The Effect of the Use of Durian Seed Flour (*Durio zibethinus* Murr) as a Filler on the Physiochemical and Microbiology of Salami

S.M. Sembor[1], H. Liwe[1], N.N. Lontaan[1], D.R.B. Rumondor[1], N.L. Wuntu[1]

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ABSTRACT

Background: Durian (*Durio zibethinus* Murr) is a very popular fruit in Indonesia. People consume durian flesh because it has high nutritional value and delicious taste, while the durian skin and seeds are thrown away as waste. Durian seed flour contains quite a lot of starch, so it can be used as a filling ingredient for salami. The purpose of this study was to determine the physicochemical and microbiological quality of salami using durian seed flour as a filler.

Methods: The research has been successfully carried out using a completely randomized design with five treatments P0(0%), P1(5%), P2(10%), P3(15%), P4(20%) each treatment was repeated 4 times. Parameters measured include physical and chemical and microbiology. The data used by ANOVA and continued with the Tukey test.

Result: The research results show that the cooking losses decreased from 24.48%-8.31% and #39; Water Holding Capacity increased from 27.78%-51.97%; softness increases from 13.94-22.39 mm/g/10 seconds. While the chemical quality of the water content increased from 40.46%-44.84%; fat decreased from 19.95-13.75%; protein decreased from 24.90-20.74%; carbohydrates increased from 40.46-44.84%; microbiological such as pH 4.07-4.08; total plate count decreased from 3.57×10^3 CFU/gram - 3.038×10^3 CFU/gram. The study concluded that Durian seed flour cat a level of 15% could be used as a filler which improved the physicochemical and microbiological quality of salami.

Key words: Chicken laying hens, Durian seed flour, Salami.

INTRODUCTION

Durian (*Durio zibethinus* Murr) is a very popular is fruit in Indonesia and also called The King of fruits which is found in many tropical areas. According to Prasetyaningrum (2010), people usually consume durian flesh due to high nutritional value and tastes good, while the durian skin and seeds of durian are disposed of as waste. The percentage of the weight of the coated part of the fruit or the flesh is low, namely only 20-35%. The nutritional content of this fruit is carbohydrates, protein and vitamins B and C (Feng *et al.,* 2016). This fruits contain 51.1% water, 46.2% carbohydrates, 2.5% protein and 0.2% fat. The carbohydrate content is higher than cassava (34.7% carbohydrates) or sweet potatoes (27.9% carbohydrates). Durian seeds can be used as an alternative to processed food in the form of flour which can add information about nutrition to the community and create a clean environment (Setio *et al.*, 2013). Durian seed starch has similarities to tapioca starch such as starch content, namely amylose and amylopectin, so it can be combined with tapioca starch as a salami filler.

The use of durian seed flour as a mixture in food processing has not been widely used. To increase of using durian seeds as a food source, it is necessary to know the maximum limit for adding durian seed flour to the dough, so that it can produce quality products, even though they contain aspects of toxicity, especially the content of cyclopropene fatty acids and possibly HCN. Cyclopropene fatty acids can be anesthetic and affect the body's metabolism dizziness, thin body and reduced fertility. The polysaccharide content of durian seed extract can be used as an alternative source of food additives in the food industry (Bronikowska *et al.*, 2012; Ho and Bhat, 2015).

Durian seeds contain two main components, namely starch and gum which contain carbohydrates and protein (Amid and Mirhosseini, 2012). The filler material that is generally used in the manufacture of sausages is tapioca flour. Tapioca flour has a high level of elasticity and starch content (Melia *et al.,* 2010). Durian seed flour contains starch as high as tapioca flour. Tapioca flour has a starch content of around 90% (Reputra, 2009) and durian seed flour contains starch of 88.68% (Malini, 2016), so durian seed

[1]Faculty of Animal Science, Sam Ratulangi University, Manado 95115, Indonesia.

