Maize and indigenous African teff starches modified with stearic acid as potential fat replacer in low calorie mayonnaise-type emulsions

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ABSTRACT

Fat replacers have been developed to produce low-calorie foods due to the association of fat-rich diet to lifestyle diseases. Teff (an underutilized and under-researched cereal), and maize starch pastes modified with stearic acid could be used as fat replacers because of their reduced gelling ability and higher viscosity. The effect of teff and maize starch pastes modified with stearic acid on the rheological properties, microstructure, freeze-thaw and high temperature stability of low-calorie mayonnaise type emulsions (LCMTE) was investigated. Starch suspensions (10% w/v) containing stearic acid (1.5%) were pasted for an extended holding time (2 hr at 91 °C) in a Rapid Visco Analyzer (RVA) and used to prepare LCMTE with 50% and 80% oil replacement. LCMTE with modified teff and maize starches had lower yield stress and viscosity and larger oil droplets compared to LCMTE with unmodified teff and maize starches. Increasing oil replacement level (50% to 80%) increased the viscosity. LCMTE with maize starch had higher yield stress and viscosity and smaller oil droplets than LCMTE with teff starch. All samples showed shear thinning behaviour ($n < 1$). All the LCMTE were more stable to freeze-thaw cycles and high temperature storage than full fat mayonnaise. At 50% oil replacement, unmodified and modified teff and maize starch with stearic acid could produce LCMTE. When the oil content was further decreased to 80% only the LCMTE with modified starches were similar to the full fat.

Keywords: Fat replacer; Low-calorie mayonnaise type emulsions; Amylose-stearic acid complex; Teff starch

1. Introduction

Production of low-calorie foods has been developed due to the positive relation between lifestyle diseases such as diabetes and cardiovascular diseases and consumption of high fat foods [1, 2, 3]. The design of low-calorie food necessitates the use of fat replacers to mimic some of the functional properties of fat. Otherwise the low calorie foods will have inferior sensory properties. Mayonnaise is considered as a high calorie food as it contains over 70% oil [4, 5]. It is used extensively as a sauce and condiment in various dishes and in most fast food outlets.

Starch, a natural biopolymer, can be used as fat replacer. Teff [*Eragrostis tef* (Zucc.) Trotter], a highly underutilized tropical crop indigenous to Ethiopia, is a potential cereal source of starch. Teff starch has small granules (2-6 μm) and has slightly different properties compared to other tropical cereals (e.g. maize). Teff has been suggested to be a good fat mimetic [6]. Even though native starch can sometimes be used as fat replacer, it is usually modified by physical or chemical methods for this purpose. This is because native starch has some limitations for food use like low thermal resistance, high tendency towards retrogradation and rubbery weak bodied gels [7, 8]. Chemical and physical modification of starch can improve its properties such as thickening, binding, mouthfeel, gelling, dispersion or cloud formation [7]. Stearic acid has been found to modify maize and teff starch pasting properties [9]. Stearic acid added to a level of about 1.5% (w/w of starch) can increase the pasting viscosity twice during pasting for more than 1 hr. [9] also found teff and maize starch pastes modified with stearic acid to have a non-gelling behaviour compared to the gelling behaviour of the unmodified starches. [9] proposed that the modified teff starch with stearic acid can be used as fat replacer in high fat foods because of its non-gelling behaviour and increased viscosity. However research has not been conducted to test this proposition.

The objective of this study was to determine the effect of teff and maize starch pastes modified with stearic acid on the rheological properties and microstructure of low-calorie mayonnaise type emulsion.

2. Materials and Methods

2.1 Samples

A white teff variety (Witkop) was obtained from PANNAR, Kroonstad, South Africa. Commercial maize starch, Amyral (from a white maize variety) was obtained from Tongaat Hulett®, Edenvale, South Africa. The starch, protein, ash, crude fat of the commercial maize starch were 91, 0.54, 0.13, and 0.01% (dry basis) respectively. Pasteurized and spray dried egg yolk powder, product code YLKPW 500, was obtained from Ovipro (PTY) Ltd., Bronkhorstpruit, South Africa. Stearic acid was obtained from Sigma-Aldrich (product code S 4751 – 25 G). Sunflower oil, white spirit vinegar, salt and sugar were bought from a local supermarket. All chemicals other than those bought from local supermarket were of analytical grade unless specified otherwise.

2.2 Starch extraction

Teff starch was extracted by the methods of [9]. Sieved teff grain was milled in a laboratory hammer mill to pass through an 800 μm screen. The flour was defatted by adding hexane at 25^{o}C and stirred for 1 h (1 part flour: 3 parts hexane). The slurry was then wet milled with a Retsch Mill ZM 200 (Haan, Germany) and passed through a 250 μm screen. The filtrate was retained and filtered with 75 μm and 38 μm hand sieves sequentially. The filtrate was then centrifuged at 8000 rpm for 10 min at 25^{o}C to remove the brown protein layer. This procedure was repeated by adding distilled water to the remaining starch pellet until a white starch pellet was obtained. The white starch pellets were then freeze dried. The starch, protein, ash and crude fat of the extracted teff starch were 88, 1.3, 0.38, and 0.03% (dry basis) respectively.

