and 5%, respectively). In contrary, Ca, Cu and Na contents decreased slightly after fermentation process either by *R. oligosporus* or by *R. arrhizus*. The decrement of Ca, Cu and Na in tempe produced by *R. oligosporus* (about 19%, 5.5% and 9%, respectively) was higher than that in tempe fermented by *R.arrhizus* (about 16 %, 5% and 6 %, respectively). However, fermentation process either by *R. oligosporus* or by *R. arrhizus* caused no apparent changes in the concentrations of Mg, Mn, and Zn. Gandjar *et al.* (1977) reported that Fe in the solid refuse of soybean was slightly increased after fermentation, while Ruiz-Teron and Owens (1996) observed that minerals content of soybean did not alter appreciably after fermentation.

Nitrogenous constituents

The results in Table (5) illustrate the changes in nitrogenous constituents of dehulled soybean after soaking and autoclaving processes as well as fermentation process. It was evident that, soaking and autoclaving processes as well as fermentation process increased significantly ($P \leq 0.05$) contents of total nitrogen (TN) and non protein nitrogen (NPN). Conversely, they decreased significantly ($P \leq 0.05$) contents of protein nitrogen (PN) and hence true protein (TP). The nitrogenous constituents content of tempe produced by *R. arrhizus* was higher than those of fermented one by *R. oligosporus*. *The increases of TN, NPN and PN in tempe produced by R. arrhizus* as compared with tempe fermented by *R. oligosporus* were 8.34, 6.81 and 9.06%, respectively.

Table (5): Nitrogenous constituents of dehulled soybean and tempe products (g/100g dry weight)

Sample	Total nitrogen (TN)	Total non nitrogen protein (NPN)	Protein nitrogen (PN)*	True protein (TP)**
Dehulled (raw)	6.07 d	0.65 d	5.42 a	33.89 a
Soaked and autoclaved	6.16 c	0.77 c	5.39 a	33.69 b
Fermented by *R. Oligosporus*	6.81 b	2.19 b	4.62 c	28.88 d
Fermented by *R. arrhizus*.	7.43 a	2.35 a	5.08 b	31.75 c

In a column, means followed by the same letter are not significantly different at P ? 0.05.
*** PN = TN-NPN ** TP = PN x 6.25**

The increase in NPN content and the decrease in PN content of tempe produced either by *R. oligosporus* or *R. arrhizus* are due to hydrolysis of soybean proteins by proteolytic fungi enzymes during fermentation (Agosin *et al.* 1989). *These results are in agreement with those findings of Agosin et al.* (1989) and El-Bagoury (1996).

4.6. Amino acid composition

Table (6) shows amino acids content of dehulled soybean and tempe products as well as casein as a reference protein. The major essential amino acids in dehulled soybean were leucine and lysine, but glutamic and aspartic were the predominant among the non essential amino acids. Moreover, most of essential amino acids were close to the amounts needed for protein balance according to the FAO/ WHO (1973) reference protein. The lowest limiting amino acid was cystine (0.97g /100g protein). On the other hand, it is clear that little or no change in the essential amino acids content occurred after fermentation process either by *R. oligosporus* or *R. arrhizus*. The total essential amino acids content of tempe produced by *R. arrhizus* (37.49 g/ 100g protein) was higher than that of tempe fermented by *R.oligosporus* (36.87g /100g protein). The ratio of total essential amino acids to total amino acids (E/T) was not changed for tempe produced by *R. arrhizus* (0.39), while it was increased slightly to 0.40 for tempe fermented by *R. oligosporus*. These findings are in harmony with those of Nout and Rombouts (1990) and El Bagoury (1996).

4.7. Chemical scores of essential amino acids

The results obtained in Table (7), illustrate the influence of fermentation process either by *R.oligosporus* or *R. arrhizus* on chemical scores of essential amino acids of dehulled soybean. It was observed that dehulled soy bean and tempe products were lower in chemical scores of the most essential amino acids than needed for balance comparing with casein as a reference protein with the exception of threonine in all samples and tryptophan in tempe produced by *R. arrhizus*. On the other hand, chemical scores of essential amino acids were affected by tempe production either by *R.oligosporus* or *R. arrhizus*. The chemical scores of lysine, methionine, cystine, threonine and tryptophan were increased, while those of isoleucine, leucine, valine, phenylalanine and tyrosine were decreased by tempe production.