Corresponding Author: S.M. Sembor, Faculty of Animal Science, Sam Ratulangi University, Manado 95115, Indonesia.
Email: semborsofi@yahoo.com

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flour can be used as a filler for sausage dough. Salami (fermented sausage) is a processed meat product mixed with fat and meat through a fermentation process using lactic acid bacteria cultures such as *Lactobacillus acidophylus* and *Lactobacillus plantarum.* Based on the above background, a study has been carried out to examine the effect of waste-based durian seed starch (*Durio zibethinus* Murr) as filler in terms of physical, chemical and microbiological quality.

MATERIALS AND METHODS

The equipment used is Philips HR 7620 food, thermometer, harner, sleeve, hand stuffer, gloves, electric scale, knife and cutting board. The materials used include 96 weeks old Isa Brown culled laying hens, which were obtained from a livestock company in Tetey Village, Dimembe District, Minahasa Regency and durian seeds (*Durio zibethinus* Murr). Spices for making salami such as garlic, ginger, pepper, nutmeg, sugar and salt, flour, skim milk, fat, ice, or ice water. The starter cultures for the fermentation of *Lactobacillus plantarum* and *Lactobacillus acidophillus* were obtained from the Food and Nutrition Center, Gadjah Mada University, Yogyakarta.

The study was carried out using a completely randomized design (CRD) with 5 treatments and each treatment was repeated 4 times so that 20 treatment combinations were obtained (Steel and Torrie, 1995). The main ingredients in making salami consist of a chicken laying hens meat and fat in a ratio of 80:20. The meat and fat are ground together, then frozen for 24 hours. The ground and frozen meat are then again ground using a food processor along with spices, salt, sugar, garlic, ginger, pepper, nutmeg and starter cultures of *Lactobacillus plantarum* and *Lactobacillus acidophilus* in a ratio of 1: 1. As a treatment, durian seed flour (*Durio zibethinus* Murr) namely 0% (P0), 5% (P1), 10% (P2), 15% (P3) and 20% (P4) were incorporated as a filler. To make sausage dough skim milk, fat, ice or ice water and vegetable protein were added and mixed well. The dough was then put into a casing with a diameter of 30 mm, then tied at suitable distance which was then hung on a rack and conditioned for 24 hours at room temperature (Arief *et al.,* 2008). Salami underwent a conditioning process, then fermented for 6 days at room temperature. Fermentation was interspersed with a smoking process for 1 hour per day. The temperature during smoking was maintained at 30-35°C. The fuel used is dry coconut shells and the variables measured consist of physical qualities such as water holding capacity, cooking loss (Soeparno, 2005) and tenderness (Muchtadi and Sugiyono, 1992) and Chemical quality such as proximate analysis (the content of water, protein, fat, carbohydrates) (AOAC, 2005) as well as Microbiology measurements (pH and Total Plate Count) (Lukman and Trioso, 2009). The process of making salami is in the flow chart (Fig 1).

RESULTS AND DISCUSSION

Effect of treatment on cooking loss

The results of the research on the effect of treatment on the physical properties of salami using durian seed flour (*Durio zibethinus* Murr) on cooking loss (%), water holding capacity (%) and tenderness (%) are presented in Table 1.

The data in Table 1 shows that cooking loss decreased from 24.48% to 8.31% along with the increasing level of use of durian (0%, 5%, 10%, 15%, 20%,) thus the use of durian seed starch flour durian (*Durio zibethinus* Murr) affects the cooking losses of salami (fermented sausage) it is suspected that durian seed starch has the ability to bind water or the amount of water bound between the muscle fibers contained in the product salami causes cooking shrinkage to be small. Low cooking losses in a food product means good quality. If a food product has a cooking loss low means the product is of good quality. This is supported by Soeparno, (2005) that meat or processed meat products with low cooking losses have better quality than meat with high cooking losses because the loss of nutrients during cooking will be less.

Effect of treatment on water holding capacity

The increasing water holding capacity of the research results from 27.78%-51.97% indicates the effect of treatment using durian seed starch flour on salami binding free water, especially during the meat emulsion formation process because both can grow well in a medium with sufficient water content (Fardiaz, 1992). Soeparno (2005) defined WHC as the ability to bind water or added water during influence of external forces and affected by the state of meat protein, although only less than 5% of water binds directly to the hydrophilic group of meat protein (Bintoro, 2008). Furthermore, it is said that WHC, juices and texture are interrelated which is a determining factor for meat quality.

