PJN
ISSN 1680-5194

PAKISTAN JOURNAL OF NUTRITION

ANSInet
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorpjn@gmail.com

OPEN ACCESS Pakistan Journal of Nutrition

ISSN 1680-5194
DOI: 10.3923/pjn.2017.221.226

Research Article
Development and Properties of Tegurt, a Yogurt-like Tempe Product

[1]Siti Harnina Bintari, [2]Nyoman Suci Widyastiti, [3]Natalia Desy Putriningtyas, [4]Rebriarina Hapsari and [5]Kartika Nugraheni

[1]Department of Biology, Faculty of Mathematics and Natural Science, Semarang State University, Semarang, Central Java, Indonesia
[2]Department of Clinical Pathology, Faculty of Medicine, Diponegoro University, Semarang, Central Java, Indonesia
[3]Department of Nutrition, Faculty of Health Science, Respati University of Yogyakarta, Yogyakarta, Indonesia
[4]Department of Clinical Microbiology, Faculty of Medicine, Diponegoro University, Semarang, Central Java, Indonesia
[5]Department of Nutrition, Faculty of Nursing and Health Science, Muhammadiyah University of Semarang, Semarang, Central Java, Indonesia

Abstract

Background: Soybeans and their derivatives are a major component of Indonesia's domestic food landscape and have been consumed in several forms. Tempe was originally intended to be eaten as plant-based protein source. In recent years, there have been some new innovations to make tempe more consumable as it has a high nutritional value and antioxidant content. **Objective:** This study aims to develop a yogurt that is partially substituted with tempe flour and to analyze its acceptability, nutritional content and antioxidant capacity. **Materials and Methods:** Tempe yogurt was made with varying amounts of tempe flour (2.5, 5, 7.5 and 10%). Fresh tempe was made using a double-pasteurisation method and dried to form flour. Tempe yogurt was analyzed for acceptability and nutrient and antioxidant contents. **Results:** Yogurt with 10% added tempe flour had the highest fat, protein, carbohydrate and flavonoid contents, while yogurt with 7.5% added tempe flour was the most acceptable according to hedonic tests. **Conclusion:** Tempe yogurt shows potential as a functional food, especially for consumers with metabolic diseases.

Key words: Food functional, soybean, tegurt, tempe, yogurt

Received: December 12, 2016 Accepted: February 17, 2017 Published: March 15, 2017

Citation: Siti Harnina Bintari, Nyoman Suci Widyastiti, Natalia Desy Putriningtyas, Rebriarina Hapsari and Kartika Nugraheni, 2017. Development and properties of tegurt, a yogurt-like tempe product. Pak. J. Nutr., 16: 221-226.

Corresponding Author: Siti Harnina Bintari, Department of Biology, Faculty of Mathematics and Natural Science, Semarang State University, Indonesia

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Soybeans and their derivatives are a major component of Indonesia's domestic food landscape. Indonesia's high soy consumption makes it one of the largest soy markets in Asia. Indonesians consume soybean products in several forms: Up to 50% as Tempe, 40% as tofu and 10% in other forms (fermented soy paste, soy sauce, etc.)[1]. Tempe is made of whole soybeans fermented with fungi, such as *Rhizopus oligosporus*, *Rhizopus oryzae*, *Rhizopus stolonifer*, *Rhizopus chlamydosporus* and *Rhizopus arrhizus*. The Indonesian National Standards Authority (SNI) defines tempe as a soybean product that is fermented with *Rhizopus* sp. and formed into compact solid blocks and is generally white with a slight greyish tint and a distinct tempe aroma[2].

Soybean tempe is an Indonesian fermented specialty and an icon of Javan cuisine. It is also rich in active nutritional ingredients. The soybean fermentation process causes changes in taste, aroma, texture, nutritional content and digestibility[3]. The principal advantages of soybean lie in its nutritional composition, easily digested protein content and high levels of essential amino acids. Tempe also has lower concentrations of anti-nutritional components, such as antitrypsin and phytic acid, than do raw soybeans. The main disadvantage of fresh tempe is its relatively short shelf life. One method commonly used to extend its shelf life is grinding it into flour[1].

