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The effect of *Moringa oleifera L.* leaf extract supplementation on the chemical and sensory quality of beef patties cooked to a well-done level

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ABSTRACT: This study aimed to evaluate the effect of *Moringa oleifera L.* leaf extract supplementation on the chemical composition and sensory quality of beef patties cooked to a well-done level. This study used a completely randomized design (CRD) in one direction with five treatments and five replications. The research treatments included the supplementation of *Moringa oleifera L.* leaf flour extract (MOE) 0% (P0), 1% (P1), 2% (P2), 3% (P3), and 4% (P4). The variables observed included water content, fat, protein, collagen, and sensory attributes. A 15-cm line scale was used to evaluate the intensity of the attributes tested on beef patties by 30 semi-trained panelists. Chemical and organoleptic quality test data were analyzed using Analysis of Variance. If the treatments had a significant effect, then the analysis was continued using the Tukey Test. The results showed that the addition of MOE had a very significant effect (P<0.05) on the chemical quality of beef patties (water, fat, and protein content). The use of MOE at the 2% level produces higher protein and fat levels than the use of 4%. Adding MOE up to 4% can increase the intensity of beef patties' color, aroma, and tenderness (P<0.05). The supplementation of MOE at the 2% level can produce beef patties with relatively high protein and fat content and can maintain sensory quality.

Keywords*:* *Moringa oleifera L.*; Beef patties; Chemical quality; Sensory quality

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INTRODUCTIONS

Fresh meat is a perishable food that is very important because of its chemical composition, which is rich in protein, omega-3 polyunsaturated fats, vitamins, and minerals. Meat also contains various endogenous antioxidants and other bioactive substances, including carnitine, taurine, carnosine, ubiquinone, and creatine (Elsharawy et al., 2019). This meat's chemical components vary according to animal species, age, breed, sex, feed, and body weight. Generally, meat contains 70-78% water, 17.4-21.1% protein, 2.74-6% fat, and 1.04-1.87% ash (Alamin, 2019). Because of its high nutritional value, meat is a very favorable substrate for the growth of microorganisms. Also, meat is susceptible to spoilage due to chemical and enzymatic activity (Dave and Ghaly, 2011). Therefore, meat must be processed immediately after slaughter to reduce damage and extend the shelf life of meat.

Meat processing aims to increase food diversity, preserve and retain original nutritional values, increase shelf life (Amit et al., 2017), increase the nutritional value of meat, and promote food choices for improved health. One popular processed meat product and a central part of hamburger products is patties. Patties are generally made using beef, chicken, or pork as the main ingredient. Patties processing can increase and improve the nutritional quality of the product (Daiyan et al., 2020). Beef patties are generally sold as fast food, which tends to be unhealthy, so it is necessary to add ingredients that can increase the nutrition of beef patties, for example, natural extracts that are high in antioxidants (Valencia et al., 2008). Meat and processed meat products could contain harmful elements to human health under certain circumstances and in inappropriate proportions (Jiménez-Colmenero et al., 2001). Several methods can achieve healthier meat and processed meat products. Two methods can be applied, namely avoiding unwanted substances or reducing these substances to the limit and increasing levels (naturally or by programmed addition) of other substances with beneficial properties (Arihara, 2006). Adding ingredients such as vegetable oils and various natural extracts with antioxidant content to raw meat or processed products can increase its functional properties (Valencia et al., 2008).

Moringa (*Moringa oleifera L.*) leaves are currently known as a "functional food" because of their unique nutritional profile and potential as an alternative for health purposes (Razis et al., 2014). Moringa leaves are a plant that contains high levels of phytochemicals. Moringa leaves grow widely in tropical areas, including Indonesia. It has significant nutritional characteristics, including high levels of protein, fiber, minerals, and vitamins, as well as bioactive nutrients such as antioxidants (ascorbic acid, flavonoids, phenolics, and carotenoids), which are beneficial for health (Trigo et al., 2023). Thus, it can be used to improve the quality of meat products. Using moringa leaves in processed meat products is one way to improve meat quality from a physical, chemical, microbiological, and sensory perspective. Moringa leaves contain very high macronutrients (González-Burgos, 2021). Moringa leaf extract can neutralize free radical compounds and inhibit rancidity in processed meat products (Sánchez-Machado et al., 2010). However, moringa leaf extract could change meat's sensory quality if used excessively (Apriantini et al., 2022). Usually, factors that can influence meat quality are changes in its chemical components: protein, water, fat, and ash (Elsharawy et al., 2019).