Table (6): Amino acid composition of dehulled soybean and tempe products as well as casein as reference protein (g/100g protein)

Amino acid	Dehulled soybean	Fermented by		Casein	FAO/WHO Pattern (g / 100g protein)
		R.oligosporus	*R.arrhizus*		
Essential amino acids (E)					
Lysine	5.11	5.15	5.25	7.50	5.50
Methionine	1.36	1.49	1.52	2.96	3.50
Cystine	0.97	0.98	1.05		
Threonine	3.98	4.19	4.24	3.43	4.00
Isoleucine	4.88	4.68	4.71	5.01	4.00
Leucine	7.15	7.10	7.08	9.20	7.00
Valine	4 76	4.66	4.69	5.42	5.00
Phenylalanine	3 89	3.95	3.75	9 81	6.00
Tyrosine	3.75	3.47	3.77		
Tryptophan	1 17	1.20	1.23	1.21	1.00
Total (E)	37.02	36.87	37.49	44.54	36.00
Non-essential amino acids (NE)					
Alanine	4.40	4.31	4.37	2.65	
Arginine	7.09	6.69	6.80	4 22	
Aspartic	12 11	11.65	11.74	5.97	
Glutamic	17.12	15.70	15.89	17.53	
Glycine	4.31	3.98	4 14	1.78	
Histidine	2.45	2.36	3.38	2.63	
Proline	5.05	4.95	4.99	5.92	
Serine	5 35	5.25	5.13	5.59	
Total (NE)	57.88	54.89	55.44	46.29	
Total amino acids (T)	94.90	91.72	92.93	90.83	
E/T	0.39	0.40	0.39	0.49	

The results in the same Table showed that the sulfur-containing amino acids gave the lowest score, although they were increased by fermentation process. These results clarify that strains of *Rhizopus* may consumed a part of the essential amino acids (El-Bagoury 1996). However, the chemical scores of essential amino acids of tempe produced by *R. arrhizus* were higher than those of tempe fermented by *R. oligosporus* with the exception of chemical score of leucine.

Table (10): Microbial contents (CFU/g)* of dehulled soybean and tempe products

Sample	Yeasts	Lactic acid bacteria	Total bacterial spores
Deulled soybean (raw)	? 9	1.2×10^3	? 8
Soaked -autoclaved	? 7	< 10	? 8
Tempe produced by *R.oligosporus*	? 7	3×10^4	? 9
Tempe produced by *R.arrhizus*	? 7	3×10^4	? 9

* CFU/g = Colony forming unit per one gram.

Table (11): Sensory evaluation of fried soybean tempe products

Sample	Properties (score out of 9)					
	Color	Texture	Odor	Taste	Appearance	Overall acceptability
1- with flavoring mixture						
Control*	6.7 a	6.5 a	6.7 a	6.6 a	6.6 a	6.6 a
Fermented by *R.oligosporus*	6.3 a	6 0 a	6.0 a	6.3 a	6.5 a	6.2 a
Fermented by *R arrhizus*	6.4 a	6.1 a	6.2 a	6.4 a	6.6 a	6.3 a
2- with blanched potatoes						
Control*	6 5 a	6 6 a	5 1 b	6.9 a	6.7 a	6.4 a
Fermented by *R oligosporus*	6.2 a	6 4 a	6.4 a	6.6 a	6.4 a	6.4 a
Fermented by *R.arrhizus*	6 1 a	6.4 a	6.3 a	6.4 a	6.5 a	6.3 a
3- with flavoring mixture of taamia						
Control**	7.7a	7.5a	7.6a	7.6a	7.8a	7.6a
Tempe produced by *R.oligosporus*						
25% of tempe instead of broad beans	7.2 a	7.1 a	7.2 a	7.3 a	7 4 a	7.2 a
50% of tempe instead of broad beans	6.7 b	6.6 b	6.7 b	6.5 b	6.8 b	6.6 b
Tempe produced by *R.arrhizus*						
25% of tempe instead of broad beans	7.3 a	7.0 a	7 3 a	7.2 a	7 4 a	7.2 a
50% of tempe instead of broad beans	6.6 b	6 5 b	6.8 b	6 7 b	6.9 b	6.7 b
4- with flavoring mixture of kofta						
Control***	7 9 a	8.2 a	8.0 a	7.9 a	8.1 a	8.0 a
Tempe produced by *R.oligosporus*						
25% of tempe instead of broad beans	7 3 a	7.5 a	7.5 a	7.3 a	7.5 a	7.4 a
50% of tempe instead of broad beans	6.4 b	6.2 b	6.0 b	6.0 b	6.2 b	6.2 b
Tempe produced by *R.arrhizus*						
25% of tempe instead of broad beans	7 2 a	7 4 a	7.2 a	7.2 a	7.4 a	7.3 a
50% of tempe instead of broad beans	6.3 b	6 1 b	6.1 b	6 0 b	6.1 b	6.1 b