2.3 Incorporation of stearic acid into starch

Stearic acid (1.5% on the weight basis of dry starch) was dissolved in ethanol in a beaker. Teff or maize starch was then added to the solution in a 1: 3 (wt: vol.) starch: ethanol ratio. The beaker was then covered with Parafilm and foil and placed in a shaking water bath at 50°C for 30 min. The ethanol was then evaporated off in a force draught oven at 40°C. The control starch samples were treated exactly the same, but without the addition of stearic acid.

2.4 Starch pasting (extended holding time)

Starch (2.8 g dry basis) was suspended in distilled water to get a total of 28 g starch-water suspension. The suspension was pasted using a Rapid Visco Analyser (RVA model 3D) (Newport Scientific, Warriewood, Australia). The pasting condition used was: initial stirring at 960 rpm at 50^{o}C for 30 s and thereafter at 160 rpm for the entire period. The temperature was stabilized at 50^{o}C for 2 min and was increased from 50^{o}C to 91^{o}C at a rate of 5.5^{o}C/min, and then held at 91^{o}C for 1 h 51 min. The total time for the whole process was 2 h. The weight of the paste was readjusted to the original weight (28 g) with distilled water and used to replace oil in low-calorie mayonnaise type emulsions (LCMTE).

2.5 Emulsion preparation

The aqueous phase (vinegar, egg yolk, salt and sugar and water or starch paste for low-calorie mayonnaise type emulsions) was mixed first and oil was added little by little while continuously homogenising it at 8000 rpm using an Ultra Turrax T25 (JANKE & KUNKEL IKA® Labortechnik) for 5 min at room temperature. The mayonnaise formulations were similar to [10], where only the percentages of the ingredients were adjusted as in Table 1 because of the level of oil replacement. In the low-calorie mayonnaise type emulsions the oil was replaced at 50 and 80% levels. The mayonnaise samples were allowed to settle for 30 min before analysis.

2.6 Flow Property measurement

Steady shear flow measurements were conducted with a Physica MCR 301 Rheometer (Anton Paar, Ostfildern, German) using a vane method. The mayonnaise samples were allowed to equilibrate for 30 min at 25°C. Shear rate was increased from; 0.01 s^{-1} to 1000 s^{-1} and reduced back from 1000 s^{-1} to 0.01 s^{-1}. The measurements were taken at 25°C. The experimental data were fitted to the Herschel-Bulkley model:

$$\sigma = \sigma_o + K (\dot{\gamma})^n$$

Where σ_o is the yield stress, σ is the shear stress (Pa), $\dot{\gamma}$ is the shear rate (1/s), K is the consistency index ($Pa.s^n$) which is an indication of the viscosity, and n is the flow index where n=1 for Newtonian fluid, n<1 for shear thinning and n>1 for shear thickening materials.

2.7 Confocal Laser Scanning Microscopy (CLSM)

Nile Red (Sigma Aldrich) was used to stain the oil phase and Nile blue (BDH Chemicals ltd, Poole, England) was used to stain the aqueous phase. Mayonnaise samples were prepared as described above, but this time a 0.01% (w/v) aliquot of Nile Red solution was dissolved in the oil, and 0.01% (w/v) of Nile blue solution in the vinegar before the mayonnaise preparation.

From each sample a small amount of mayonnaise was placed on a concave microscope slide, covered with cover slip and analyzed on a Zeiss LSM 510 META Confocal Laser Scanning Microscope (Zeiss SMT, Jena, Germany) at 40X magnification. Plane neoflar 100 x and Numerical aperture (N.A) 1.4 was used. The pixel time for both tracks 1 and 2 was 12.8 µs.

Picture size was 512 x 512 pixels. The excitation and emission spectra for Nile Red were 488 nm and 550 – 603 nm respectively and for Nile blue 633 nm and 668 – 753 nm.

2.8 Freeze thaw stability

Forty grams of each sample was weighed into 100 ml plastic centrifuge tubes. The mayonnaise samples were centrifuged at 500 rpm for 30 min. The amount of separated liquid was weighed. The samples were then frozen at -18^{o}C for 24 h then thawed at 30^{o}C for 1 h and centrifuged at 500 rpm for 30 min. The amount of liquid separated was weighed. This procedure was repeated for ten consecutive cycles and freeze thaw stability at each cycle was calculated as:

$$\frac{\text{(Original weight)} - \text{(Weight of separated liquid)}}{\text{Original weight}} \text{X100}$$

