CODEX ALIMENTARIUS COMMISSION



Food and Agriculture Organization of the United Nations



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# Agenda Item 8

CX/ASIA 10/17/9 September 2010

# JOINT FAO/WHO FOOD STANDARDS PROGRAMME FAO/WHO COORDINATING COMMITTEE FOR ASIA

Seventeenth Session

Yogyakarta, Indonesia, 22 – 26 November 2010

# DISCUSSION PAPER ON TEMPE AND TEMPE PRODUCTS

Prepared by Indonesia

# Background

At the 16<sup>th</sup> session FAO/WHO Coordinating Committee for ASIA, the Delegation of Indonesia introduced the proposal for new work on the development of a standard for tempe and tempe products.

One delegation, while not objecting to considering the proposal, recalled the concern expressed at the 15<sup>th</sup> Session of the CCASIA regarding the late availability of project documents and expressed the view that no decision should be made at the current session because the proposal was made available only at the meeting thus making it difficult to consult with relevant national stakeholders. It was also noted that project documents for new work should be prepared according to the format set out in the current revision of the Procedural Manual.

The Coordinating Committee agreed to request Indonesia to prepare a comprehensive discussion paper to justify the need for new work and including a detailed project document, as per the above comments, for consideration at its next session.

Asia Pacific J Clin Nutr (2000) 9(4): 322–325 contained a review article on "Tempe, a nutritious and healthy food from Indonesia" written by Mary Astuti (Centre of Women's Studies, Gadjah Mada University, Yogyakarta, Indonesia), Andreanyta Meliala, Fabien S Dalais and Mark Lwahlqvist (*International Health and Development Unit, Faculty of Medicine, Monash University, Melbourne, Victoria, Australia*). The information below are taken from the article as a scientific source.

# Introduction

# History

Tempe is a widely consumed Indonesian traditional fermented food, which is principally made with soybeans, but can also be made from a variety of legumes and seeds. In Indonesia, soybeans are consumed mostly in the form of traditional foods, consisting of fermented and non-fermented products. Commercially fermented soybeans include tempe, soy sauce and soy paste. Historical evidence shows that soybean tempe is a fermented product originally made by Central Javanese people and appeared in their food pattern around the 1700s.<sup>1,2</sup> In addition, this finding is supported by the presence of non-salted fermented soybeans in Asian countries, for example, *natto* in Japan, *dau chi* in China and *kinema* in Nepal and India, which are products fermented by bacteria *Bacillus* sp.1 Tempe, however, is fermented by the *Rhizopus* sp.

# How is it made?

There are basically six steps in the tempe manufacturing process, namely hydration, skin removing, heat treatment, mold inoculation, wrapping and incubation. Tempe in Indonesia is fermented with *Rhizopus* sp. mould, especially *Rhizopus oligosporus*, *R. oryzae*, *R. arhizus*, *R. stolonifer* and *R. microsporus* (I Ganjar, unpubl. data, 1995). Traditional inoculum is prepared in Hibiscus or teak leaf and inoculum powder is prepared from cooked rice. Tempe producers in Indonesia do not use the pure culture of *R. oligosporus*, but they use a mixed culture of *Rhizopus* sp. There is no standard process for tempe making, which is one of the reasons why there is a lot of variation in tempe making from one region and one producer to another.

