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PHYSICOCHEMICAL, TEXTURAL AND SENSORY PROPERTIES OF YOGHURT AS EFFECTED BY CARBOXYMETHYL CELLULOSE FROM SHORT, MEDIUM AND LONG RICE HUSKS

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Abstract

Carboxymethyl cellulose (CMC), a derivative of commercial cellulose, is a popular additive used in commercial yoghurt preparation. Rice husk has been recently considered as an important source of cellulose. In this study, CMC synthesised from cellulose extracted from short (SM), medium (MS) and long (LP) grain rice varieties were applied to yoghurt and compared with a control sample containing commercial CMC. The three CMCs showed different levels of carboxymethyl substitution. CMC from LP rice husk showed the strongest gel formation without any negative impact on sensory acceptance of the yoghurt. It had a significantly ($p<0.05$) high water holding capacity (46.6%), viscosity (4134.3 cP) and hardness (1584) when compared with the control sample giving values of 57.8 %, 8826.5 cP and 1935 respectively. The sample also exhibited very low syneresis over storage (0.098 %). From the results, it could be opined that the LP husk CMC may be effective as stabilizer and thickener in yoghurt and other such food systems with superior physicochemical, textural and sensory properties.

Key words : Cellulose, Viscosity, Stabilizer, Gel, Thickener.

Introduction

Dairy products are known for their inherited 'healthy' properties, both physiological and nutritional. They are also rich sources of vitamins and minerals (Sakandar *et al.,* 2014). Yoghurt is a coagulated dairy product obtained as a result of controlled lactic acid fermentation of milk by the action of selected thermophilic bacterial strains, namely *Streptococcus thermophilus* and *Lactobacillus bulgaricus* which are also responsible for its characteristic clean mild lactic flavour and aroma (Alankali *et al.,* 2008, Athar *et al.,* 2000). Yoghurt is an important and dominant consumable dairy fermented product across the world and is also categorized as a functional food (Weerathilake *et al.,* 2014). Yoghurt is a rich source of energy and nutrition as it contains milk fat, vitamins, unfermented lactose and protein (Bhattarai *et al.,* 2015). The ease of digestion and assimilation of yoghurt by the body is due to the predigestible nutrients formed by the yoghurt strains and the transformation of complex protein fractions into simpler forms during the fermentation (Athar *et al.,* 2000). The superior nutritional status of yoghurt and functionality has attracted consumer interests and expectations that have consequently created the urgency for maintaining its shelf stability and quality. Aside from its enhanced nutritional properties, the attainment of consumer acceptance endorses good health imparted by yoghurt in several ways, including boosting of immune system, improving the lactose digestion, improvement in digestive health, better absorption of nutrition, controlling body weight, eliminating harmful bacterial count in the gut, maintaining gastrointestinal function, treating constipation and diarrhoea, reduction of disease risk, contribution to body detoxification management, administrating blood pressure, cholesterol and prevention of osteoporosis and cancer (Faisal *et al.,* 2019, Abdalla *et al.,* 2015, Vahed *et al.,* 2008, Andronoiu *et al.,* 2011). The gastrointestinal functions introduced by yoghurt consumption are mostly acceptable due to the effects arbitrated through the gut microflora, bowl transit, improvement of gastrointestinal innate and robust immune responses (Adolfsson *et al.,* 2004). The yoghurt body often has limited shelf stability. A common issue

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experienced by yogurt manufacturers during its refrigerated storage and transportation to far sales points is its textural deformation coupled with change in viscosity, water holding capacity, and syneresis which create obstacle in its large scale production and handling (Hematyar *et al.*, 2012, Makit and Bakirci, 2017). To inhibit these quality deteriorations over time, stabilizers are added to the yoghurt during preparation (Athar *et al.,* 2000, Mohammadifar *et al.,* 2007, Macit and Bakirci, 2017). Stabilizers are generally hydrocolloids which have thickening or gelling properties for improving and retaining structures of foods giving bulky mouthfeel (Macit and Bakirci, 2017). Stabilizers may have plant, animal or microbial origins (Imeson, 2011). Stabilizers are meant for improving texture and viscosityby formation of stronger and stable gel, preventing syneresis, maintaining homogeneity and increasing its firmness thereby stabilizing the overall body of the yoghurt matrix (Alankali *et al.,* 2008, Bhattarai *et al.,* 2015). In dairy products like yoghurt, it is very essential that the addition of additives should not mask the natural flavour of the product. Also, such functional additive should be stable and active within the pH range 4.0-4.6, which is specific for yoghurt (Andiç *et al.,* 2013). Pectin, gelatin, carboxyl methyl cellulose (CMC) and alginate are the stabilizers and thickeners used under such conditions (Andiç *et al.*, 2013). Sodium carboxymethyl cellulose or CMC is a water soluble cellulose gum that is actually a cellulose derivative obtained by alkalization followed by etherification (carboxymethylation) of cellulose using an etherifying agent which consequently substitutes the hydroxyl groups by the carboxymethyl group presented in the schematic Fig. 1. The protein dispersion is stabilized by CMC particularly at its isoelectric pH, thus providing enhanced stability against casein precipitation and loss of structural integrity (Andic *et al.,* 2013). Tamime and Robinson (2000) mentioned the recommended level of carboxymethylcellulose used in production of yoghurt as 0.1-0.2%. CMC is known to possess a variety of suitable properties including a broader pH range (3-10), hydrophilicity for both cold and hot water and a viscosity (at 2% concentration) ranging between 10 to 50000 cP or even higher (Alakali *et al.,* 2008). These properties relate to its broad spectrum of applications such as emulsifier, thickener and protective colloid, in flavour emulsions and salad dressings, as a thickener in fruit juices for the prevention of fruit settling or floating during preparation as well as giving a clear and bright appearance, used as a stabilizer in ice cream, ice pops, sherbets and other frozen confectionary products for prevention of ice crystal formation, formingdesirable gel texture and reducing syneresis, and also as a thickening agent in sour cream (Alakali *et al.,* 2008). Usually commercial CMC is prepared from cellulose extracted from popular bioresources like palm kernel cake (Bono *et al.,* 2009), sago waste (Pushpamalar *et al.*, 2006), suagr beet pulp (Toðrul and Arslan, 2003), pomelo peel (Chumee and Seeburin, 2011), corn cob (Jia *et al.*, 2016), banana pseudo stem (Adinuugrana *et al.*, 2005), durian rind (Rachtanapun *et al.*, 2012). Husk is a major waste of the rice milling industry. The husk consists of about 38% (db) cellulose (Shukla *et al.*, 2013). In another study, cellulose was extracted from husks of a long grain, a medium grain and a short grain variety. It was observed that major structural differences occur amongst the husks and celluloses extracted therefrom. From that, it was hypothesized that derivative products made from the three celluloses might also bear different extent of functionalities. In this study, the three rice husk celluloses were processed to CMCs, analysed for their degrees of substitution and applied in yoghurt samples. The physicochemical, textural and sensory attributes of the prepared yoghurt samples were assessed and compared with control yoghurt containing a commercial grade CMC.

