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FULL PAPER

The equivalence test of functional properties and sensory characteristics of transgenic and nontransgenic soybean-based soy flour

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Abstract

In Indonesia, approximately 70% of soybeans are imported to fulfill domestic demands. Most of the imported soybeans are transgenic soybeans or Genetically Modified Organisms (GMO). The soybeans are judged to have a better grain quality, are relatively uniform in grain size and colour and are more readily available. One of the local soybeans in Indonesia having the potential to compete with the imported soybean quality is Grobogan soybean. One of the processed soybean products having good potential to be developed is soy flour. This study was conducted to determine the effect of the soybean variety types (imported transgenic, imported nontransgenic and local grobogan) on functional properties and sensory characteristics within the produced flour. The soy flour production process began with the soybean washing and soaking phase, followed by boiling, dehulling, drying, milling using a pin disc mill and sifting using a size 60 mesh. The soy flour was subjected to analyses of the functional properties and sensory characteristics. The local grobogan soy flour has a water absorption capacity (2.66 g/g), oil absorption capacity (0.91 g/g), foaming capacity (107.14%), emulsion capacity (100%) and PDI (11.63%) equal to those of the imported soy flour (transgenic and nontransgenic). The local grobogan soy flour is even superior in the foam stability and emulsion stability parameters. The local grobogan soy flour also has a more preferable score in all of the sensory attributes tested (colour, texture, aroma and overall) than those of imported soy flour (both transgenic and nontransgenic), even though they were not significantly different statistically, which is as good as the flour made from imported soybeans.

1. Introduction

Soybeans (*Glycine max* L.) are one of the field crops most commonly cultivated by Indonesian farmers. The average demand for soybeans in Indonesia is 2.9 million tons/year with a soybean consumption of 6.43 kg/capita/year and it is projected that these numbers will continue to rise in the period 2020–2024 (Pusdatin, 2020). Indonesian soybean production is only approximately 424 thousand tons/year in 2019. These days, most of the imported soybeans (up to 93.2% of the demand) originate from the US, Argentina and Brazil which are the main transgenic or Genetically Modified Organism (GMO) soybean producers (Mantalean, 2022).

Imported soybeans are judged to have better grain quality, relatively uniform grain size and colour, and are more readily available (Ginting and Tarmizi, 2021; Sutawi, 2022). Indonesia has several local soybean varieties. One of which is the Grobogan variety which is known to have the potential to compete with the quality of imported soybeans in terms of colour, grain size, protein content, fat content, antioxidant capacity and consumer preference level (Wresdiyati *et al*., 2018; Wresdiyati *et al*., 2021).

Soybeans are usually traded and used in the form of whole grains as raw ingredients in various products such as tempe, tofu, soy sauce, tauco and soymilk (Amar *et al*., 2021; Ginting and Tarmizi, 2021; Kamble *et al*., 2021; Romulo and Surya, 2021). Soybeans can also be traded in the form of flour. Soybean in the form of flour is more easily stored and has a relatively longer shelf life compared to its fresh form because it has lower moisture content (Astawan *et al*., 2020a). Soy flour can be used as

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a wheat flour substitute in the production of various foods, such as bread, cakes, drinks and weaning foods (Aboge *et al*., 2021; Filipini *et al*., 2021; Mai *et al*., 2021).

The soy flour utilization in various foodstuff products is related to its fairly good functional properties, such as its water absorption capacity, oil absorption capacity, foaming capacity and stability, emulsion capacity and stability, gel strength, protein dispersibility index (PDI) and nitrogen solubility index (NSI). Soy flour also has fairly good sensory characteristics, making it readily acceptable to consumers.

To date, no studies have reported the comparison of functional properties and sensory characteristics of flour made from imported transgenic soybeans, nontransgenic imported soybeans, and local grobogan soybeans (nontransgenic Indonesian soybeans). This study was designed to measure the equivalence of the three types of soy flour by testing their protein's functional properties and sensory characteristics.

