

Impact of lard-based diacylglycerols on the quality and sensory characteristics of emulsion-type sausage

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Abstract: The objective of this study was to evaluate the effects of fat levels (200 g·kg⁻¹ meat and 500 g·kg⁻¹ meat) and types [lard, glycerolised lard (GL), and purified glycerolised lard (PGL)] on the quality and sensory characteristics of emulsion-type sausages. As observed, at the same type of fat, the low-fat sausage (200 g·kg⁻¹ meat) had a significantly higher *L**-value (lightness) and lower cooking loss and total expressible fluid ($P < 0.05$) than the high-fat sausage (500 g·kg⁻¹ meat) and exhibited a denser and more homogeneous microstructure. Additionally, T_{23} (relaxation time) of the low-fat sausage shifted toward a slower relaxation time, and higher A_{23} (peak area) was found, which suggested the water mobility was restricted. However, at the same fat content, the low-GL and low-PGL sausages showed better textural properties and superior overall acceptability from sensory evaluation compared with the low-lard sausage ($P < 0.05$). Still, they have no significant differences ($P > 0.05$). Therefore, lard-based diacylglycerol could be effectively applied as a fat replacer in emulsion-type sausages with low-fat contents to produce healthier meat products.

Keywords: instrumental texture; microstructure; sensory; evaluation; cooked sausage

In the production of meat emulsions, in addition to playing a critical and direct role in flavour, fat can also stabilise the dissolving of protein gels and act as a filler to help prevent protein from contracting during cooking (Lurueña-Martínez et al. 2004). Whereas, because of high cholesterol and calorie contents, high-fat diets can increase the development of hypertension and cardiovascular disease (Nejat et al. 2010) and raise the risk

of obesity and certain cancers (Botchlett and Wu 2018). The increasing requirement of consumers for healthy meat products has urged meat manufacturers to develop new or higher-quality low-fat products continuously. However, reducing total fat from the product formulation would make the product lose its flavour and mouth-feel and decrease sensory texture properties, resulting in low acceptability by consumers (Choi et al. 2010).

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Some vegetable oils involving a lot of unsaturated fatty acids have been used as animal fat substitutes. They showed positive results in sensorial and textural characteristics when compared to products without fat reduction, such as pumpkin seed oil in bologna-type sausages (Uzlasir et al. 2020), peanut oil in emulsified sausages (Nacak et al. 2021), and wheat germ oil in beef patties (Khalid et al. 2021). However, few research was found on applying diacylglycerol (DAG) oil in meat products.

Nowadays, DAG has been confirmed to inhibit abdominal and visceral fat accumulation, halt the rise in obesity, and decrease postprandial serum triacylglycerol levels (Taguchi et al. 2000). Additionally, the security of DAG has been demonstrated through many animal and human research, and it may be used as an edible oil for human consumption (Morita and Soni 2009). Therefore, a variety of oils, i.e. palm oil (Xu et al. 2016), rapeseed oil (Weber and Mukherjee 2004), and soybean oil (Zhong et al. 2019), have been utilised to produce DAG in recent years. Our previous study found that GL (lard, glycerolised lard), and PGL (purified glycerolised lard) containing DAG can form emulsions (Diao et al. 2016a) and gels (Diao et al. 2016b) since DAG has hydrophilic groups in its molecular structure and shows interfacial properties. However, so far, no study has reported the application of lard-based DAG as a fat replacer in emulsion-type sausages.

Hence, this study aimed to assess the impact of lard-based DAG as a pork-fat substitute on cooking loss, texture profile, colour difference, water distribution, microstructure, and sensory characteristics of emulsion-type sausages.

MATERIAL AND METHODS

Preparation of lard diacylglycerol (DAG). Lard was obtained by heating the backfat and filtering with four layers of gauze. Lard DAG was prepared using an enzymatic glycerolysis reaction, according to Diao et al. (2017). After the reaction was finished, the enzyme was filtered to obtain the glycerolised lard (GL), and the GL was purified using molecular distillation to yield purified glycerolised lard (PGL). The contents of DAG in the GL and PGL were 61.76% and 82.03%, respectively.