An increase in water holding capacity will be followed by an increase in tenderness. Besides being influenced by the addition of filler, the tenderness is also influenced by the water-holding capacity. The high water holding capacity results in a small amount of water being lost during the smoking process of the salami, resulting in a better texture and tenderness of the salami. Meat tenderness is determined by at least three meat components, namely myofibrils and their contraction status, connective tissue content and degree of cross-linking air binding capacity by meat protein and meat structural juices (Soeparno, 2005).

Effect of treatment on tenderness

The data from the analysis of tenderness on salami with the use of durian seed starch flour (*Durio zibethinus* Murr) gave a significant effect ($P<0.01$) on tenderness. There was an increase in the tenderness of salami P0 (13.94) mm/g/10 seconds (without durian seed meal) to P4 (20% durian seed meal) tenderness reaching 22.39 mm/g/10 seconds. This shows that the use of durian seed flour in salami process can increase tenderness because the increasing use of durian seed starch will cause more water to be bound so

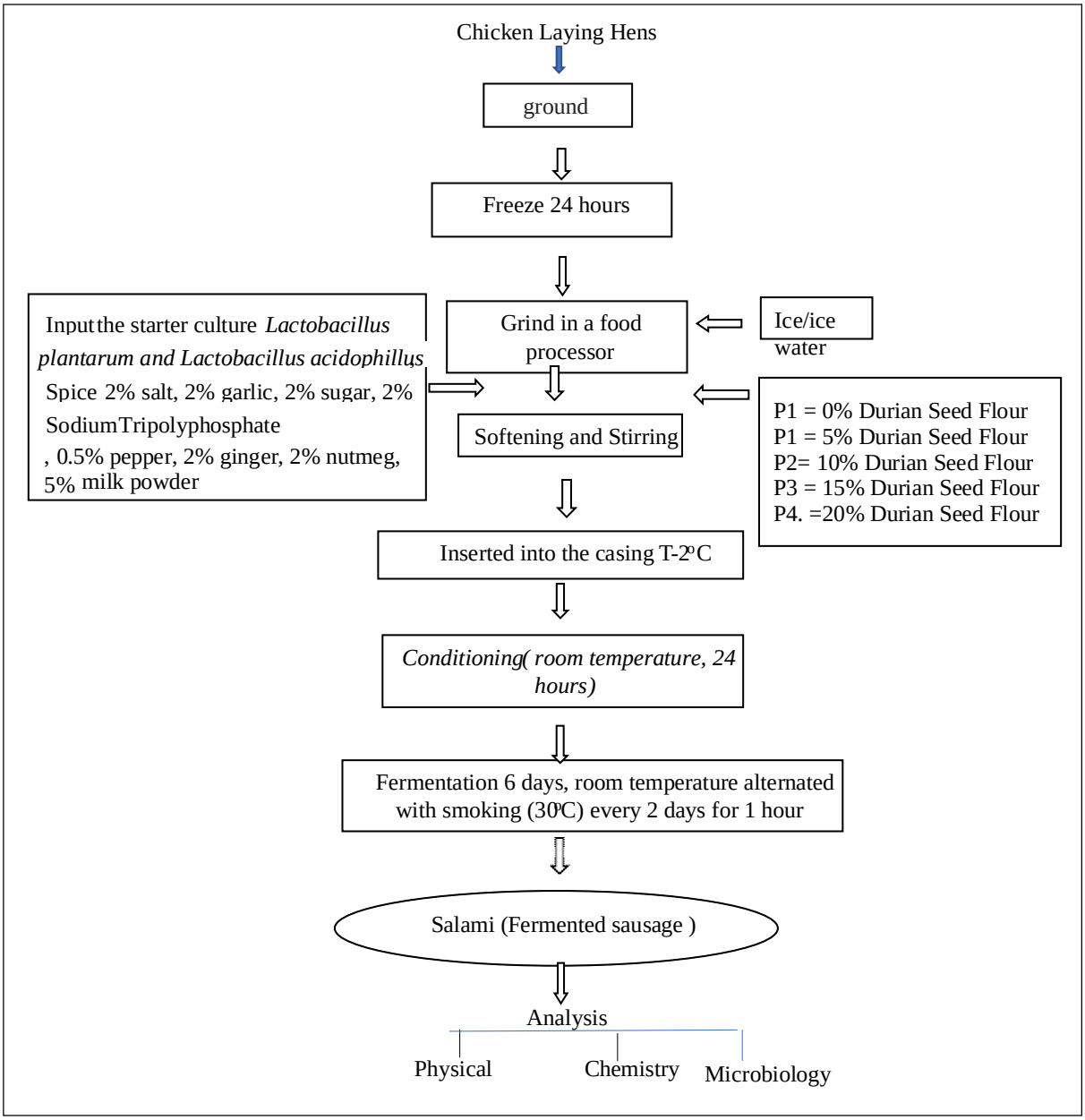

Fig 1: Making Salami (fermented sausage).

Table 1: Average cooking loss (%), water holding capacity (%) and tenderness (mm/g/10 seconds) salami using durian seed starch (*Durio zibethinus* Murr) as filler.

Treatment	Average (%)		
	Cooking loss	Water holding capacity	Tenderness mm/g/10 sec
P0	24.48[a]	27.78[a]	13.94[a]
P1	22.96[a]	34.90[b]	15.78[a]
P2	18.49[b]	43.59[c]	17.79[b]
P3	13.41[c]	47.17[c]	20.45[c]
P4	8.31[d]	51.97[d]	22.39[c]

Note: Different superscript in the same column indicate a significant difference (P<0.05).

that tenderness increases. According to Ockerman (1983) the tenderness of the meat is largely determined by at least three components of meat, namely the myofibril structure and contraction status, the content of connective tissue and the level of cross-linking and the water-binding capacity of meat proteins and meat juices. Factors that affect meat tenderness are classified into antemortem factors such as genetics, race, age and sex, livestock stress and postmortem factors such as withering, freezing, processing methods including cooking and the addition of tenderizers (Soeparno, 2005).

Effect of treatment on water content