Materials and Methods

Materials

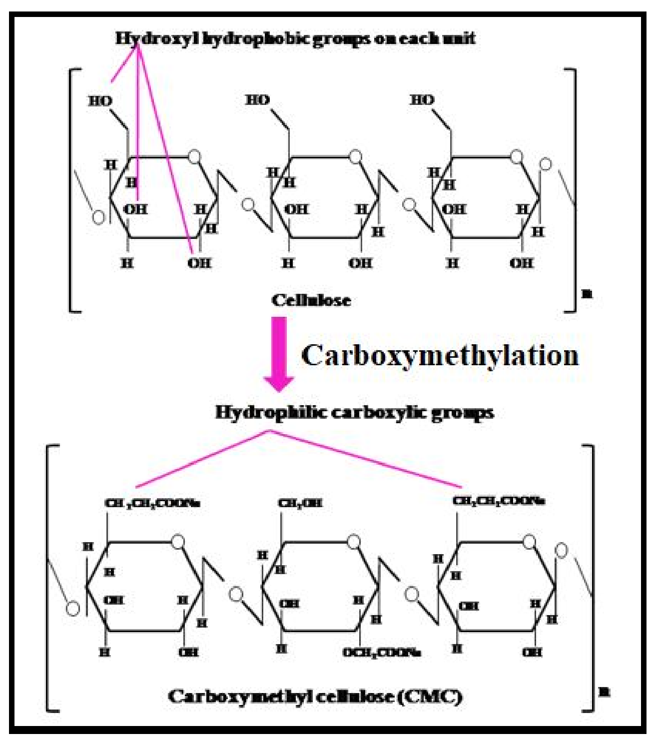

Fig. 1: Schematic picture showing the presence of hydroxyl hydrophobic groups which renders the cellulose insoluble in water and the formation of CMC introduces carboxymethyl hydrophilic groups which modifies the cellulose to act as a stabilizer.

Cultivated mature paddy samples of P10 (long grain), SR1 (medium grain) and Mushkbudij (short grain) rice varieties were procured from the store of Central Institute of Temperate Horticulture, India. Husks were obtained after dehulling the paddy grains in a laboratory rice huller (Satake, Japan) and washed thoroughly 6 times with distilled water to remove any adhering foreign particle. After drying in a tray dryer at 60 °C for 12 h, a Philips mixer grinder was used for grinding the samples to particle sizes of 100 to 240 mm. The powdered samples were stored in airtight containers. A final moisture content of 5-6% (wb) was recorded. Freeze dried yoghurt starter culture (Yogourmet, Canada), skim milk powder (Foodvit, India) and commercial edible grade CMC (HiMedia) used for the preparation of yoghurt were purchased. Fresh raw cow milk was procured from a local dairy farm. Analytical grade reagents and glassware were purchased from HiMedia and Borosil (India), respectively.