2.9High Temperature Stability

Each mayonnaise sample was weighed six times into six eppendorf tubes with 2 g in each tube and stored at 50^{o}C for 0 h, 24 h, 7 days, 14 days, 21 days, and 28 days. Each time the samples were centrifuged at 3000 rpm for 10 min. The amount of liquid separated was weighed and the high temperature stability was calculated as:

$$\frac{\text{(Original weight)} - \text{(Weight of separated liquid)}}{\text{Original weight}} \text{ X 100}$$

2.10 Statistical analysis

Main effects and interaction effects of treatments [type of starch used to replace the oil (teff and maize), oil replacement level (0%, 50% and 80%), and modification of starch (unmodified and modified with 1.5% stearic acid)] were calculated by Analysis of Variance (ANOVA). Means were then compared using Tukey (HSD) test. A t-test was then performed to compare the mean value at D_0 (30 min after preparation) and D_8 (after 8 days of storage at room temperature). The experiments were repeated three times unless stated otherwise.

3. Results

3.1 Flow Properties

The flow properties of low-calorie mayonnaise type emulsions (LCMTE) measured 30 min and 8 days after preparation is shown in supporting information Figure and Table 2-4. Full fat mayonnaise (reference sample) and a 50% oil reduced product with no starch added were also used for comparison purposes.

The apparent viscosity versus shear rate graphs (supporting information Figure) showed a shear thinning behaviour of all samples studied. The apparent viscosity decreased as the shear rate increased for all samples. The 50% oil reduced product with no starch added had exceptionally low viscosity. This product also showed visual phase separation immediately after preparation. Thus, this emulsion was not considered for further discussion. To characterize the flow properties of mayonnaise, the Herschel-Bulkley model [5, 11] was used. The Herschel-Bulkley model has been largely used in mayonnaise as yield stress is considered [12]. The Herschel-Bulkley model parameters are summarized in Tables 2 to 4 (and the supporting information table).

The experimental data showed a good fit to the model as the determination coefficients (R^2) were higher than 0.90 (0.94 – 0.99). When level of oil replacement increased from 50% to 80% the LCMTE yield stress generally decreased (Table 2). LCMTE with stearic acid-modified starches had lower yield stress ($P<0.05$) compared to LCMTE with unmodified starches (Table 2 - main effect starch modification). LCMTE with teff starches had lower yield stress than LCMTE with maize starch (Table 2 - main effect type of starch). After 8 days storage, the yield stress of most LCMTE increased except for LCMTE with modified teff starch at 80% oil replacement and LCMTE with modified maize starch at 50% oil replacement.

The consistency coefficient (K) value increased with increase in the level of oil replacement for all the samples (Table 3). The use of modified starches with stearic acid seemed to decrease the consistency coefficient (K) value compared with unmodified starches. LCMTE with teff starches had lower ($P<0.05$) consistency coefficient (K) value than LCMTE with maize starches (Table 3 - main effect type of starch). Storage for 8 days seemed to increase the consistency coefficient (K) value of most LCMTE except a decrease ($P<0.05$) for LCMTE with modified teff starch at 80% level of oil replacement.

All samples studied had a value of $n<1$ (supporting information Table) indicating all samples were shear thinning. The apparent viscosity versus shear rate curves also showed shear thinning behaviour (supporting information Figure). Generally the n-values were not affected significantly by level of oil replacement, type of starch (supporting information Table - main effect type of starch) and modification with stearic acid (supporting information Table - main effect starch modification) except a significant decrease ($P<0.05$) for modified teff starch with stearic acid. Storage for 8 days seemed to decrease the n-value of most LCMTE except LCMTE with modified teff starch at 80% oil replacement level.

All the mayonnaise samples studied were found to have a thixotropic behaviour. The magnitude of hysteresis loop area corresponds to an ability to rebuild the damaged structure after the removal of shear forces. A lower hysteresis loop area of mayonnaise corresponds with its ability to rebuild the damaged structure faster after removal of the shear force and vice versa [13]. Generally the hysteresis loop area increased (became less stable) with increase in level of oil replacement (Table 4). The addition of stearic acid-modified starch decreased the hysteresis loop area (become more stable). Type of starch did not affect the hysteresis loop area significantly (Table 4 - main effect type of starch), but it seems that teff starches had lower hysteresis loop.

Storage for 8 days increased ($P<0.05$) the hysteresis loop area (became less stable) of most LCMTE except no significant change for modified teff and maize starches with stearic acid at 80% replacement level.

3.2 CLSM of LCMTE

Full fat mayonnaise after 24 hr of preparation showed closely packed and uniformly distributed oil droplets of about 5-10 μm (Figure 1). The 50% oil-reduced product with no starch added had large non uniform oil droplets of about 20-40 μm, and larger proportion of aqueous phase compared to full fat mayonnaise. This suggests phase separation. When the level of oil replacement increased from 50% to 80%, the oil phase decreased because of decrease in oil content (Figure 1). Compared to full fat mayonnaise, LCMTE with 50% and 80% oil replacement level had smaller oil droplets (1-10 μm) except LCMTE with modified teff starch (10-20 μm) (Figure 1). LCMTE with modified starches at 50% and 80% oil replacement levels had larger size oil droplets compared to LCMTE with unmodified starches. LCMTE with maize starches seem to have smaller oil droplets compared to LCMTE with teff starches (Figure 1) especially with the modified starches. After storage for 8 days, the oil droplets size increased in full fat mayonnaise, the 50% oil-reduced product with no starch added and LCMTE with modified teff starch at 50% and 80% oil replacement levels (Figure 2).