Table 2 indicated that the water content of salami was found to be increased with increase in Durian (*Durio zibethinus* Murr) seed flour. Based on the data (Table 2), it shows that the water content of the research results is still within the standard water content set by SNI No. 3820:2015, which is a maximum of 67%. The use of durian seed flour up to 20% does not significantly affect the water content of salami. The water content will greatly affect the quality of the salami. The high water content will make it easier for microbes (bacteria, molds and yeast) to breed, affecting the salami's quality.

Opinion Winarno (2007) that the water content in food ingredients determines the freshness and durability of food. This opinion was clarified by Buckle *et al.* (2010) that water content is important in determine the durability of food ingredients. Until now, the water content of fermented sausages has not been determined, but with the acquisition of water content ranging from 40.47 (P0) to 44.84% (P4) it is close to the results of Soeparno (2005), which explains that dry sausage has a moisture content of around 25 - 45%, while the dry sausage has a water content of around 55-60%; The water content of the research results is between 40.47%-44.84% which still meets the requirements of SNI 3820-2015 with the water content of meat sausages being 67%. Salami research results are a dry sausage category. According to Hui *et al.* (2001), fermented sausage (dry sausage) has a moisture content of 30%-40%; while Pratama *et al.* (2021) reported that the water content of fermented lamb sausage was 54.93%.

Effect of treatment on protein levels

Based on the data in Table 2, it can be seen that the protein content of salami using durian seed flour decreased from 24.90%-20.74%. While the protein content of sausage according to SNI 01-3820-2015 is at least 13%, while the results of this study are far above the recommendation of the Indonesian National Standard. The decrease in protein content was due to protein denaturation which caused the protein to lose its secondary and tertiary structure due to external pressure. Similar to previous studies protein decreased from 20.42% to 18.54%. using 3% yeast starter culture and 2% lactic acid bacteria each (Sembor, 2017). It can be stated in this study, using 2% lactic acid bacteria (*L. acidhophyllus* and *L. plantarum)* to produce 24.90% protein content indicating the effect of lactic acid bacteria in increasing protein levels.