Methods

Cellulose isolation and CMC synthesis

Husk particles in each sample was dewaxed using a 1:2 (v/v) solvent mixture of methanol and benzene for 9 h at 75 °C (Bano and Negi, 2017). The dewaxed sample was then treated with 3% (w/v) sodium hydroxide at 45 °C for 8 h with rigorous mechanical agitation. The alkali treated samples were then autoclaved at 121 °C ± 2 °C under 15 psig pressure for 8h. This step of steam explosion was repeated thrice for maximizing efficiency of delignification and breakdown of bonds between the cellulosic non cellulosic components. The obtained fibers were washed several times to remove any trace of alkali. Remaining parts of lignin and hemicellulose were removed by washing bleaching with peracetic acid (6.5:2.0, acetic acid:hydrogen peroxide, v/v) at 45 °C for 7 h under mechanical agitation (Nascimento *et al.*, 2016). After washing several times with water, white and pure cellulose fibers were obtained. Samples were oven dried at 40 °C and stored in airtight containers. Carboxymethyl cellulose was prepared by following the procedure of Salama et al. (2018) with slight modifications. Briefly, 5 g cellulose was mercerized by the addition of 100 mL isopropanol under continuous vigorous agitation. Subsequently, 14.15g of 40% NaOH (w/w) was added drop wisely. The mixture was filtered and activated cellulose was obtained after drying at 70 °C for 8 h. To a 5 g of the activated cellulose 150 mL of isopropanol and 15 mL of 40% NaOH were added under continuous agitation for 30 min at room temperature. 7 g monochloroacetic acid was added to the mixture at 55 °C for and let the reaction continue for 4 h. Precipitation was carried out using ethanol. After filtering, precipitate was washed 6 times with 70% ethanol. The precipitate was then washed with absolute ethanol and oven dried at 60 °C for 6 h to obtain CMC.

Degree of substitution

The degree of substitution of the obtained CMCs as well as commercial CMC was done according to the acid wash method reported by Schuerch (1968). 10 mL of 2M nitric acid was added to 4 g of CMC under mechanical agitation for 2 min. The supernatant liquid was discarded so as to filter the solution and the resultant precipitate was heated at 60 °C until all the nitric acid is removed. 1 g of CMC was mixed with 100 mL of distilled water and 15 mL of 0.3N NaOH. Using phenolphthalein as an indicator the titration of excess amount of sodium hydroxide was done with 0.3N HCl. The degree of substitution was determined as:

$$\text{Degree of substitution (DS)} = 0.162A/(1 - 0.058A) \quad (1)$$

$$A = BC - DE / F \quad (2)$$

Where,

A = milliequivalent of acid consumed per gram of sample,

B = mL of NaOH solution added.

C = normality of NaOH, D = mL of HCl required for titration of excess sodium hydroxide. E = normality of HCl, F = g of acid CMC used, 162 = g molecular weight of the anhydroglucose monomer unit of cellulose and 58 = net increase of anhydroglucose for any substituted carboxymethyl group.

Proximate and chemical analysis

Protein, crude fat, titrable acidity and ash were determined as per AOAC (2000). pH was determined by a digital pH meter (Eutech pH meter). Lactose content was determined as per the method given by Triebold and Aurand (1963). Total soluble solids content was calculated by hot air oven drying of samples at 70°C under vacuum until constant weight.

Yoghurt preparation and coding

The preparation was followed as per Bhattarai et al. (2015). A 2% skim milk was added to the raw milk preheated at 45 °C and further heated to 70 °C. Under constant stirring, 4% sugar and 0.2% CMC were added. The pasteurization was completed after keeping at 85 °C for 30 min. Cooling was done at 43 °C followed by inoculation with 2% starter culture. The mixture was poured in an 80 ml glass beaker and incubated at 43 °C for 3-4 h. The obtained yoghurt was then cooled to 7°Cin order to cease the fermentation and stored at this temperature further to carry out the periodic physicochemical, textural and sensory analysis on 0^{th}, 3^{rd}, 7th and 14th day. The yoghurt samples were coded as SMY (Short Mushkbudij CMC incorporated) MSY (Medium *SR1* CMC incorporated) and LPY (Long P10 CMC incorporated). Control yoghurt was coded as CY.

Water holding capacity (WHC)

2 g of yoghurt sample was centrifuged under refrigeration (REMI, India) at 13500 ´ g for 30 min at 10 °C. WHC was calculated by the following equation (Andric *et al.,* 2013).

$$WHC (\%) = W_2 - W_1 / W_1 ´ 100 \quad ....(1)$$

Where,

W_2 = weight after sample after centrifugation and W_1 = weight of sample before centrifugation.

Syneresis

For determining syneresis, 10 g of yoghurt sample was centrifuged at 222 × g at 10 °C for 10 min (Andic *et al.,* 2013). Syneresis was calculated as the percentage of supernatant per initial weight of the yoghurt sample.

Viscosity

Static viscosity determination of stored yoghurt samples was performed using a rheometer (HAAKE™ Viscotester™C, Thermo Fisher Scientific, USA) equipped with a helipath stand and a spindle (L_3) rotating at a speed of 20 rpm

Color

A CIE color measuring instrument (Ultrascan Vis, Hunter Lab, USA) was used to evaluate the L*(lightness to darkness), a*(redness to greenness) and b*(yellowness to blueness) color values of the yoghurt samples. The machine was calibrated for pure whiteness and pure blackness before each operation using standard color surfaces provided by the supplier.

Texture Profile Analysis (TPA)

TPA of the yoghurt samples was carried out using a texture analyzer (TA-Hdi, Stable Micro systems, U.K) fitted with a 5 Kg load cell. Samples were kept in beakers of 45mm diameter. A two-cycle penetration test was carried out with a cylindrical probe of 35 mm diameter allowing 4 mm penetration at a speed of 1mm/s during both cycles. Parameters namely hardness, cohesiveness and springiness were obtained using the inbuilt TPA macro software. The maximum peak force obtained during the first cycle of penetration represented the hardness (given in g). The ratio of the positive force area during the second penetration to the positive force area during first penetration gives the values of cohesiveness. The height that the food retrieves during the time between the end of the first penetration and the start of the second penetration represents the value of springiness.