3.3 Freeze-thaw and high temperature stability

Figure 3 shows the amount of phase separation during nine consecutive freeze-thaw cycles. 50% oil-reduced product with no starch added showed phase separation (60% phase separation) at the initial stage, i.e. before any freeze-thaw cycle (Figure 3). Full fat mayonnaise showed phase separation at the 1st freeze-thaw cycle (60% phase separation). Generally with increase in oil replacement level from 50% to 80% the phase separation had decreased at each freeze-thaw cycle. LCMTE with modified starches showed more phase separation than LCMTE with unmodified starches at 50% oil replacement level. At 80% oil replacement level all samples except LCMTE with modified teff starch showed no phase separation after 10 freeze-thaw cycles. LCMTE with maize starches showed less phase separation than LCMTE with teff starch at 50% oil replacement level.

Figure 4 shows the high temperature stability of the mayonnaise type emulsion. The 50% oil-reduced product with no starch added showed phase separation (50.5% phase separation) after 10 min storage at 50^{o}C (Figure 4). Full fat mayonnaise also showed some phase separation after 10 min storage at 50^{o}C (9.5% phase separation). LCMTE with modified teff starch showed phase separation after 14 days storage at 50^{o}C (10.5% phase separation) at 50% oil replacement level. LCMTE with modified teff starch at 80% oil replacement level and all the other LCMTE at both levels of oil replacement did not seem to have phase separation.

4. Discussion

LCMTE with modified and unmodified maize starch at 50% and 80% oil replacement levels had higher yield stress and consistency coefficient (K) value except LCMTE with modified teff starch (Tables 2 and 3) compared to the full fat mayonnaise. The possible reason could be the increase in viscosity of the continuous phase. Starch polymers can interact with water due to its hydrophilic nature and form structural/entangled network to increase viscosity and body [14, 15]. The smaller oil droplet size of LCMTE with teff and maize starches compared to full fat mayonnaise can also be related to the higher viscosity of the LCMTE compared to full fat mayonnaise (Figure 1). Fine emulsions have much higher viscosities than the corresponding coarse emulsions [16].

The use of stearic acid-modified starches produced LCMTE with lower yield stress, lower consistency coefficient K and a lower hysteresis loop compared with LCMTE made with unmodified starches (Tables 2, 3 and 4). This suggests that the addition of the modified starches produce a more flowable mayonnaise with better stability. This could be because of the properties of the modified starch pastes. D'Silva et al.[9] found addition of stearic acid to teff and maize starches led to formation of a soft, non-gelling paste while the control teff and maize starch pastes formed a gel upon cooling. They also suggested that addition of stearic acid may result in the formation of amylose-lipid complex which could hinder the re-association of amylose molecules. The lower viscosity of the LCMTE with modified starches compared to the LCMTE with unmodified starches can also be related to the microscopy. LCMTE with unmodified starches had smaller fat globules and as stated above this can be related to the higher viscosity.

Teff and maize starches modified with stearic acid seem to be better stabilizers resulting in LCMTE with lower hysteresis loop area compared to LCMTE with unmodified teff and maize starches (Table 4). This could be due to the less gelling tendency of the starch pastes modified with stearic acid compared to unmodified starch pastes [9]. In LCMTE with unmodified teff/maize starch pastes (which have more gel-like structure), an increase in shear rate will break down the gel structure and rebuilding the gel structure will take more time (higher hysteresis loop area). Santipanichwong and Suphantharika [17] added curdlan to an oil-in-water emulsion and the emulsion had gel-like strength and a very high hysteresis loop area.

LCMTE made with teff starches had lower yield stress, lower consistency coefficient K, lower shear thinning index n and lower hysteresis loop compared to LCMTE made with maize starches (Tables 2-4). These lower viscosities of LCMTE made with teff starches can also be related to the larger oil droplets when compared to LCMTE made with maize starches. The possible reason for these lower viscosities could be the lower retrogradation tendency of teff starch compared to maize starch to form a relatively weaker network [6]. A lower retrogradation tendency can produce a weaker continuous phase and hence lower viscosity [18]. The lower shear thinning behaviour (higher n-value) of LCMTE with teff starch compared to LCMTE with maize starch (Supporting information Table) could be due to lower shear thinning [9] and the higher resistance of teff starch to breakdown compared to maize starch [19]. LCMTE with teff starch had lower hysteresis loop area (more stable) than LCMTE with maize starch (Table 4) and this can be related to the higher stability of teff starch to breakdown compared to maize starch [19].