Tempe is traditionally eaten with rice but in recent years there have been some new innovations such as tempe burgers, tempe salads and various third-generation products[3]. Tempe burgers and salads are generally regarded as second-generation derivatives. In these products, the shape, texture and taste of the tempe component are no longer apparent as the processing blends the tempe with other ingredients. Third-generation tempe derivatives are characterized by the effort to extract, isolate and purify the bioactive ingredients in Tempe[1]. They take the form of useful bioactive compounds, such as antioxidants, that can be used to preserve foods, prevent cancer and reduce blood glucose levels. This study aims to explore tempe yogurt's technical characteristics and evaluate it for use as a probiotic product and functional food with health benefits.

MATERIALS AND METHODS

The production of tempe as the main ingredient of Tegurt is based on the tempe fermentation method described by Bintari[4]. This method involves pasteurization with two heating stages: One before and one after steeping. The first heating stage runs at 100°C for 30 min, while the second runs at 60-70°C for 15-20 min. This pasteurization kills pathogenic bacteria, reduces the concentration of anti-nutrients and prevents protein denaturation[4].

The two-stage cooking process generally begins with sorting the soybeans, followed by the first heating step to soften and remove the seed coat. The de-hulled beans are then washed and steeped before the second heat step (Pasteurization and leaking); fermentation is then started with a *Rhizopus* strain.

The pasteurization step makes use of a double pan, where a small plastic pan is inserted into a larger metal container that has been filled with water. This second heating step improves protein quality by reducing the concentration of trypsin inhibitors and increasing the amount of vitamin B12, maintaining the tempe's freshness for 80 h[4].

The next stage is making the tempe yogurt, which we prepare with several different amounts of tempe flour (Table 1). Fresh tempe is dried, ground into flour and mixed with fresh milk. The mixture is strained through cheesecloth and then warmed to 43-45°C, while sugar is added to a concentration of 6.5%. The milk mixture is heated to 85-90°C and kept at that temperature for 30 min under constant stirring. After 30 min, the mixture is cooled to 45°C and inoculated with a yogurt starter culture. The culture is made of *Lactobacillus acidophilus* and *Lactobacillus casei* in a 50:50 milk mixture, incubated for 4-6 h and chilled for 12 h to stop the fermentation process. The resulting tempe yogurt is tested by semi-trained panelists prior to initial organoleptic testing.

The formulation for the tempe yogurt was defined in the Microbiology Laboratory of the Semarang National University and the Food Processing Laboratory at the Muhammadiyah University in Semarang. Proximate analysis and flavonoid content measurements were performed in the Food and Nutrition Laboratory at Gadjah Mada University. Organoleptic tests (of appearance, taste, aroma, texture and color) were performed in the Clinical Pathology Laboratory at the University of Diponegoro in Semarang using a 9 point hedonic scale with scores ranging from 0-9 (from "dislike extremely" to "like extremely"). The results of these tests were analyzed using SPSS and p-values of less than 0.05 were considered statistically significant.

Table 1: Possible compositions of tempe yogurt

Groups	Ground tempe flour (%)	Cow's milk (%)	Sucrose (%)
T0	0.0	100.0	5
T1	2.5	97.5	5
T2	5.0	95.0	5
T3	7.5	92.5	5
T4	10.0	90.0	5

RESULTS AND DISCUSSION

Raw soybeans have a mildly bitter taste but the steeping, washing and enzymatic modifications during the fermentation process reduce the beany flavor. Tempe is a fermented soybean product cultured with *Rhizopus* sp. This microorganism can grow at low concentrations (0-2%) at an optimum temperature of 37°C[5]. The fermentation process decreases the concentration of anti-nutrients, such as phytic acid; deactivates the lipoxygenase enzyme and increases nutrient bioavailability. Phytic acid is reduced to one-third of its original concentration in raw soybeans under the tempe fermentation process with the bacteria *Rhizopus oligosporus*[6]. Tempe is a traditional food produced through the fermentation of soybeans with *Rhizopus* moulds of the species *Rhizopus oligosporus*, *R. oryzae* and *R. stolonifer* (known as tempe moulds). Soybeans fermented by the mould *Rhizopus oligosporus* experience a change in protein structure, while *Rhizopus oryzae* can produce amylase and *Rhizopus stolonifer* can produce pectinase. Soybeans fermented with *Rhizopus oligosporus* are bound into a solid cake by the mould's mycelium[7].