Microbial evaluation

The dilution of samples was done in the ratio of 1:10 (v/v) and used for the enumeration. The inoculation was done by standard pour plating method. For the enumeration of *S. thermophilus* count and *Lactobacilli* count, M17 and MRS agar medium were respectively used. The inoculated plates were accordingly incubated at 37 °C for 48 h under aerobic conditions for *S. Thermophilus* and at 37 °C for 72 h under anaerobic conditions for *Lactobacilli* (Macit and Berkirci, 2017). Coliform count was enumerated by using MacConkey agar medium and the plates were incubated at 32 °C for 48 h (Abdalla *et al.,* 2015). Yeast and mold count was determined using potato dextrose agar media and subsequently the plates were incubated at 25 °C for 5 days (Abdalla *et al.,* 2015).

Sensory evaluation

Sensory evaluation was done by using 5 point hedonic scale with the categories defined as, 1= More like, 2= Like, 3= Neither like nor dislike, 4= Dislike and 5= More dislike. Appearance, taste, flavour, mouth feel and general acceptability were determined by a panel of 28 semi-expert judges of both genders falling under the age group of 18 to 30 years. It was ensured that the panel members rinsed their buccal cavity after testing each sample and immediately recorded their scores.

Statistical analysis

All the experiments were carried out in triplicates and mean values were reported. Tests of significant differences between the mean values were analyzed by Duncan's multiple range tests at a significance level of 95% using SPSS 11.5 software (SPSS Inc., USA).

Results and Discussions

Degree of substitution (DS)

The degree of substitution of rice husk CMC was SMCMC = 0.42 (Short grain), MSCMC =0.53 (Medium grain) and LPCMC = 0.87 (Long grain). The degree of substitution of commercial CMC was 2.5. The average number of hydroxyl groups substituted with carboxymethyl groups at carbon 2, 3 and 6 represents the degree of substitution. The According to Asl *et al*. (2017) the hydrophilicity of CMC increases with the increase in degree of substitution and CMC with DS > 0.4 is completely soluble while DS< 0.4 is swellable but not soluble. Pushpamalar *et al.*, (2006), the implementation of high polarity solvent in the carboxymethylation process cater greater efficiency due to the accessibility of the etherifying reagent to the reaction centre of the cellulose chain instead of forming glycolate side chain. The lowest degree of substitution was observed in the SMCMC which is assigned to the less exposure of the –OH groups to get substituted by the carboxymethyl groups and greater interactions with the non cellulosic components that was indicated by the composition of MSC. The reason was that the NaOH concentration used in the initial stage of carboxymethylation process reacted inevitably with the non cellulosic components of the cellulose. Subsequently leading to the lesser concentration of NaOH available for the reaction with cellulose to expose the –OH groups for the substitution reaction. The initial stage being important for the action of the etherifying reagent in the final stage of carboxymethylation process thereby affects the final properties of the of the carboxymethyl cellulose (Candido and Goncalves, 2016).

Proximate and chemical analysis

The reason behind the addition of stabilizers to the milk base for the production of yoghurt is to preserve and sustain the enticing properties in yoghurt without presenting any hurdle in the aesthetic appeal of the product including, texture and body, mouthfeel, appearance and viscosity. Stabilizers have been mentioned to have a negligible effect on the chemical composition except pH, acidity and lactose content (Athar *et al.,* 2000). The values of proximate composition and chemical status of raw milk and yoghurt samples are given in Table 1.Fermentation of lactose to lactic acid decreased the pH of the yoghurt, ultimately increasing acidity of the product. The post fermentation pH is the most essential factor that affects the growth and survival of the lactic acid bacteria in the culture. The apprehension for the decrease in shelf life of the probiotic organisms is associated with the decrease in pH of the medium and formation of organic acids during to their growth and fermentation process (Faisal *et al.,* 2019).The control sample CY showed the lowest acidity value (0.743%), highest pH (4.25) and lactose content (3.82%). The highest value of acidity was shown by SMY (1.105%) and the lowest value by LPY (0.874%). Titrable acidity depicted significant variations amongst the yoghurt samples with rice husk CMC during the storage period. This significant difference could be assigned to the production of highly viscous matrix which causes the resistance of the reactants to diffusion, restricting their movement. Consequently, this decreased the rate at which the reacting species including starter culture of yoghurt and lactose interact for the fermentation process to take place (Alakali *et al.,* 2007). The acidity value of LPY was observed to be nearer to the value presented by CY. The changes in acidity, pH and lactose content during the storage of yoghurt are presented in Fig 2a, 2b and 2c respectively. The decrease in acidity parameter during the storage was observed in the order of SMY>MSY>LPY>CY. The lactose content also decreased upon storage which is associated with the utilization of the lactose by the yoghurt culture (Athar *et al.,* 2000). According to Andic *et al*., (2013) the addition of stabiliser may result in decreased acidity as a result of dilution and acting as buffer between the yoghurt culture and lactose, thereby reasonably preventing the lactic acid fermentation. The decrease in pH was found to be within the range of 4.25-3.84. The highest pH observed in LPY. The pH values of the samples gradually decreased with the storage period due to increasing concentration of lactic acid (Andic *et al.,* 2013). It could be concluded that addition of CMC had no significant effect on the pH of the yoghurt samples during storage, which is essential to maintain the proper level of fermentation.