In a column under each recipe, means followed by the same letter are not significantly different at $P \leq 0.05$

.* Control was unfermented soybean grits

** Control was taamia with broad bean (without tempe)

*** Control was kofta with minced meat (without tempe)

REFERENCES

Agosin, E.; D.Diaz; R. Aravenia and E.Yanez (1989). Chemical and nutritional characterization of lupine tempeh. J. Food Sci., 54:102-104,107.

A.O.A.C. (1990). Official Methods of Analysis. Association of Official chemists, 15th Ed., Washington, D.C., USA.

Ashenafi, M. and M. Busse (1991a). Production of tempeh from various indigenous Ethiopian beans. World J. Microb. Biotechnol., 7: 72-79.

Ashenafi, M. and M. Busse (1991b). Development of microorganisms during cold storage of pea and chickpea tempeh and effect of *Lactobacillus plantarum* on storage microflora. J. Sci. Food Agric., 56: 71-78.

Asker, A. (1986). Faba beans (*Vicia faba* L.) and their role in the human diet. Food Nut. Bull., 8(3): 15-24.

Berghofer, E. and A. Werzer (1986).Herstellung von tempeh mit einheimischhen bohnen. Chem. Mikrob. Technol. Lebensm., 10: 54-62.

De-Reu, J.C.; R. M. T. Wold; J. De-Groot; M. J. Nout; F.M. Rombouts and H. Gruppen (1995). Protein hydrolysis during soybean tempe fermentation with *Rhizopus oligosporus*. J. Agric. Food. Chem., 43: 2235-2241.

Djien, K.S. and C. W. Hesseltine (1979). Tempe and related foods. In: Economic Microbiology. Vol.4, Microbial biomass, A.H. Rose (Ed.), Academic press, New York, pp.115-140.

El-Bagoury, A.A. (1996). Chemical and technological studies on some fermented foods. Ph.D. Thesis, Fac. Agric., Kafr El-Sheikh, Tanta Univ., Egypt.

FAO/WHO (1973). Energy and protein requirements. World heath Org. Technol. Rept. Series No. 522, Geneva. Food and Agric. Org. Rept. Series 53, Rome.

Gandjar, I.; D.S. Slamet; I. Ekasari; D. Kartouswondo and M. Rasyidi: (1977). Tempe from the solid waste of Hunkwe factory, Symposium on Indigenous fermented food, Bangkok, Thailand. In: Handbook of Indigenous fermented foods, K.H. Stein Kraus; R.E.; Cullen; C.S. Pederson; L.F. Nellis and B.K.Gavitt (Eds.), Marcel Decker, Inc., New York, (1983).

Gomez, K.A. and A. A. Gomez (1984). Statistical procedures for agricultural research. 2nd Ed., John Willy and Sons, New York, USA, pp.180.

Gorczyca, G.C. and M. Zabik (1979). High fiber sugar snap cookies containing cellulose and coated cellulose products. Cereal Chem., 56: 537-541.

Hsu, H.W.; N.E. Sutton; M.O. Banyo; L.D. Satterlee and J.G.Kendrick (1978). The C-PER and T-PER assays for protein quality. Food Technol., 69-73.

Kakade, M.L.; N.R. Simons and L.E. Liener (1969). An evaluation of natural versus synthetic substrates for measuring the antitryptic activity of soybean samples. Cereal Chem., 46: 518-526.

Kaul, M. and M.Bajwa (1987). Effect of heat and natural fermentation on trypsin inhibitor and haemagglutinin of black gram (*Phaseolus mungo*) J. Nut. Diet. ,24: 40-44.