Effect of treatment on fat content

Table 2 indicated that the fat content of salami durian seed flour salami decreased from 19.95 to 13.75, with increase in the levels of This could be due to the fact that Durian seed flour had very low fat (1.18%) and therefore the higher the level of durian seed flour lower the fat content of salami. The higher the use of durian seed starch causes the fat content of salami to be low, this is because durian seed starch has lower fat compared to chicken meat fat so the addition of durian seed starch to salami products causes the fat content to decrease. The fat content of culled laying hens meat is fat 1.3 to 7.3% (Mountney and Parkhurst, 1995). while the fat content of durian seed flour is 1.18%. The decrease in fat content as a result of research suggests that fat degradation is carried out by lipase enzymes produced by lactic acid bacteria and produces a series of free fatty acids which characterize taste, aroma and texture

Table 2: Average water content (%), protein (%) and fat (%) and carbohydrates (%) salami using durian seed starch flour (*Durio zibethinus* Murr) as filler.

	Average (%)			
Treatment	Water content	Protein	Fat	Carbohydrate
P0	40.47^{c}	24.90^{a}	19.95^{a}	40.46^{c}
P1	42.64^{b}	22.11^{bc}	19.90^{a}	42.64^{b}
P2	43.44^{ab}	21.88^{bc}	17.06^{ab}	43.44^{ab}
P3	45.18^{a}	21.74^{c}	16.70^{b}	45.18^{a}
P4	44.84^{a}	20.74^{c}	13.75^{c}	44.84^{a}

Note.

P0= Salami without durian seed flour.

P1= Salami with the addition of 5% durian seed flour.

P2= Salami with the addition of 10% durian seed flour.

P3= Salami with the addition of 15% durian seed flour.

P4= Salami with the addition of 20% durian seed flour.

Different superscript in the same column indicate a significant difference (P<0.01).

which is highly dependent on type of fatty acid produced and amount of concentration. Pratama *et al.* (2021) reported that lamb sausage fermented with 2% Candida apicola yeast obtained 9.21% fat.

Effect of treatment on carbohydrate levels

Based on analysis of variance, it was shown that salami using durian (*Durio zibethinus* Murr) starch flour had a significant effect (P<0.01) on carbohydrate content (%). The data in Table 2 showed an increase in carbohydrate content along with an increase in the percentage of durian seed flour from 41.26 (PO) to 44.41% (P2). Durian seeds consist of two main components, namely starch and gum. The crude polysaccharide extract content of durian seeds has the effect of being used as an alternative source of food additives in the food industry (Bronikowska *et al.*, 2012); Ho and Bhat (2015). Raw durian seeds can be processed into flour and used as a substitute for carbohydrates because durian seeds contain as much as 43.6 grams of carbohydrates per 100 grams (Zuhri, 2015). The high carbohydrate content in salami products is accompanied by a higher level of durian seed flour, up to 20%. Durian seed flour is a source of carbohydrates so the levels of carbohydrates in salami also increase. The ratio of amylose and amylopectin greatly determines the final product of a food ingredient. The amylograph properties of foodstuffs indicate the selection of varieties according to the desired product. The composition of amylose and amylopectin affected the starch profile. Ratnayake *et al.* (2002) stated that amylopectin affects the starch granule development process. Amylose can inhibit the development of starch granules by forming a complex with fat which inhibits the increase in peak viscosity at high temperatures (Sang *et al.,* 2008; Singh *et al.,* 2010).

Effect of treatment on pH (Degree of acidity)

The results of the research on the effect of treatment on pH salami using durian seed flour (*Durio zibethinus* Murr) as a filler has been presented in Table 3.

The data from the analysis of varians (ANOVA) showed that the utility of durian seed starch flour as a filler or filler in the processing of chicken laying hens salami did not show significant effect (P>0.5) on acidity (pH) as shown in the Table 3.

