



A systematic, comparative study on the beneficial health components and antioxidant activities of commercially fermented soy products marketed in China



Lu Xu^a, Bin Du^b, Baojun Xu^{a,*}

^a Food Science and Technology Program, Beijing Normal University-Hong Kong Baptist University United International College, Zhuhai, Guangdong 519085, China

^b Analysis and Testing Center, Hebei Normal University of Science and Technology, Qinhuangdao, Hebei 066600, China

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Malonylglycitin (PubChem CID: 23724657)

Malonylgenistin (PubChem CID: 25245762)

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Glycitein (PubChem CID: 5317750)

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ABSTRACT

The objectives of this study are to systematically assess the bioactive substances and overall antioxidant capacities of commercially fermented soy products and to find the relationships between the presence of beneficial components in different types of soybean fermented products. The results show that phenolic profiles increased significantly after fermentation as compared with raw yellow soybeans. Among all the samples, the douchi and fermented black bean sauce had the highest detected antioxidant profiles. Even though the total isoflavone content was reduced in fermented soybean products (794.84 µg/g on average) as compared with raw yellow soybeans (3477.6 µg/g), there was an obvious trend of conversion of the glucoside form in raw soybeans into the aglycone-form isoflavones in the fermented soybean products. The highest daidzein and genistein values were found in the “Yangfan” black bean douchi, i.e. 860.3 µg/g and 1025.9 µg/g, respectively. The amounts of essential amino acids also were improved in most fermented soybean products. The douchi and black bean fermented products are recommended for consumption due to their abundant bioactive substances.

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1. Introduction

Soybeans and their derived food products play significant roles in the current food market. Long term cultivation and consumption make soybeans well accepted as functional foods with high contents of amino acids, isoflavones, vitamins, minerals, and proteins, which could serve as a great substitute for meat proteins (Kwak, Lee, & Park, 2007; Wood, 1998). Large numbers of studies have investigated the bioactive compounds in soybeans. It has long been recognised that the large groups of phenolic derivatives and flavonoids in soybeans have overall beneficial effects on breast and

prostate cancer inhibition (Messina, Persky, Setchell, & Barnes, 1994; Yin, Li, Li, & Saito, 2004), cardiovascular diseases, and osteoporosis risk reduction (Ishida et al., 1998). Epidemiologic studies also confirm the health benefits of soy products. The results show that the high level of soy food consumption leads to low incidence of some hormone-dependent diseases among people living in Asian countries (Adlercreutz et al., 1992).

Even though soybeans have a high nutritional value, raw soybeans with simple boiling preparation have low bioavailability. In this condition, fermentation has been widely applied in the food industry to improve texture and mouthfeel by removing undesirable flavours with soaking and heat treatments together with adding food spices. After fermentation, large molecules have been broken down into small molecules; thus, they can be easily digested. With the improvement of sensory characteristics and

* Corresponding author at: 28, Jinfeng Road, Tangjiawan, Zhuhai, Guangdong Province 519085, China. Tel.: +86 756 3620636; fax: +86 756 3620882.

E-mail address: baojunxu@uic.edu.hk (B. Xu).

certain nutritious components, fermentation has become a beneficial and safe method in product manufacturing. There are various commercially fermented products currently available in markets. For example, China is well known for its douchi, stinky tofu, and sufu; natto and miso are a daily necessity for Japanese families; tempeh is consumed by Indonesians; and Cheonggukjang and Doenjang are consumed by Koreans. These are famous and popular commercially fermented soybean products; they act as widely popular flavourings in Asian diets (Kwon, Daily, Kim, & Park, 2010).

Improvement of antioxidant profiles in soy products after fermentation has sparked great attention. Apart from the texture improvements, the significant increases of antioxidant substances found in fermented soy products have been reported (Huberta, Bergerb, Nepveuc, Paula, & Daydéb, 2008; Kwak et al., 2007; Ping, Shih, Rong, & King, 2012). Raw soybeans are rich in glucoside-form isoflavones, polypeptides, and saponins; these components in raw soy materials are called “primary physiologically active ingredients.” Some new antioxidants or antioxidant substances with less content in raw soybeans can be generated or enhanced during fermentation. These substances, such as aglycone-form isoflavones generated by hydrolytic action of enzymes during food processing, are called “secondary physiologically active ingredients” (Liao et al., 2012). After the fermentation of soybeans, the contents of the secondary physiologically active ingredients are increased, especially the aglycone-form isoflavones and free amino acids. There is an increasing interest in finding the contents and composition changes of bioactive substances with fermentation in order to find the relationships between antioxidants and consequences of relevant diseases.