Previous research has highlighted the positive impact from incorporating *Moringa oleifera L.* on improving meat quality. For instance, Hazra et al. (2012) found that adding 1.5% *Moringa oleifera L.* extracts reduced water-holding capacity and lower cooking loss. Moringa within the meat matrix was also documented to provide protection against cellular damage and inhibition from pro-oxidants, demonstrating strong antioxidative properties (Cui et al., 2018; Fakurazi et al., 2008; Hazra et al., 2012). Despite the health benefits and nutritional value, using *Moringa oleifera L.* seed flour as a binding agent in beef patty production may decrease sensory levels, excluding emotionality (Al-Juhaimi et al., 2016). Adding *Moringa oleifera L.* leaf flour (MOE) to goat meat patties, on the other hand, can increase protein, fat, and ash content. At a 0.6% level, it doesn't alter taste and color but enhances juiciness and tenderness. However, exertion at 0.9% level was proven to reduce taste, color, and overall preference (Khomola et al., 2021).

Chemical changes in meat can occur due to molecular interactions during the addition of food additives, heat treatment application, or prolonged storage. These changes, such as denaturation, hydrolysis, and gelation of proteins, can impact organoleptic characteristics and nutritional value, influencing consumer acceptance and a balanced diet (Diéguez et al., 2010). Consequently, ensuring food safety and meeting consumer demands without compromising the nutritional value of meat products requires advanced meat processing technology. According to the backgrounds mentioned earlier, this study aims to investigate beef patties' chemical and organoleptic quality containing MOE at different levels, prepared with well-done cooking. The goal is to develop beef patty products with favorable chemical quality that meets consumer acceptance.

MATERIALS AND METHODS

Design and material preparation

The research design in this study used a completely randomized design (CRD) with a unidirectional pattern. This research used MOE. The nutritional values of MOE included protein content (3.96%), lipid content (0.08%), ash content (0.95%), total phenolic (541 mg/100 g), and antioxidant activity (65.59% wb). The treatments included four different levels of MOE, consisting of MOE 0% (P1), MOE 1% (P2), MOE 2% (P3), MOE 3% (P4), and MOE 4% (P5) cooked to a well-done level. The flour and seasonings used in making beef patties are food-grade commercial products in the form of MOE, tapioca flour, ice cubes, chicken eggs, beef stock powder, salt, sugar, garlic powder, stabilizers, and ground pepper. The composition of the ingredients used in this research treatment was the same except for the level of moringa leaf flour extract that was added. The elemental ingredient composition consists of beef (81.5%), ice (7.0%), whole egg (5.0%), stabilizer (0.5%), spices (3%), and tapioca flour (3%)

Beef patties processing

The process for making beef patties is as follows: weigh all the ingredients and spices used and mix the ground beef, moringa leaf flour extract, and spices using a mixer for 35 seconds. The dough is molded with a weight of 60 g and a thickness of 1 cm, and then the patties are cooked to a well-done level using a grill pan with an internal temperature of around 72-80°C for 7-8 minutes (Rosli et al., 2011). Some modifications include using electric tools like a 1-mm grinder and a rotary combination mash system mixer.

Chemical quality testing of beef patties

The beef patties samples were wrapped in aluminum foil and put in a plastic clip, then stored in the freezer at 20°C until testing for chemical quality and sensory properties. Testing the chemical quality of beef patties includes testing water, fat, protein, and collagen content using the FOSS FoodScan Meat Analyzer (NIRs spectroscopy) with duplo testing of each treatment sample. Beef patties were weighed at 50 g, ground with a meat grinder, placed in sample cups, and then put into the scanner. The results that came out were recorded (AOAC, 2007).

Panelist selection

The selection was conducted by recruiting 60 students from the UNS Faculty of Animal Husbandry. Prospective panelists were then given a questionnaire to collect information regarding the consumption of beef patties. The questionnaire data was then analyzed based on scoring to select 30 semi-trained panelists. Selected panelists will be given a 'willingness as a panelist' form and information regarding how to conduct QDA organoleptic testing of research products.

