

## SHORT COMMUNICATION

# Tempeh Waste as a Natural, Economical Carbon and Nutrient Source: ED-XRF and NCS Study

SITI KHODIJAH CHAERUN

*School of Life Sciences and Technology, Bandung Institute of Technology, Ganesha 10, Bandung 40132, Indonesia*

*Phone: +62-22-2511575, Fax: +62-22-2534107, E-mail: [skchaerun@sith.itb.ac.id](mailto:skchaerun@sith.itb.ac.id), [skchaerun@gmail.com](mailto:skchaerun@gmail.com)*

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The purpose of this study was to determine the elemental composition of three types of waste from tempeh production. They are soybean hull “tempeh waste” after dehulling soybeans, tempeh wastewater after soaking dehulled soybeans in water for 24 h, and tempeh wastewater after boiling dehulled soybeans in water for 30 min. By using ED-XRF analyzer, it was revealed that tempeh waste contained Mg, Si, P, S, K, Ca, Mn, Fe, and Zn. The highest elemental content was K, followed by Ca, P, and Mg. NCS analysis showed that tempeh waste was composed of C, N, and S with C/N ratio of 11.20. The present study provides evidence that both tempeh waste and wastewater are rich in carbon and nutrient contents, thus their potential for both inorganic and organic nutrient and carbon sources for microbial growth in bioremediation or as natural NPK fertilizers is promising.

Key words: bioremediation, NPK fertilizer, tempeh wastewater, tempeh waste

### INTRODUCTION

Tempeh is a traditional Indonesian fermented soybean food that is gaining acceptance elsewhere due to its low-cost, superior nutritive qualities and metabolic regulatory functions (van der Riet *et al.* 1987; Matsumoto & Imai 1990; Baumann *et al.* 1991; Friedman & Brandon 2001). Studies on organic characteristics of tempeh (such as protein, lipids, vitamin, phytate, isoflavone, and carbohydrates) have been published elsewhere (Kirk *et al.* 1999; Murphy *et al.* 2002; Cuevas-Rodriguez *et al.* 2004; Nakajima *et al.* 2005; Eklund-Jonsson *et al.* 2006; Azeke *et al.* 2007; Angulo-Bejarano *et al.* 2008). The microbiological quality of the water used for soybean cooking ( $1.9 \times 10^4$  CFU/g) for tempeh production has also been evaluated (Ashenafi 1994). However, the entire elemental composition of both tempeh and tempeh waste is poorly documented.

Tempeh in Indonesia is usually produced by fermenting soybeans (i.e., one of the most important grains of legume family) with *Rhizopus oligosporus*, but other pulses or solid substrates such as barley kernels (Feng *et al.* 2005; Feng *et al.* 2007) and chickpeas (Reyes-Moreno *et al.* 2000) can be used as well. Correspondingly, one of the major problems of tempeh industries in Indonesia is the direct disposal of tempeh processing wastewaters to aquatic ecosystems, therefore posing significant environmental problems. Since interest is rising in developing countries in low-cost, beneficial materials, as replacements for or supplements to the expensive and frequently scarce materials, the tempeh wastewater should deserve considerable attention, and hence the exploration of novel uses for tempeh wastewater is required to convert it into a renewable natural resource prone to sustainable use.

The purpose of this study was to determine the entire elemental composition of three kinds of waste from tempeh production, such as soybean hull “tempeh waste” after dehulling soybeans, tempeh wastewater after soaking dehulled soybeans in water for 24 h, and tempeh wastewater after boiling dehulled soybeans in water for 30 min by using ED-XRF (Energy Dispersive-X-Ray-Fluorescence) and NCS analyzers.