Among all the bioactive components, isoflavones play important roles in the inhibition of osteoporosis and cardiovascular diseases and the alleviation of women's menopausal symptoms (Yin et al., 2004). Great beneficial properties, such as antimutagenic and anticancer cell proliferative activities of isoflavones are also found (Song, Barua, Buseman, & Murphy, 1998). In general, isoflavones can be divided into aglycone, 7-O- β -glucoside, 6''-O-malonylglucoside, and 6''-O-acetylglucoside forms (Kudou et al., 1991). Studies show that many fermented soybean products have high antioxidant activities, including natto in Japan and Taiwan, tempeh in Indonesia, chungkookjang in Korea, as well as douchi in China. Daidzein and genistein as aglycone-form soy isoflavones were assumed to contribute to beneficial health effects (Ping et al., 2012). It has been reported that the aglycone-form of isoflavones contribute to the significant part of biologic activities because they can be easily absorbed in a greater amount as compared with other glycoside forms (Cheng, Lin, & Liu, 2011).

Soybeans are a rich source of protein with not only great quantity but also good quality. Free amino acids are protein hydrolysates, which also have important beneficial health properties. For example, soy amino acids help to reduce blood cholesterol and coronary heart diseases (Anderson, Johnstone, & Cook-Newell, 1995). Effects on antimutagenicity and antiobesity of free amino acids also were found (Allison et al., 2003; Lee, Park, Jung, Park, & Kim, 2005). Amino acids, such as tyrosine, methionine, histidine, and lysine, are considered to have antioxidant properties (Saito et al., 2003). During fermentation, proteolytic degradation is a major reaction to release more free amino acids in the products, which greatly improves the nutritional values of fermented soy foods (Dajanta, Apichartsrangkoon, Chukeatirote, & Frazier, 2011).

Extensive studies have already been carried out to investigate the phenolic substances in various raw soybeans, including black and yellow soybeans (Xu & Chang, 2008), and also targeted on specific fermented soybean products, such as chungkookjang (Kwak et al., 2007), natto (Kuo, Cheng, & Wu, 2006), and douchi (Liu et al., 2009). However, there is still a lack of an overall investigation into antioxidant ability and identification and quantification of bio-

active components from diverse, commercially fermented soy products. The objectives of this study are to systematically assess the phenolic compounds and bioactive substances to obtain the overall antioxidant capacities of fermented soybean products. Specifically, this study focuses on isoflavone profiles and free amino acids to find the relationships between the generation and distribution of beneficial ingredients in different types of fermented products. An overall measurement to identify and quantify the antioxidants and their activities in fermented soybean products will be established in this study. Findings from the present study are expected to provide consumers with suggestions to choose commercially fermented soybean products marketed in China.

2. Materials and methods

2.1. Commercially fermented soybean products

Twenty-seven fermented soy products along with one raw yellow soybean sample were randomly selected from local or online markets. Sample information is shown in Table 1. The samples cover a wide range of fermented soy products, which include five kinds of douchi, three kinds of natto, two kinds of miso, four kinds of sufu, two kinds of black bean sauce, two kinds of fried yellow soybean sauce, one stinky tofu, and one Huizhou mouldy tofu. The remaining seven samples were yellow soybean paste processed by different methods. The production areas of these products were also carefully selected to cover a broad range in domestic Chinese markets, such as Liaoning, Beijing, Shandong, Hunan, Guangdong, Chongqing, Anhui, and Zhejiang. The country of origin of doenjang and fried bean sauce is Korea. The country of origin of “Ba Ding” miso, rice miso, and “Mito” natto is Japan. The wide range of sample collection provides reliable reference values on commercially fermented soy products. A sample of raw yellow soybeans was also tested at the same conditions as a reference.

2.2. Chemicals and reagents

Phenylisothiocyanate (PITC) was purchased from Aladdin Industrial (Shanghai, China). Two external isoflavone standards, malonyldaidzin (MD) and malonylgenistin (MG), were provided by Wako Pure Chemical Industries, Ltd. (Osaka, Japan). Six of the isoflavone standards (daidzin, glycitin, genistin, daidzein, glycitein, and genistein), 2,4,6-tris (2-pyridyl)-1,3,5-triazine, 2-diohenyl-1-picrylhydrazyl (DPPH), catechin, gallic acid, and Folin-Ciocalteu reagent were purchased from Shanghai Yuanye Biological Technology Co., Ltd. (Shanghai, China). A mixed standard of 16 amino acids was purchased from Agela Technologies Co., Ltd. (Tianjin, China). Acetonitrile (of HPLC grade) and methanol (HPLC grade), triethylamine, dimethylsulfoxide, fluorescein disodium, and Trolox were obtained from Sigma-Aldrich (St. Louis, MO, U.S.A.). Sodium carbonate was provided by Nuoke Technology Development Co., Ltd. (Tianjin, China). The 2,4,4-trihydroxybenzoic acid (THB) was synthesised by the corresponding author Dr. Xu. All chemicals were of analytical grade unless specially stated.

2.3. Extraction of total phenolics

Fermented soy products underwent freeze-drying by a freeze-dryer (FreeZone Benchtop, Labconco Corporation, Kansas City, MO, U.S.A.) and then were finely ground by a grinding machine and screened through a mesh size of 420 μ m. The sample of raw yellow soybeans was directly ground without freeze-drying. The ground flours were subjected to extraction, and the extraction procedure was carried out according to our previous study (Xu &

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