Preparation of emulsion-type sausage. Fresh lean meat from pork hindquarters without visible connective tissue was obtained from a local supermarket (Jinzhou, China) and transferred to the meat laboratory on ice. Lean pork was ground through a grinder (JRJ-B1; Beijing Liren Technology Co., China) with an 8 mm plate. Lean pork mince (500 g) was blended with salt (15 g), sugar (7.50 g), sodium nitrite (0.05 g), erythorbic acid (0.25 g), and compound sodium phosphate (2.00 g). The mixture was marinated for 12 h at 4 °C, and then corn starch (50 g) and soy isolate protein (10 g) were added. Afterwards, six types of emulsion sausages were manufactured by adding different kinds of fat. The three high-fat treatments (lard, GL, and PGL) prepared with fat (500 g·kg⁻¹ meat) were used as control groups, and the other three low-fat treatment groups (lard, GL, and PGL) were prepared with fat (200 g·kg⁻¹ meat), respectively. Ice water (300 g·kg⁻¹ meat) was added to each formula treatment group, and the mixing was chopped to get a uniform mass using a chopper. Chopped batters were stuffed into PE/PA/PE (polyethylene/polyamides/polyethylene) casing, and both ends were tightly sealed. The samples were cooked at 85 °C for 30 min and then cooled in an ice-water bath for 20 min. Cooked sausages were maintained under 4 °C until analysis.

Cooking loss. The cooking loss of emulsion-type sausage was calculated as follows [Equation 1]:

$$\text{Cooking loss (\%)} = \frac{W_0 - W_1}{W_0} \times 100 \quad (1)$$

where: W_0 – weight of samples before cooking; W_1 – weight of samples after cooking.

Total expressible fluid. The full expressible fluid of emulsion-type sausage was determined using a strain-controlled unconfined compression apparatus (NX-YYW-2, Hengao Instrument Co., Beijing, China). The emulsion-type sausage (diameter 20 mm, height 10 mm) was wrapped with four layers of gauze and ten layers of filter paper and then pressed on the platform for 10 min under the force of 69.44 N. Subsequently, the gauze and filter paper were removed, and the total expressible fluid was calculated as follows [Equation 2]:

$$\text{Total expressible fluid (\%)} = \frac{\text{weight before pressing} - \text{weight after pressing}}{\text{weight before pressing}} \times 100 \quad (2)$$

Textural profile analysis (TPA). TPA was performed via a TA-XT plus texture analyser (Stable Micro Systems, Surrey, England) with a 25 kg load cell according to the procedure of Maqsood et al. (2012). Hardness (g), springiness, cohesiveness, chewiness (g), adhesiveness, and resilience were recorded.

Colour determination. The colour of the emulsion-type sausage was determined from 5 different positions via a ZE-6000 colourimeter (Nippon Den-shoku, Kogyo Co., Japan). A white plate was employed as a reference ($L^* = 95.26$, $a^* = -0.89$, $b^* = 1.18$), and L^* (lightness), a^* (redness), and b^* (yellowness) values were recorded during measurement.

Water distribution. Low-field nuclear magnetic resonance (LF-NMR) measurement was conducted via an mq-20 NMR analyser (Bruker Optik GmbH, Germany). Approximately 2.0 g of the minced emulsion-type sausage was placed in an 18 mm-diameter NMR tube. The transverse relaxation time (T_2) was determined by the Carr-Purcell-Meiboom-Gill (CPMG) sequence. The relaxation data were treated using the CONTIN algorithm, and the relaxation time (T_{2i}) and amplitude (A_{2i}) were analysed for every detected population in CONTIN (Han et al. 2014).

Scanning electron microscopy (SEM). The microstructure of emulsion-type sausage was evaluated via scanning electron microscopy (S-3400N, Hitachi, Japan). After the sample from each treatment group was cut into small squares ($3 \times 3 \times 3 \text{ mm}^3$), they were fixed with 2.5% glutaraldehyde in phosphate buffer (0.2 M, pH 7.0) for 12 h. Subsequently, fixed samples were washed with distilled water, and dehydration was carried out via a series of ethanol (50, 70, 80, 90, and 100%) for 15 min at every concentration. Finally, every sample was lyophilised and sputter-coated with 10 nm of gold. The prepared samples were observed with an SEM at an accelerating voltage of 5.0 kV. Photographs of each sample were taken.

Sensory analysis. Sensory evaluation of sausages was performed by ten untrained panellists who regularly eat sausages. The sausage was cut into 2 mm thick slices and placed on a white polystyrene plate. Panellists evaluated sausage texture, elasticity, taste, odour, and overall acceptability and scored using a 9-point sensory test (1 = extremely unpleasant, 9 = extremely pleasant).

