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Interplay Role of Heat-Moisture Treatment and Lipid from Egg yolk and Margarine on Functional and Pasting Properties of Banana Flour

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Egg yolk and margarine are commonly applied in starchy food processing as a lipid source. Both may affect the starch properties to different extent. Therefore, the effects of adding egg yolk (0.3, 1.0, and 1.7 g/100 g of flour) and margarine (1, 3, and 5 g/100 g of flour) to banana flour on the complex formation of gelatinized starch–lipid and on paste functional properties were investigated in this study. Native banana flour was used as a control. The complexing index (CI) value increased with the increasing egg yolk or margarine content in the paste. Egg yolk lipids formed complexes with flour starch more favorably than margarine lipids. The heat-moisture treatment (HMT) of banana flour increased its ability to form a complex. Complex formation may partly affect the swelling volume (SV) and freeze-thaw stability (FTS) of pastes. The decrease in SV was smaller in the paste with egg yolk than with margarine. Egg yolk also led to a lesser syneresis and solubility than margarine. For freeze-thaw stability, the interplay roles between the reducing effect of lipid addition and the increasing effect of HMT led to an increased syneresis suggesting that HMT factor prevailed. The reducing effect of lipid addition on egg yolk complex solubility predominated the increasing impact of HMT. Lipid source did not affect pasting properties except set back viscosity. HMT-pretreatment played a major role in the alteration of pasting properties compared to the lipid content.

ABBREVIATIONS

HMT – heat-moisture treatment, SV – swelling volume, FTS – freeze-thaw stability, CI – complexing index.

INTRODUCTION

Banana starch application in food has been explored in several studies [Cahyana *et al*., 2020; Fida *et al*., 2020]. In order to improve its properties in food application, native starch is commonly modified by various methods including physical [Adebowale *et al*., 2005; Bian & Chung, 2016], chemical [Aparicio-Saguilán *et al*., 2014; Cahyana *et al.*, 2018; Handarini *et al.*, 2020b; Wattanachant *et al*., 2003], and enzymatic ones [do Prado Cordoba *et al*., 2016], of which the physical treatment is promising due to the absence of chemical residues following the treatment. Amongst physical treatments, heat-moisture treatment (HMT) is of particular interest. HMT is carried out by heating starch above its gelatinisation temperature (normally ≥100°C) in a limited moisture content (normally ≤30%) [Marta *et al*., 2020]. The HMT treatment to banana flour results in remarkable changes of its pasting (an increase in pasting temperature but a decrease in peak, breakdown, and setback viscosity) and functional properties (a decrease in swelling volume and water absorption capacity) compared to the native starch [Cahyana *et al*., 2019; Cordeiro *et al*., 2018]. Therefore, the application of HMT-starch or flour in food application would be very advantageous.

When either native or HMT-starch is applied in food, other major components like lipid are often added as a product ingredient. A number of studies have reported the formation of complexes between lipid and starch. Fatty acids, such as lauric, myristic, palmitic, and stearic acids, form complexes with starch [Qin *et al*., 2019]. The lipid-starch complex was reported to affect starch properties, including a reduction in the final viscosity and *in vitro* starch digestibility [Mapengo & Emmambux, 2020].

Margarine and egg yolk are those which are often mixed with starch as a dough mixture prior to baking. The presence of both lipids and starch in a food matrix is expected to affect certain properties of starchy food due to the lipid-starch interaction. Given that both margarine and egg yolk are oftent present altogether in starchy food as the ingredients, it is interesting to study starch interaction with their lipids and its effect on complex formation as well as on starch properties. Considering that modified starch is favourably used in food application compared to the native starch, it is important to examine the effect of starch modification particularly that modified with HMT on its interaction with lipid using native starch as a control. Different content of margarine and egg

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yolk were applied to comprehend the effect of the content on studied parameters.

This study aimed to reveal the effect of lipid addition from egg yolk and margarine on functional properties, such as swelling volume, solubility and freeze-thaw stability, as well as pasting properties of HMT banana flour. Complexing index was analysed to measure the extent of complex formation between banana flour and lipids.

MATERIALS AND METHODS

Material

Banana cultivar Kapas harvested at 8–10 weeks after flowering was used in the present study (ripening stage 1, entirely green). The bananas were obtained from a local farmer in Jati Gede, Indonesia. Fruit fingers were selected for uniformity of colour, size, and shape. Margarine Palmia and egg of laying hens were purchased at a local market (Bandung, Indonesia). All chemical reagents used in this work were of analytical grade (SIGMA, Singapore) and used directly without further purification.