Preparation and organoleptic evaluation

The organoleptic testing method was performed according to Kartikasari et al. (2020) with several modifications. The plastic container containing the beef patties was removed from the freezer and moved to the refrigerator 24 hours before testing. After the beef patties were at room temperature, they were cooked using a grill pan for 4 minutes on each side and then divided into four parts. The patties were warmed at 35-40°C using a bain marie for 10 minutes. After that, the sample was placed in an aluminum cup coded with three random numbers (3-digit numbers) to be tested by the panelists. The organoleptic testing of beef patties started by explaining the procedure to the panelists. The panelists were then given a consent form and asked to sign the form. The panelists were given an assessment sheet regarding the characteristics of beef patties using a 15-cm line scale. The panelists marked a line from low to high intensity according to their assessment of the attribute being tested. The attributes evaluated in the organoleptic test are color, aroma, tenderness, texture, juiciness, taste, flavor, and aftertaste. The panelists were given one minute between two samples to drink water and consume plain crackers to neutralize the taste.

Data analysis

This research used analysis of variance (ANOVA) to determine the effect of using *Moringa oleifera L.* leaf flour extract cooked to well-doneness on beef patties' chemical and organoleptic quality. If there was a significant effect on the treatments, the test was continued with the Tukey Test with a significant difference of $P<0.05$.

RESULTS AND DISCUSSION

The results of the chemical quality analysis of beef patties supplemented with MOE at different levels are presented in Table 1.

Table 1. Values of water, fat, protein, and collagen content of beef patties supplemented with *Moringa oleifera L.* leaf flour extract at different levels

Parameter (%)	Treatment (mean ± standard deviation)					*P value*
	MOE 0% (P1)	MOE 1% (P2)	MOE 2% (P3)	MOE 3% (P4)	MOE 4% (P5)	
Water	66.53±1.836^{a}	65.99±1.070^{ab}	65.51±1.723^{ab}	68.20±1.842^{a}	63.62±0.386^{b}	0,002
Lipid	3.53±0.155^{ab}	4.09±0.550^{a}	4.18±0.292^{a}	4.08±0.607^{ab}	3.28±0.351^{b}	0,010
Protein	24.22±0.356^{a}	24.19±0.523^{a}	24.75±0.469^{a}	23.89±0.568^{ab}	23.04±0.354^{b}	0,001
Collagen	2.88±0.238	2.83±0.257	2.71±0.331	2.38±0.354	2.76±0.208	0,084

Values with different superscripts on the same row indicate significant differences between treatments with P<0.05. P1 (without *Moringa oleifera L.*), P2 (1%), P3 (2%), P4 (3%), P5 (4%), supplementation with *Moringa oleifera L.* flour extract cooked to a well-done level.

Water content

The analysis revealed a highly significant impact of incorporating MOE on the water content of beef patties ($P<0.01$). The inclusion of MOE led to a notable reduction in the water content of the patties, reaching the lowest level at 4% concentration (P5). This decrease in water content at the 4% level is likely attributed to the interaction of soluble fiber with the water in the beef patties. Dissolved solids, known to contribute to decreased water content, play a role in influencing the water content of patties (Mashau et al., 2021b).

Adding moringa leaf extract resulted in low water binding in the meat matrix (Aleson-Carbonell et al., 2005). This occurs because the water in the meat matrix is not tightly bound; instead, it binds to hydrophilic molecules such as proteins, carbohydrates, pectin, and starch through intermolecular hydrogen bonds (Kusnandar, 2019). These findings align with that of Mashau et al. (2021a), which observed a reduction in the water content of mutton patties by adding moringa leaf extract.

The water content of beef patties in this study ranged from 63.6% to 68.4%, which is relatively higher than the maximum water content specified in the standard quality criteria for meat burgers (SNI 01-6683-2002), set at 60%. Adding plant flour to meat products increases the fiber and protein content, which can contribute to higher water content (Guevara et al., 2020). This may explain the elevated water content in beef burgers compared to SNI standards, as noted in studies by Carvalho et al. (2019) (65.55-71.24%), Bahnyar et al. (2020) (61.54-70.82%), and Nadia et al. (2018) (66.37-72.72%).