Based on the research results, salami using durian seed flour as a filler has low acidity (pH), ranging from 4.04 to 4.08. In addition, salami processing is added with bacteria such as L. *plantarum* and L. *acidophillus* each 2% capable of producing lactic acid so that the pH decreases. The decrease in the pH of salami is due to the accumulation of lactic acid as a result of glucose and sucrose metabolism by lactic acid bacteria (Gonzales-Fernandez *et al*. (2006). This is the opinion of Fardiaz (1992) and Gonzalez-Fernandez *et al.* (2006) that LAB (L. *acidophyllus* and L. *plantarum*) prefer to grow in acidic conditions, namely at pH 4-4.5. The results showed that the level of acidity in salami affects the number of microorganisms. Statement from Harmain *et al*. (2012) that changes in the pH value of fermented catfish sausages is caused by a fermentation process by the lactic acid bacteria *Lactobacillus plantarum.* The change in pH value was caused by the presence of lactic acid produced from carbohydrate metabolism by lactic acid bacteria L. *plantarum*. The presence of lactic acid bacteria such as L. *plantarum* and L. *acidophylus* also causes a decrease in pH during fermentation. The lactic acid produced by the lactic acid bacteria will be excreted outside the cells and accumulate in the fermentation media, thus increasing the acidity and decreasing the salami's pH while Pratama *et al.* (2021) reported that the pH value of fermented sausages using yeast *Candida apicola* was 4.83.

Effect of treatment on TPC (total plate count) (CFU/gram)

The data in Table 4 shows that the total plate count of salami using durian seed flour was up to 20%, with the addition of 2% lactic acid bacteria (L. *acidophyllus* and L. *plantarum*), the total plate count decreased.

The decrease in the total plate count (TPC) of salami was due to the use of durian seed flour as a filler, besides that the microorganisms L. *acidophyllus* and L. *plantarum* were also added 2% each for each treatment. The treatments using 10%, 15% and 20% durian seed powder each showed no significant difference, although the treatment using 20% durian seed powder had the lowest total plate count (3.038×10^3 CFU/gram). This is due to the influence of the viability of lactic acid bacteria cultures used such as L. *plantarum* and L. *acidophyllus*, causing the total bacteria to be low. In addition to producing lactic acid, lactic acid bacteria produce bacteriocins which can inhibits the growth of spoilage and pathogenic bacteria, thereby increasing the shelf life of salami. Under tightly controlled conditions, the cultured strains of these microorganisms can induce enzyme activity specifically to modify substrates. This substrate change can

Table 3: Average pH of salami using durian seed starch as a filler.

Replication	Treatment					
	P0	P1	P2	P3	P4	Total
1	4.10	4.01	4.04	4.11	4.08	81.01
2	4.05	4.05	4.04	4.10	4.06	
3	4.10	3.99	4.05	3.99	4.09	
4	4.03	3.97	4.02	4.04	4.09	
Total	16.28	16.02	16.15	16.24	16.32	
Average	4.07	4.005	4.04	4.06	4.08	

Table 4: Analysis of the variety of effects of treatment on total lactic acid bacteria (CFU/gram) using durian seed flour (*Durio zibethinus* Murr) as filler.

	Treatment (.....× 10^3 CFU/gram)					
Replication	P0	P1	P2	P3	P4	Total
1	3.579	3.491	3.431	3.342	3.255	
2	3.568	3.477	3.447	3.301	3.176	
3	3.579	3.505	3.431	3.301	2.400	
4	3.556	3.556	3.415	3.322	3.322	
Total	14.282	14.029	13.724	13.266	12.153	67.454
Average	3.571^{a}	3.507^{a}	3.431^{ab}	3.316^{ab}	3.038^{b}	

eliminate the potential for harmful microorganisms such as *Salmonella, Staphylococcus* and *Clostridia* (Abunyewa *et al.,* 2000). Rosahdi *et al.* (2022) reported that the number of bacteria in jackfruit seed flour was 3.24×10^{10} cells/ml, while in durian seed flour it was 3.04×10^{10} cells/ml.

CONCLUSION

The conclusion of this study shows that durian seed flour can be used as a filler in salami products so that it can improve the physicochemical and microbiological quality with a level of 15%.

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Conflict of interest

All authors declared that there is no conflict of interest.

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