The main benefits of soybean fermentation are improvements in organoleptic qualities and nutritional value and a reduced need for preservatives. The steeping, boiling, cooking and fermentation of soybeans in tempe production greatly decrease phytic acid and antitrypsin concentrations. Reducing phytic acid is very important for increasing the bioavailability of minerals. Fermentation can reduce phytic acid concentration to a low enough level that calcium, zinc and iron become significantly more available[8].

One common issue with fresh tempe is its short shelf life; grinding tempe into flour is one approach for extending its shelf life[1]. Fresh tempe has a very high water content (55-65%) that supports a living and growing microbial community; thus, reducing this water content is a major issue. One kilogram of fresh tempe can be expected to yield approximately 700 g of tempe flour[9].

Only 100 g of tempe can provide 37% of the recommended daily intake of protein. This study's analysis of tempe flour suggests an even higher protein content of 41.08% (Table 2). The variety of amino acids in tempe is also quite complete. Its most abundant amino acids are glutamic acid, aspartic acid and leucine. The fermentation process also decreases the level of fat saturation; thus, tempe contains a good amount of polyunsaturated fatty acids (PUFAs)[1].

Tempe contains 220 mg of omega-3 fatty acids and 3590 mg of omega-6 fatty acids per 100 g. Tempe is also a plant source of vitamin B12 (cyanocobalamin), which is more commonly found in animal food products. Other vitamins found in tempe include B2, B6, B1, niacin, folic acid and fat-soluble vitamins (vitamins A, D, E, K). Vitamin B12 levels can increase up to 33-fold during fermentation; riboflavin increases 8 to 47-fold; pyridoxine increases 4 to 14-fold; niacin increases 2 to 5-fold; folic acid increases 4 to 5-fold; and pantothenic acid increases two-fold. The vitamins produced during tempe fermentation are not by-products of the mould but rather of contaminating bacteria, such as *Klebsiella pneumoniae* and *Citrobacter freundii*[1].

The fermentation agent in tempe is a fungus that breaks down the soy proteins into amino acids, making them easier to digest[3]. This alters the overall protein content in tempe and especially impacts the absorption process. Tempe has a higher nutritional value and can be consumed as an affordable protein source[8]. This increased digestibility leads to a qualitatively better nutritional value than that of unprocessed soybeans. The activity of proteolytic enzymes increases the water solubility of the proteins[10].

Table 2 shows the nutritional composition of tempe flour. Raw soybeans contain approximately 38-44% protein[11]. By comparison, the protein content of the tempe flour used in this research is 41.08%. Fermentation during tempe preparation increases the amount of individual free amino acids between 1 and 85 times above the levels in raw soybeans[12]. Other nutritional components show no significant changes.

Table 3 shows the results of a nutritional analysis on tempe yogurt. The tempe yogurt formulations with the highest fat, protein, carbohydrate and flavonoid contents are, respectively; T4, T1, T4 and T4. According to these results, the T4 formulation is most recommended for consumption, although organoleptic tests with semi-trained panellists show that most of the panellists preferred "tegurt" with a composition of 7.5% tempe flour and 92.5% cow's milk (T3).

Do Amaral Santos *et al.*[13] developed a new functional fermented beverage from peanut and soy using mixed cultures, including *L. acidophilus* and showed that it can be consumed by all ages. Shori[14] showed that soybean yogurt made from cow's and camel's milk has antioxidant activity. This study also revealed that tempe yogurt shows promise as an antioxidant-rich beverage.

Table 2: Analysis of contents in tempe flour

Sample	Water content (%)	Ash content (%)	Fat content (%)	Protein content (%)	Flavonoid (ppm)
Tempe flour	5.41	2.66	22.46	41.08	70.85

Table 3: Analysis of components in tempe yogurt

	Nutrition						
Groups	Water (%)	Ash (%)	Fat (%)	Protein (%)	CH (%)	Flavonoid (%)	Calories (cal g^{-1})
T0	83.68	0.78	2.24	2.04	11.26	0.006870	861.06
T1	83.18	0.66	2.65	3.01	10.51	0.008444	821.98
T2	83.66	0.66	3.02	2.58	10.09	0.009173	825.14
T3	83.40	0.65	2.91	2.61	10.43	0.009606	889.69
T4	82.94	0.68	3.12	2.70	10.55	0.011146	907.66