Lipid content and fatty acid profile determination

Lipid content and fatty acid profile of margarine and egg yolk were analysed at a commercial laboratory service of PT Saraswanti Indo Genetech (Jakarta, Indonesia). The laboratory has been certified by the National Accreditation Committee (KAN). Lipid content in margarine was determined using the Soxhlet method while lipids in egg yolk before extraction were hydrolysed (Weibull-Stoldt method) [Kolar *et al*., 1993]. Fatty acids were analysed using gas chromatography with flame ionization detection (GC FID Clarus 680 Perkin Elmer, USA). The analysis was carried out by injecting 1 μL of methylated (using BF_3-methanol reagent) samples at injection temperature of 240°C into the DB Fast-FAME (30 m × 0.25 mm, film thickness 0.25 μm) capillary column (Agilent, Santa Clara, CA, USA). Helium (H_2) was used as a carrier gas. The H_2 and air flow rates were 30 and 300 mL/min. An oven was set at temperature gradient of 50––230°C with running time being 24.67 min.

Banana flour preparation

Banana flour was prepared according to the previous method [Marta *et al*., 2019b]. The pulp from unripe banana fruit was sliced into pieces approx. 2 mm thick, dipped in water for 15 min, and then drained. The banana slices were then dried in a drying oven at 50°C for 24 h followed by milling to produce flour using a miller machine FCT Z-300 (Fomac, Jakarta, Indonesia). The flour was then passed through 100 mesh screens. Banana flour was stored in a polypropylene plastic bag as a primary packaging using aluminum foil along with silica gel as the secondary packaging, and stored at room temperature (26±2°C) for later analysis.

Heat-moisture treatment of banana flour

Heat-moisture-treated banana flour was prepared according to the previous method [Marta *et al*., 2020]. The flour was placed in a pan and its moisture content was adjusted to 30% by spraying distilled water homogenously. The pan was then covered and allowed to equilibrate at 4°C for 24 h in a refrigerator followed by heat treatment at 100°C for 8 h. The preparation was then dried in an oven at 50°C for 24 h, and ground prior to sieving through 100 mesh screen.

Formulating egg yolk or margarine mixtures with banana flour and measuring pasting properties

Rapid visco analyzer (RVA-SM2; Perten, Warriewood, Australia) was used to measure pasting properties of the mixture of egg yolk or margarine with banana flour. Margarine or egg yolk was weighed accurately on flour weight basis (1, 3, and 5 g/100 g of flour for margarine and 0.3, 1.0, and 1.7 g/100 g of flour for egg yolk) and placed in a viscometer followed by the addition of distilled water (25 mL) and 3.5 g of banana flour (native or HMT-flour). The mixture was then agitated by raising and lowering the plastic paddle through the canister 10 times before the canister was inserted into the instrument. Viscosity was recorded with the following profile: holding at 50°C for 1 min, heating from 50°C to 95°C for 3.7 min, holding at 95°C for 2.5 min, and cooling down to 50°C for 3.8 min. The gel was maintained for 2 min at 50°C with rapid stirring at 960 rpm for the first 10 s to disperse the starch sample, under a constant paddle rotating speed of 160 rpm throughout the entire analysis. The total run time for each sample was 13 min. Parameters of pasting properties were measured. The final resulting paste was used for other analyses.

Complexing index evaluation

The complexing index (CI) of gelatinised starch with lipids was determined using the method of Handarini *et al*. [2020a] with slight modifications. Immediately after the completion of the parameters of pasting properties, 5 g of starch paste was removed from the RVA canister and placed in a 50-mL capped tube. Distilled water (25 mL) was added into the tube and mixed with the paste at 50°C. The tube was the homogenised using vortex for 2 min, and 100 μL of the resulting dispersion was mixed with 15 mL of distilled water, followed by the addition of 2 mL of an iodine solution (2.0% KI and 1.3% of I_2 in distilled water). The absorbance was measured at 690 nm with a spectrophotometer (Rayleigh UV-9200, Beijing, China). Pastes without lipid addition were used as a reference. The experiment was carried out within 60 min to avoid starch retrogradation, and CI was calculated using the following equation:

$$CI\ (\%) = \frac{(A_{ref} - A_{sample})}{A_{ref}}\ x\ 100\% \quad (1)$$

where: CI is complexing index, A_{ref} and A_{sample} are respectively absorbance of reference and sample.