Storage for 8 days at room temperature increased the viscous properties in terms of higher yield stress and consistency coefficient (K) values of LCMTE with unmodified teff and modified and unmodified maize starches (Tables 2 and 3). During storage further retrogradation will occur mainly due to the re-crystallization of the amylopectin fraction [20] and retrograded starches generally increase viscosity. However, LCMTE with teff starch modified with stearic acid behaved differently. This needs further investigation.

LCMTE with modified and unmodified starches had better freeze-thaw stability than the full fat mayonnaise (Figure 3). The possible reasons for this could be the hydrophilic characteristics of the starch pastes may stabilize water from crystallization and the starch pastes could also keep the oil droplets far apart from each other protecting possible droplet coalescence. Thanasukarn et al. [21] studied the freeze-thaw stability of palm oil-in-water emulsion stabilized with different emulsifiers, Tween 20, casein and whey protein isolate and found emulsions stabilized with both casein and whey protein were stable to freeze-thaw cycles compared to emulsion stabilized with Tween 20. These differences were attributed to the fact that Tween 20 forms a thin interfacial membrane, whereas the proteins form relatively thick membranes. The non-gelling behaviour of modified teff and maize starches with stearic acid could have allowed the oil droplets to come together and promote droplet-droplet interaction which leads to coalescence and phase separation.

The LCMTE with modified and unmodified starches were more stable to high temperature storage (lower phase separation) compared to the full fat mayonnaise (Figure 4). The possible reason for this could be the addition of the starch pastes which increase the viscosity of the aqueous phase which slows down the droplet motion and could also form a protective layer around the oil droplets protecting the solubilisation of the surfactant. Mun et al. [11] used 4αGTase-modified rice starch and xanthan gum in reduced fat mayonnaise and found the reduced fat mayonnaise with 4αGTase-modified rice starch to be more stable to high temperature storage than the full fat mayonnaise. They proposed that this was due to slow oil droplet movement because of viscosity increase of the aqueous phase by the addition of the starch. Ogawa et al. [22] added a secondary chitosan layer above the lecithin layer in oil-in-water emulsions and found the emulsion with the secondary layer to be stable to high temperature storage up to 90^{o}C.

5. Conclusions

LCMTE can be produced with modified and unmodified teff and maize starches, but the properties depend on the level of oil reduction. Modified and unmodified teff and maize starch with stearic acid can produce LCMTE at 50% oil replacement. When the oil content is further decreased to 80% only LCMTE made with stearic acid-modified starches are the most similar to the full fat product in terms of physical properties. Generally LCMTE can be produced with the modified starches at 80% of the oil replacement and this can substantially decrease the calorific value of mayonnaises by up to 76.44% compared to the full fat mayonnaise.

