FINAL REPORT FOR DEMAND-DRIVEN INNOVATION PROJECT BY PUBLIC-PRIVATE RESEARCH NETWORK (PPRN)

CMSS (CARBOXYMETHLY SAGO STARCH) AS HALAL THICKENER FOR FOOD AND BEVERAGES INDUSTRY

Dr. Farhan Binti Mohd Said
AP. Dr. Fatmawati Adam
Nur Syuhada Binti Asmar
Dr. Ruwaida Abdul Rashid
AP. Dr. Sumaiya Zainal Abidin
Hj. Mohd Noor Nawi
Johanizal Johari

RESEARCH VOTE NO:
UIC150704

FAKULTI TEKNOLOGI KEJURUTERAAN KIMIA DAN PROSES
UNIVERSITI MALAYSIA PAHANG

2019

ACKNOWLEDGEMENT

Bismillahirrahmanirrahim

In the name of Allah S.W.T and Rasulullah S.A.W., I would like to express my sincere appreciation to UMP for giving me opportunity to grant this project. My sincere appreciation also extends to all technical staff of FTKKP Lab for the technical support. Special thanks go to the team members and students for the help to complete this research. Special dedicated to my beloved family members for their full support through the research. Finally, to all those involved directly and indirectly in realization of this research, on which I could not mention one by one, thank you. May Allah bless us.

ABSTRACT

(Keywords: sago starch, temperature, time, bread improver)

The issue of the industry is to diversify the use of Carboxymethly Sago Starch (CMSS) in food products. From the study, researchers managed to formulate CMSS in bakery products. Parameters such as temperature and time of baking play important roles to produce good bread. CMSS is able to be used as a bread improver in the bakery industry.

Email: farhan@ump.edu.my/ farhan_msaid@yahoo.co.uk
Tel.: 09-5492887
Vote no.: UIC150704

ABSTRAK

Isu industri adalah untuk mempelbagaikan penggunaan karboksilmetil kanji sagu (CMSS) dalam produk makanan. Dari kajian tersebut, penyelidik Berjaya merumuskan formulasi penggunan CMSS dalam produk roti. Parameter seperti suhu dan masa pembakar memainkan peranan penting untuk menghasilkan roti yang baik. CMSS dapat digunakan sebagai penambah baik roti dalam industri roti.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

CMSS	carboxymethyl sago starch
SEM	Scanning Electron Microscopic
ICPMS	Inductively Coupled Plasma Mass Spectrometry
CFU	colony forming unit
h	hour
ppm	part per million

CHAPTER 1

INTRODUCTION

1.1 Background of study

In Southeast Asia especially, sago starch has been used in the cooking of various types of dishes such as jellies, puddings, soups, noodle, biscuit, sago pearls, and many more. Sarawak is now the world's biggest exporter of sago. Annually export 25000 to 40000 tonne of sago product to peninsular Malaysia, Japan, Taiwan, Singapore and other countries. In Malaysia, the sago industry (in the State of Sarawak) is well established and has become one of the important industries contributing to export revenue. Sago palm has a main advantage of the ability to thrive in the harsh swampy peat environment (Ruddle 1977), which covers an area of 1.5 million ha, 12% of Sarawak's total land area (Tie and Lim 1977). The sago palm (*Metroxylon* spp.) is exploited as both a staple and a cash crop in Sarawak.

Sago is almost pure starch, being composed of 88 percent carbohydrate, 0.5 percent protein, and minute amounts of fat, and contains only a trace of B vitamins. It is a basic food of the southwest Pacific area, where it is used in meal form to prepare soups, cakes, and puddings. Elsewhere, its use in cookery is mainly as a pudding and sauce thickener. Food thickeners frequently are based on either polysaccharides (starches, vegetable gums, and pectin), or proteins. Different thickeners may be more or less suitable in a given application, due to differences in taste, clarity, and their responses to chemical and physical conditions.

The demand of Halal products has been increasing recently. Therefore, there is a need to formulate products especially on the modified sago starch known as carboxymethyl sago starch (CMSS) on food and beverages product. The product should be non-toxic and not harmful to human health.