# **Tempe in Indonesia**

Apart from soybean-based tempe, there are many other kinds of tempe in Indonesia and their name is derived from the raw material in the tempe. They include sword beans, velvet beans, pigeon peas, *leucaena lecocephala* and tofu waste material (left over soy-pulp after tofu preparation). As soybean tempe is the most popular, the word tempe usually refers to soybean-tempe. The local name of tempe can be used for international and regional publications, as there are no common English names available.<sup>3</sup> Tempe is produced mostly by small household industries with a production range of 10 kg–4 metric tons of tempe per day.<sup>4</sup> It is estimated that there are more than 100 000 tempe producers spread out in the provinces of Indonesia. Urban and rural populations, especially in Java, generally consume tempe as a part of their food pattern. As a source of protein, tempe is consumed in greater quantities than other protein sources. Tempe supplies at least 10% of the current protein consumption, while chicken egg supplies 1.25%, meat supplies 3.15% and cereals supply around 60%.<sup>4</sup> Tempe is not consumed as a fresh food but in the form of cooked tempe and served as a delicacy or a side dish, often fried, boiled, steamed or roasted. Tempe is categorised as a low social value food, which means that tempe is only served at home as domestic food and through stall food-vendors, although tempe is consumed by people of various ages of different socioeconomic status.

#### Biochemical changes from soybean to tempe

Research in the tempe field has shown its potential health benefit, possibly due to biochemical changes during soybean fermentation. During tempe processing, there are valuable changes not only in the increase of nutritional values of some nutrients in soybeans, but also in the development of vitamins, phytochemicals and antioxidative constituents.

#### Protein

Although the protein content of tempe and unfermented soybeans is almost the same, the soluble protein content increases sharply due to the action of protease enzyme produced by mould during fermentation. Furthermore, the quality of protein in tempe is slightly higher than in unfermented soybean.<sup>5</sup> The soluble nitrogen content in unfermented soybeans is 3.5 mg/g, compared to 8.7 mg/g in tempe. Additionally, following 48 h of fermentation, most amino acids decrease in the range of 3.62-27.9%. According to Murata *et al.* total amino acids content decrease but the free amino acids increase sharply, possibly due to strains of *Rhizopus* using amino acids as a source of nitrogen for growth (AJ Graham *et al.*, unpubl. data, 1995).<sup>6</sup>

# Lipid

The lipid content of tempe is lower than that of unfermented soybeans. It has been shown that during soybean fermentation the lipase enzyme hydrolyses triacylglycerol into free fatty acids. These fatty acids are used as a source of energy for the mould resulting in lower lipid content in tempe. During soybean fermentation the lipid contents decrease about 26%. A study by Graham *et al.* shows that the mould of *R. oligosporus* and *R. stolonifer* use linoleic acid, oleic acid, palmitic acid as energy sources, therefore during fermentation, palmitic acid, stearic acid, and linoleic acid rapidly decrease 63.4, 59.25 and 55.78, respectively (AJ Graham *et al.*, unpubl. data, 1995).<sup>5</sup>

# Minerals

Trace mineral (iron, calcium and cuprum) levels were not influenced by the fermentation process; even so, their solubility increased sharply. Most of the iron in soybean is present as organic iron, which is bound to protein and other organic compounds. Total soluble iron increases from 24.29% in unfermented soybean to 40.52% in tempe and during fermentation soluble iron increases 66.51%.<sup>5</sup> Protein is broken down resulting in free amino acids, peptide or simple proteins. As a result, the iron is liberated from the iron protein complex, thus increasing soluble iron. Astuti *et al.* have shown that tempe is a good source of available iron.<sup>5</sup> Calcium content decreases during the fermentation, but is not clearly understood. Calcium is possibly released from the bridge of phytate–protein during digestion of complex compound and lost together with bound water, which may be released during fermentation.

# Vitamin B, B12 and tocopherol

The levels of vitamin B-complexes increase except for thiamine. Okada reported that vitamin B12 content of fresh tempe bought from Indonesia was 4.6  $\mu$ g/100 g, the range from tempe prepared with *R. oligosporus* NRRL 2710 in Japan was 0.03–0.06  $\mu$ g/100 g<sup>8</sup>. It seems that vitamin B12 is mainly produced by bacteria other than mould. During the soaking of soybeans, *Klebsiella pneumonia* is developed and produces vitamin

B12.<sup>8</sup> Tocopherol composition changes during fermentation. Except for  $\alpha$ -tocopherol, the levels of beta, gamma and delta tocopherol increases. Even though  $\beta$ -tocopherol has only 40% of the biological activity of  $\alpha$ -tocopherol, an increase of 222.5% in beta tocopherol adds value to the natural antioxidant activity of tempe.