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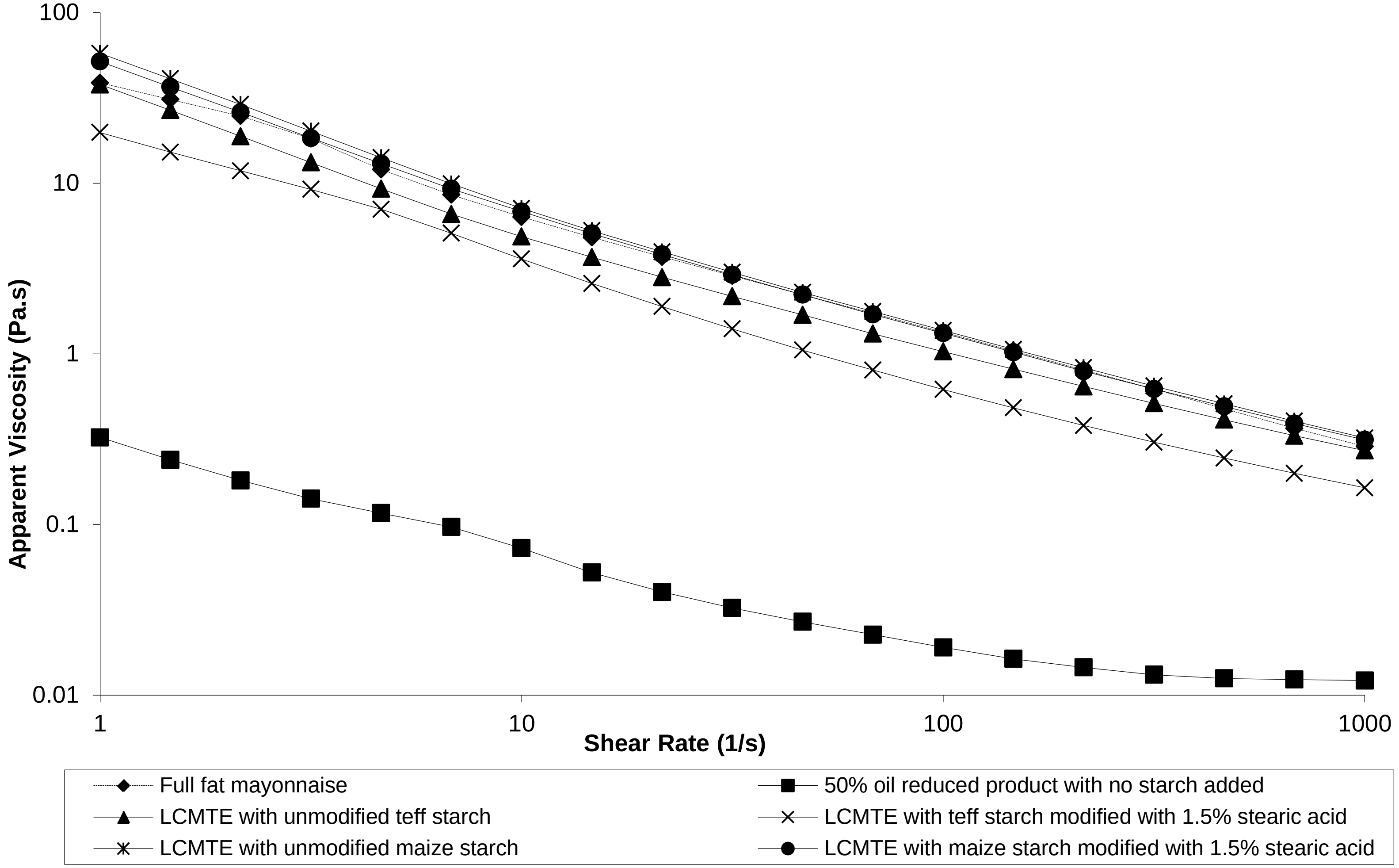
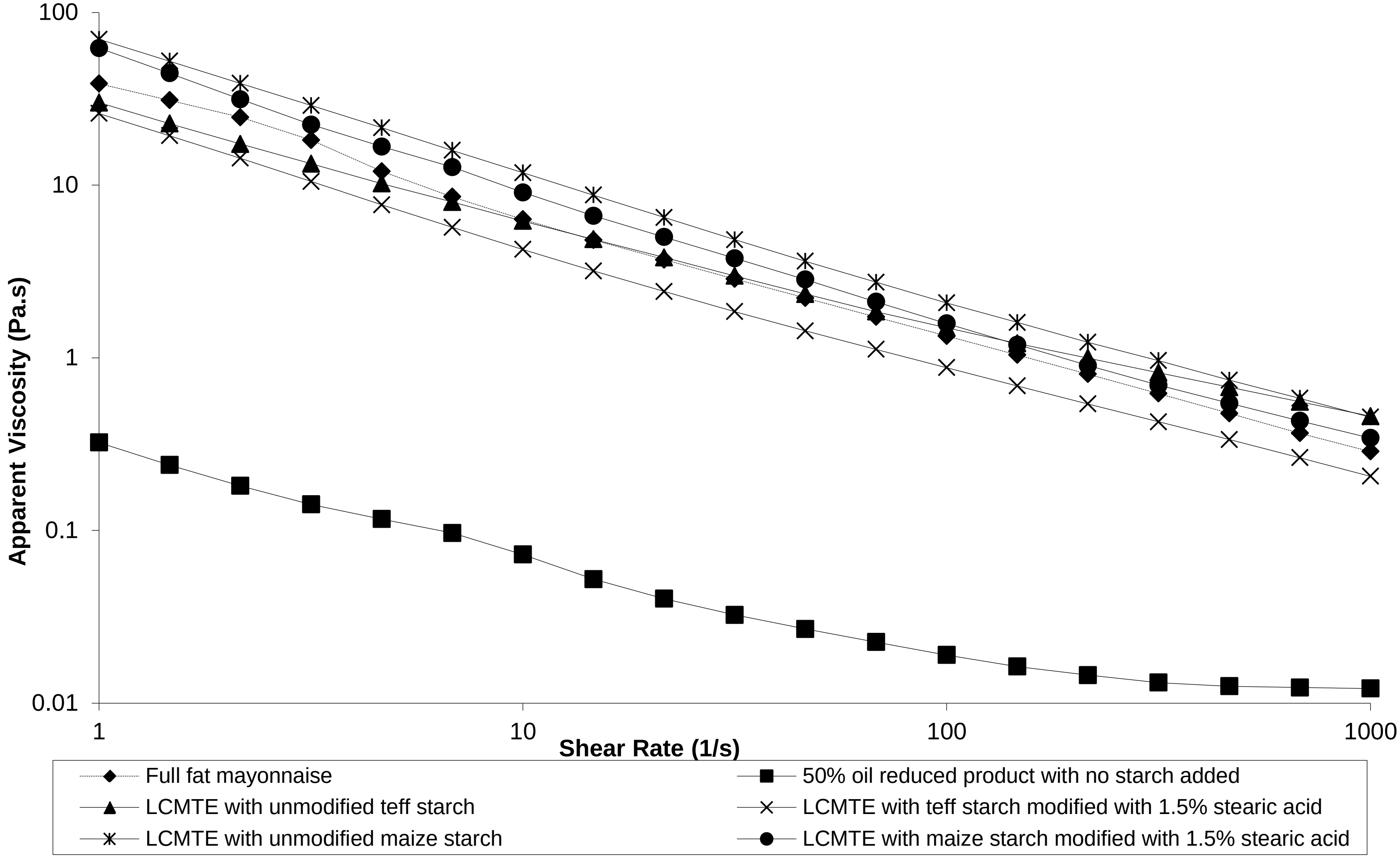