This project was done to enhance the applications of CMSS in the food and beverage product, particularly in bread. Problems regarding the uncertainty to choose Halal bread thickener/improver can be solved especially in Malaysia that consist larger Muslims population.

1.2 Objective

The objective of this research is to formulate Carboxymethly Sago Starch (CMSS) as Halal bread thickener/improver for food product (bread)

CHAPTER 2

MATERIALS AND METHODS

2.1 Materials

The materials comprised of commercial bread flour (44%), commercial wheat flour (11%), water (28%), salt (0.9%), sugar (11%), yeast (0.9%), shortening (2.8%), commercial bread improver and CMSS.

2.2 Raw Material Characterization

CMSS characterization analysis for Scanning Electron Microscopic (SEM) and Inductively Coupled Plasma Mass Spectrometry (ICPMS) can be obtained in this section.

2.3 Scanning Electron Microscopic (SEM) Analysis

In analysing the morphology of CMSS, scanning electron microscope method was used (Carl Zeiss EVO50). In order to eliminate the moisture, the CMSS was stored in desiccators for overnight. Then by using a carbon conductive pad, the samples were mounted on a metal stub. The silver metals which acts as conductor were placed at both sides the sample. Lastly, under vacuum state and in an argon atmosphere, the samples were coated with gold and were observed. Figure 1 shows the Scanning Electron Microscope (Carl Zeiss EVO50) instrument.

Figure 1: Scanning Electron Microscope (Carl Zeiss EVO50)

2.3 Inductively Coupled Plasma Mass Spectrometry (ICPMS) Analysis

Inductively Coupled Plasma Mass Spectrometry (ICPMS) Analysis was done by using ICPMS Agilent 7500 series (Figure 2). The application of ICPMS is for the confirmation and identification elemental analysis in solid, liquid and semi liquid. Each 0.5 ml of sample solution was diluted into 2% HNO_3 solution up to 50 ml total volume in volumetric flask prior to the ICPMS run. Then the sample of CMSS was analyzed using ICPMS.

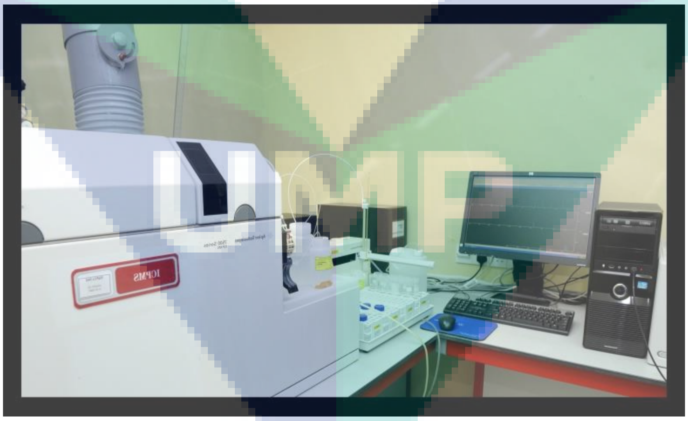


Figure 2: Inductively Coupled Plasma Mass Spectrometry (ICPMS)

2.4 Bread making procedure

The dough was prepared separately based on different amount of CMSS and bread improver, respectively. Control dough was prepared without bread improver or CMSS. All of the ingredients were mixed with a Kenwood Mixer (Chef Premier KM C560 4.6L). Flour was premixed for 30 minutes with low speed prior other ingredients. The dough was baked at 175°C for 30 minutes (FABER Microwave) after rested 30 minutes prior baking. Later, the bread was kept in a sealed container at room temperature.

2.4.1 Bread quality

Physical characterizations of bread included volume, height and mass were determined. The volume of bread was determined by green beans displacement method, modified from Sahraiyan et al. (2013). Height of the bread was measured at the centre of the loaf. Mass of the bread was also recorded in accordance with previous studies (Mariotti et al., 2013).

2.4.2 Moisture content of bread

To study the moisture content on bread, a moisture analyzer brand AND MF-70 was used. The result in percentage was recorded.