Swelling volume and solubility determination

The paste was cooled in iced water for 1 min and centrifuged at 3500 rpm for 30 min. The volume of the supernatant was measured, and solubility was determined from the supernatant previously dried in a hot air oven. The total volume was determined separately by measuring the mixture of the same quantity of flour, lipid, and distilled water as carried out in

RVA. The swelling volume and solubility were calculated according to Equations (2) and (3):

$$Swelling\ volume\ \left(\frac{mL}{g}\right) = \frac{(\text{Total volume} - \text{Supernatant volume})}{\text{Sample weight (dry basis)}} \quad (2)$$

$$Solubility\ (\%) = \frac{\text{Dried supernatant weight}}{\text{Sample weight}}\, x\ 100\% \quad (3)$$

Freeze-thaw stability determination

A 10 g of paste from RVA was taken, placed in a centrifuge tube, and cooled to room temperature in an iced shaking water bath. The tube was then subjected to a freeze–thaw cycle by storing at 4°C for 24 h, freezing at -15°C for 48 h, thawing at 25°C for 3 h, and centrifuging at 3500 rpm for 15 min. The supernatant removed from the gel was weighed. The extent of syneresis was expressed as the percentage of separated liquid per total weight of the sample in the centrifuge tube.

Statistical analysis

The experiments were conducted in triplicate. Statistical analysis was performed using Statistical Package for the Social Sciences (IBM, New York, USA). Data were expressed as mean ± standard deviation of triplicate determinations and were compared through one-way ANOVA using Duncan's Multiple Range test at $p<0.05$ significance level.

RESULTS AND DISCUSSION

Total lipid content and fatty acid profile

The egg yolk and margarine used in experiments were determined for their total lipid content and fatty acid profile. The total lipid content was 33.3 g/100 g and 81.0 g/100 g, respectively. Fatty acid profile of egg yolk and margarine is tabulated in Table 1. Approximately 25 different fatty acids were found in either egg yolk or margarine. Palmitic, stearic, oleic, and linoleic acids were the major fatty acids of egg yolk and margarine. However, unsaturated fatty acids were predominant in egg yolk and accounted for approximately 62.2% of total fatty acids. Meanwhile, 55.4% of total fatty acids of margarine were saturated fatty acids. Fatty acids of ω-9 and ω-6 were the unsaturated fatty acids predominantly present in both egg yolk and margarine.

Complexing indexes

Previous study has demonstrated that the maximum complex formation between starch and lipids takes place with the optimal ratio of both components [Tang & Copeland, 2007]. Lipids tend to self-associate when their addition to starch is above the optimal ratio. Therefore, it is important to choose an appropriate amount of lipid added to starch in experimental system used. Our preliminary study on the addition of egg yolk and margarine to banana flour at 1, 2, and 3 g/100 g showed that egg yolk at 2 g/100 g was above the optimal value while margarine at 3 g/100 g was below the optimal value. Further experimental work found that the addition of egg yolk and margarine less than 1.7 and 5 g/100 g, respectively, was within the acceptable range. To compare the effectiveness of margarine with egg yolk, their addition of 1 g/100 g of flour was selected.

TABLE 1. Fatty acid profile (% of total fatty acids) of egg yolk and margarine.