Effect of oil replacement with teff and maize starch pastes on the viscous properties of low calorie mayonnaise type emulsions (LCMTE) at 50% oil replacement level at different shear rates.



Effect of oil replacement with teff and maize starch pastes on the viscous properties of low calorie mayonnaise type emulsions (LCMTE) at 80% oil replacement level at different shear rates.

***the viscosity vs shear rate curves for full fat mayonnaise and 50% oil reduced product with no starch added was repeated for comparison**.

Extra Table: Flow behaviour index (n) values of mayonnaise samples, measured 30 minutes after preparation (D_0) and after 8 days of storage (D_8) at room temperature

Storage time				
Day 0				
Full Fat Mayonnaise		0.43 (0.02)		
Low calorie mayonnaise type emulsions				
Type of starch	Level of oil replacement	Unmodified starch	Modified starch with stearic acid	***Main effect types of starch***
Teff	50	0.54 (0.02) A	0.44 (0.003) A	0.48 (0.01) A
	80	0.51 (0.01) A	0.42 (0.01) A	
Maize	50	0.49 (0.01) A	0.51 (0.01) A	
	80	0.39 (0.02) B	0.45 (0.07) A	0.46 (0.02) A
Main effect modification			Teff	Maize
	Unmodified		0.52 (0.01) A	0.44 (0.03) A
	Modified with stearic acid		0.43 (0.01) B	0.48 (0.03) A
Day 8				
Full Fat Mayonnaise		0.44 (0.01)		
Low calorie mayonnaise type emulsions				
Type of starch	Level of oil replacement	Unmodified starch	Modified starch with stearic acid	***Main effect types of starch***
Teff	50	0.44 (0.03) A	0.41 (0.01) A*	0.38 (0.03) A*
	80	0.23 (0.01) B*	0.44 (0.02) A	
Maize	50	0.40 (0.01) A*	0.44 (0.01) A*	
	80	0.16 (0.03) B*	0.20 (0.03) B*	0.30 (0.04) A*
Main effect modification			Teff	Maize
	Unmodified		0.33 (0.05) A*	0.28 (0.06) A*
	Modified with stearic acid		0.43 (0.01) A	0.32 (0.06) A*

- ***Values in parenthesis indicate standard error of the mean***
- ***Means followed by the same letter/s within a block are not significantly different (($P < 0.05$), Tukey (HSD) test.***
- ***n - values followed with * are significantly different from the corresponding values measured at D_0***

Table 1: Formulations of full fat mayonnaise and low-calorie mayonnaise type emulsion samples (wt %)

	Full fat	Low-calorie without fat replacer	Low-calorie 50% oil replacement	Low-calorie 80% oil replacement
Sunflower oil	70	35	35	14
Egg yolk	12	12	12	12
Vinegar	15	15	15	15
Salt	2	2	2	2
Sugar	1	1	1	1
Water	-	35	-	-
Starch pastes*	-	-	35	56

*Teff and commercial maize starch pastes, unmodified and modified with 1.5% stearic acid pasted for 120 min (solid content is paste was 10% w/v).

'-' indicates 0% of the ingredient was added

Table 2: Yield stress values (Pa) of mayonnaise samples measured 30 minutes after preparation (D_0) and after 8 days of storage (D_8) at room temperature

Storage time

Day 0				
Full Fat mayonnaise	28.09 (1.23)			
Low calorie mayonnaise type emulsions				
Type of starch	**Level of oil replacement**	**Unmodified starch**	**Modified starch with stearic acid**	***Main effect types of starch***
Teff	50	28.91 (2.9) A	16.56 (0.8) A	
	80	18.99 (2.2) A	7.72 (1.2) B	18.04 (2.42) B
Maize	50	46.78 (1.8) A	44.46 (3.0) A	
	80	30.64 (1.2) B	19.44 (1.2) B	35.33 (3.44) A
Main effect modification		Teff	Maize	
	Unmodified	23.95 (2.75) A	38.71 (3.74) A	
	Modified with stearic acid	12.14 (2.07) B	31.95 (5.78) A	