2. Materials and methods

2.1 Materials

The main material used in this study was local soybeans from the grobogan variety obtained from farmers in Grobogan, Central Java, Indonesia, transgenic imported soybeans (SWAN brand USA soybeans) and nontransgenic imported soybeans (SB&B Foods, SB90 Lot No. RF 17046A34) obtained from Indonesian Tempe and Tofu Producer Cooperative (KOPTI) Bogor.

2.2 Soy flour production

Soy flour was produced based on the method described by Omosebi and Otunola (2013) with slight modifications. The soy flour production began with selecting the soybean grains manually to discard any damaged, spoiled and unacceptable soybeans and to remove any impurities, such as twigs, pieces of gravel and corn grains. The soybeans were then weighed, washed with clean water three times and soaked in water for 8 hrs (1:5 w/v). The soybeans were then boiled for 30 min, shelled, split into two pieces using a dehuller (KOPTI Bogor, Indonesia) and dehulled. The boiled, dehulled soybeans were then dried using a cabinet dryer (Pilot Plant Engineering and Equipment GmbH 6072 Dreieich, Germany) at 60°C for 6 hrs until the moisture content was <12% in accordance with the National Oilseed Processors Association standard (Van Eys, 2011). The dried soybeans were then ground using a pin disc mill having a 60-mesh sieve attached. The soy flour obtained was then wrapped and stored in a refrigerator (4^oC) for functional property and sensory characteristic analyses. The production of soy flour was conducted in two batches and each analysis was in triplicate.

2.3 Moisture content analysis and yield

The moisture content analysis was conducted using the AOAC (2019) method. The yield analysis was done by comparing the weight of the produced soy flour and the weight of the soybean grains used multiplied by 100%.

2.4 The functional properties analysis

The soy flour functional properties were analysed for their water absorption capacity by measuring the weight of additional water bound by flour after centrifugation (Ikegwu *et al*., 2021), oil absorption capacity by measuring the weight of additional oil bound by flour (Raigar and Mishra, 2021), foaming capacity by measuring the percentage increase in volume of flour suspension after stirring (Kambabazi *et al*., 2022), foaming stability was determined as a function of time after 2 hrs by monitoring the fall in the foaming capacity value (Biswal *et al*., 2021), emulsion capacity by measuring the ratio between emulsified volume of oil–flour suspension and total volume of mixture (Ayo-Omogie *et al*., 2021), emulsion stability by measuring the volume of oil–flour suspension after 0.5, 1, 2, 4 and 6 hrs after mixing (Tafadzwa *et al*., 2021), protein dispersibility index by measuring the ratio between the protein content in the supernatant after extraction of soy flour and the protein content in total material (Cai *et al*., 2021) and nitrogen solubility index by measuring the ratio between the nitrogen content in the supernatant after extraction of soy flour and the nitrogen content in total material (Singh and Koksel, 2021).

2.5 Sensory analysis

The sensory analysis was conducted according to the BSN method (2006). The analysis was conducted to determine the consumers' acceptance of the soy flour samples with the help of a questionnaire. The sensory test was done with the help of seventy untrained panelists aged 18 – 25 years old. The samples were prepared as homogeneous as possible and then served one by one randomly to the panelists to be scored without comparing them to other samples. The tests used were the acceptance tests (hedonic rating) of the aroma, colour, texture and overall attributes, each using a scale of 1 = very unacceptable, 2 = unacceptable, 3 = neutral, 4 = acceptable, and 5 = very acceptable.

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2.6 Experimental design and statistical analysis of the data

The statistical analysis of the moisture content, yield and functional properties of the soy flour was conducted using a Completely Randomized Design. The statistical analysis of the sensory characteristics was conducted using a Completely Randomized Group Design. The results of the soy flour functional properties and sensory characteristics analyses were processed using the SPSS 20.0 program with an analysis of variance (ANOVA). If the ANOVA showed a significant difference ($p<0.05$), it would be followed with Duncan's Multiple Range Test (DMRT).