Water holding capacity (WHC)

The water holding capacity of yoghurt is primarily the quantity of water carried by the protein structure and the increase in WHC means increased hydration of the protein network (Landge, 2009). The effect of rice husk CMC on the WHC of the yoghurt samples and storage is shown as patterns in Fig. 3. The highest WHC was shown by CY sample (57.8%). Among the samples incorporated with rice husk CMC, the LPY sample showed the higher WHC value (46.6%) nearest to the control. The increased WHC of the samples could be attributed to the increased viscosity of the yoghurt matrix. Also the increased WHC in all the samples was found to be consistent with the increasing viscosity. The increase was consistent upto 7th day. The increase in the WHC was due to the increased casein micelle interaction (Sodini *et al.,* 2004). The WHC was observed to be decreasing on 14th day which could be the lactic acid fermentation leading to damage in the stabilizer and casein micelle interaction (Shirkhani *et al.,* 2012)

Syneresis

Syneresis occurs in coagulated milk products and is caused by the aggregation and sedimentation of casein particles during the storage period (Kokroy and Kilic, 2004). Syneresis is generally considered as a defect in the quality yoghurt and is observed to be more prominent if no stabilizer is added to the yoghurt matrix (Mohsin *et al.,* 2019). The effect of addition of rice husk CMC and storage period upon the values of syneresis is presented in (Table 2). The effect of the stabilizer on the syneresis of the yoghurt was well established from the results. No syneresis occurred during the initial stages of storage in any of the samples. The highest value among the rice

Table 1: Proximate composition of milk and yoghurt samples.

Parameters	Milk	CY	SMY	MSY	LPY
Total soluble solids (%)	12.83 ± 0.16^{ab}	12.83 ± 0.17^{b}	12.81 ± 0.15^{a}	12.81 ± 0.15^{a}	12.82 ± 0.14^{a}
Titrable acidity(%)	0.132 ± 0.09^{cd}	0.743 ± 0.04^{ab}	1.105 ± 0.10^{e}	1.045 ± 0.08^{c}	0.874 ± 0.03^{a}
pH	6.5 ± 0.11^{e}	4.25 ± 0.09^{cd}	4.15 ± 0.06^{a}	4.18 ± 0.07^{ab}	4.22 ± 0.08^{bc}
Lactose (%)	4.83 ± 0.13^{b}	3.82 ± 0.11^{a}	3.81 ± 0.12^{ab}	3.81 ± 0.12^{ab}	3.82 ± 0.15^{c}
Fat (%)	4.58 ± 0.12^{d}	3.94 ± 0.05^{a}	3.91 ± 0.06^{ab}	3.91 ± 0.06^{ab}	3.92 ± 0.07^{c}
Protein (%)	3.52 ± 0.35^{ab}	3.51 ± 0.33^{a}	3.44 ± 0.44^{e}	3.45 ± 0.43^{cd}	3.46 ± 0.40^{c}
Ash (%)	0.63 ± 0.02^{a}	0.83 ± 0.06^{c}	0.72 ± 0.02^{a}	0.77 ± 0.03^{ab}	0.82 ± 0.06^{c}

The significance level was set at 0.05. The values were presented as average mean ±standard deviation.

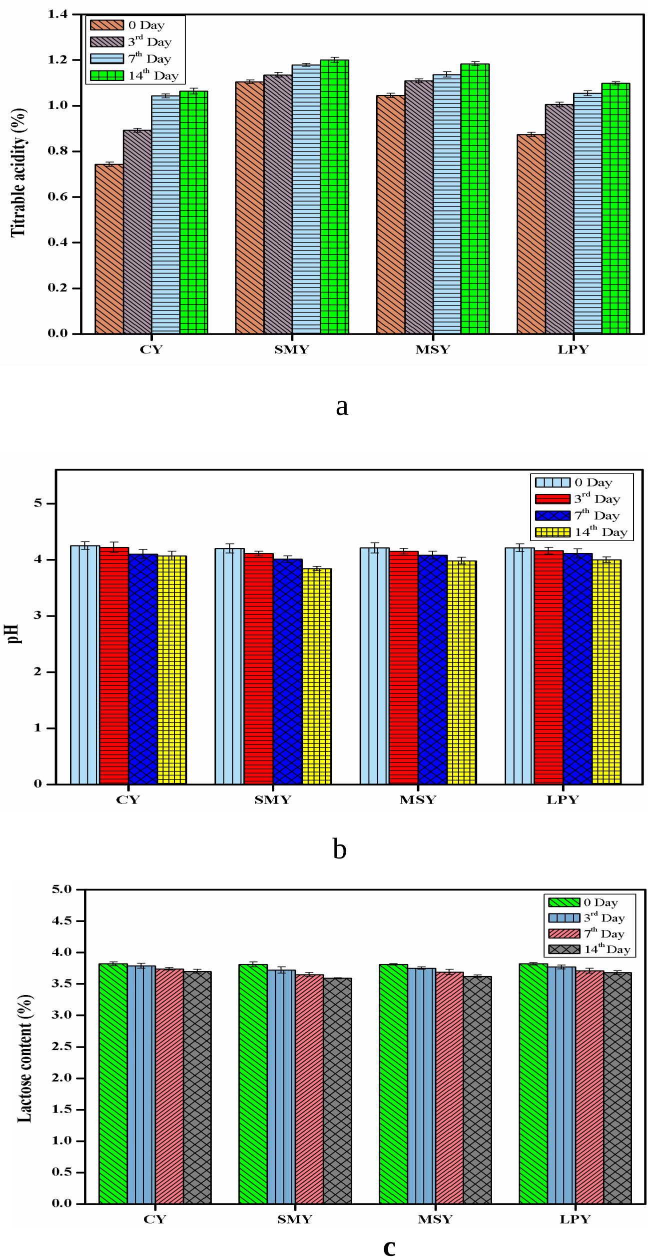

Fig. 2: Effect of addition of rice husk CMC and storage period on the titratable acidity (a), pH (b) and lactose content (c) of yoghurt samples.