Lipid content

The results showed that adding *Moringa oleifera L.* leaf flour extract significantly affected the fat content of beef patties (P<0.01). Further tests showed that the addition of *Moringa oleifera L.* leaf flour extract at levels of 1% (P2) and 2% (P3) resulted in a higher fat content compared to the 4% treatment (P5). Adding moringa leaf flour extract at the 4% level (P5) reduced the fat content. The increase in fat content in beef patties with MOE to a level of 2% was probably due to the sample having high-fat retention. Fat retention in meat is a complex phenomenon resulting from several chemical and physical mechanisms. High-fat retention is caused by the ingredients' expansion of starch and fiber and the fat's interaction in the patties with denatured proteins. According to Alkali et al. (2010), protein denaturation in soluble fiber during cooking can expose non-polar residues inside the protein molecule, increasing the fat-binding sites. The results of this study are similar to the research of Al-Juhaimi et al. (2018), who reported that adding argel (*Solenostemma argel*) leaf flour could increase fat retention and fat content in camel patties. The fat content of beef patties in this study ranged from 3.3-4.3%. Hazra et al. (2012) also reported a similar result, stating that the lipid content in the *Moringa oleifera L.* added ground meat was 9.60%. Similarly, study by Khomola et al. (2021) suggested the fat percentage of the *Moringa oleifera L.* added mutton patties was at 5.17-7.21% in raw condition, while it ranged between 12.10-18.32% after subjection to cooking. Further, this present study generated beef patties that meet the standard criteria for beef burger quality (SNI 8503:2018), namely a maximum fat content of 20% (BSN, 2018).

Protein and collagen content

The study demonstrated a significant impact on protein levels upon the addition of *Moringa oleifera L.* leaf flour extract (P<0.01). Subsequent examinations revealed that incorporating moringa leaf flour extract at the 2% level (P3) led to a relatively high protein content, whereas the protein content decreased at the 4% addition level (P5). No significant change in protein content was observed when *Moringa oleifera L.* leaf flour extract was added at the 3% level (P4). This lack of change could be attributed to the release of water-containing proteins from the patties during cooking, subsequently binding to the soluble and insoluble fiber in moringa leaf flour extract. Kusnandar (2019) elucidated that water in food quickly exits tissue capillaries and binds to materials containing hydrophilic molecules.

The result of this study confirms prior study by Al-Juhaimi et al. (2016), wherein protein content remained relatively stable in beef patties supplemented with moringa seed flour. However, in the present study, adding MOE extract at the 4% level (P4) decreased protein levels. This decline could be attributed to the denaturation of water-soluble moringa leaf extract. Higher water content facilitates protein denaturation, with increased water causing penetration and hydration in cavities on the protein's surface. Consequently, protein swelling occurs, reducing the denaturation temperature (Estiasih et al., 2022). This is consistent with findings that beef patties have a high water content. The protein content of beef patties in this study ranged from 23.12% to 24.62%, meeting the quality criteria for beef burgers (SNI 8503:2018), which mandates a minimum protein content of 8%.

The collagen content did not show variation across treatments, likely because *Moringa oleifera* L. leaf flour extract lacks collagen. As Soeparno (2015) outlined, collagen is a structural protein exclusive to meat connective tissue. This perspective is reinforced by Hall (2023), who notes that plants do not naturally produce collagen. Collagen is characterized by three polypeptides intertwining to form a triple helix, comprising the amino acids glycine, proline, and alanine (Safithri et al., 2019). Moringa leaves contain 1.5% glycine, 1.2% proline, and 3% alanine of the total amino acids (Moyo et al., 2011).

Sensory quality of beef patties

Table 2 presents the results of the quantitative and descriptive analysis (QDA) test conducted on beef patties enriched with *Moringa oleifera L.* leaf flour extract. The analysis indicated a significant impact (P<0.05) of moringa leaf flour extract on the color and aroma intensity of well-done beef patties (Figure 1). Specifically, the supplementation of moringa leaf flour extract at the 4% level notably enhanced beef patties' color, aroma, and tenderness intensity compared to the control group without the extract. However, there was no discernible increase in color, aroma, and tenderness intensity when comparing treatments with 1% (P2) to 4% (P5) moringa leaf extract. Panelists provided average scores for color and aroma intensity in the range of 7.96 to 9.41 and 8.21 to 9.91, respectively.

The heightened color intensity observed in beef patties can be attributed to the moringa leaf flour extract's elevated chlorophyll content. As the concentration of moringa leaf flour extract increases, so does the color intensity of the processed product (Figure 1). This aligns with the findings of Munthe (2022), who suggests that moringa leaves' chlorophyll content serves as a natural green coloring for processed products. The increased color intensity in beef patties supplemented with moringa leaf flour extract, as opposed to the group without its use, is consistent with the study by Apriantini et al. (2022), indicating that supplementation with moringa leaf extract up to 2% enhances the color intensity of ground beef and kofta meat. These results are further supported by Dianti et al. (2023), who assert that adding moringa leaf flour to crawfish-based nuggets increases color intensity.