3. Results and discussion

3.1 Moisture content and yield

The material's moisture content is one of the important parameters in determining the shelf life of a food product (Villanueva and Trindade, 2010). All the soy flour produced fulfilled the maximum moisture content standard, which is a maximum of 12% (Van Eys, 2011). The soybean variety treatment had a significant effect ($p<0.05$) on the soy flour moisture content (Table 1). The flour made from local grobogan soybeans had a moisture content which was significantly lower ($p<0.05$) than flour made from imported soybeans (both transgenic and nontransgenic). Therefore, it could be assumed that flour from the local grobogan soybeans would have a longer shelf life in similar storage conditions. The difference in moisture content was caused by differences in soybean handling, drying, storage and distribution by the suppliers. The soaking and boiling steps in the soy flour production process also affected the moisture content due to the difference in water penetration to the seed matrix and differences in seed expansion (Astawan *et al*., 2020b).

Table 1. Moisture content and soy flour yield.

Soy flour based on the variety	Moisture (%)	Yield (%)
Transgenic imported soybeans	4.3±0.1^b	58.3±0.6^b
Nontransgenic imported soybeans	4.5±0.1^b	51.3±1.1^a
Local grobogan soybeans	3.8±0.3^a	54.5±0.5^{ab}

Values are presented as mean±SD (n = 3). Values with different superscripts within the same column are statistically significantly different ($p<0.05$).

The soy flour yield was calculated by comparing the weight of the flour produced and the weight of the dry soybeans used as the raw ingredient. The soybean variety treatment had a significant effect ($p<0.05$) on the soy flour yield (Table 1). The flour yield of nontransgenic imported soybeans was significantly lower ($p<0.05$) than that of the transgenic imported soybeans but was not significantly different from that of the local grobogan soybeans. It may be caused by the higher moisture content of nontransgenic and local grobogan soybeans, compared to transgenic soybeans (Astawan *et al*., 2013), which can affect on decreasing the yield after soybean drying process to flour. The flour yield from transgenic imported soybeans was also not significantly different from that of the local grobogan soybeans. Hence, the yield of flour made from local grobogan soybeans was equivalent to flour from both transgenic and nontransgenic imported soybeans.

3.2 Soy flour functional properties

3.2.1 Water absorption capacity

Water absorption capacity has an important role in the food system because it could affect other functional properties and sensory characteristics. The application of flour as the main ingredient for foods mostly depends on its interaction with water (Sreerama *et al*., 2012). The treatment of different soybean varieties did not have a significant effect ($p>0.05$) on the water absorption capacity. The water absorption capacity of soy flour ranged from 2.6 – 2.7 g/g flour (Table 2), which is better than the water absorption capacity of kidney beans (2.2 g/g flour), adzuki beans (2.6 g/g flour), cranberry beans (2.4 g/g flour) and black beans (2.2 g/g flour) (Siddiq *et al*., 2010).

A higher water absorption capacity indicates a greater quantity of polar amino acid residues in a certain protein, causing the flour to have a higher affinity to water molecules (Kambabazi *et al*., 2022). The soy flours produced in this study had good water absorption capacities, giving them the potential to produce moist and soft products and making them suitable for use in thick products, such as soups, dough, custards and bakery products (Sreerama *et al*., 2012).

Table 2. Functional properties of soybean flour.

Soy flour based on the variety	Water absorption (g/g)	Oil absorption (g/g)	Foaming capacity (%)	Emulsion capacity (%)	PDI (%)	NSI (%)
Transgenic imported	2.6±0.3	0.8±0.1	110.7±7.1	100.0±0.0	16.8±0.8	11.8±1.4^b
Nontransgenic imported	2.6±0.2	0.8±0.1	103.6±7.1	100.0±0.0	14.8±3.7	12.9±0.6^b
Local grobogan	2.7±0.2	0.9±0.0	107.1±8.2	100.0±0.0	11.6±1.7	9.5±0.3^a

Values are presented as mean±SD (n = 3). Values with different superscripts within the same column are statistically significantly different ($p<0.05$). PDI;: Protein dispersibility index, NSI: Nitrogen solubility index