2.4.3 Microbiological test

Microbiological test has been tested on bread based on the total plate count method. Constant volume of nutrient agar was on plate agar for microbiological test. Small portion of bread was blended in a 10 ml sterilized distilled water. Later, 1 ml of blended bread was spread on the plate agar. The spread plate was then incubated at 37°C. The colony forming unit (CFU) on the plate was measured after 24 h.

2.4.4 Nutritional content

The nutritional content of bread has been tested at Food Analysis Unit, Faculty of Food Science and Technology, Universiti Putra Malaysia, Serdang Selangor. Five tests has been done which included crude protein (UPM/ FSTM/ BC/ T1 based on Nielsen, S.S (2010) Food Analysis Laboratory Manual), crude fat (JSTM/ FBC/ T4-Gerber), crude fibre (JSM/ FBC/ T5), moisture content (JSM/ FBC/ T2), ash content (JSM/ FBC/ T3) and carbohydrate content (JSM/ FBC/ T6).

2.4.5 Scanning Electron Microscopic (SEM) Analysis

Small portions of bread samples were cut and prepared for SEM analysis. The bread samples were mounted on specimen stand with double stick tape. The samples were viewed with a Hitachi S4500 scanning electron microscope operating at an accelerating voltage of 3 kV.

CHAPTER 3

RESULTS AND DISSCUSSION

3.1 Raw Material Characterizations

3.1.1 Scanning electron microscopy (SEM) Analysis

Scanning electron microscopy (SEM) analysis is a method for examining granules morphology. The SEM micrograph of CMSS shows intact and distorted granules compared to the native sago starch by Yaacob et al. (2010) (Figure 3). This observation generally is found in acid or enzyme-treated starch granules or in the gelatinization form of starch granules where small amount of amylose is being leached. However, CMSS by Yaacob et al. (2010) were more roughened, grooved and cracked.

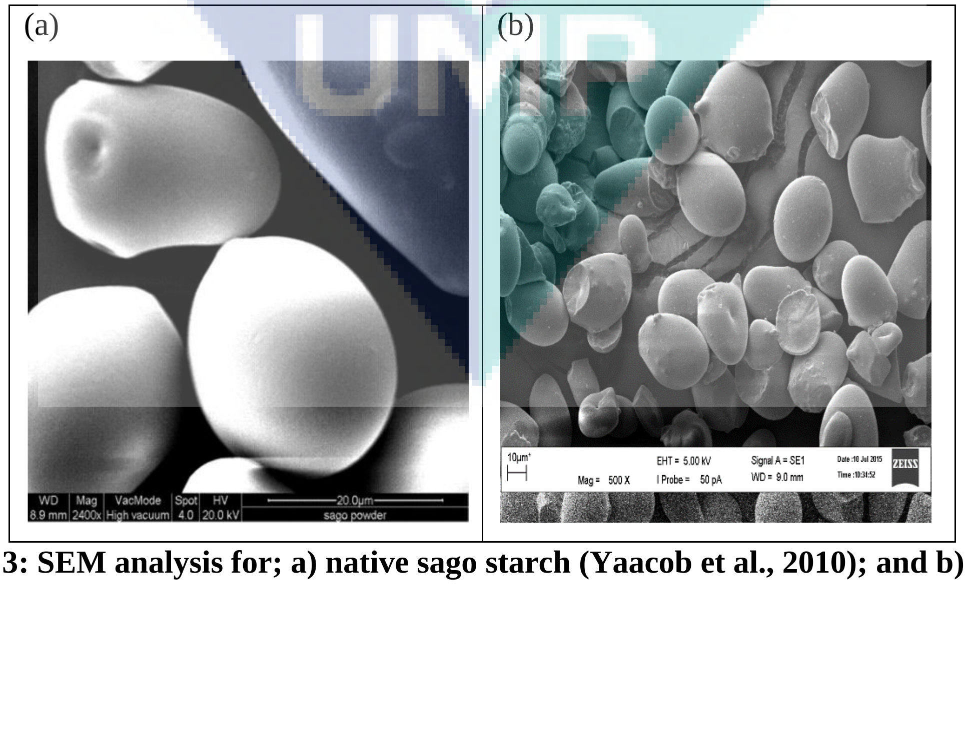


Figure 3: SEM analysis for; a) native sago starch (Yaacob et al., 2010); and b) CMSS