Table 2. Average value of quantitative descriptive analysis for beef patties supplemented with *Moringa oleifera L.* leaf flour extract cooked to a well-done level

Parameter	Treatment (mean ± standard deviation)					*P-value*
	MOE 0% (P1)	MOE 1% (P2)	MOE 2% (P3)	MOE 3% (P4)	MOE 4% (P5)	
Color	7.96±2.070^{b}	9.19±1.976^{ab}	8.89±1.974^{ab}	8.51±2.007^{ab}	9.41±2.020^{a}	0,050
Aroma	8.21±1.954^{b}	8.99±2.284^{ab}	9.21±1.885^{ab}	8.85±2.027^{ab}	9.91±2.297^{a}	0,040
Tenderness	8.92±2.630^{ab}	8.18±2.790^{b}	9.44±2.333^{ab}	9.61±2.943^{ab}	10.18±2.612^{a}	0,052
Texture	7.05±2.611	7.86±2.931	7.62±2.440	7.35±2.946	6.36±2.858	0,262
Juiciness	7.29±2.089	6.96±2.240	7.90±1.592	7.70±2.022	7.76±2.124	0,357
Taste	8.80±2.222	8.82±1.933	9.17±2.060	8.81±2.177	8.80±2.437	0,953
Aftertaste	8.77±2.177	9.03 ±2.720	8.62±3.036	9.01±2.536	8.95±2.565	0,970
Flavor	8.72±1.884	8.83±2.385	9.39±1.735	9.44±1.812	9.44±2.278	0,452

$^{a.b}$ Different superscripts on the same row indicate significant differences P<0.05

The heightened aroma intensity observed in beef patties containing *Moringa oleifera* L. leaf flour extract may be attributed to the extract's distinct and pungent aroma, influenced by the presence of the lip oxidase enzyme (Mardiyah and Astuti, 2019). Furthermore, the aroma intensity could result from the evaporation of aromas from both the meat and the moringa extract during the cooking process, a phenomenon noted by Blackmon et al. (2015) associated with high-temperature cooking and aroma evaporation. This notion is also supported by Widyawatiningrum et al. (2018), who suggest that the aroma in nugget products after cooking is derived from the primary ingredients, namely meat and moringa leaves. The increase in aroma intensity in beef patties supplemented with moringa leaf flour extract, compared to those without the extract (P1), corresponds with the findings of Beti et al. (2020), who propose that the addition of moringa leaf extract at concentrations of 5%, 10%, and 15% to beef results in a distinctive moringa leaf aroma. This observation aligns with Mardiyah and Astuti's (2019) findings that higher concentrations of moringa leaves in nuggets produce a more intense moringa leaf aroma. The average tenderness panelists' scores for treatments P1-P5 ranged from 8.59 to 10.32, indicating a relatively high tenderness intensity in the beef patties. This tenderness is likely a result of the cooking temperature and method employed baking at 80°C to achieve a well-done level, as suggested by Jamhari et al. (2007). Lumbung et al. (2017) also propose that grilling methods can evaporate water, causing connective tissue to expand, ultimately enhancing meat tenderness and juiciness.

In this study, the intensity of texture, juiciness, taste, aftertaste, and flavor in beef patties supplemented with *Moringa oleifera L.* leaf flour extract did not show significant differences (P>0.05) (Table 2). The similar texture intensity may be attributed to the nearly identical percentage of moringa leaf flour extract in the supplemented beef patties and the absence of gluten in the MOE protein. Consequently, increasing the supplementation of moringa leaf flour extract does not significantly change texture intensity. This is in line with Gracia's (2009) perspective that the type of protein in a food product can influence its texture. Soeparno (2005) further explained that while moringa leaves have a high protein content, the absence of gluten, which contributes to texture formation, leads to relatively consistent texture in the product.

The taste intensity of beef patties supplemented with MOE remained consistent, as the moringa leaf extract imparts a bland taste, and the addition percentage was nearly uniform across treatments. This results in the predominant taste of the beef patty remaining unchanged. These findings support Krisnandi's (2013) assertion that moringa leaves have a bland taste and that increasing the use of moringa leaf flour extract does not significantly impact the taste intensity of beef patties. The study's supplementation with moringa leaf extract was minimal, reaching a maximum of 4%, in line with Widyawatinigrum's (2018) observation that smaller percentages of moringa leaves in nuggets lead to a savorier taste due to the meat content.