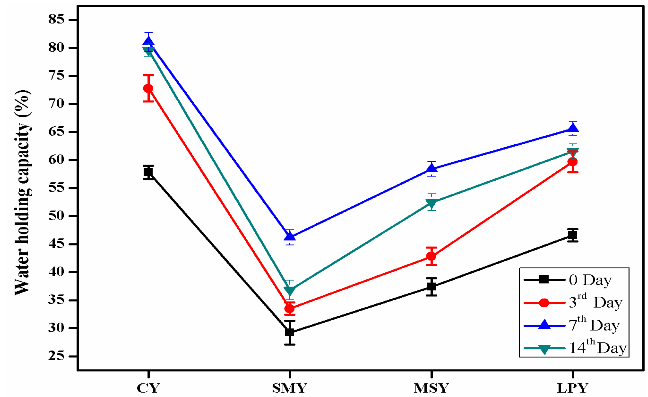

Fig. 3: Water holding capacity of stored yoghurt samples.

husk CMC added yoghurts was shown by the SMY sample which could be attributed to the decreased pH and increased acidity values due to lactic acid fermentation (Athar *et al.*, 2000, Bhattarai *et al.*, 2015).In CY, MSY and LPY samples, the syneresis was observed on 14th day with the lowest values presented by the CY (0.049%). Nearest value to the value of CY sample was shown by LPY (0.098%).

Viscosity

The measure of viscosity is given by the ability of a material to resist the distortion by shear stress as a result of intermolecular cohesive forces. The effect of addition of rice husk CMC and storage period on the viscosity of yoghurts is presented in Fig. 4. Among the rice husk CMC incorporated samples, the highest viscosity values (4143.3 cP) were shown by LPY. This could be due to the addition of CMC with highest degree of substitution. The higher degree of substitution revealed the presence of higher number of hydrophilic groups within the cellulose structure which could enhance the viscosity of the yoghurt (Asl *et al.*, 2017). Another explanation can be that the use of 0.2% concentration of CMC caused the adsorption of CMC on the casein micelles, resulting in flocculation (Andic *et al.*, 2013).Similar explanation was also proposed by Maroziene and Kruif (2000) regarding the interaction of pectin stabilizer with casein micelle. During storage, viscosity increased till 7^{th} day but decreased on 14^{th} day which could be associated with the development of acidity and decrease in pH**.** According to Aini et al. (2017) the viscosity of yoghurt is affected by the hydrophobic interactions of casein micelles. Production of lactic acid that lowers the acidity upon storage disrupt these interactions and consequently could lead to coagulation of casein and decrease the viscosity. Macit and Bakirci (2017) reported that the reduced values of WHC and increased values of syneresis could also lead to the decrease in viscosity.

Color

The values of L^*, a^* and b^* are given in (Table 2). No significant difference in the values were observed amongst the yoghurt sample, indicating no effect of addition of rice husk CMC over the commercial CMC. Although no change was observed in the color parameters during the initial period of storage, but a decrease was

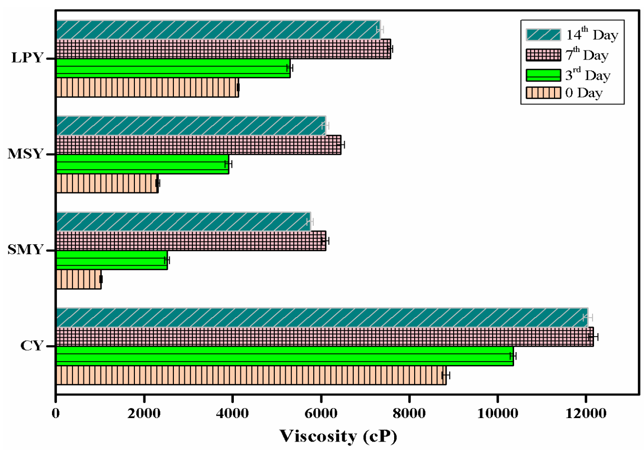

Fig. 4: Viscosity of stored yoghurt samples.

observed in L^* values on 14^{th} day of storage. A parallel decrease was also shown by a^* and b^* values. The changes can be assigned to the result of syneresis shown by the yoghurt samples (Macit and Bakirci, 2017).

Texture profile analysis

The TPA values of yogurt samples are presented in (Table 2). In TPA parameters, an increase was observed in almost every parameter and in every sample, becoming more noticeable with further storage. The highest increase in TPA parameters was presented by CY which was due to the higher increase in viscosity of the control CY. According to Andicetal. (2013), the increase in viscosity and hardness was related to the usage of proper amount of CMC (0.2% w/w) that caused stable protein network. The increase in the hardness values were due to occurrence of flocculation caused by the adsorption of CMC on the casein micelles. The increase in the hardness values were in the order CY<LPY<MSY<SMY. In case of SMY sample, the hardness values were not increased to such extent which might be related to the acid development and decreases the viscosity by damaging the casein micelle structure. Similarly, the cohesiveness and springiness values followed an increase upon storage as does the hardness values due to the adsorption of the CMC on the casein micelles.