3.1.2 Inductively Coupled Plasma Mass Spectrometry (ICPMS) Analysis

The Inductively Coupled Plasma Mass Spectrometry (ICPMS) analysis was conducted to monitor and control metal impurities in food application (i.e: raw material for bread). According to International/National Standards for Heavy Metals in Food, maximum permitted concentration for heavy metals is 20 ppm and Arsenic 1 ppm. The highlighted parameter in Table 1 shows the heavy metals reading for the CMSS and all of them have the reading less than 20 ppm.

Table 1: Inductively Coupled Plasma Mass Spectrometry (ICPMS) result for CMSS

No	Parameter	Results	Unit
1	Berylium (Be)	Not Detected (Less than 0.5)	ppb
2	Sodium (Na)	911.29	ppm
3	Magnesium (Mg)	2206.04	ppm
4	Aluminium (Al)	446.38	ppm
5	Potassium (K)	8329.18	ppm
6	Calcium (Ca)	7641.93	ppm
7	Vanadium (V)	0.020	ppm
8	**Chromium (Cr)**	**0.1111**	**ppm**
9	Manganese (Mn)	5.378	ppm
10	Iron (Fe)	30.18	ppm
11	Cobalt (Co)	0.226	ppm
12	Nickel (Ni)	0.366	ppm
13	Copper (Cu)	1.527	ppm
14	Zinc (Zn)	2.515	ppm
15	**Arsenic (As)**	**Not Detected (Less than 0.5)**	**ppb**
16	Selenium (Se)	Not Detected (Less than 0.5)	ppb
17	Molybdenum (Mo)	Not Detected (Less than 0.5)	ppb
18	Silver (Ag)	424.05	ppm
19	**Cadmium (Cd)**	**0.0316**	**ppm**
20	**Antimony (Sb)**	**0.0011**	**ppm**

21	Barium (Ba)	43.994	ppm
22	**Lead (Pb)**	**1.573**	**ppm**
23	Thorium (Th)	Not Detected (Less than 0.5)	ppb

3.2 Application of CMSS in food (bread)

3.2.1 Effectiveness of CMSS as a bread thickener/ improver

Based on the study, parameters such as temperature and time of baking bread play important roles to produce good and quality bread. Thus, preliminary studies need to be done to identify the suitable temperature and time for baking the bread. The replacement of commercial bread improver to CMSS on the bread making has given a significant improvement to the bread performance, especially on their texture, mass, volume and the colour of bread. Table 2 and Figure 4 show the comparison of the bread performance and texture of bread with CMSS and commercial bread improver.

Table 2: Comparison of the bread performance with CMSS and commercial bread improver

AMOUNT OF FILLER (g)	MEASUREMENTS					
CMSS/ BREAD IMPROVER	HEIGHT (cm)	WIDTH (cm)	VOLUME (cm^3)	MOISTURE CONTENT		MASS (g)
				CRUST (%)	INSIDE (%)	
0g CMSS	8.0	18 .0	1320	17.534	33.786	750.6
3g CMSS	8.4	17.6	1510	16.188	29.351	752.9
5g CMSS	8.5	18.0	1512	18.915	35.356	762.6
7g CMSS	8.0	18.5	1257	16.543	28.694	756.8
10g CMSS	9.0	18.5	1320	19.436	27.786	755.9
5 g commercial bread improver	8.7	17.0	1351	20.534	25.786	753.6