# Phytic acid

Phytic acid is reduced by about 65% as a result of the action of phytase enzyme produced by *R. oligosporus.*<sup>9</sup> Phytic acid is known as an antinutrient factor which is able to bind divalent minerals, thus lowering the mineral bioavailability. Therefore, the decrease in phytic acid has a beneficial effect on mineral bioavailability.<sup>5</sup>

# Oligosaccharide

The level of glucose increases sharply during tempe fermentation, possibly a product of the digestion of complex to simple carbohydrate. Starch, stacchyose, raffinose and sucrose in soybean are all decreased during tempe processing.<sup>10</sup>

# Isoflavones

Isoflavones are oestrogenic compounds which have been reported to have many health beneficial effects. György *et al.*, Zilliken and Murakami *et al.* reported that tempe contains isoflavones.11–13 Hutchins *et al.* compared the urinary isoflavonoid recovery between groups fed with fermented soybeans in the form of tempe and unfermented soybean in the form of tofu. A greater increase in urinary isoflavonoid recovery in the fermented-soybean group suggests that fermentation increases the availability of isoflavones in soy.<sup>14</sup> Dalais (FS Dalais pers. comm., 1998), Wuryani, Wang and Murphy, Dwyer *et al.* and Pillow *et al.*, have measured the isoflavone levels in tempe and found that the values are relatively high compared to other soybean products such as tofu and soy beverages.<sup>15–18</sup>

# Superoxide dismutase

Superoxide dismutase (SOD) is a new enzyme, found in 1969. All biochemical living cell systems produce superoxide radicals, which cause a toxic effect to cells, thus every normal cell will have a defense system to protect the cells against the action of free radicals, such as superoxide anions.19,20 The free radical scavenger, SOD, is present during the fermentation process (M Astuti *et al.*, unpubl. data, 1996). At the early state of fermentation, no SOD is present, but after 24 h of fermentation it gradually increases until up to 60 h of fermentation then starts to decrease, possibly due to a decrease in mould growth which is influenced by environmental conditions, such as pH. The presence of SOD in tempe is concomitant with the mould growth.

# Health aspects of tempe

# Flatulence and diarrhoea

A decrease in oligosaccharide, especially raffinose content during tempe processing eliminates the flatulence problems observed when consuming soybeans. Tempe is essentially non-flatulent when it is fed to human subjects.<sup>20</sup> Van Veen and Schaffer observed the health benefits of tempe in preventing diarrhea problems in prisoners in Java. Individuals who did not consume tempe in their diet suffered from diarrhoea due to the bad sanitation of the prison.<sup>21</sup> A relevant thesis paper studied the effect of tempe on *E. coli* infection in a rabbit model. One group was given a diet containing tempe, the other were fed on a diet without tempe. After 4 weeks of feeding, both groups were infected with *E. coli* and observed for 14 days. Diarrhea symptoms occurred in 36% of the rabbits in the tempe group and 64% in the group without tempe.<sup>22</sup>

# Lipid-related health matters

# Lipid-lowering effect

The effect of a tempe-rich diet on cholesterol levels was reported by Mangkuwidjoyo *et al.* Tempe had a positive effect on cholesterol level and histopathological changes in liver and arteries of rats after a 4-month feeding trial.<sup>23</sup> Tempe constituents inhibit the enzyme which is responsible for biosynthesis of cholesterol and prevent the oxidation of low-density lipoprotein (LDL) thus minimize the production of plaque in arteries (M Astuti, unpubl. data, 1997).<sup>1</sup> The role of tempe on lipid profiles and lipid peroxidation has been studied by Astuti, using three groups of anemic rats fed with casein, unfermented soybean and tempe as a source of protein and iron.<sup>5</sup> Protein source affected the serum lipids. Unfermented soybean and tempe groups tended to have lower levels of total cholesterol and triacylglycerol. Tempe feeding depressed lipid peroxides