Microbial evaluation

The microbiology of the yoghurt is presented in (Table 2). The addition of stabilizer did not have notable effect on the microbiology of the yoghurt but the storage period affected significantly. The initial bacterial count of the yoghurt was found to be increasing increased upto 7^{th} day but on 14^{th} day the count was found to be decreasing in all the samples. The decrease was accredited to the decrease in pH and the development in acidity (Macit and Bakirci, 2017). Coliform and yeast and mould count were found to be nil in all the samples throughout the storage period (Abdalla *et al.*, 2015).

Sensory analysis

The results of sensory attributes (flavour, aroma, texture, taste and appearance) are presented in (Table 3). The results of sensory analysis depicted that the addition of rice husk CMC had almost a little significant effect on the sensory attributes of the yoghurt. The highest sensory scores in flavour, aroma, texture, taste and appearance were given to CY sample followed by the LPY sample given the nearest value to the control. The addition of stabilizers provides the desirable sensory attributes to the yoghurt. The sensory attributes were not significantly affected by the addition of stabilizer upto 7^{th} day gaining the scores. However on 14^{th} day changes in the sensory attribute scores were given to all the samples in the order, CY<LPY<MSY<SMY. The lowest scores in SMY sample could be related to the acid development that affect the flavour (score=3.8), aroma (score=3.8), taste (score=3.9), texture (score=3.5), and appearance (score=3.9). The difference among the samples would be attributed to the rate at which the lactose and yoghurt culture comes together for the fermentation process to take place (Alakali *et al.*, 2007).

Conclusion

The results revealed that the incorporation of rice husk CMC during the preparation of yoghurt enhanced the physicochemical and textural properties of yoghurt. The addition of CMC also showed no significant negative effect on the sensory properties of yoghurt. The LPY

Table 2: Values of syneresis, color, texture parametersand microbiological status during storage of yoghurt samples.

Parameters		0 Day				3rd Day			
		CY	**SMY**	**MSY**	**LPY**	**CY**	**SMY**	**MSY**	**LPY**
Syneresis (%)		-	-	-	-	-	-	-	-
Color	L*	88.34±1.25^{de}	88.25±2.0^{b}	88.32±1.23^{cd}	88.33±1.35^{jk}	88.33±1.33^{hi}	88.24±1.31^{g}	88.31±1.21^{bc}	88.32±1.29^{f}
	A*	-3.15±0.12^{fg}	-3.10±0.09^{de}	-3.10±0.13^{gh}	-3.12±0.14^{i}	-3.15±0.08^{cd}	-3.09±0.05^{ab}	-3.10±0.11^{f}	-3.13±0.13^{gh}
	B*	7.06±0.39^{e}	7.16±0.35^{cd}	7.14±0.47^{g}	7.11±0.33^{b}	7.07±0.23^{a}	7.18±0.61^{l}	7.12±0.44^{f}	7.11±0.35^{cd}
TPA	Hardness	1935±21.2^{a}	956±43.1^{ij}	1173±36.0^{gh}	1584±42.2^{i}	5467±34.7^{ef}	1784±24.2^{bc}	2567±56.1^{k}	3985±29.7^{d}
	Cohesiveness	0.634±0.042^{m}	0.234±0.015^{h}	0.311±0.032^{l}	0.478±0.010^{e}	0.657±0.011^{ef}	0.232±0.003^{a}	0.314±0.017^{hi}	0.494±0.011^{ef}
	Springiness	0.942±0.036^{ef}	0.445±0.049^{j}	0.582±0.045^{i}	0.731±0.064^{n}	0.976±0.023^{ab}	0.456±0.063^{lm}	0.596±0.034^{d}	0.765±0.035^{de}
Microbiology (log cfu/g)	*S.thermophilus*	7.89±0.36^{fg}	7.82±0.28^{bc}	7.84±0.33^{d}	7.87±0.39^{hi}	7.92±0.24^{a}	7.88±0.45^{jk}	7.89±0.36^{fg}	7.90±0.44^{j}
	Lactobacillus bulgaricus	7.56±0.55^{l}	7.45±0.37^{ef}	7.47±0.43^{ij}	7.50±0.39^{fg}	7.69±0.28^{ab}	7.49±0.27^{a}	7.51±0.41^{gh}	7.61±0.34^{d}
	Coliform	-	-	-	-	-	-	-	-
	Yeasts and moulds	-	-	-	-	-	-	-	-

table 2 continue ....