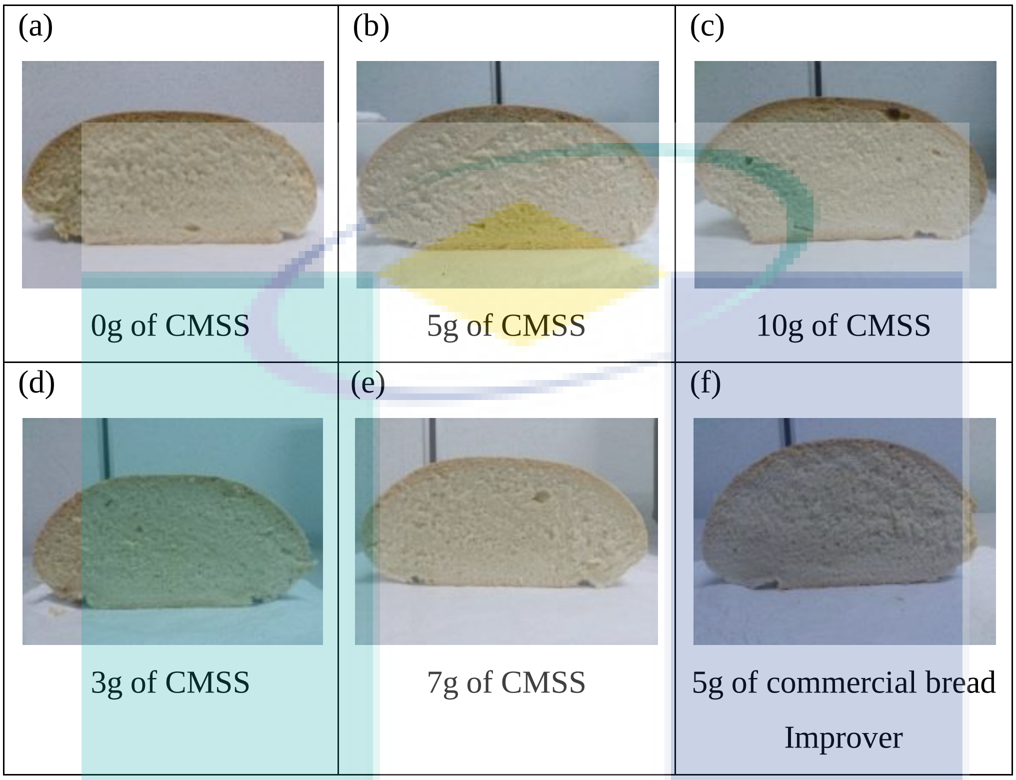


Figure 4: Texture of bread with CMSS and commercial bread improver; a) 0 g CMSS; b) 5 g CMSS; c) 10 g CMSS; d) 3 g CMSS; e) 7 g CMSS; f) 5 g commercial bread improver

Significant improvement on bread performance was shown with 5 g CMSS, compared to the other formulas. The improvements were clearly seen on their weight and volume of the breads (Table 2). The mass and volume of the bread with 5 g CMSS were 762.6 g and 1512 cm^3, respectively, heavier than with commercial bread improver, which the weight and volume of the bread were 753.6 g and 1351 cm^3, respectively (Table 2). Internal structure of the breads of 5 g CMSS and commercial bread improver, did not show any significant different. Where, both breads produced nice pore and the breads were slightly dense (Figures 4b and 4g).

The moisture content of the breads produced with CMSS were less than 36% for both crust and internal structure of bread (Table 2), which followed the standard requirement of bread in Food Act 1983 Regulation. Based the Food Act 1983 Regulation, the maximum water content of any part of the loaf is 45%.

Consequently, the application of CMSS on the food product (specifically on bread) was effective. Thus, it can be concluded that the application of food product (CMSS) can be used as (an alternative) to the commercial bread improver.

3.2.2 Stability of the bread using CMSS as a bread improver

Stability of CMSS on the bread was tested in terms of the microbiological test. Figure 5 shows the microbiological test of bread with CMSS and commercial bread improver at day 2 and day 3.

Figure 5 shows, after 2 days of bread making, small white spots were spotted on each formula. Spotted white spots show that there were microorganisms on the bread after 2 days baked. The spotted white spots appeared ~~perhaps~~ due to the effect of the storage containers, which were not fully sealed. After 3 days of baking, bread with commercial bread improver shows massive ~~of~~ white spots compared to other CMSS formulas (Figure 5). These results were due to the advantages of CMSS as a bread improver and can be acted as preservative agent. The spotted white spots were approximately $2x10^4$ cfu/g which under microbiological standard of 10^5 cfu/g. Additionally, the bread that produced with CMSS and commercial bread improver did not contain any preservative, thus shortened the shelf life of the bread. Spores were seen after 5 days baking (data not shown).

