



UNIVERSITI PUTRA MALAYSIA

***PREPARATION AND CHARACTERIZATION OF STEAMED BUNS
INCORPORATED WITH CROSS-LINKED RESISTANT STARCH TYPE-4***

FARAH SYAHIRAH BINTI ABDUL SHUKRI

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By

FARAH SYAHIRAH BINTI ABDUL SHUKRI

**Thesis Submitted to the School of Graduate Studies, Universiti Putra
Malaysia, in Fulfilment of the Requirements for the Degree of Master of
Science**

November 2017

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in
fulfilment of the requirement for the degree of Master of Science

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November 2017

Chair : Radhiah Shukri, PhD
Faculty : Food Science and Technology

Steamed buns were prepared from different percentages (5%, 10%, and 15%) of three types of cross-linked (CL) starches as wheat flour substitution. The objectives of the research were to study effects of three types of CL starches at different levels (5%, 10%, and 15%) on physicochemical properties and sensory acceptance of steamed buns. The storage study was also conducted to determine the shelf life of the fortified steamed buns at refrigeration temperature ($\pm 4^{\circ}\text{C}$) for 15 days. The study was carried out to evaluate the potential of RS as a dietary fibre in a conventional steamed bun and its potential for health benefits in food application without affecting the appearances of the steamed buns produced. Three types of CL starches which were cross-linked potato starch (CLPS), cross-linked corn starch (CLCS) and cross-linked tapioca starch (CLTS) were produced by cross-linking with a combination of 12% sodium trimetaphosphate and sodium tripolyphosphate (99:1) for 3 h at 45°C under pH 11.5. Cross-linking process demonstrated significant increase of RS and TDF content in the three different types of CL starches (CLPS, CLCS and CLTS) more than 70% compared to their native starches. For pasting properties, the viscosity of the CL starches displayed a significant decrease as CL starches resist swelling much greater than their native starches. In contrast, CL starches had higher transition temperatures but lower enthalpy values of gelatinization as compared to their native starches for the thermal properties. The gelatinization enthalpy of the CLCS was 13.03 J/g which was lower than CLPS and CLTS (15.27 J/g and 16.21 J/g, respectively). The cooked steamed buns were characterized by physicochemical and sensory properties. The volumes were positively correlated with the protein content, indicating lower gluten formation in fortified steamed buns. Fortification of CL starches in steamed buns had minimized the distribution of air cells in the buns, lowered the volume of

expansion by making them slightly denser. The CL starch-enriched steamed buns had high TDF and RS content (up to 3.8% and 3.05% respectively), improved textural properties and similar sensory acceptance as the control. The addition of 5%, 10% and 15% substitution of CL starch showed no statistically significant on the moisture content of steamed buns but was observed to increase slightly around 0.27-0.7% from the positive control (PF) for CL potato starch enriched steamed buns. Among all the starches, CL corn and CL potato starches had a better overall effect on the physicochemical and textural properties, as well as the sensory acceptance of the steamed buns. For the microbial storage study, the a_w and pH of the fortified steamed buns showed no significant changes. The steamed bun samples kept for 15 days still had total plate count lower than 4.00 log CFU/g which implied that they were safe to consume. Formation of molds only been detected in some samples on day 13 and day 15. The results indicated that CL starches, particularly CLCS and CLPS have potential to improve the nutritional properties of steamed buns with less detrimental sensorial and textural effects as compared to whole wheat flour.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia
sebagai memenuhi keperluan untuk Ijazah Master Sains