Fatty acid	Egg yolk	Margarine
α-Linolenic acid (18:3ω-3)	0.238	0.105
γ-Linolenic acid (18:3ω-6)	0.133	–
Linoleic acid (18:2ω-6)	16.3	8.32
Oleic acid (18:1ω-9)	41.1	35.9
Heptadecenoic acid (17:1)	0.159	0.017
Palmitoleic acid (16:1)	0.423	0.116
Arachidonic acid (20:4ω-6)	2.18	–
Caprylic acid (8:0)	–	0.675
Pentadecenoic acid (15:1)	0.090	–
Eicosatrienoic acid (20:3ω-6)	0.246	–
Myristoleic acid (14:1)	0.126	–
Eicosadienoic acid (20:2)	0.288	–
Docosahexaenoic acid (22:6ω-3)	0.469	–
Lignoceric acid (24:0)	–	0.077
Caproic acid (6:0)	–	0.049
Stearic acid (18:0)	9.34	5.57
Heptadecanoic acid (17:0)	0.213	0.099
Palmitic acid (16:0)	27.6	41.0
Pentadecanoic acid (15:0)	0.096	0.046
Behenic acid (22:0)	–	0.063
Myristic acid (14:0)	0.568	2.61
Heneicosanoic acid (21:0)	0.063	–
Lauric acid (12:0)	0.033	4.36
Eicosenoic acid (20:1)	0.303	0.120
Capric acid (10:0)	–	0.537
Arachidic acid (20:0)	0.027	0.386
Sum of ω-3 fatty acids	0.709	0.105
Sum of ω-6 fatty acids	18.8	8.32
Sum of ω-9 fatty acids	41.1	35.9
Sum of monounsaturated fatty acids	42.3	36.2
Sum of polyunsaturated fatty acids	19.8	8.42
Sum of saturated fatty acids	37.8	55.4
Sum of unsaturated fatty acids	62.2	44.6

The values of complexing indexes (CI) of starch of banana flour and lipids of egg yolk and margarine are presented in Table 2. The CI value for complexes of lipid-HMT flour starch increased with the increase in lipid content. The formation of lipid-starch complexes has been reported in previous studies using another lipid source such as palm oil [Farooq *et al*., 2018;

TABLE 2. Complexing indexes of gelatinised native and HMT-banana flours with various contents of egg yolk or margarine.

Lipid source	Content (g/100 g of flour)	Complexing index (%)	
		Native flour	HMT–flour
Egg yolk	0.3	–	2.03±0.32[c]
	1.0	2.56±0.18[B]	8.23±0.19[b,A,α]
	1.7	–	9.07±0.32[a]
Margarine	1	–	2.80±0.26[c,β]
	3	9.38±0.30[B]	11.2±0.65[b,A]
	5	–	15.9±0.72[a]

Uppercase letters compare values in the same row; lowercase letters compare values in the same column for egg yolk and margarine separately; Greek letters compare values for HMT-flours with egg yolk or margarine at the same addition level (1 g/100 g). Different letters denote significant differences at $p<0.05$.

Handarini *et al*., 2020a]. Our finding on the increase of the complexing index value with the increase in lipid content is in line with a previous study [Handarini *et al*., 2020a]. As mentioned above, when the ratio of lipids mixed with starch is above optimal value, CI may decrease due to the preference of lipids to self-associate [Tang & Copeland, 2007]. In the present study, the CI increased as the lipid content increased, confirming that the lipid contents in pastes did not exceed the optimal value.

In an inclusion model, the aliphatic part of lipids forming complexes with starch is located inside the helical cavity of amylose, whilst the polar group remains outside due to the steric and electrostatic repulsions [Godet *et al*., 1993]. Amylose can interact with two fatty acids in which the polar part of fatty acids is located at the end of each helix. Long amylose chains can even accommodate more than two fatty acids. The chains, however, are distorted to accommodate polar groups of fatty acids. The increase in CI in the present study with the increase of contents of egg yolk or margarine in pastes may indicate that the amylose chains might not be in a saturated state with the lipid so that they could accommodate more lipid to form a complex. At the content of 1 g/100 g, CI in egg yolk-HMT flour mixture was 8.23% which was much higher value than that of margarine-HMT flour (2.80%) (Table 2), suggesting that egg yolk containing more unsaturated fatty acids formed complexes with starch of banana flour more favourably than margarine. The CI value may also be affected by the presence of protein. Study on the effect of protein on starch-lipid complexes showed that the addition of protein promoted the formation of starch-lipid complexes [Cai *et al*., 2021]. Unfortunately, the content of protein in both egg yolk and margarine was not analysed in the present study. It is, therefore, difficult to assess the role of protein in the present study. The role of protein present in egg yolk on starch-lipid complex formation is worth further investigation.