Statistical analysis. Three independent batches of sausages (replicates) were prepared, and a total of six types of sausages were prepared in each batch. For each batch of sausages, measurements of related traits were conducted three times, and the results were expressed as mean \pm standard errors (SE). Statistical analysis of data was carried out using the general linear model

procedure in the Statistics 8.1 software package (Analysis Software, USA). The Tukey's honestly significant difference (HSD) test was used for significant difference ($P < 0.05$) analysis, and the sigmaplot 11.0 software was used for drawing.

RESULTS AND DISCUSSION

Cooking loss and total expressible fluid. The cooking loss reflects the ability of products to retain their water and fat in cooking. When moisture is lost during cooking, product tenderness and flavour decrease (Barbut 2007). Figure 1 shows that high-fat samples' cooking loss and total expressible fluid were higher than low-fat samples. The phenomenon may be because the fat contained between myofibrils was saturated, and the excess fat was lost during the cooking or compression process, increasing the cooking loss or total expressible fluid of emulsion-type sausages (Hopkins et al. 2006). Another reason may be that excess fat affects the capacity of proteins to form emulsions, thus reducing water-holding (Abbasi et al. 2019). In addition, as shown in Figure 1, the emulsion-type sausages with GL or PGL compared with that with lard had a lower cooking loss and total expressible fluid, which may be the hydrophilic groups in DAG can more strongly associate with water during the emulsification process, improving the water and oil retention capacity of emulsion-type sausages (Nakajima 2004).

Textural profile analysis. Textural profile analysis parameters for emulsion-type sausages with lard, GL, or PGL were listed in Table 1. At the same type of fat, textural properties of hardness, springiness, cohesiveness, chewiness, and resilience from high-fat samples were significantly lower than from low-fat samples, which indicated that adding more fat harmed the texture of products. The difference may be explained by the fact that muscle protein in high-fat samples could not completely wrap all fat, resulting in the outflow of some fat during cooking, which destroyed the spatial structure of emulsion-type sausages to a certain extent. Additionally, the emulsion-type sausages with GL or PGL significantly increased hardness, springiness, cohesiveness, chewiness, and resilience compared with the emulsion-type sausage with lard at the same fat levels. The discrepancy may be because the particle of DAG is smaller than lard and can positively stabilise the batter by acting as a filler in the protein network (Diao et al. 2016a).

In addition, the high adhesiveness could indicate undesirable behaviour because the high stickiness of products may make them difficult to slice (Álvarez and

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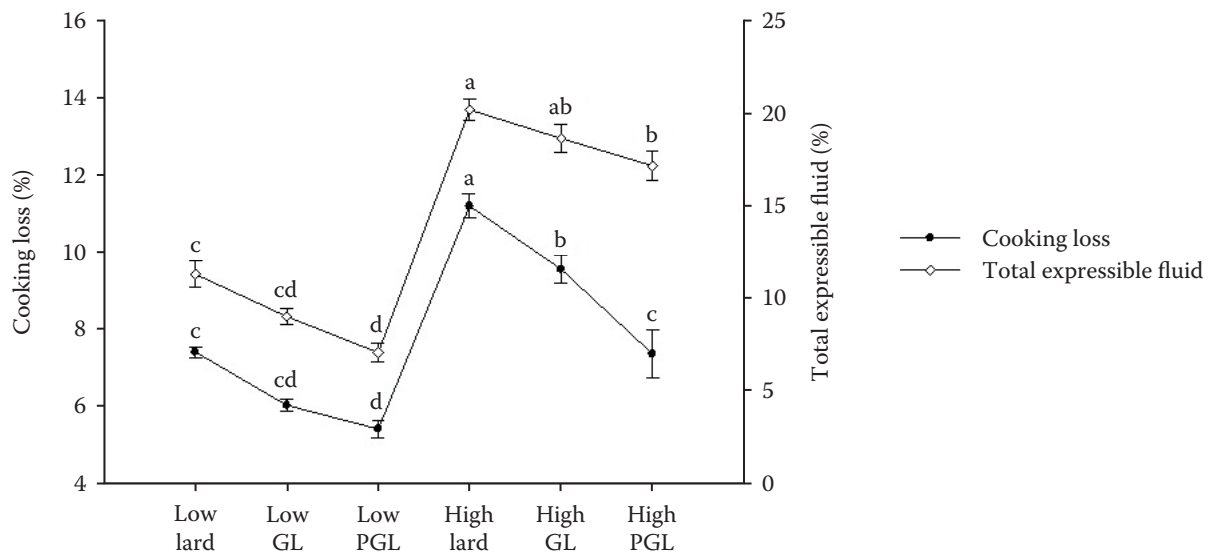