**PENYEDIAAN DAN ANALISA SIFAT-SIFAT ROTI PAU DENGAN
PENAMBAHAN KANJI RINTANG HABA HADAM JENIS-4**

Oleh

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Kuih pau disediakan dari tiga jenis kanji rintang haba untuk menggantikan tepung gandum dalam peratusan yang berbeza (5%, 10% dan 15%). Objektif kajian ini adalah untuk menyediakan tiga jenis kanji rintang haba dan mengkaji kesan kanji rintang haba tersebut dalam jumlah peratusan yang berbeza ke atas kesan fizikokimia dan penilaian penerimaan deria roti pau. Kaedah penyimpanan turut dikaji untuk melihat jangka hayat bagi roti pau tersebut. Kajian ini dijalankan untuk mengenalpasti potensi kanji rintang haba untuk penambahbaikan dari segi kesihatan dalam penghasilan makanan tanpa mengubah ciri fizikal pau yang terhasil. Tiga jenis kanji rintang haba iaitu kanji rintang haba kentang, kanji rintang haba jagung dan kanji rintang haba ubi kayu telah dihasilkan dengan kombinasi 12% natrium trimetafosfat dan natrium tripolifosfat (99:1) selama 3 jam pada 45 °C dalam pH 11.5. Proses perintang haba telah menunjukkan peningkatan jumlah RS dan TDF dalam tiga jenis kanji rintang haba (kanji rintang haba kentang, kanji rintang haba jagung dan kanji rintang haba ubi kayu) melebihi 70% berbanding kanji asal. Untuk keupayaan kepekatan, kelikatan kanji rintang haba telah memaparkan penurunan kerana kanji rintang haba lebih menolak proses gumpalan berbanding kanji asal. Selain itu, kanji rintang haba juga menunjukkan suhu transisi yang lebih tinggi tetapi rendah nilai entalpi jika dibandingkan dengan kanji asal dalam keupayaan termal haba. Nilai entalpi bagi kanji rintang haba jagung adalah 13.03 J/g iaitu lebih rendah daripada kanji rintang haba kentang dan kanji rintang haba ubi kayu (masing-masing dengan 15.27 J/g dan 16.21 J/g). Roti pau yang telah dimasak disifatkan oleh ciri-ciri fizikokimia dan penilaian penerimaan deria. Kadar pengembangan roti pau dikaitkan dengan kandungan protein, menunjukkan pembentukan gluten yang rendah dalam roti

pau kanji rintang haba. Penambahan kanji rintang haba telah mengurangkan ruang-ruang udara di dalam roti pau malah melemahkan kadar pengembangan roti pau dengan menjadikan ia lebih padat. Kanji rintang haba yang diperkayakan dalam roti pau mempunyai kandungan TDF dan RS yang tinggi (masing-masing dengan 3.8% dan 3.05%) serta tekstur roti dan penerimaan penilaian deria yang baik. Penambahan 5%, 10% dan 15% kanji rintang haba ke dalam formulasi roti pau telah menambahbaik dari aspek kelembapan dalam lingkungan 0.27-0.7% daripada roti pau kawalan positif terutamanya bagi pau kanji rintang haba kentang. Di antara semua kanji, kanji rintang haba jagung dan kanji rintang haba kentang mempunyai kesan keseluruhan (sifat-sifat fizikokimia dan tekstur, serta penerimaan penilaian deria) yang lebih baik kepada roti pau. Untuk kajian penyimpanan mikrobiologi, a_w dan pH bagi roti pau yang ditambah baik menunjukkan tiada perubahan ketara. Kesemua sampel roti pau yang disimpan selama 15 hari di dalam suhu penyejukan (± 4 °C) masih menunjukkan jumlah kiraan plat lebih rendah daripada 4.00 log CFU/g yang menggambarkan sampel roti pau masih selamat untuk dimakan. Pembentukan kulat hanya dapat dikesan di beberapa sampel roti pau pada hari ke 13 dan hari ke 15. Kesemua hasil menunjukkan bahawa kanji rintang haba, khususnya kanji rintang haba jagung dan kanji rintang haba kentang mempunyai potensi untuk memperbaiki sifat pemakanan roti pau dengan mengurangkan pengubahan tentang penerimaan penilaian deria dan kesan tekstur jika dibandingkan dengan tepung gandum bijian sempurna.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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LIST OF ABBREVIATIONS

ANOVA	Analysis of variance
CL	Cross-linked
CLPS	Cross-linked potato starch
CLCS	Cross-linked corn starch
CLTS	Cross-linked tapioca starch
CS	Corn starch
DSC	Differential scanning calorimeter
EtOH	Ethanol
g	Gram
H ₂ O	Water
HCl	Hydrochloric acid
HNO ₃	Nitric acid
KOH	Potassium hydroxide
MeOH	Methanol
NaNO ₂	Sodium nitrate
NaOH	Sodium hydroxide
Na ₂ SO ₄	Sodium sulphate
NH ₄ NO ₃	Ammonium nitrate
P	Phosphorus
PF	<i>Pau</i> flour
PFCLPS	<i>Pau</i> flour mixed cross-linked potato starch
PFCLCS	<i>Pau</i> flour mixed cross-linked corn starch
PFCLTS	<i>Pau</i> flour mixed cross-linked tapioca starch
PS	Potato starch
RS	Resistant starch
RVA	Rapid Visco Analyzer
SEM	Scanning Electron Microscopy
STMP	Sodium trimetaphosphate
STPP	Sodium tripolyphosphate
TDF	Total dietary fibre
TS	Tapioca starch
U	Enzyme unit
v/v	Volume per volume
w/w	Weight per weight
WWF	Whole wheat flour
mg	Miligram
μL	Microlitre
μM	Micromolar
°C	Degree Celcius
%	Percentage
min	Minute
h	Hour
pH	Potential of Hydroxide