in the serum and liver, which could be correlated with the action of natural antioxidants in tempe. Isoflavonoids are able to form chelate complexes with iron which are efficient in inhibiting ferrousiron induced lipid peroxidation (HC Jha et al., unpubl. data, 1990). The hypocholesterolaemic effect of tempe was investigated by Astuti et al. using hyperlipidaemic rats, fed with tempe as a source of protein at varying concentrations (0, 25, 50, 75 and 100%) for 2 months. Lipid profile, lipid peroxide and superoxide dismutase activity were evaluated in rats' serum. Bile was then collected to evaluate a potential mechanism for cholesterol reduction. The result showed that tempe feeding lowered the cholesterol level in the tempe group, possibly due to the high content of cholesterol released from the liver through the bile (M Astuti, unpubl. data, 1997). According to Gorcia Hermosilla et al. free fatty acids in tempe inhibited the action of hydroxymethyl glutaryl CoA reductase, an enzyme which is responsible for cholesterol synthesis in the liver.<sup>25</sup> Hypocholesterolaemic properties of tempe in human subjects were studied by Astuti et al. in a feeding-trial of instant tempe-formula on 24 (8 male, 16 female) volunteers. Each respondent drank the formula daily for 3 months. The lipid profile, malondyaldehyde (MDA) and uric acid levels were measured in serum of each respondent as baseline before the feeding trial, every month during the feeding and 2 months after the feeding trial. (Malondyaldehyde is one of the products from the decomposition of fatty acid in lipid peroxidation. It is able to reach cell and tissue, thus resulting in cell damage. It does not only damage lipid molecules, but also non-lipid biomolecules, such as protein and nucleic acid. Damaged nucleic acid, especially in the nucleus, may cause gene mutation, which is able to promote cancer.) It showed that during the feeding trial, total cholesterol decreased 8.6% and 10.25% in males and females, respectively, but then increased in the same level of initial stage for both male and female respondents after 2 months of not consuming tempe formula. The LDL cholesterol levels decreased 12% in male and 9.67% in female respondents and then increased 9% in males and 15.5% in females after 2 months of not consuming the tempe formula. Lipid peroxidation which is expressed as MDA decreased 23% for both male and female respondents and then increased 13% in males and 15% in females after 2 months of not consuming the tempe formula. Uric acid level did not differ from baseline in the male group, but decreased about 14% in the female group, then increased in the same level of initial stage after 2 months of not consuming the tempe formula. Even though this was an uncontrolled study, these results are encouraging. The effect of tempe on SOD modulation was studied by Astuti, by using 45 copper-deficient male Wistar rats which were divided into five groups of nine rats and were fed with diets of different tempe concentrations (0, 25, 50, 75 and 100%, respectively) for 45 days. Copper is known as an important trace mineral acting as a cofactor of SOD. The activities of SOD and lipid peroxidation were evaluated from the serum. The highest inhibition of SOD against lipid peroxidation and the lowest level of MDA were found in rats on the 100% tempe diet as a source of protein and copper.<sup>28</sup> Copper as a component of SOD plays a dual role in SOD activity, as a cofactor as well as regulator.<sup>29,30</sup>

# Menopausal symptoms

There are studies reporting a lower incidence of menopausal symptoms in Asian populations consuming high levels of soy, such as Japan, China, Korea and Indonesia. These oestrogenic compounds may play an important role in the prevention of menopausal symptoms.<sup>31,32</sup> Trials to date have not been properly designed to determine whether these compounds act similarly to oestrogen in alleviating menopausal symptoms. There is no epidemiological data specifically on menopausal disorders in populations with a very high intake of tempe.