Figure 1. Cooking loss and total expressible fluid of different emulsion-type sausages prepared with lard, glycerolised lard (GL), or purified glycerolised lard (PGL)

^{a–d} statistically significant differences ($P < 0.05$)

Barbut 2013). In our study, emulsion-type sausages with GL or PGL showed a lower adhesiveness, which may be because DAG has higher crystallisation points than triacylglycerol (TAG) (Miklos et al. 2013). In conclusion, the emulsion-type sausage with PGL showed the best texture characteristics due to its high content of DAG.

Colour evaluation. Lightness (L^*), redness (a^*), and yellowness (b^*) are regarded as the most informative parameters for colour changes in meat products (Savadkoobi et al. 2014). At the same type of fat, the L^* from high-fat samples was higher than low-fat samples (Table 2), which indicated that fat could in-

Table 1. Textural parameters of different emulsion-type sausages prepared with lard, glycerolised lard (GL), and purified glycerolised lard (PGL)

Fat type and content	Hardness (g)	Adhesiveness	Springiness	Cohesiveness	Chewiness (g)	Resilience
Low lard	2 560.2 ± 57.52 ^c	–48.89 ± 4.02 ^d	0.72 ± 0.01 ^b	0.55 ± 0.03 ^c	1 473.4 ± 48.08 ^c	0.25 ± 0.03 ^b
Low GL	2 886.8 ± 52.27 ^b	–36.63 ± 0.61 ^{ab}	0.80 ± 0.04 ^a	0.64 ± 0.04 ^{ab}	1 837.4 ± 26.40 ^b	0.32 ± 0.02 ^a
Low PGL	3 989.5 ± 60.65 ^a	–32.52 ± 2.67 ^a	0.83 ± 0.03 ^a	0.68 ± 0.01 ^a	2 052.9 ± 33.63 ^a	0.34 ± 0.01 ^a
High lard	549.29 ± 47.93 ^e	–52.63 ± 2.75 ^e	0.67 ± 0.03 ^c	0.40 ± 0.06 ^d	159.71 ± 11.73 ^e	0.11 ± 0.04 ^c
High GL	2 390.7 ± 73.97 ^d	–44.03 ± 2.34 ^c	0.74 ± 0.03 ^b	0.56 ± 0.04 ^c	840.73 ± 38.93 ^d	0.24 ± 0.01 ^b
High PGL	2 511.2 ± 73.72 ^{cd}	–41.78 ± 1.11 ^{bc}	0.75 ± 0.01 ^b	0.53 ± 0.01 ^c	741.82 ± 21.26 ^d	0.25 ± 0.03 ^b

^{a–d} statistically significant differences ($P < 0.05$); values are given as the mean ± standard errors (SE) from triplicate determinations

Table 2. The internal colour L^* (lightness), a^* (redness), and b^* (yellowness) values of different emulsion-type sausages prepared with lard, glycerolised lard (GL), or purified glycerolised lard (PGL)

Fat type and content	Lightness (L^*)	Redness (a^*)	Yellowness (b^*)
Low lard	72.59 ± 1.32 ^b	6.84 ± 0.22 ^a	10.35 ± 0.45 ^c
Low GL	73.83 ± 0.20 ^{ab}	6.01 ± 0.35 ^{ab}	10.67 ± 0.50 ^{bc}
Low PGL	74.15 ± 0.39 ^{ab}	5.89 ± 0.28 ^b	12.29 ± 1.11 ^{ab}
High lard	72.79 ± 0.49 ^{ab}	5.24 ± 0.25 ^{bc}	11.16 ± 0.48 ^{bc}
High GL	73.97 ± 0.62 ^{ab}	4.65 ± 0.49 ^c	12.09 ± 0.10 ^{abc}
High PGL	74.57 ± 0.14 ^a	4.29 ± 0.33 ^c	13.75 ± 0.85 ^a

^{a–c} statistically significant differences ($P < 0.05$); values are given as the mean ± standard errors (SE) from triplicate determinations

crease the lightness value of products. However, they had no significant difference ($P > 0.05$).