CHAPTER 1

INTRODUCTION

According to the World Health Organisation (WHO) Non-Communicable Disease Country Profile in 2011, Malaysians are the most obese amongst citizens of South-East Asian countries leading by 44.2 % (Alwan A., 2011). The numbers are alarming as Malaysians that have body mass index (BMI) of over 25 are considered as overweight or obese. Malaysia is now facing an upward rush of non-communicable diseases such as diabetes and cardiovascular diseases (Kiew and Chong, 2013). Malaysians enjoys roti canai, nasi lemak, capati and traditional kuih as their staple food that contain high carbohydrates, sugar, and oil but low in fibre intake. Malaysia Reference Nutrient Intake (2005) recommended 20-30 g of dietary fibre per day for all age groups, but still Malaysians cannot achieve this. Therefore, it is crucial to incorporate more dietary fibre into Malaysians daily diet.

Dietary fibre is known to positively influence the functioning of digestive tract, microbial flora and blood cholesterol level. Being included in the fibre group, resistant starch (RS) displays hypoglycaemic effect that is similar to the health benefit of fibre. RS also has low water holding capacity, fine texture, white colour and tasteless, as opposed to other conventional fibre (Fuentes-Zaragoza et al., 2010). Resistant starch (RS) has received attention for both its potential health benefits and functional properties. Another positive advantage is its lower impact on the sensory properties of food compared with traditional sources of fibre, as whole grains, fruits or bran (Fuentes et al., 2010).

RS is categorized into five types, RS1, RS2, RS3, RS4, and RS5. RS1 is the physically inaccessible starch normally found in whole grains; RS2 consists of ungelatinized resistant granules, and RS3 is retrograded or recrystallized starch. RS4 is chemically modified starch with additional chemical bonds formed (Sajilata et al., 2006) and RS5 is amylose-lipid complex (Brown et al., 2006).

Enrichment of famous Malaysian foods with resistant starch is a good way to ensure Malaysian community consumes high fibre food for prevention and management of overweight problems. As being a well-known delicacy in Malaysia among all races, steamed bun or pau is selected to be enriched with resistant starch. In Malaysia, steamed bun is usually eaten for breakfast or tea time. The popularity of steamed bun is due to the fact that the cooking process does not involve cooking oil and other means of introducing 'fats' into the food. It is perceived as a healthy food because it

is cooked by steaming method. Basically, steamed bun is made from soft wheat flour, water, yeast, and vegetable shortening and originated from northern China where the supply of wheat is high because of wheat growing areas (Rubenthaler et al., 1990). Beside wheat flour, there is an attempt to produce steamed bun using whole wheat flour that has gained much attention among health-conscious consumers but it showed poor appearances of the bun such as darker in colour and crumbly texture.

Health-conscious people always find new ways to improve health. The awareness has become a trend due to the aging process of the population and limitations of modern medicine. Doubts surrounding lifestyle and diet along with the growing interest in functional foods and nutraceuticals also have contributed to this trend. Researches on functional foods in the past decades were focusing on vitamin fortification. Later, the focus has changed to enrich food with dietary fibre (Foschia et al., 2013). However, usage of traditional fibre, for example, whole wheat flour and bran in a steamed bun may cause undesirable changes in the physical properties of the product such as harder texture, darker colour, have distinct fibre taste and gives out a very low specific volume (Seyer and Gélinasy 2009). These changes will negatively affect the overall acceptability of consumers. Hence, fortification of steamed bun with RS is a good option because RS has a fine texture, white in colour and tasteless. The appearance of steamed bun would be more appealing to consumers with the benefits of additional fibre.