#### Possible role of tempe in cancer prevention

Recently, attention has also focused on the potential role of soybean products in reducing cancer risk. Asian countries have among the lowest rates of common cancers in Western society such as breast, prostate and colon cancer.<sup>33</sup> The protective effect of a diet high in soy may partly explain it. An epidemiological study on colorectal cancer in Japan found that frequent consumption of soybeans and tofu markedly decreased both rectal and colon cancer risk.34 Kiriakidis *et al.* demonstrated that tempe, especially its glucolipids, inhibits the proliferation of tumor cell in mice.<sup>35</sup> Indonesians are known as the largest soybean-consumers, especially in the form of tempe and tofu, in the South-East Asian countries. However, epidemiological studies relating to tempe consumption and the prevalence of cancer, particularly in Indonesia, have not yet been conducted.<sup>36</sup>

#### Conclusion

Diet as a part of lifestyle plays an important role in maintaining nutrition and health. Tempe is considered as a good source of protein, vitamin B12, antioxidants, phytochemical and other bioactive substances.

Numerous studies to date strongly indicate that soybean-based tempe offers positive nutritional and health benefits. However, the recommendation of tempe consumption should be based on and supported by scientific experiments which show that tempe has indeed specific beneficial effects in human health. Continued multidisciplinary scientific research will provide a better understanding and further knowledge on the identification of the beneficial components and mechanisms of action, function, nutritional and health aspects of tempe. Furthermore, contribution from nutrition and the food-science community from all over the world to develop tempe from a variety of legumes as a raw material that are nutritious, tasty, acceptable and affordable will help us meet the challenge of health for all towards the 21st century.

#### Necessity to develop the standards

Indonesia is the biggest tempe consumers and producers which annually around 1.5 million ton of soybeans have been processed into 2.42 million ton of tempe. Tempe production is scattered through out the country and it is possible to distribute regionally since it already consumed in other countries such as Malaysia, Singapore and Japan. Therefore, the development of Codex standard is needed to assure its safety and quality.

Indonesia is of the opinion that the scope of the tempe products will cover a wide range of products, for which some provisions such as food additives should be different, and that many of certain products were consumed only in a few countries or weer significantly traded. Indonesia proposed to exclude tempe products from the scope of the standard and the Coordinating Committee should focus to develop Proposed Draft Codex Regional Standard for Tempe.

#### Recommendation

Indonesia invites the Coordinating Committee to support the proposal to develop the Codex Regional Standard for Tempe and to consider the attached project document (Annex).

Indonesia noted that the 33rd Session of the Codex Alimentarius Commission has adopted a proposal of the Executive Committee (ALINORM 10/33/3 Appendix II) on amendment to the *Criteria for the establishment of work priorities* and inclusion of new *Guidelines on the application of the Criteria for the establishment of work priorities applicable to commodities*. We would like to suggest the Coordinating Committee to consider the new guidelines in considering the project document.

Indonesia also invites the Coordinating Committee to provide some additional information such as volume of production and consumption, and trade volume to complete the project document before submitting to Executive Committee for critical review.

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# Annex

# **PROJECT DOCUMENT**

# PROPOSAL FOR UNDERTAKEN NEW WORK ON THE STANDARD FOR TEMPE

# 1. The purpose and scope of the Standard for tempe

The purpose is to establish a regional standard for tempe that provides essential guidance related to food safety and quality for protecting the health of consumers and ensuring fair practices in food trade. This standard would cover tempe, a specific product prepared by the fermentation of soybeans by the mold of *Rhizopus* sp and intended for human consumption.

# 2. Relevance and timeliness

Tempe is originally from Indonesia and currently is produced and consumed in other countries such as Malaysia, Singapore and Japan. The health benefit of tempe is well known, resulting in increasing tempe consumption globally. Codex regional standard for this product will protect consumers and ensure fair trade practices by harmonizing quality and safety requirements at regional level.

# 3. The main aspects to be covered

This standard will cover essential quality and safety aspects.