Furthermore, the L^* from emulsion-type sausage with PGL was higher than with lard at the same fat levels. The finding was in accord with the result of the cooking loss. In addition, compared with the high-fat sample, the higher a^* value in the low-fat sample could be attributed to the higher amounts of lean (i.e. higher myoglobin pigment). However, the emulsion-type sausages with GL or PGL decreased a^* -values but increased b^* -values compared with the sausage with lard at the same fat levels, which may be due to the natural yellow colour of GL or PGL. Although the b^* -values

of the sausage with PGL containing more DAG were higher than those with GL, no difference was observed ($P < 0.05$).

Low-field nuclear magnetic resonance (LF-NMR) analysis. Figure 2A shows the distribution curves of relaxation times (T_{21} , T_{22} , T_{23} , and T_{24}) of different emulsion-type sausages prepared with lard, GL, or PGL. Four populations of water were observed in curves, including bound water with a relaxation time of T_{21} (1–5 ms), moderately bound water with a relaxation time of T_{22} (6–16 ms), immobilised water with a relaxation time of T_{23} (40–61 ms), and free water with a relaxation time of T_{24} (100–1 000 ms). Pearce

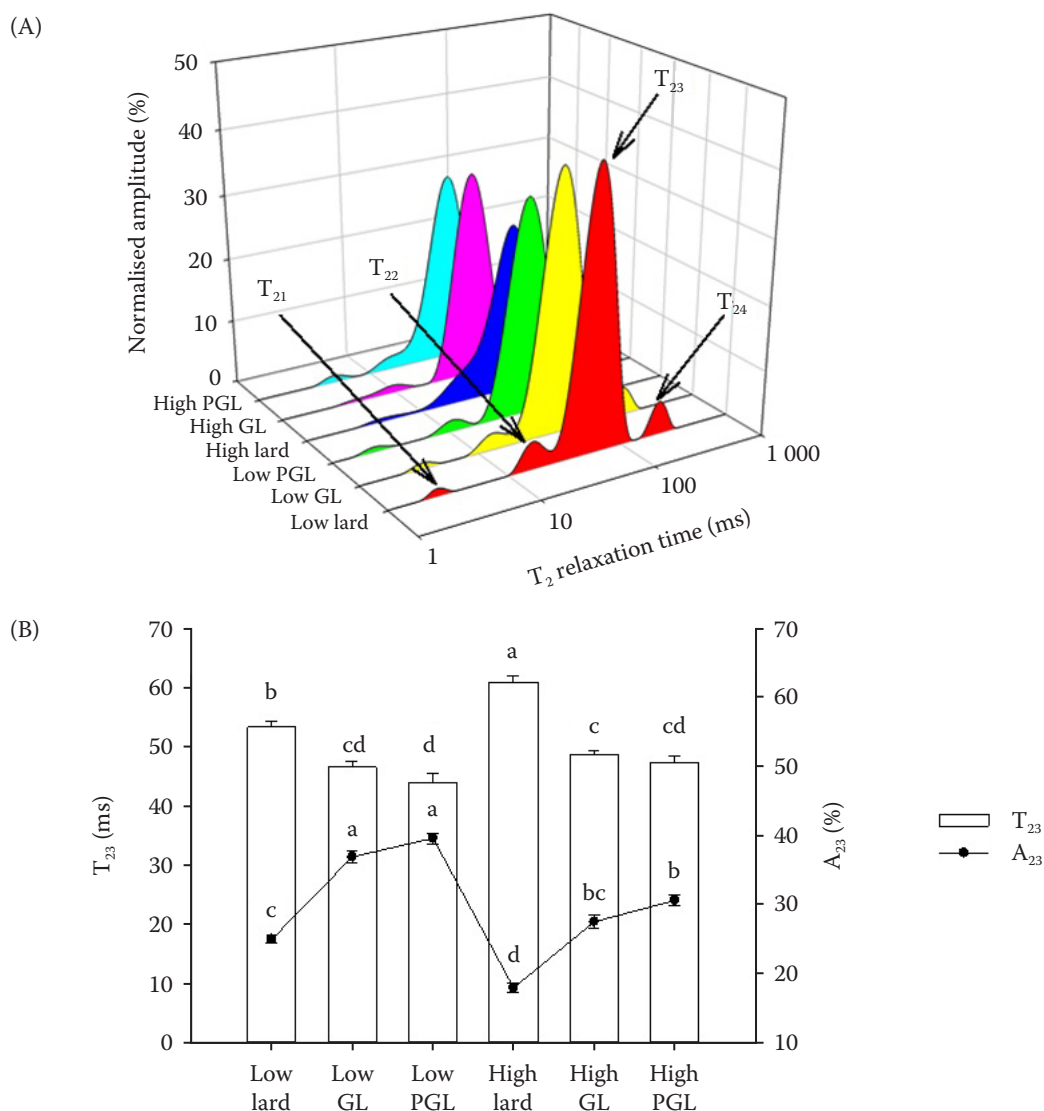


Figure 2. Distributions of low-field nuclear magnetic resonance (LF NMR): (A) T_2 relaxation times of different emulsion-type sausages prepared with lard, glycerolised lard (GL), or purified glycerolised lard (PGL) and (B) relaxation time T_{23} and the corresponding peak area A_{23}

^{a-d} statistically significant differences ($P < 0.05$)

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et al. (2011) reported that sausage water distribution was principally immobilised, which played a vital role in meat quality.

Our experimental result also indicated that the water distribution in emulsion-type sausages prepared with lard, GL, or PGL was mainly immobilised. In contrast, the water content of the other three types was relatively small. Therefore, the changes in relaxation time T_{23} and the corresponding peak area proportions A_{23} are shown in Figure 2B. At the same kind of fat, compared with high-fat samples (control), the T_{23} from low-fat samples moved toward the direction of slower relaxation time, suggesting that water mobility in the low-fat samples was confined. Furthermore, the corresponding areas A_{23} of high-fat samples were lower than low-fat samples at the same type of fat (Figure 2B), which was also closely related to higher cooking loss.