The CI values of the complexes formed in pastes of native banana flour and egg yolk or margarine were 2.56 and 9.38%, respectively (Table 2). The CI increased to 8.23 and 11.2%, respectively, when HMT-flour was used at a corresponding lipid content. It suggests that more starch polymers are available to form complexes with lipids. HMT alters crystallinity and starch structure from B to A crystalline type [Cahyana *et al*., 2019; Marta *et al*., 2020]. These changes, however, may not be the driving force of the complex formation as the starch was mixed with lipid at high temperature (95°C) where the crystallites were melted and in amorphous phase when starch formed a complex with lipid. HMT also changes the content and chain lengths of the amylose fraction [Silva *et al*., 2017; Singh *et al*., 2011]. These changes may facilitate complex formation with lipid. Another study found that the other components present in the flour underwent an alteration following HMT treatment [Puncha-arnon & Uttapap, 2013], suggesting that apart from starch *per se*, the other components might affect the CI value of lipid-HMT banana flour compared to that of lipid-native flour.

Functional properties

The functional properties of pastes of native and HMT banana flour with different contents of lipids from two sources are presented in Table 3.

Swelling volume (SV)

The SV of pastes of HMT-flour and egg yolk or margarine decreased with the increase of their lipid contents (Table 3). The SV of HMT-flour with egg yolk ranged from 10.1 to 11.6 mL/g while that of HMT-flour with margarine from 8.27 to 9.25 mL/g. It suggests that the presence of both egg yolk and margarine lipids diminishes starch capacity to swell. The decrease in SV was concomitant with the increase in CI, therefore SV might be partly related to CI. A similar observation was also made for arrowroot starch-palm oil [Handarini *et al*., 2020a] and maize starch-stearic acid [Raphaelides & Georgiadis, 2006] mixtures.

The SV of paste of HMT-banana flour with egg yolk (1 g/100 g) was only slightly higher than that of HMT-flour mixed with margarine in the same proportion, even though the CI for the egg yolk complexes was significantly higher than that with margarine. This finding suggests that SV may be driven not only by complex formation but also by the nature of the lipid *per se*. Lipids in egg yolk are more hydrophilic due to the content of phospholipids. The presence of phospholipids may lead to higher water absorption and swelling volume.

Compared to the paste of native banana flour, the paste made of HMT- flour had lower SV values, *i.e.* 12.1 *vs.* 10.2 mL/g, respectively, when egg yolk was added and 11.6 *vs.* 8.92 mL/g, respectively, for pastes with margarine. This finding suggests that HMT plays a role in decreasing the swelling volume regardless of lipid type. Another study revealed that HMT of banana flour decreased its ability to swell compared to its native form along with a decrease in water absorption capacity [Cahyana *et al*., 2019], suggesting a link between the water absorption capacity and SV. In the present study, the CI values increased when the flour was pre-treated (HMT). This finding confirms that starch-lipid complex formation may play a role in decreasing SV of paste of banana flour with egg yolk or margarine.

Solubility

Trends in solubility of pastes of HMT-flour with egg yolk or margarine varied (Table 3). In the egg yolk-flour pastes,

TABLE 3. Functional properties of native and HMT-banana flour pastes with various contents of egg yolk or margarine.

Paste properties	Lipid source	Content (g/100 g of flour)	Banana flour	
			Native	HMT
Swelling volume (mL/g)	Egg yolk	0.3	–	11.6 ± 0.96^{a}
		1.0	12.1 ± 0.91^{A}	$10.2\pm0.19^{b,B,\alpha}$
		1.7	–	10.1 ± 0.73^{b}
	Margarine	1	–	$9.25\pm0.10^{a,\beta}$
		3	11.6 ± 0.32^{A}	$8.92\pm0.17^{b,B}$
		5	–	8.27 ± 0.10^{c}
Solubility (%)	Egg yolk	0.3	–	4.44 ± 0.28^{a}
		1.0	5.16 ± 0.95^{A}	$1.50\pm0.58^{b,B,\beta}$
		1.7	–	0.65 ± 0.48^{b}
	Margarine	1	–	$10.3\pm0.29^{b,\alpha}$
		3	11.5 ± 0.44^{A}	$10.7\pm0.49^{b,A}$
		5	–	12.4 ± 0.98^{a}
Freeze-thaw stability (% syneresis)	Egg yolk	0.3	–	32.4 ± 0.86^{a}
		1.0	8.34 ± 0.24^{B}	$20.9\pm0.45^{b,A,\beta}$
		1.7	–	21.0 ± 0.62^{b}
	Margarine	1	–	$42.8\pm1.37^{a,\alpha}$
		3	39.7 ± 0.53^{B}	$42.5\pm1.74^{a,A}$
		5	–	41.9 ± 1.45^{a}

Uppercase letters compare values in the same row; lowercase letters compare values in the same column for egg yolk and margarine separately; Greek letters compare values for HMT-flours with egg yolk or margarine at the same addition level (1 g/100 g). Different letters denote significant differences at $p<0.05$.

the solubility decreased from 4.44 to 0.65% with the increase in egg yolk content. Meanwhile, an opposite trend in solubility was observed with the increase in margarine content. Considering that CI of the mixture increased with the increase of egg yolk or margarine content, this opposite trend in solubility of egg yolk-banana flour pastes compared to margarine-banana flour pastes indicates that the solubility may not be linked to CI.