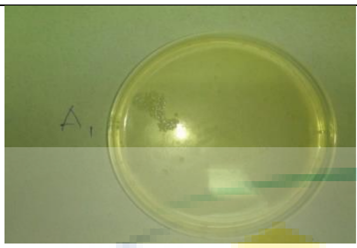
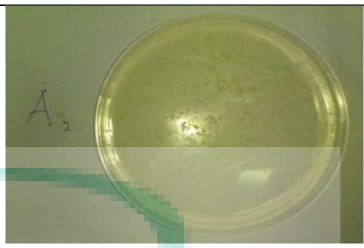
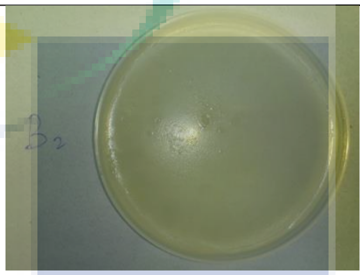
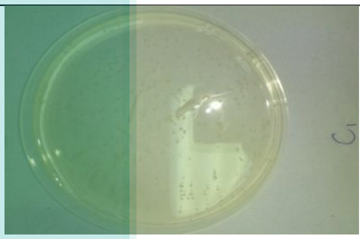
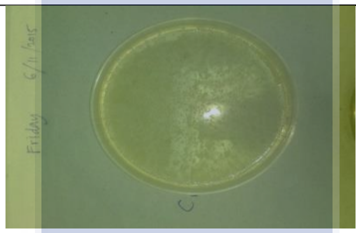
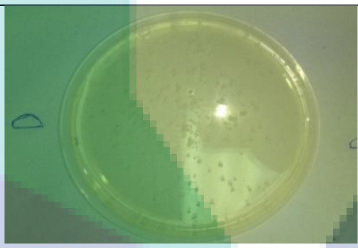
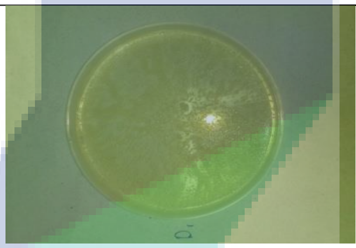
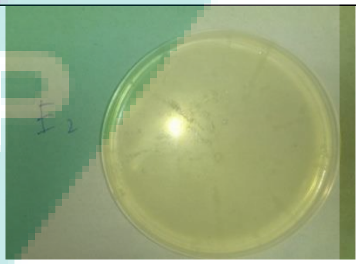
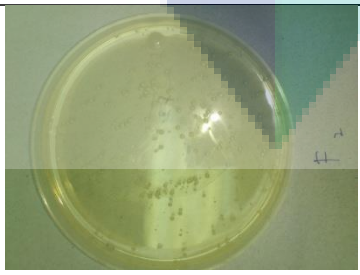
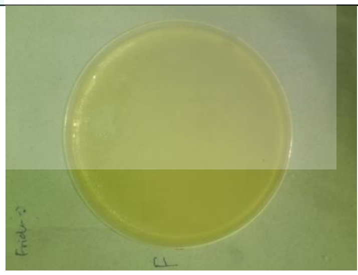
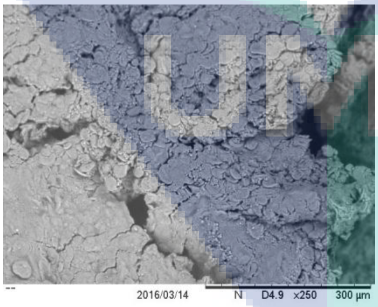
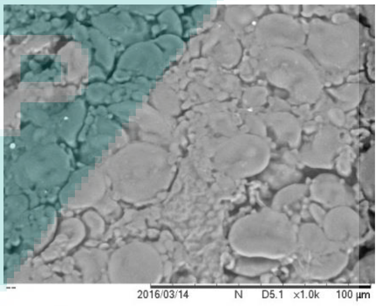
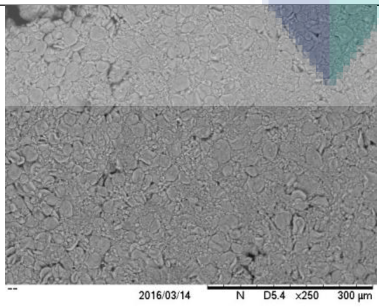
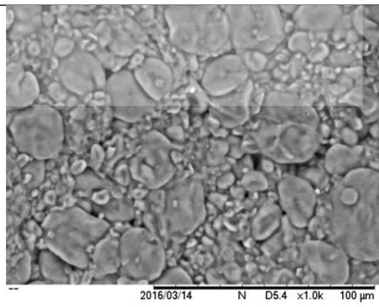
Sample	Day 2	Day 3
0g CMSS		
3g CMSS		
5g CMSS		
7g CMSS		
10g CMSS		
5 g commercial bread improver		