Table 4: Microbiological tests on tempe yogurt

	Microbiological analysis			
Groups	pH	Bacterial TPC (CFU mL^{-1})	Fungal TPC (CFU mL^{-1})	Yeast TPC (CFU mL^{-1})
T0	4.60	1.48×10^{8}	$<10^{1}$	3.5×10^{6}
T1	4.49	2.42×10^{8}	$<10^{1}$	9.0×10^{6}
T2	4.41	2.60×10^{8}	0	4.7×10^{7}
T3	4.40	2.60×10^{8}	0	3.9×10^{7}
T4	4.43	3.59×10^{8}	0	3.4×10^{7}

Yogurt is viewed as a highly nutritious dairy food product due to its low carbohydrate content and high levels of protein, vitamin B-complex, phosphorus, magnesium and potassium. Yogurt's lactic acid bacterial cultures can also improve digestive conditions, including gut microflora growth, bowel transit time and immune response in the alimentary tract[15].

Table 4 shows an increase in lactic acid bacteria in all groups of tempe yogurt. It also shows that the *Lactobacillus* are clearly stimulated by the addition of tempe flour to the yogurt. The growth of mould-a detrimental microorganism in yogurt-is inhibited by the addition of tempe flour via this induction of lactic acid bacteria. The yeast and fungi in these tempe yogurt products were by-products of the tempe fermentation process; yeast was detected in the tempe yogurt because it grows well in the yogurt's acidic environment.

Lactobacillus is known to modify gut flora and affect metabolic enzymes, such as bile hydrolase, azoreductase and β-glucuronidase. Previous study also shows that fermented milk containing *Lactobacillus* can reduce cholesterol levels in mice serum and liver[15,16].

Phytoestrogens are found in various fruits, beans and vegetables. There are three broad classes of phytoestrogens, namely, isoflavones, lignans and coumestans. Isoflavones are flavonoid compounds commonly found in beans, especially soybeans. Phytoestrogens are structurally similar to oestradiol, with a non-steroidal structure and a phenolic ring that enables binding to oestrogen receptors (ERs) as well as the ability to act as either an agonist or an antagonist oestrogen[17].

Flavonoids are bioactive phenols commonly found in fruits, vegetables and parts of plants and have a major role as antioxidants[18]. Fermentation increases antioxidant activity and thus the functional value of the foodstuff in question. A study by McCue and Shetty[19] shows that fermentation causes a transformation of glycosides accompanied by an increase in glycosidase and glucuronidase activity along with the release of potential antioxidants through flavonoid transformation. Table 4 shows that the T4 group has the highest flavonoid content among the tempe yogurt formulations tested.

These results show that fermentation has increased the bioavailability of isoflavones during the digestion process. The high bioavailability comes from the activity of microorganisms, which degrade macromolecular compounds into simpler forms that are easier to digest and absorb. During fermentation, isoflavones change from glycosides into aglycones, while proteins are broken up into peptides and amino acids. This change increases the anti-diabetic activity of the fermented soy product[20]. Isoflavones as a flavonoid component, can influence cellular insulin secretion capacity[18]. Insulin secretion in DM type 1 patients has been shown to improve under glucose control[21].

There are two main forms of isoflavones in soybeans: β-D-glycosides known as genistein and daidzein. These glycosides are biologically inactive. Isoflavone glycosides are hydrolysed by β-glycosidase bacteria on the intestinal wall into bioactive aglycone forms (genistein and daidzein). The aglycone forms can be absorbed by the intestines, which improve their biological activity. Daidzein can be rapidly metabolized into equol and O-demethyangolensin, while genistein is metabolised into p-ethyl phenol. Genistein, daidzein, equol and O-demethyangolensin are the primary isoflavones found in human and animal blood and urine[21]. Healthy adult humans can absorb isoflavones quickly and efficiently[22]. The effective half-lives of daidzein and genistein-9.3 and 7.1 h, respectively indicate that isoflavones and their metabolites are rapidly excreted[23].

Polyphenolic flavonoids display strong antioxidant activity[24]. Genistein and daidzein can increase plasma insulin levels in test animals with type 1 diabetes. Isoflavones are active flavonoids that can increase antioxidant activity by increasing the level of cellular antioxidant enzymes, such as superoxide dismutase (SOD), catalase and glutathione peroxidase[25].