Additionally, at the same fat levels, the emulsion-type sausage with GL or PGL had significantly lower T_{23} and higher A_{23} than that prepared with lard, which indicated that GL and PGL could hold water in a less mobilised state. One possible explanation was that GL and PGL contained more hydrophilic polar groups in their molecular structure. However, T_{23} and A_{23} of the samples with GL and PGL showed no significant difference ($P < 0.05$).

Microstructure of emulsion-type sausages. The low-fat samples exhibited a compact and homogeneous network structure, but the high-fat samples showed many pores or void spaces (Figure 3).

This may be because proteins could not wrap more fat during heating, which made fat overflow and formed holes. In the low-fat samples, the sausage with PGL had a more compact structure with fewer voids than those with GL and lard. The result indicated that increasing DAG concentrations resulted in the formation of smaller and more uniform lipid droplets. In addition, our previous experiment also revealed that PGL could evenly disperse in the protein network of gels prepared with myofibrillar protein and PGL, leading to a compact and homogeneous microstructure (Diao et al. 2016b). These structural features also explained why emulsion-type sausages with PGL have stronger hardness and water binding.

Sensory analysis. At the same type of fat, all sensory parameters of high-fat samples gained lower scores than the low-fat samples (Figure 4), which was to the TPA test result of the sausages. The lower odour scores of high-fat samples could be due to higher fat making the sausages greasy.

Additionally, a higher concentration of fat would make the product soft, resulting in a lacking texture of the meat itself. Therefore, the odour, texture, and overall acceptability of high-fat sausages were disliked by panellists, which may result in declining their market value. At the same fat levels, all sensory attributes in the sausage added with lard were lower than those with GL and PGL, but no significant difference ($P > 0.05$) was observed. Usually, higher overall acceptability was found in the sausage with low PGL, sug-