Figure 5: Microbiological test of bread with CMSS and commercial bread improver at day 2 and 3.

From the results obtained, it can be summarised that application of CMSS in the bread was relatively stable within 4 days. To prolong the shelf life of the bread, permitted preservative can be introduced to the bread.

3.2.3 Microscopic analysis and proximate analysis of CMSS in the bread

Microscopic analysis of bread was analyzed using scanning electron microscope (SEM). Figure 6 shows the microstructure of internal bread at 250x and 1000x of different formulation. From the microstructure, the qualitative observations about the pore characteristics of the bread can be seen. From the figure, more pores can be seen at 5 g CMSS. The pores were smaller, well distribution and more spherical (Figure 6). While, the pores without CMSS cannot be seen clearly, the pores are so close to each other. On the other hand, bread baked with commercial bread improver, the pores were less developed than with 5 g CMSS (Figure 6). Overall, very significant pore size distributions exist in all bread (with CMSS and commercial bread improver), except without CMSS. Thus, it can be concluded that CMSS can be used as a bread improver or thickening agent.

Formula	**Surface morphology of bread**	
	Surface morphology for Control at 250x	**Surface morphology for Control at 1000x**
0g CMSS		
3g CMSS		

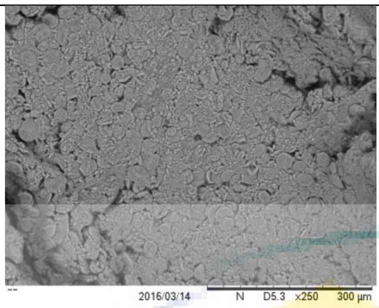
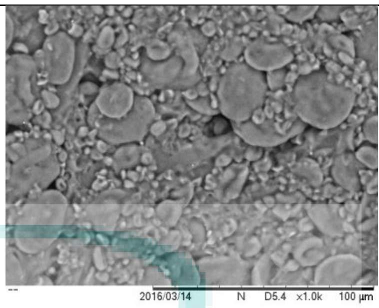
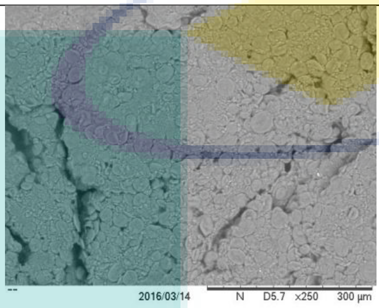
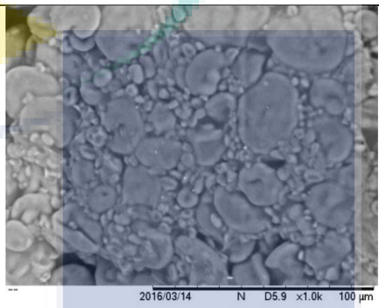
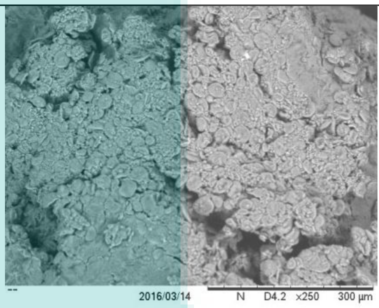
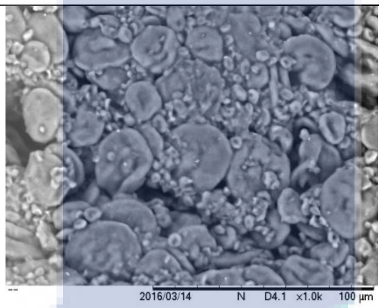
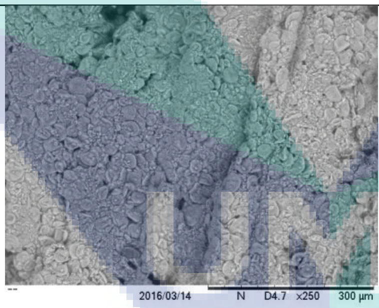
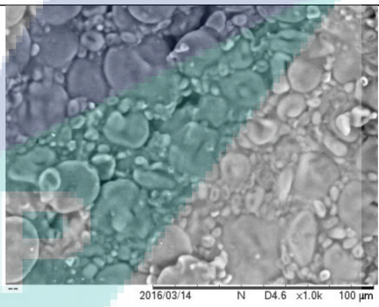
5g CMSS		
7g CMSS		
10g CMSS		
5g commercial bread improver		