The flavonoid and lactic acid bacteria content in this tempe yogurt gives it potential as a functional food product. Flavonoids, especially tempe isoflavones, have antioxidant activity that can help protect against free radicals and prevent clinical complications from metabolic diseases, such as diabetes mellitus, dyslipidemia and metabolic syndrome[26-28]. A high-isoflavone diet is also possibly beneficial for accelerating the apoptosis of cancer cells[29]. The lactic acid bacteria in tempe yogurt can provide relief from diarrhoea and other digestive tract diseases[30].

This study shows that tempe can be used to create a nutritious and healthy beverage. Tempe yogurt is suitable for consumption not only by healthy people but also by those who have metabolic diseases, such as dyslipidemia and diabetes as it contains isoflavones, fibre, protein and lactic acid bacteria that could help decrease blood sugar, cholesterol and triglyceride levels. This study has many limitations that could be improved by further research, especially regarding the safety and recommended dosages for certain health conditions.

CONCLUSION

Tempe yogurt, containing many beneficial and nutritious components, has a good chance of getting a positive reception from the public. It shows potential as a functional food that could be used as part of a person's daily intake or as a dietary intervention for free radical-related diseases.

ACKNOWLEDGMENT

This study received grant funding from the University of Diponegoro budget in 2016 with award number 1051-23/UN7.5.1/PG/2016 in the name of Dr. Nyoman Suci Widyastiti, Sp.PK, M.Kes.

REFERENCES

1. Astawan, M., 2008. Sehat Dengan Tempe: Panduan Lengkap Menjaga Kesehatan Dengan tempe Panduan Lengkap Menjaga Kesehatan Dengan Tempe. Dian Rakyat, Jakarta, Indonesian.
2. Nasional, B.S., 2012. Tempe: Persembahan Indonesia Untuk Dunia. Badan Standardisasi Nasional, Jakarta, Indonesian.
3. Nout, M.J.R. and J.L. Kiers, 2005. Tempe fermentation, innovation and functionality: Update into the third millennium . J. Applied Microbiol., 98: 789-805.
4. Bintari, S.H., 2013. Pasteurization for hygienic tempe: Study case of Krobokan Tempe yesterday and today. GSTF Int. J. BioSci., 2: 39-44.
5. Babu, P.D., R. Bhakyaraj and R. Vidhyalakshmi, 2009. A low cost nutritious food Tempeh: A review. World J. Dairy Food Sci., 4: 22-27.
6. De Arruda Oliveira, E., L.M.G. Cheim, R.V. Veloso, V.C. Arantes and M.A. de Barros Reis *et al*., 2008. Nutritional recovery with a soybean flour diet improves the insulin response to a glucose load without modifying glucose homeostasis. Nutrition, 24: 76-83.
7. Osundahunsi, O.F. and A.C. Aworh, 2002. A preliminary study on the use of tempe-based formula as a weaning diet in Nigeria. Plant Foods Hum. Nutr., 57: 365-376.
8. Cuevas-Rodriguez, E.O., N.M. Verdugo-Montoya, P.I. Angulo-Bejarano, J. Milan-Carrillo, R. Mora-Escobedo and L.A. Bello-Perez, 2006. Nutritional properties of tempeh flour from quality protein maize (*Zea mays* L.). LWT-Food Sci. Technol., 39: 1072-1079.
9. Bintari, S.H., 2007. The effect of tempe isoflavone on proliferation and apoptosis of breast cancer cell in Mus musculus strain C3H on AgNORs, p53, Cas-3 anf Bcl-2. Ph.D. Thesis, Diponegoro University, Semarang.
10. Wulan, S.N., M. Astuti, Y. Marsono and Z. Noor, 1999. The examination of hypoglycemic effect of soybean, soybean protein fraction and tempe on diabetic rats. Jurnal Teknologi Pertanian, 3: 94-102 (In Indonesia).
11. Pohl, J., 1988. H. E. Snyder und T. W. Kwon: Soybean Utilization. 346 Seiten, zahlr. Abb und Tab. An AVI Book, published by Van Nostrand Reinhold Company, New York 1987. Preis: 39,95 £. Mol. Nutr. Food Res., 32: 408-408.
12. Farnworth, E.R., 2008. Handbook of Fermented Functional Foods. CRC Press, USA., ISBN: 9781420053265, Pages: 600.
13. Do Amaral Santos, C.C.A., B. da Silva Libeck and R.F. Schwan, 2014. Co-culture fermentation of peanut-soy milk for the development of a novel functional beverage. Int. J. Food Microbiol., 186: 32-41.
14. Shori, A.B., 2013. Antioxidant activity and viability of lactic acid bacteria in soybean-yogurt made from cow and camel milk. J. Taibah Univ. Sci., 7: 202-208.
15. Georgakouli, K., A. Mpesios, D. Kouretas, K. Petrotos, C. Mitsagga, I. Giavasis and A.Z. Jamurtas, 2016. The effects of an olive fruit polyphenol-enriched yogurt on body composition, blood Redox status, physiological and metabolic parameters and yogurt Microflora. Nutrients, Vol. 8. 10.3390/nu8060344