Similarly, the flavor of beef patties did not differ between treatments, as taste intensity is one of the flavor determinants. Since taste did not vary significantly, the flavor intensity remained constant. This is in accordance with the opinion of Winarno (2004) that the flavor of food products is influenced by taste and mouth stimulation. Additionally, the consistent use of spices in processing beef patties across treatments contributed to the uniform flavor intensity. Tinangon et al. (2014) emphasized that the use of spices generally enhances the flavor of processed food, and in this study, the consistent spice composition resulted in uniform flavor intensity. Soeparno (2005) also noted that employing the same spice composition leads to consistent flavor in processed meat products.

Furthermore, the flavor of a product is influenced by its ingredients, with Guichard (2007) reporting that the physical state of carbohydrates and the addition of fat can significantly impact flavor perception.

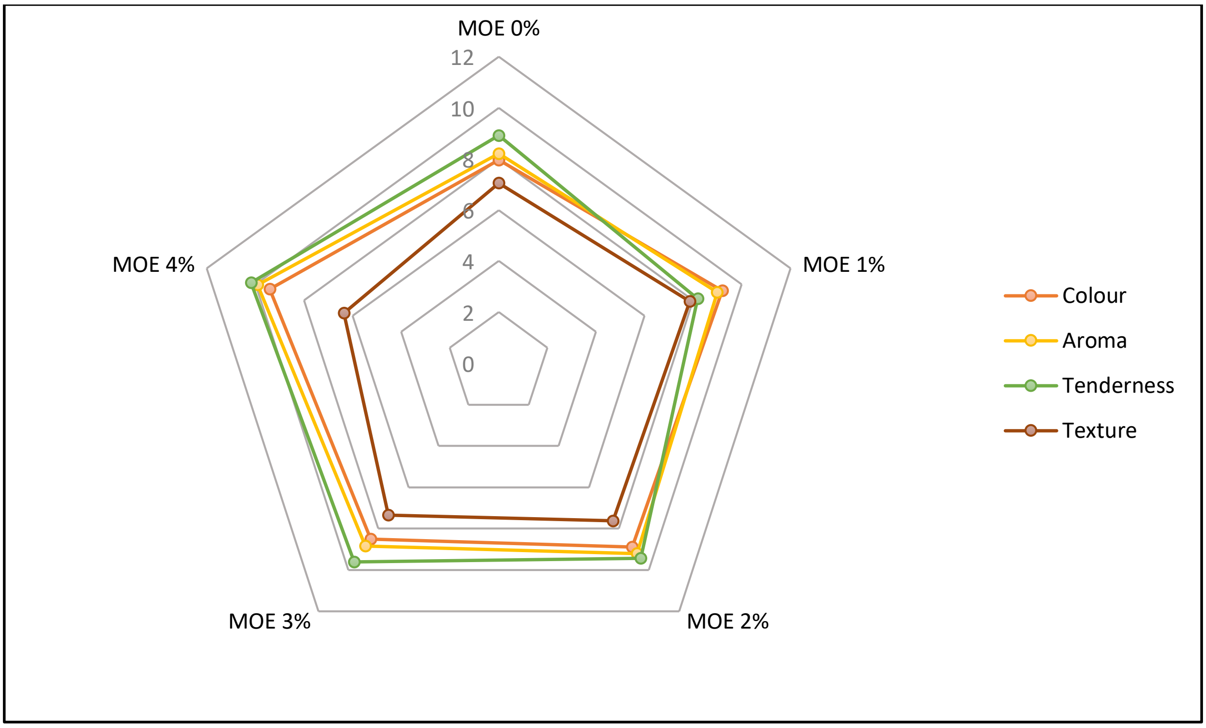

Figure 1. The effect of *Moringa oleifera L.* leaf extract (MOE) supplementation ranged from 1 to 4% on the sensory characteristics of beef patties cooked to a well-done level. The use of MOE at a level of 4% significantly increased color, aroma, tenderness, and texture ($P<0.05$).

CONCLUSION

Based on the results obtained, the use of *Moringa oleifera L.* leaf flour extract at the 2% level can be applied in the processing of beef patties by producing relatively high protein and fat content and can maintain sensory quality.

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