Figure 6: Surface morphology of bread.

Nutritional content is important in the food product. Table 3 shows the nutritional content of bread with CMSS, without CMSS and commercial bread improver. Overall, bread without CMSS or commercial bread improver content higher crude protein, crude fibre, ash and carbohydrate, compared to with bread improver or CMSS (Table 3). Significant results show for 5 g CMSS and commercial bread improver which resulted approximate similar content of crude fat and ash. No significant differences on the crude protein, crude fibre and carbohydrate contents for both formulas of 5 g CMSS and commercial bread improver. Where, the crude protein, crude fibre, and carbohydrate contents were 0.0842 g/g, $1.8x10^{-3}$ g/g, and 0.578 g/g, respectively for 5 g CMSS. And for commercial bread improver, the contents were 0.076 g/g, $2.3x10^{-3}$ g/g, and 0.598 g/g of crude protein, crude fibre, and carbohydrate, respectively.

Table 3: Nutritional Content of bread

RESULT (g/ 100g)	BREAD					
	Control	**3g CMSS**	**5g CMSS**	**7g CMSS**	**10g CMSS**	**Commercial bread improver**
Crude Protein	8.45	7.66	8.42	7.26	8.39	7.60
Crude Fat	2.56	7.32	2.70	7.32	6.56	2.85
Crude Fibre	0.36	0.17	0.18	0.17	0.16	0.23
Ash	1.51	1.36	1.29	1.36	1.32	1.28
Carbohydrate	60.67	58.32	57.83	58.32	53.90	59.75

CHAPTER 4

CONCLUSION AND RECOMMENDATION

4.1 CONCLUSION

From the data obtained, CMSS shows great potential as a bread improver / halal thickener food product. The replacement of CMSS to the commercial bread improver seems to produce slightly better/similar product. The application of CMSS on the food product (specifically on bread) was effective and can be used as (an alternative) to the commercial bread improver. However, the application of CMSS in the bread was relatively stable in 4 days. Thus, to prolong the shelf life of the bread; permitted preservative can be introduced to the bread.

4.2 RECOMMENDATION

There are some aspects of the research that might be useful to be investigated in future studies. Recommendation for future study is for specific application of CMSS on the food product. Special analysis is needed to validate the function of CMSS in the food product.

REFERENCES

1. Bayor, M. T. Tuffour, E. and Lambon, P. S. Evaluation of Starch From New Sweet Potato Genotypes for use as A Pharmaceutical Diluent , Binder or Disintegrant, *J. Appl. Pharm. Sci.*, vol. 3, no. 8, pp. S17–S23, 2013.

2. Yaacob, B., Amin, M. C. I. M., Hashim, K., & Bakar, B. A. 2011. Optimization of Reaction Conditions for Carboxymethylated Sago Starch. *Iranian Polymer Journal*, 20 (3): 195-204.