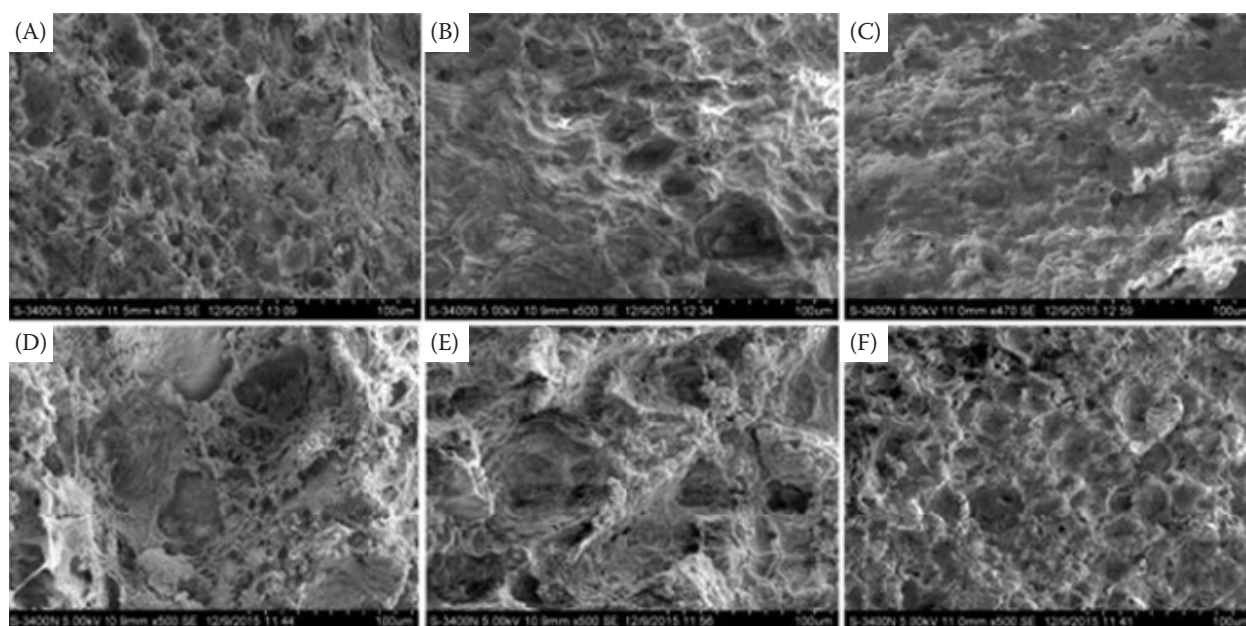


Figure 3. Scanning micrographs of different emulsion-type sausages prepared with lard, glycerolised lard (GL), or purified glycerolised lard (PGL): (A–C) low lard, GL, or PGL; (D–F) high lard, GL, or PGL

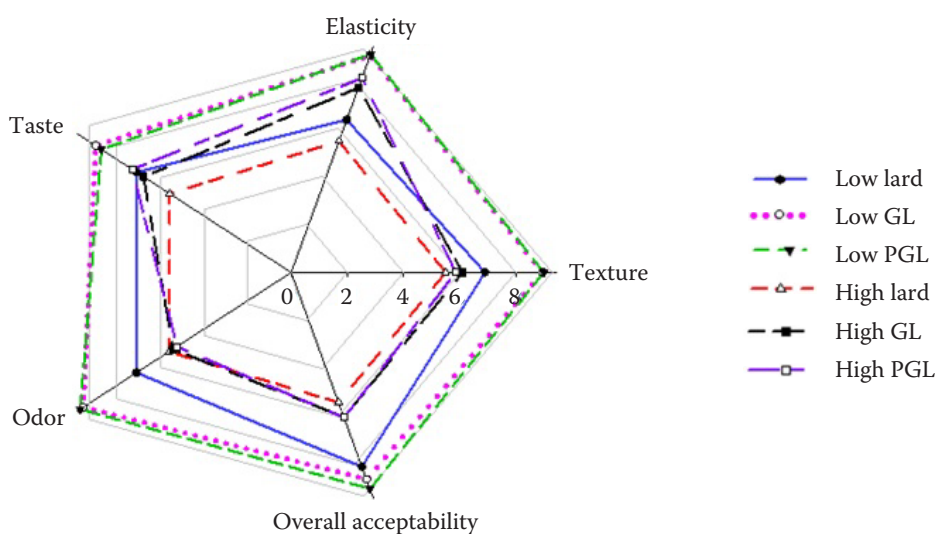


Figure 4. Sensory evaluation of different emulsion-type sausages prepared with lard, glycerolised lard (GL), or purified glycerolised lard (PGL)

gesting that the sausage is the most acceptable product to the panellist.

CONCLUSION

The types and concentrations of fat significantly affect the quality characteristics and sensory properties of emulsion-type sausages. The sausage prepared with low GL or PGL exhibited the desirable quality and sensory characteristics compared with the other formulations. Moreover, consumers prefer foods with lower fat. Therefore, results from this study indicated that adding a small amount of lard DAG to meat products has good application potential.

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