### MATERIALS AND METHODS

Samples of tempeh wastewater/waste were taken from tempeh home industry located in Rancaekek, Bandung Regency, West Java Province, Indonesia. They were collected from three different processing units of tempeh production: soybean hull “tempeh waste” after dehulling soybeans (TWA), tempeh wastewater after soaking dehulled soybeans in water for 24 h (TWB), and tempeh wastewater after boiling dehulled soybeans in water for 30 minutes (TWC) (Figure 1). Elemental contents of tempeh waste and wastewater were estimated using the ED-XRF (Energy Dispersive-X-Ray-Fluorescence) and NCS analyzer. Measurement was conducted in triplicate. Three replicates of each sample were prepared, and the elemental content was estimated in all replicates. For measurement of elemental compositions in TWA, the samples were dried to constant weight at room temperature, homogenized in an Agate mortar, made into thin pellets, mounted on Mylar films, and analyzed by an energy dispersive X-ray fluorescence spectroscopy (ED-XRF; JEOL JSX-3201), using Rh-K $\alpha$  generated at 30 kV under vacuum. For measurement of elemental compositions in TWB and TWC, 400  $\mu$ l of solution was mounted on Mylar films, and

allowed to evaporate overnight, then analyzed by an energy dispersive X-ray fluorescence spectroscopy (ED-XRF; JEOL JSX-3201), using Rh-K $\alpha$  generated at 30 kV under vacuum. In addition, the elemental composition of N (nitrogen), C (carbon), and S (sulphur) in the wastes was also determined by an automatic gas chromatographic elemental analyzer (CE Instruments NA 2500-NCS) at 1000 °C with 20 ml oxygen.

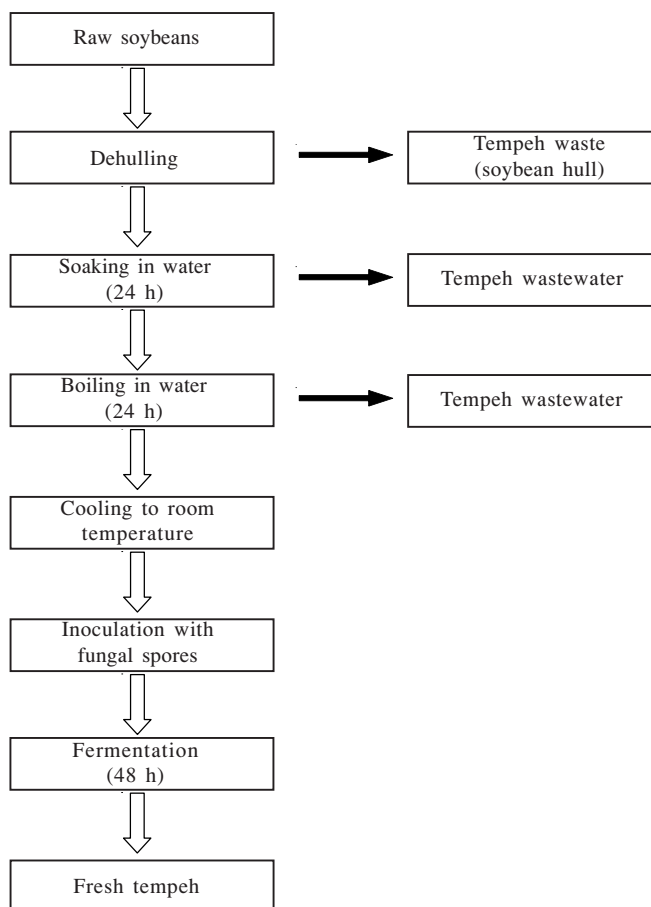


Figure 1. Flow chart of tempeh production from which tempeh wastewater/waste is discharged (Azeke *et al.* 2007 with modification).

## RESULTS

ED-XRF analysis revealed that both tempeh wastewater and waste (TWA, TWB, TWC) contained elements of Mg, Si, P, S, K, Ca, Mn, Fe, and Zn, except for TWC which did not contain Mn (Table 1). TWA contained the following elemental order: K>Ca>S>P>Fe>Si>Mg>Mn>Zn. TWB comprised the following elemental order: K>Ca>Mg>P>S>Si>Fe>Mn>Zn, while TWC was composed of the elements in the following order: K>Ca>P>Mg>S>Fe>Si>Zn. The highest elemental content in all samples was K, followed by Ca, suggesting that all processes discharge the waste in which K and Ca elements predominate. Both TWA and TWB contained lowest elemental concentration of Zn, while TWC contained no detectable Mn.