16. Sakin-Yilmazer, M., S.N. Dirim, D. Di Pinto and F. Kaymak-Ertekin, 2014. Yoghurt with candied chestnut: Freeze drying, physical and rheological behaviour. J. Food Sci. Technol., 51: 3949-3955.
17. Makela, S., R. Santti, L. Salo and J.A. McLachlan, 1995. Phytoestrogens are partial estrogen agonists in the adult male mouse. Environ. Health Perspect., 103: 123-127.
18. Pinent, M., A. Castell, I. Baiges, G. Montagut, L. Arola and A. Ardevol, 2008. Bioactivity of flavonoids on insulin secreting cells. Comprehens. Rev. Food Sci. Food Safety, 7: 299-308.
19. McCue, P. and K. Shetty, 2003. Role of carbohydrate-cleaving enzymes in phenolic antioxidant mobilization from whole soybean fermented with *Rhizopus oligosporus*. Food Biotechnol., 17: 27-37.
20. Kwon, D.Y., J.W. Daily III, H.K. Kim and S. Park, 2010. Antidiabetic effects of fermented soybean products on type 2 diabetes. Nutr. Res., 30: 1-13.
21. Setchell, K.D., 1998. Phytoestrogens: The biochemistry, physiology and implications for human health of soy isoflavones. Am. J. Clin. Nutr., 68: 1333S-1346S.
22. Setchell, K.D., 2001. Soy isoflavones-benefits and risks from nature's Selective Estrogen Receptor Modulators (SERMs). J. Am. College Nutr., 20: 354S-362S.
23. Kao, T.H., Y.F. Lu., H.C. Hsieh and B.H. Chen, 2004. Stability of isoflavone glucosides during processing of soymilk and tofu. Food Res. Int., 37: 891-900.
24. Bhonde, R., R.C. Shukla, M. Kanitkar and R. Shukla, 2007. Isolated islets in diabetes research. Indian J. Med. Res., 125: 425-440.
25. Choi, M.S., U.J. Jung, J. Yeo, M.J. Kim and M.K. Lee, 2008. Genistein and daidzein prevent diabetes onset by elevating insulin level and altering hepatic gluconeogenic and lipogenic enzyme activities in Non Obese Diabetic (NOD) mice. Diabetes/Metabol. Res. Rev., 24: 74-81.
26. Galleano, M., V. Calabro, P.D. Prince, M.C. Litterio and B. Piotrkowski *et al*., 2012. Flavonoids and metabolic syndrome. Ann. NY. Acad. Sci., 1259: 87-94.
27. Babu, P.V.A., D. Liu and E.R. Gilbert, 2013. Recent advances in understanding the anti-diabetic actions of dietary flavonoids. J. Nutr. Biochem., 24: 1777-1789.
28. Vinayagam, R. and B. Xu, 2015. Antidiabetic properties of dietary flavonoids: A cellular mechanism review. Nutr. Metabol., Vol. 12. 10.1186/s12986-015-0057-7
29. Messina, M., 2016. Soy and health update: Evaluation of the clinical and epidemiologic literature. Nutrients, Vol. 8. 10.3390/nu8120754
30. Soenarto, Y., I. Sudigbia, H. Herman, M. Karmini and D. Karyadi, 2001. Antidiarrheal characteristics of tempe produced traditionally and industrially in children aged 6-24 months with acute diarrhea. Paediatrica Indonesiana, 41: 88-95.