7th Day				14th Day			
CY	**SMY**	**MSY**	**LPY**	**CY**	**SMY**	**MSY**	**LPY**
-	0.361±0.32c	-	-	0.049±0.28ab	0.712±0.56e	0.254±0.43cd	0.098±0.17a
88.29±1.34^{ij}	88.03±1.45l^{m}	88.27±1.16^{a}	88.29±1.33^{hi}	88.01±1.43^{l}	85.26±1.45^{lm}	86.69±1.32^{gh}	87.45±1.47^{n}
-3.15±0.07^{bc}	-3.03±0.12^{fg}	-3.05±0.03^{a}	-3.02±0.05^{ab}	-3.14±0.12^{fg}	-3.05±0.05^{ab}	-3.07±0.07^{bc}	-3.10±0.12^{fg}
7.10±0.55^{k}	7.19±0.67^{m}	7.13±0.49^{hi}	7.09±0.34^{bc}	7.11±0.35^{cd}	7.23±0.52^{j}	7.18±0.48^{gh}	7.13±0.45^{fg}
3985±29.7^{d}	6157±66.5^{l}	1997±73.2^{m}	2798±23.4^{ab}	4346±43.5^{ij}	6311±56.5^{k}	2009±35.1^{fg}	2827±33.2^{e}
0.667±0.004^{ab}	0.240±0.001^{a}	0.335±0.012^{fg}	0.523±0.022^{jk}	0.683±0.005^{bc}	0.252±0.021^{j}	0.341±0.012^{fg}	0.537±0.006^{cd}
0.988±0.027^{c}	0.459±0.042^{h}	0.602±0.051^{j}	0.772±0.039^{g}	0.997±0.021^{a}	0.463±0.027^{c}	0.613±0.062^{l}	0.782±0.055^{k}
.90±0.44^{j}	7.89±0.37^{gh}	7.75±0.46^{kl}	7.81±0.35^{ef}	7.85±0.45^{jk}	7.0±0.54^{m}	6.88±0.26^{ab}	6.95±0.34^{de}
7.77±0.47^{j}	7.53±0.34^{d}	7.58±0.48^{jk}	7.70±0.48^{jk}	7.74±0.42^{hi}	7.01±0.39^{fg}	7.23±0.35^{de}	7.56±0.29^{bc}
-	-	-	-	-	-	-	-
-	-	-	-	-	-	-	-

The significance level was set at 0.05. The values were presented as average mean ±standard deviation.

Table 3: Values of sensory parameter of yoghurt samples during storage.

	0 Day				3rd Day			
	CY	**SMY**	**MSY**	**LPY**	**CY**	**SMY**	**MSY**	**LPY**
Flavor	4.6±0.11^{cd}	4.0±0.17^{g}	4.4±0.09^{ab}	4.4±0.07^{a}	4.7±0.22^{jk}	4.0±0.24^{lm}	4.2±0.14^{ef}	4.5±0.13^{e}
Aroma	4.7±0.12^{bc}	4.2±0.14^{cd}	4.2±0.16^{ef}	4.5±0.17^{fg}	4.7±0.12^{bc}	4.1±0.16^{ef}	4.2±0.18^{gh}	4.5±0.14^{cd}
Texture	4.8±0.15^{cd}	4.5±0.18^{f}	4.5±0.15^{ef}	4.6±0.09^{a}	4.8±0.22^{hi}	4.6±0.14^{c}	4.6±0.25^{k}	4.7±0.22^{hi}
Taste	4.7±0.12^{b}	4.1±0.26^{hi}	4.2±0.25^{gh}	4.4±0.28^{jk}	4.8±0.31^{l}	4.2±0.27^{ij}	4.3±0.28^{jk}	4.5±0.09^{a}
Appearance	4.8±0.21^{de}	4.5±0.27^{ij}	4.5±0.24^{g}	4.7±0.28^{jk}	4.8±0.26^{hi}	4.6±0.22^{ef}	4.6±0.20^{cd}	4.7±0.24^{g}

table 3s continue ....

	7th Day				14th Day			
	CY	**SMY**	**MSY**	**LPY**	**CY**	**SMY**	**MSY**	**LPY**
Flavor	4.7±0.18^{gh}	3.9±0.35^{p}	4.3±0.37^{o}	4.5±0.19^{hi}	4.6±0.10^{bc}	3.8±0.27^{n}	4.0±0.21^{j}	4.3±0.23^{kl}
Aroma	4.7±0.15^{de}	3.9±0.26^{j}	4.2±0.29^{k}	4.5±0.09^{a}	4.6±0.11^{b}	3.8±0.39^{l}	4.1±0.12^{bc}	4.3±0.19^{hi}
Texture	4.8±0.21^{gh}	4.2±0.19^{fg}	4.6±0.16^{de}	4.7±0.18^{f}	4.8±0.10^{ab}	3.5±0.23^{ij}	4.2±0.26^{kl}	4.4±0.27^{m}
Taste	4.8±0.16^{c}	4.1±0.18^{cd}	4.4±0.23^{fg}	4.6±0.22^{ef}	4.8±0.31^{l}	3.9±0.33^{m}	4.2±0.27^{ij}	4.4±0.21^{e}
Appearance	4.8±0.32^{l}	4.5±0.25^{gh}	4.5±0.27^{ij}	4.7±0.24^{g}	4.8±0.14^{a}	3.9±0.29^{jk}	4.3±0.19^{c}	4.5±0.15^{ab}

The significance level was set at 0.05. The values were presented as average mean ±standard deviation.

also presented lower acid development upon storage. Although among the three varieties the yoghurt (LPY) incorporated with CMC of long grain husk showed the enhanced properties. The LPY sample showed lower syneresis and high water holding capacity upon storage. The increased viscosity and hardness values improved the body and texture of yoghurt. This concluded that the use of long grain CMC can be used as a convenient food ingredient in fermented milk products especially yoghurt so as to overcome the storage problems.

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