The solubility of gelatinised banana flour with a margarine content of 1 g/100 g was significantly higher than that with egg yolk added in the same amount. When measuring the solubility, supernatant was separated from the solid fraction following centrifugation, and then dried. Therefore, the remaining dry solid content following oven drying represents solids from margarine and amylose leaching into supernatant. The higher solubility of margarine-banana flour paste may be due to the formation of a lipid-starch complexes, which seems to be readily separated when solubility test was applied.

The solubility of pastes of native banana flour with lipids was higher compared to the solubility of pastes of HMT-flour either with egg yolk or margarine. In the absence of lipid, the solubility of breadfruit starch and banana flour was reported to increase following HMT [Cahyana *et al*., 2019; Marta *et al*., 2019a]. The solubility of HMT-banana flour in the presence of lipid may be affected by interplay factors of lipid and HMT effect. The interplay factors between HMT and lipid presence resulted in the decrease in solubility of egg yolk-HMT flour pastes suggesting that the lipid effect counteracted significantly the HMT effect. Meanwhile, in margarine-HMT flour pastes, the lipid effect on solubility was not statistically significant at margarine content below 3 g/100 g.

Freeze-thaw stability (FTS)

The increase of egg yolk content in the pastes of banana flour reduced significantly mixture syneresis from 32.4 to 21.0% (Table 3). However, the decrease of syneresis was not significant in the pastes of banana flour and margarine. The decrease of syneresis indicates an increase of FTS. Therefore the finding of syneresis reduction with the increase of lipid content, particularly egg yolk lipids, suggests that complex formation between lipids of egg yolk and starch of banana flour led to an increase of FTS.

The addition of egg yolk at 1 g/100 g to the banana flour caused 20.9% syneresis while the addition of margarine at the same content caused 42.8% syneresis, suggesting that egg yolk was more effective in increasing FTS than margarine. The higher FTS of pastes with egg yolk compared to these with margarine might be attributed to the extent of complex formation between banana flour and egg yolk or margarine. The CI of egg yolk-HMT flour was much higher than that of margarine. This higher CI means there were more egg yolk lipids to form a complex with starch polymers (amylose or amylopectin). During storage of the pastes at low temperature (as indicated in the FTS experiment), amylose and/or amylopectin move closer expelling water from the gel system (paste), which is quantified as a syneresis. The extent of complex formation may partly play a role in hampering the starch polymer from moving closer, hence in lower syneresis. In this context, egg yolk, which form complexes with banana flour starch with the higher CI than that of margarine, is effective in lowering syneresis. The effectiveness of egg yolk in reducing syneresis may also be linked to the different lipid composition in egg yolk compared to margarine.

The syneresis of paste of egg yolk-banana flour at the content of 1 g/100 g increased from 8.34% in the native flour to 21.0% in HMT-flour. A similar finding was also reported for margarine, suggesting that HMT of flour decreased FTS. Our finding is in line with previous studies on banana flour [Cahyana *et al*., 2019] and breadfruit starch in the absence of lipid [Marta *et al*., 2019a].

Pasting properties

The pasting properties of the mixtures of banana flour with egg yolk or margarine are presented in Figure 1 and tabulated in Table 4. The result shows that the increase in neither egg yolk nor margarine content affected pasting point. Similarly, the pasting points of mixtures with either egg yolk or margarine at 1 g/100 g did not differ significantly. This finding suggests that the addition of lipid did not play any role in determining the pasting point, and was in line with another study on arrowroot starch [Handarini *et al*., 2020a].