# 4. Assessment against Criteria for Establishment of Work Priorities

a. Volume of production and consumption in individual countries and volume and pattern of trade between countries

No	Year	Soybean for tempe (MT)
1	2006	1,442,000
2	2007	1,514,000
3	2008	1,362,000
4	2009	1,512,000

Total soybean used for tempe are described below:

Source: ASA IM (American Soybean Association International Marketing) and Indonesian Tempe Forum

By assuming that 1 kg soybean can be processed to yield 1.6 kg tempe, the total amount of tempe production are described below:

No	Year	Production of tempe (MT)
1	2006	2,307,200
2	2007	2,424,200
3	2008	2,179,200
4	2009	2,419,200

It is around 500-700 ton per year, which is approximately valued US\$ 1-1.4 million was traded abroad such as to Malaysia and Singapore.

b. Diversification of national legislations and apparent resultant or potential impediments to international trade.

Does not apply

c. International or regional market potential

Considering the shifting of the consumption pattern towards healthier life, tempe as one of vegetable protein source has high opportunity to be traded regionally. The increasing of vegan and vegetarian

make a chance for tempe to be a good food for their consumption. It is estimated that 10 % of world population are vegetarian.

d. Amenability of the commodity to standardisation.

Soybean and *Rhizopus* sp have always been the most important raw materials for making tempe. As soybean tempe is the most popular, the word tempe usually refers to soybean-tempe. The local name of tempe can be used for international and regional publications, as there are no common English names available. The characteristic of tempe are described below:

- Physical: white color, surface covered internally by mold mycelia, compact and soft. If it is cut with a knife, it will results in sharp edge cake like (not disintegrated)
- organoleptic: slightly beany flavor, strong tempe characteristic (aroma of mold mycelia)
- Microbiology: luxury mold growth no yellow spot. Black spot may appear on uncovered part
- Increasing soluble protein, sugar and FFA but decreasing phytic acid
- e. Coverage of the main consumer protection and trade issues by existing or proposed general standards

A specific codex standard for this product is needed to avoid fraudulent practices and to protect consumers' health by meeting the safety and quality requirements for the product.

f. Number of commodities which would need separate standards indicating whether raw, semiprocessed or processed

Scope of the tempe and tempe products will cover a wide range of products, for which some provisions such as food additives should be different, and that many of certain products were consumed only in a few countries or weer significantly traded. This standard will exclude tempe products from the scope and focus to develop standard for tempe.

g. Work already undertaken by other international organization in this field and/or suggested by the relevant international intergovernmental body(ies).

This new work does not duplicate work undertaken by other international organizations.

# 5. Relevance to Codex strategic objectives

This proposal is relevant to goal 1.2 of the Codex Alimentarius Strategic Plan 2008 - 2013 relating to the review and development of Codex Standards and related texts for food quality by the commodity committees and the regional coordinating committees.

# 6. Information on the relation between the proposal and other existing Codex documents.

The proposal will take into account Recommended International Code of Practice General Principles of Food Hygiene (CAC/RCP 1-1969) rev 3 1997) and relevance Recommended International Code for Hygienic Practice, Codex General Standard for the Labeling of Prepackaged Foods (Codex Stan 1-1985. Rev 3-1999) and Codex General Guidelines on Sampling (CAC/GL 50-2004).

# 7. Identification of any requirement for and availability of expert scientific advice

None identified

# 8. Identification of any need for technical input to the standard from external bodies

None identified

# 9. Proposed time-line for completion of the new work, including the start date, the proposed date for adoption at Step 5, and the proposed date for adoption by the Commission.

It is expected that the work could be completely done within the remaining time frame of 5 years. If the proposed new work is recommended by the 17th Session of Codex Coordinating Committee for Asia (November 2010) and adopted as a new work by the 34th Session of the Codex Alimentarius Commission (CAC) in July 2011, a proposed draft standard would be presented to the next session of CCASIA (2012) for consideration at Step 4. The proposed draft standard is expected to be adopted by the CAC at step 5 in 2013 and step 8 in 2015.