Khalil, M.M. (1996). Chemical and biological evaluation on mung beans compared with faba beans affected by soaking, germination, autoclaving and cooking. J. Agric. Sci. Mansoura Univ., 21(12): 4451-4461.

Miller, E.L. (1967). Determination of the tryptophane content in feeding stuffs with particular reference to cereals. J. Sci. Food Agric., 18: 381-387.

Mital, B.K. and S. K. Grag (1990). Tempe-Technology and food value. Food Rev. Intern. , 6:213-217.

Mitchell. H.H. and R.J. Block (1946). A method of determining the biological value of protein. J. Bio. Chem., 163: 599.

Moeljopawiro, S.; D.T.Gordon and M.L.Fields (1987). Bioavailability of iron in fermented soybeans. Food Sci., 52(1): 102-105.

Mohamed, A.; P.A. Perera and Y.S. Hafez (1986). New chromophore for phytic acid determination cereal Chem., 63(6): 475-478.

Moore, S. and W.H. Stein (1963). Chromatographic determination of amino acids by the use of automatic recording equipment. In: Methods in Enzymology, vol.6, S.P. Colowick and N.O. Kaplan (Eds), Academic press, New York, pp. 819.

Murata, K.; H. Ikehata and T. Miyamoto (1967). Studies on the nutritional value of tempeh. J. Food Sci., 32: 580-586.

Njoku, H.O.; C.O. Ofuya and J. N. Ogbulie (1991). Production of tempeh from the African yam bean (*Sphenostylis stenocarpa Hans*). Food Microb., 8(3): 209-214.

Nout, M.J. R.; G. Beernink and M.G. Bononts- Van Laarhoven (1987). Growth of *Bacillus cereus* in soybean tempeh. Intern. J. Food Microb., 4: 293-301.

Nout, M.J. R.; S. Notermans and F.M. Rombouts (1988). Effects of environmental conditions during soybean fermentation on the growth of *staphylococcus aureus* and production and thermal stability of enterotoxins A and B. Intern. J. Food Microb., 7: 299- 309.

Nout, M.J. R. and F.M. Rombouts (1990). Recent development in tempe research (Review). J. Appl. Bact., 69: 609-633.

Paredes-Lopez, O. and G.I.Harry (1989). Changes in selected chemical and antinutritional components during tempeh preparation using fresh and hardened common beans. J. Food Sci., 54(4): 968- 970.

Paredes-Lopez, O.; G. I. Harry and J. Gonzalez-[1]Castaneda (1990). Sensory evaluation of tempeh produced by fermentation of common beans .J. Food Sci., 55: 123- 126.

Pellet, P.L. and ver Young (1980). Nutritional evaluation of protein foods. WHTR-3UnμP-129. The United Nations University.

Ruiz- Teron, F. and J.V. Owens (1996). Chemical and enzymatic changes during the fermentation of Bacteria -free soybean tempe. J.Sci. Food Agric., 71: 523-528.

Salem, F.A.: M.i.Shehata; E.Abd El-Latif and S.M. Abou El-Maati (1994). Effect of pre-treatments and some processing techniques on the antinational properties of some legumes. Egypt J. Agric. Res., 72 (4): 1101- 1111.

Salgo, A.; K. Granzler and J. Jecsal (1984). Simple enzymatic methods for prediction of plant protein digestibility. Proc. Intern. Assoc. Cereal Chem. Symp. R. Lasztity and M. Hidvegi(Eds.), Akademiai Kiado, Budapest, PP. 311.

Shurtleff.W. and A. Aoyagi (1979). The Book Tempeh, a Super Soy food from Indonesia. New York: Harper and Row, USA.

Sutardi, A. and K. A. Buckle (1985). Phytic acid changes in soybeans fermented by traditional inoculum and six strains of *Rhizopus oligosporus*. J. Appl. Bact., 58: 539- 543.

Wang, H.L.; E.W. Swain and C.W. Hesseltine (1980). Phytase of mold used in Oriental food fermentation. J. Food Sci., 45 (5): 1262- 1266.

Wang, H.L.; E.W. Swain L.L.Wallen and C.W. Hesseltine (1975). Free fatty acid identified as antitryptic factor in soybean fermented by *Rhizopus oligosporus* . J. Nut., 105: 1351- 1355.