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3.2.2 Oil absorption capacity

Oil absorption capacity is an important parameter in developing a new food product. Oil absorption capacity is also important in maintaining flavour and preventing oxidative rancidity in the product during storage. The oil-retaining characteristic is closely related to the protein content and protein types contained in the foodstuff (Raigar *et al*., 2021). The soybean variety treatment did not have a significant effect (p>0.05) on the oil absorption capacity. The oil absorption capacity of the soy flour in this study ranged between 0.8 and 0.9 g/g flour (Table 2), which was still lower than that of flours made from kidney beans (1.5 g/g), adzuki beans (1.2 g/g), cranberry beans (1.5 g/g) and black beans (1.3 g/g) (Siddiq *et al*., 2010).

The oil absorption of a product depends on the availability of hydrophobic amino acids on the surface (Benítez *et al*., 2013). Besides being affected by the availability of hydrophobic amino acids, oil absorption capacity is also influenced by amylose and amylopectin contents. Flour with a lower oil absorption capacity is suitable for making crispy fried products which are fried at a high temperature, approximately 180°C (Vongsawasdi *et al*., 2008).

3.2.3 Foaming capacity and stability

Foaming characteristics depend on the interfacial protein layers which are formed to trap air bubbles in a suspension and to reduce the coalescence rate. Foaming characteristics strongly depend on the type of proteins and other components, such as carbohydrates (Sreerama *et al*., 2012). The soybean variety treatment did not have a significant effect (p>0.05) on the foaming capacity (Table 2). Good foaming characteristics are an important parameter in the formulation of bakery products (Siddiq *et al*., 2010). The foaming capacity of the soy flour produced in this study ranged between 103.6 and 110.7%, which was higher than winged-bean protein isolate (89.5%) (Budijanto and Sitanggang, 2011), wheat flour (4.1%) (Adeleke and Odedeji, 2010), sweet potato flour (1.3%) (Adeleke and Odedeji, 2010) and germinated-fermented sorghum flour (9.0%) (Elkhalifa and Bernhardt, 2018).

In addition to the foaming capacity, another parameter which is also essential in a protein-source flour is foam stability. The foam stability is influenced not only by the protein content of a certain material but also by its proteins' characteristics (Sreerama *et al*., 2012). The foam stability of the flour made from local grobogan soybeans was more stable than that of the flour made from imported soybeans (both transgenic and nontransgenic) as presented in Figure 1. It may be caused by the higher protein content of local grobogan soybean flour among others (Astawan *et al*., 2020b). Good foam stability indicates that the proteins and other components in a certain material have a better ability in forming a strong and cohesive layer around air bubbles and have good resistance against diffusion of air. Materials that have good foam stability can be applied in baked products because they can constantly trap air during baking. This mechanism helps optimize the rising in a baked product because it can trap air well during baking (Sreerama *et al*., 2012).

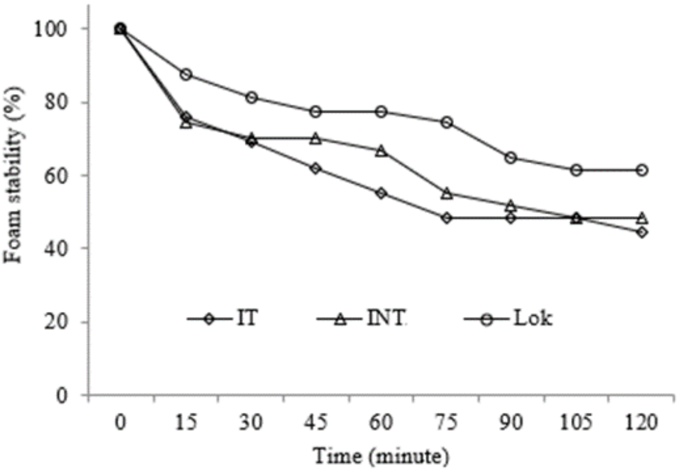

Figure 1. Foam stability of soy flour. IT: transgenic imported soybeans, INT: nontransgenic imported soybeans, Lok: local grobogan soybeans (n = 3).