Interestingly, TWC contained higher concentrations of K than TWA and TWB, indicating that boiling process releases highest concentrations of K element compared to other processes. Moreover, both soaking and boiling processes yielded wastewater that was composed of elements of Mg, Si, P, S, Ca, Fe, and Zn, although their concentrations were lower than those of TWA. In contrast, Mg concentration of TWB and TWC is greater than that of TWA. NCS analysis showed that tempeh waste (TWA) was composed of C ( $45.96 \pm 0.10$  wt.%), N ( $4.11 \pm 0.02$  wt.%) and S ( $0.03 \pm 0.02$  wt.%) with C/N ratio of  $11.20 \pm 0.04$ . Both ED-XRF and NCS analyses could detect sulphur element.

## DISCUSSION

By using ED-XRF and NCS analyses, the entire elemental composition of a material or a substance in weight percentage (wt.%) could be detected quickly and easily (Table 1). Thus, the ED-XRF and NCS study provide evidence that three successive processes of traditional tempeh production yielded both tempeh waste and wastewater that are rich in carbon and nutrient contents, particularly in C, N, Mg, P, K, and Ca. From this study, it is advisable to reuse both tempeh wastewater and waste, but not to discharge them into aquatic environment, since the discharge of such wastewater

Table 1. Elemental composition of tempeh wastewater/waste collected from three process units of tempeh production analyzed by ED-XRF (Energy Dispersive-X-Ray-Fluorescence) and NCS analyzer

| Elemental composition (wt.%) | Tempeh wastewater/waste                     |   |   |
|------------------------------|---|---|---|
|                              | Tempeh waste after dehulling soybeans (TWA) | Tempeh wastewater after soaking dehulled soybeans in water for 24 h (TWB) | Tempeh wastewater after boiling dehulled soybeans in water for 30 min (TWC) |
| Mg                           | 0.95 (0.02)                                 | 6.10 (0.03)   | 3.22 (0.06)   |
| Si                           | 2.05 (0.01)                                 | 0.81 (0.01)   | 0.41 (0.01)   |
| P                            | 6.27 (0.05)                                 | 4.60 (0.14)   | 3.25 (0.04)   |
| S                            | 6.30 (0.03)                                 | 1.37 (0.04)   | 1.83 (0.01)   |
| K                            | 49.51 (0.12)                                | 74.65 (0.21)  | 86.16 (0.25)  |
| Ca                           | 28.89 (0.26)                                | 12.11 (0.03)  | 3.46 (0.13)   |
| Mn                           | 0.56 (0.05)                                 | 0.10 (0.00)   | n.d.  |
| Fe                           | 5.11 (0.15)                                 | 0.20 (0.00)   | 1.62 (0.04)   |
| Zn                           | 0.36 (0.01)                                 | 0.05 (0.00)   | 0.05 (0.00)   |
| C                            | 45.96 (0.10)                                | -   | -   |
| N                            | 4.11 (0.02)                                 | -   | -   |
| S                            | 0.03 (0.02)                                 | -   | -   |
| C/N                          | 11.20 (0.04)                                | -   | -   |

Values were calculated as means of triplicate samples (n = 3) with standard error in parentheses. n.d.: not detected, -: not measured.

containing high organic content without prior treatment is known to adversely affect the aquatic life and water potability (Ferreira *et al.* 2005; Lefebvre & Moletta 2006); the biodegradation of organic compounds such as organic-rich tempeh wastewaters usually generates odorous compounds and dissolved oxygen depletion as well as releases organic acids, thus resulting in the pH decrease of the ambient aquatic ecosystem (Ewies *et al.* 1998).

Owing to the high carbon and nutrient contents of both tempeh wastewater and waste, their use as both carbon and nutrient sources for microbial growth in bioremediation and natural fertilizer might be economically advantageous. Due to their high contents of Mg and Ca, they can also be applied to saline-sodic soil remediation (Nelson & Oades 1998; Tejada *et al.* 2006; Ansari 2008). From an environmental point of view, soils have proved to be used as a system of assimilating, recycling or disposing off wastewaters or sewage sludge, where the interaction of the effluent with a soil-plant system can be used as a means of carrying out the final removal nutrients and organics from the liquid effluent (Di Bonito 2008). Thus, the disposal of tempeh wastewaters/wastes, therefore, presents economic advantages other than environmental problems. To our knowledge, this is the first report showing the entire elemental composition of tempeh wastewaters/wastes from traditional tempeh production.

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