Day 8				
Full fat mayonnaise	27.7 (2.1)			
Low calorie mayonnaise type emulsions				
Type of starch	**Level of oil replacement**	**Unmodified starch**	**Modified starch with stearic acid**	***Main effect types of starch***
Teff	50	55.3 (4.5) A*	21.1 (2.1) A	
	80	21.6 (3.1) B	4.0 (0.2) B*	25.50 (5.75) B
Maize	50	49.07 (2.4) A	34.6 (2.3) A	
	80	50.27 (2.3) A*	23.7 (0.4) B*	39.40 (3.42) A
Main effect Modification		Teff	Maize	
	Unmodified	38.46 (7.93) A	49.67 (1.51) A *	
	Modified with stearic acid	12.55 (3.92) B	29.14 (2.63) B	

- ***Values in parenthesis indicate standard error of the mean***
- ***Means followed by the same letter/s within a block are not significantly different ((P < 0.05), Tukey (HSD) test.***
- ***Yield stress values followed with * are significantly different from the corresponding values measured at D_0***

Table 3: Consistency coefficient (K) values ($Pa.s^n$) of mayonnaise samples measured 30 minutes after preparation (D_0) and after 8 days of storage (D_8) at room temperature

	Storage time: Day 0	Storage time: Day 8
Full Fat Mayonnaise	15.63 (0.3)	14.4 (0.5)

Low calorie mayonnaise type emulsions

Type of starch	Level of oil replacement	Day 0: Unmodified starch	Day 0: Modified starch with stearic acid	Day 0: *Main effect types of starch*	Day 8: Unmodified starch	Day 8: Modified starch with stearic acid	Day 8: *Main effect types of starch*
Teff	50	6.08 (0.73) B	6.62 (0.20) B	8.54 (0.82) B	11.38 (1.24) B*	7.32 (0.26) A	24.94 (8.68) B
	80	12.72 (0.80) A	8.74 (0.23) A		74.59 (2.1) A*	6.49 (0.38) A*	
Maize	50	9.24 (0.91) B	8.05 (0.65) B	17.31 (3.21) A	16.64 (1.83) B*	10.4 (0.65) B	88.65 (28.59) A*
	80	34.45 (1.35) A	17.51 (0.94) A		245.5 (0.3) A*	82.0 (0.65) A*	

Main effect modification	Day 0: Teff	Day 0: Maize	Day 8: Teff	Day 8: Maize
Unmodified	9.40 (1.56) A	21.84 (5.68) A	42.98 (14.17) A*	131.08 (51.19) A
Modified with stearic acid	7.68 (0.49) A	12.78 (2.18) A	6.90 (0.28) B	46.22 (15.99) A

- ***Values in parenthesis indicate standard error of the mean***
- ***Means followed by the same letter/s within a block are not significantly different ((P < 0.05), Tukey (HSD) test.***
- ***Herschel-Bulkley consistency coefficient (K) values followed with * are significantly different from the corresponding values measured at D_0***
- ***'-' Unrealistic Herschel-Bulkley consistency coefficient (K) values.***

Table 4: Hysteresis loop areas (Pa/s) of mayonnaise samples, measured 30 minutes after preparation (D_0) and after 8 days of storage (D_8) at room temperature

Storage time									
Day 0					**Day 8**				
Full Fat Mayonnaise	19124 (660)				Full Fat Mayonnaise	22728 (1203)			
Low calorie mayonnaise type emulsions					**Low calorie mayonnaise type emulsions**				
Type of starch	Level of oil replacement	Unmodified starch	Modified starch with stearic acid	**Main effect types of starch**	Type of starch	Level of oil replacement	Unmodified starch	Modified starch with stearic acid	**Main effect types of starch**
Teff	50	15963 (1674) B	8651 (503) A	21595 (5184) A	Teff	50	31628 (2443) B*	16077(750) A*	31116 (6521) B
	80	50674 (3163) A	11094 (825) A			80	65726 (2747) A*	11033 (2762)A	
Maize	50	16979 (1385) B	19540 (1340) B	36186 (5791) A	Maize	50	34773 (1451) B*	27823 (1535) B*	53206 (7782) A
	80	63623 (717) A	44601 (537) A			80	92262 (4655) A*	57968 (6327) A	
Main effect modification		Teff	Maize		**Main effect modification**		Teff	Maize	
	Unmodified	33318 (7925) A	40301 (10453) A			Unmodified	48677 (7800) A	63518 (13038) A	
	Modified with stearic acid	9873 (697) B	32071 (5641) A			Modified with stearic acid	13555 (1706) B	42895 (7343) A	

- ***Values in parenthesis indicate standard error of the mean***
- ***Means followed by the same letter/s within a block are not significantly different (($P < 0.05$), Tukey (HSD) test.***
- ***Hysteresis loop area values followed with * are significantly different from the corresponding values measured at D_0***