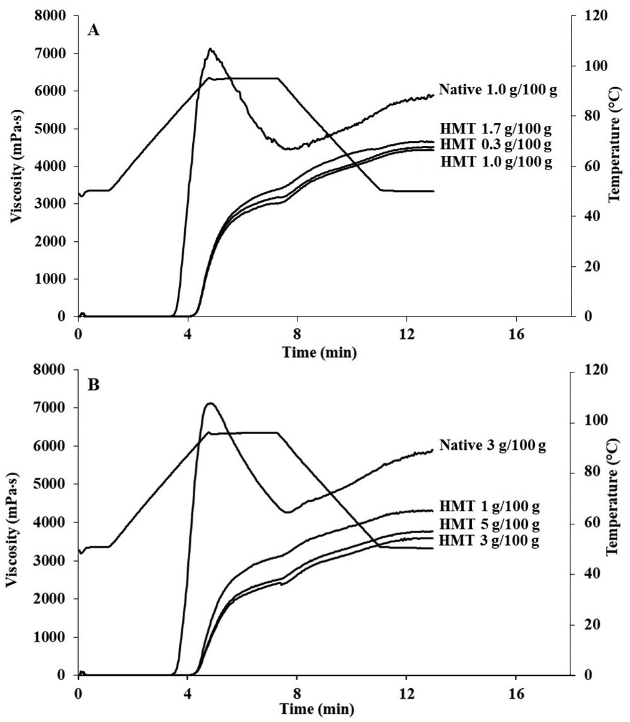

FIGURE 1. Pasting properties of native and heat-moisture-treated (HMT-flour) banana flour with various contents of (A) egg yolk (0.3–1.7 g/100 g) and (B) margarine (1–5 g/100 g).

Compared to the native flour-egg yolk mixture, the pasting point increased from 78.5°C to 87.3°C in HMT-flour. A similar finding was also made when margarine was used. It suggests that, contrarily to the lipid effect, HMT can change the pasting point. This change may relate to the modification of starch structure, which leads to the formation of more interactions and cross-links present within the granules following HMT. The more complex bonds may require higher temperature and energy to disrupt the starch structure [Huang *et al*., 2016; Zavareze & Dias, 2011]. Other studies reported that the pasting point of HMT-banana flour and breadfruit starch in the absence of lipid was higher than that of the native forms [Cahyana *et al*., 2019; Marta *et al*., 2019a].

Further examination on the other pasting properties showed that lipid addition either from egg yolk or margarine did not change the final and set back viscosity of their mixtures with banana flour. Hold viscosity was not affected by the presence of margarine while breakdown remained unchanged regardless of the egg yolk content. The addition of margarine at 5 g/100 g to HMT-flour increased the flour stability against heat and shearing during pasting, which can be attributed to the decrease in peak viscosity. The type of lipid was noted to affect the setback viscosity. Comparing the egg yolk to margarine addition at the same content (1 g/100 g), lipid from egg yolk resulted in higher set back viscosity than lipid from margarine. The other pasting properties were not affected by the type of lipid source.

In general, although a significant change was observed for several pasting properties at a certain level of lipid content, it is however considered to be small, particularly compared to the effect of HMT pretreatment. Compared to the mixture

TABLE 4. Pasting properties of native and HMT-banana flours with various contents of egg yolk or margarine.