3.2.4 Emulsion capacity and stability

Emulsion capacity is the ability of the proteins in a certain material to form an emulsion and absorb the interfacial area between water and oil in the emulsion system. The emulsion capacity of soy flour is strongly influenced by its protein content. The soybean variety treatment did not have a significant effect (p>0.05) on the emulsion capacity. All of the soy flour types resulted in a 100% emulsion capacity, indicating that all the oil, air and soy flour mixed perfectly (Table 2).

Good emulsion characteristics are an important parameter in developing the formulation of bakery products (Siddiq *et al*., 2010). Emulsion characteristics are also important in meat products, such as bologna and sausages (Astawan *et al*., 2020b; Goemaere *et al*., 2021). The emulsion system in soy flour is believed to be the oil in water type of emulsion. The ratio between water absorption to oil in the transgenic imported, nontransgenic imported and local grobogan soy flours were 3.12, 3.18 and 2.92, respectively. These ratios were obtained from the ratio between the water absorption capacity and the oil absorption capacity. The higher the ratio between water and oil absorption, the better the ability to emulsify oil in water. A ratio greater than 2.0 indicates that the hydrophilic side is more dominant than

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the hydrophobic side (Twinomuhwezi *et al*., 2020).

In addition to the emulsion capacity, the emulsion stability parameter is also needed in various food products. Emulsion stability is the ability of proteins to maintain the integrity of the emulsion system against interfacial forces and changes (Sreerama *et al*., 2012). There is a steep decline in emulsion stability from 0 min to 15th min. After 15 mins, the emulsion stability declines gradually. The local grobogan soy flour's emulsion stability was better than that of the imported soy flours (both transgenic and nontransgenic) as presented in Figure 2. All the soy flours in this study are predicted to be suitable for use as an emulsifier in products that have the oil in water emulsion type. From Figure 2, it is apparent that the emulsion of the local grobogan soy flour lasted longer and thus predicted to be suitable for soups and cakes.

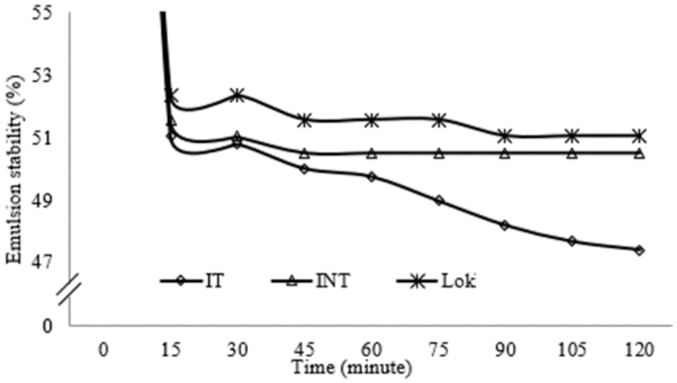

Figure 2. Emulsion stability of soy flour. IT: transgenic imported soybeans, INT: nontransgenic imported soybeans, Lok: local grobogan soybeans (n = 3).

3.2.5 Protein dispersibility index and nitrogen solubility index indexes

The Protein Dispersibility Index (PDI) and Nitrogen Solubility Index (NSI) are useful parameters in determining the characteristics of soy flour and in measuring how much protein is not denaturized. The two parameters are also important to determine soy flour's acceptance for protein isolation. The PDI is given in percentage to determine the protein dispersion rate in water. The soybean variety treatment did not have a significant effect ($p>0.05$) on the PDI.

The PDI range was 11.6–16.8% (Table 2). The PDI determines the use of soybean proteins and their functional properties. The flour from transgenic imported soybeans could be applied in products, such as crackers and cereals because it had a PDI >15%, matching the standard requirements (Riaz, 2006). The other soy flours were predicted to be suitable for use as a substitute for various flours to enrich the protein content.