Yueh, M.H.; G.V. Daravingas; F.J.Rigelhof and H.W.Mueller (1979). Process for producing a fried snack food containing tempeh. US patent, 4 151 307 [C.F., FSTA, 12(1) G2, 1980].

Youssef, M.M. (1978). A study of factors affecting the cookability of faba bean (*Vicia faba)*. Ph. D. Thesis, Food Sci. and Technol. Dept., College of Agric., Alex. Univ., Egypt.

تقييم التيمب الناتج من تخمر فول الصويا بواسطة نوعين من فطر الريزوبس

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تم إنتاج التيمب من فول الصويا منزوع القشرة بعد نقعه و أوتكلفته ثم اجري له عملية تخمر بواسطة نوعين من فطر الريزوبس و ذلك بهدف تقييمه كيماويا وغذائيا وميكروبيا وحسيا.

و قد أوضحت النتائج أن أفضل إنتاج للتيمب أمكن الحصول عليه بعد ٢٦ و ٣٠ ساعة تخمر في درجة حرارة بين ٣١- ٣٢ °م بواسطة ريزوبس أوليجوسبورس و ريزوبس اريزس علي التوالي .

و قد لوحظ أن عمليتي النقع و الأوتكلفة لفول الصويا قد أحدثتا انخفاض معنوي في محتواه من المستخلص الأثيري و الرماد و الكربوهيدرات الكلي و المواد الصلبة الكلية و المواد الذائبة في الماء و المواد الغير ذائبة و البروتين الحقيقي و المعادن وعدد الخمائر و بكتريا حمض اللاكتيك . و علي العكس وجد أن محتوي الرطوبة و البروتين الخام و النيتروجين الغير بروتيني زادوا وبدرجة معنوية في البذور المنقوعة و التي حدث لها أوتوكلفة ، بينما لم يحدث أي تغرفي

محتوي الألياف الخام و عدد الجراثيم البكتيرية الكلية في نفس البذور .وقد بينت النتائج أن عمليـة التخمر سواء بواسطة الريزوبس أوليجوسبورس أو بواسطة الريزوبس أريزس قد أحدثـت زيـادة معنوية في محتوي الرطوبة و البروتين الخام و النيتروجين الغير بروتيني و الألياف الخام و المواد الذائبة في الماء و عدد بكتريا حمض اللاكتيك و الجراثيم البكتيرية الكلية بينما على العكس بالنسـبة للمستخلص الأثيري و الكربوهيدرات الكلية و المواد الصلبة الكلية و المواد غير الذائبة و البروتين الحقيقي والتي انخفضت بدرجة معنوية . كما لوحظ زيادة طفيفة في محتوي الحديد و البوتاسـيوم ، و انخفاض في محتوي الكالسيوم و النحاس و الصوديوم بعد عملية التخمر بينما لم يتغير محتـوي الماغنسيوم و المنجنيز و الزنك و عدد الخمائر بعد هذه العملية .

كما تشير النتائج أن إنتاج التيمب لم يحدث تغيير أو كان طفيفا في محتوي الأحماض الأمينية الأساسية كما أن معظم الأحماض الأمينية الأساسية كانت مماثلة للكميات الأساسية اللازمة لتـوازن البروتين تبعا لتوصيات منظمة الأغذية و الزراعة و منظمة الصحة العالميـة (١٩٧٣) . و علـي الرغم من زيادة محتوي الأحماض الأمينية الكبريتية كنتيجة لعملية التخمر إلا أنها سجلت أقل قيمـة بالمقارنة بمقررات منظمة الأغذية و الزراعة و منظمة الصحة العالمية . كمـا وجـد أن الرقـم الكيميائي للأحماض الأمينية الموجودة في التيمب الناتج بواسطة الريزوبس اريزس كانت أعلي من الموجودة في التيمب الناتج بواسطة الريزوبس أوليجوسبورس باستثناء الليوسين. ووجد أيضـا أن عملية إنتاج التيمب قد أظهرت تحسنا معنويا في هضمية البروتين و كفاءتـه الحسـابية و القيمـة الحيوية كما أدت إلي انخفاض معنوي في محتوي حمض الفيتيك و مثبط التربسين و أخـيرا فـإن التقييم الحسي أظهر أن كل منتجات التيمب قد لاقت قبولا جيدا.