Flour properties	Lipid source	Content (g/100 g of flour)	Banana flour	
			Native	HMT
Pasting point (°C)	Egg yolk	0.3	–	87.4 ± 0.32^{a}
		1.0	78.6 ± 0.09^{B}	$87.3\pm0.23^{a,B,\alpha}$
		1.7	–	87.2 ± 1.08^{a}
	Margarine	1	–	$87.5\pm0.47^{a,\alpha}$
		3	78.5 ± 0.29^{B}	$87.5\pm0.56^{a,A}$
		5	–	87.4 ± 0.23^{a}
Peak viscosity (mPa·s)	Egg yolk	0.3	–	$3139\pm140^{a,b}$
		1.0	7288 ± 230^{B}	$3030\pm155^{b,A,\alpha}$
		1.7	–	3375 ± 113^{a}
	Margarine	1	–	$3085\pm78.5^{a,\alpha}$
		3	6207 ± 30.1^{A}	$2724\pm291^{a,b,B}$
		5	–	$2730\pm21b^{b}$
Hold viscosity (mPa·s)	Egg yolk	0.3	–	$3098\pm147^{a,b}$
		1.0	4344 ± 210^{A}	$2979\pm162^{b,B,\alpha}$
		1.7	–	3328 ± 108^{a}
	Margarine	1	–	$3047\pm86.3^{a,\alpha}$
		3	4078 ± 307^{A}	$2688\pm293^{a,B}$
		5	–	2720 ± 238^{a}
Final viscosity (mPa·s)	Egg yolk	0.3	–	4477 ± 114^{a}
		1.0	5890 ± 172^{A}	$4436\pm101^{a,B,\alpha}$
		1.7	–	4647 ± 112^{a}
	Margarine	1	–	$4303\pm158^{a,\alpha}$
		3	5619 ± 14.2^{A}	$3973\pm334^{a,B}$
		5	–	4010 ± 215^{a}
Break down viscosity (mPa·s)	Egg yolk	0.3	–	40.8 ± 7.78^{a}
		1.0	2943 ± 192^{A}	$50.5\pm6.95^{a,B,\alpha}$
		1.7	–	47.3 ± 5.48^{a}
	Margarine	1	–	$37.5\pm7.78^{a,\alpha}$
		3	2129 ± 277^{A}	$36.0\pm3.22^{a,B}$
		5	–	10.3 ± 2.80^{b}
Set back viscosity (mPa·s)	Egg yolk	0.3	–	1379 ± 33.2^{a}
		1.0	1546 ± 127^{A}	$1457\pm81.1^{a,A,\alpha}$
		1.7	–	1319 ± 57.2^{a}
	Margarine	1	–	$1256\pm71.4^{a,\beta}$
		3	1541 ± 293^{A}	$1285\pm46.0^{a,A}$
		5	–	1288 ± 26.0^{a}

Uppercase letters compare values in the same row; lowercase letters compare values in the same column for egg yolk and margarine separately; Greek letters compare values for HMT-flours with egg yolk or margarine at the same addition level (1 g/100 g). Different letters denote significant differences at $p<0.05$.

in native flour, a remarkable decrease in peak viscosity was observed in HMT-flour either with egg yolk or margarine, suggesting the preponderant contribution of HMT pretreatment on the decrease of peak viscosity. Huge decreases were also noticed for other pasting properties, such as hold and final viscosity, and particularly breakdown viscosity. Breakdown decreased incredibly from 2943 mPa·s in egg yolk or 2129 mPa·s in margarine to 50.5 mPa·s or 36.0 mPa·s when the flour was subjected to HMT pretreatment. It suggests that HMT improves heat stability of the flour. The degree of break down changes due to the difference in lipid content was much smaller than due to HMT pretreatment, suggesting the main contribution of the change in starch structure following HMT. The role of HMT on the pronounced decrease of pasting properties was demonstrated in other studies on banana flour and breadfruit starch in the absence of lipid [Cahyana *et al*., 2019; Marta *et al*., 2019a].

CONCLUSIONS

Both egg yolk and margarine are capable of forming complexes with either native or HMT-banana starch. The CI value of lipid-starch increased with the increase of margarine or egg yolk content in pastes. Lipids from egg yolk were more favorable to form complexes compared to margarine. HMT increased flour ability to form complexes with lipids, which might be attributed to reduction in amylose chain lengths and the increase in amylose content following HMT.

The increase in the content of both egg yolk and margarine in HMT-banana flour pastes decreased SV and syneresis, while the solubility decrease was only affected by the increase in the egg yolk content. Comparing the native banana flour paste and HMT banana flour paste with the same lipid content, a decrease of SV, solubility, and FTS was found. Lipid source affected the extent of the change in the functional properties of banana flour pastes in which egg yolk resulted in higher SV but lower solubility and syneresis compared to margarine pastes. SV and FTS may be partly linked to CI.

The interplay role of HMT of banana flour and lipid content in pastes was clearly observed in the HMT-flour mixed with egg yolk in which the lipid effect thwarted the opposite HMT effect significantly, hence a decreased solubility. In respect of FTS, the reducing effect of lipid on syneresis was seemingly smaller than the increasing effect of HMT leading to an increased syneresis when lipid was mixed with HMT flour.

Although a statistically significant change was observed for certain pasting properties with the increase in lipid content, it was much smaller than that due to the HMT pre-treatment. HMT plays a major role in the alteration of pasting properties. Lipid source at 1 g/100 g of neither egg yolk nor margarine affected pasting properties except set back viscosity.

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CONFLICT OF INTERESTS

The authors declare no conflict of interest

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