The use or addition of soy flour in a food product could improve the protein content, other nutrients and functional properties. The addition of 10% soy flour to yam flour in Nigeria demonstrated an increase in protein and other nutrients (Olu *et al*., 2013). The PDI of all the types of soy flours in this study has fulfilled the Fulmer standard range (10-25%), which is suitable for applications in baby foods, protein drinks, meat products with fillers, soups, gravies and HVP (Deak *et al.,* 2008).

The NSI is given in percentage to determine the amount of nitrogen dissolved in water. The soybean variety treatment had a significant effect ($p<0.05$) on the NSI. Flour from local grobogan soybeans had the lowest NSI (Table 2). This was because of the small amount of protein and nitrogen dissolved in water due to the protein denaturation process. Protein denaturation causes the formation of hydrophobic amino acid residues which are not water-soluble (Moure *et al*., 2006). The non-protein nitrogen (NPN) content in a material also influences the NSI. A lower NPN would result in a greater NSI. In the NPN measurement of many legumes, it was discovered that field peas had a lower NSI because they had a higher NPN than other legumes (Singh *et al.,* 2000).

The NSI could be used as a reference in the use of soy flour in food formulation. The flour from imported soybeans (both transgenic and nontransgenic) had an NSI range of 10–20%, making it suitable for use in crackers, drinks, cookies, cereals and baby foods according to the classification by Johnson (Shurtleff and Aoyagi, 2020). The flour from local grobogan soybeans had an NSI of less than 10%, so it did not qualify due to the range requirement but can be used as a protein enhancement for many other flours.

3.3 Sensory characteristics

Acceptance testing is important to be conducted to determine how far the consumers would accept the flour product if it is to be marketed. The scoring of the colour includes the flour colour (creamy), texture scoring includes roughness or fineness and clumping, while flavour testing includes the "beany", bean-like flavour produced by the soy flour.

The requirements for a good soy flour are that it has creamy ivory to light brown colour, tasteless and has a uniform texture with fine particles (20 mesh) (Roosendaal, 2011). The soybean variety treatment did not have a significant effect ($p>0.05$) on the acceptance of the colour, texture, flavour and overall acceptance attributes of the soy flours as presented in Table 3.

Heat treatment, the duration of heating and heating temperature influence the change in soy flour colour due to the Maillard reaction (Martínez *et al*., 2013). The

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texture is an important aspect of scoring a food product. The flavour can be formed by volatile compounds in a food product. One of the causes of changes in the flavour of soybeans is the activity of lipoxygenase enzyme which oxidizes fats and produces a beany flavour. This beany flavour is a flavour which is unfavorable in soybean products and is a parameter in the acceptance of a processed soybean product.

The scoring of the overall acceptance attributes is used to determine the acceptance of all the sample's attributes. In general, local grobogan soy flour tended to have a more preferable score in all of the sensory attributes tested (colour, texture, flavour and overall acceptance) than those of imported soy flour (both transgenic and nontransgenic), even though they were not significantly different statistically.

Table 3. Sensory characteristics of soy flour.

Soy flour based on the variety	Colour	Texture	Flavour	Overall
Transgenic imported	3.8	3.3	3.7	3.6
Nontransgenic imported	3.8	3.2	3.7	3.5
Local grobogan	3.9	3.4	3.7	3.7

No letters indicate no-significant differences among samples within the same column ($p>0.05$) ($n = 70$).

4. Conclusion

The quality of the flour made from local grobogan soybeans was equivalent to that of flours made from imported soybeans (both transgenic and nontransgenic) in the functional properties of water absorption capacity, oil absorption capacity, foaming capacity, emulsion capacity and PDI attributes, but was lower in the NSI attribute. Flour from local grobogan soybeans was also superior in the foam stability and emulsion stability parameters. The acceptance score of flour made from local grobogan soybeans was equivalent to that of flour made from imported soybeans (both transgenic and nontransgenic) in all the tested sensory attributes (colour, texture, flavour and overall acceptance).

Conflict of interest

The authors declare no conflict of interest.

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