The study was carried out to evaluate the potential of RS as a dietary fibre in a conventional steamed bun and its potential for health benefits in food application. The selection of RS4 was based on factor that RS4 is more convenient to be produce by chemical modification using cross-linking agents with almost accurate amount of RS compare to RS3. In contrast, production of RS3 requires longer process (cooking and refrigeration) and the amount of RS produced is inconsistent. RS4 were produced by cross-linking three types of starches (potato, corn, and tapioca) with a combination of 12% sodium trimetaphosphate and sodium tripolyphosphate for 3 h at 45°C under alkaline condition. The proximate composition, amylose content, RS content, total dietary fibre (TDF) content, phosphorus content, pasting properties and thermal properties of cross-linked (CL) starches were determined. Steamed buns were incorporated with 5, 10, and 15% RS4 and the physicochemical, textural, and sensory acceptances of steamed buns were evaluated.

The objectives of the study were divided into three parts:

- I. To prepare and characterize the chemical, pasting and thermal properties of three types CL starches.
- II. To evaluate the effect of CL starch at different percentages on the physicochemical properties of steamed buns fortified with CL starch.
- III. To determine the sensory attributes acceptability and storage study of steamed buns fortified with CL starch.



REFERENCES

- AACC International Approved Methods of Analysis (2010), 11th Ed. Method 54-10.01. Physical Dough Test Procedure. Minnesota: AACC International.
- AACC International Approved Methods of Analysis (2010), 11th Ed. Method 54-21.02. Constant Weight Flour Procedure. Minnesota: AACC International.
- AACC International Approved Methods of Analysis (2009), 11th Ed. Method 10-05.01. Guidelines for Measurement of Volume by Rapeseed Displacement. Minnesota: AACC International.
- Abdul-Hamid, A., and Siew Luan, Y. (2000). Functional properties of dietary fibre prepared from defatted rice bran. *Journal of Food Chemistry*, 68: 15-19.
- Adebowalea, K. O., Olu-Owolabi, B. I., Olayinkaa, O. O., and Lawalb, O. S. (2005). Effect of heat moisture treatment and annealing on physicochemical properties of red sorghum starch. *African Journal of Biotechnology*, 4(9): 928-933.
- Alcázar-Alay, S. C., and Meireles, M. A. A. (2015). Physicochemical properties, modifications and applications of starches from different botanical sources. *Journal Food Science and Technology (Campinas)*, 35(2): 215-236.
- Ali, A., Wani, T. A., Wani, I. A., and Masoodi, F. A. (2016). Comparative study of the physico-chemical properties of rice and corn starches grown in Indian temperate climate. *Journal of the Saudi Society of Agricultural Sciences*, 15(1): 75-82.
- Altuna, L., Ribotta, P. D., Tadini, C. C. (2016). Effect of a combination of enzymes on the fundamental rheological behavior of bread dough enriched with resistant starch. *Journal LWT - Food Science and Technology*, 73.
- Amaral, O., Guerreiro, C. S., Gomes, A., and Cravo, M. (2016). Resistant starch production in wheat bread: effect of ingredients, baking conditions and storage. *Journal European Food Research and Technology*, 242(10): 1747-1753.
- AOAC Official Methods of Analysis of AOAC (2005). Intl. 16th ed. Method 991.43. Maryland: Association of Official Analytical Chemists.
- Aparicio-Saguilan, A., Sayago-Ayerdi, S.G., Vargas-Torres, A., Tovar, J., Ascencio-Otero, T.E., Bello-Perez, L.A. (2007). Slowly digestible cookies prepared from resistant starch-rich lintnerized banana starch. *Journal of Food Composition*, 20:175-181.
- APHA, American Public Health Association (2005). *Compadium of Methods for the Microbiological Examination of Foods*, Washington, DC, USA.

- Armero, E. (1997). Texture properties of formulated wheat dough: Relationships with dough and bread technological quality. *Journal of European Food Research and Technology*, 204: 136–145.
- Baixauli, R., Sanz, T., Salvador, A. and Fiszman, S. M. (2008). Muffins properties of the batter. *Journal of Cereal Science*, 47: 502-509.
- Balic, R., Miljkovic, T., Ozsisli, B. and Simsek, S. (2017). Utilization of modified wheat and tapioca starches as fat replacements in bread formulation. *Journal of Food Processing and Preservation*, 41(1).
- Bao, J. and Bergman, C. J. (2004). The functionality of rice starch. *Starch in Food: Structure, Function and Applications*, (pp. 258–294). New York: CRC Press LLC.
- Bednar, G. E., Patil, A. R., and Murray, S. M. (2001). Starch and fibre fractions in selected food and feed ingredients affect their small intestinal digestibility and fermentability and their large bowel fermentability *in vitro* in a canine model. *Journal of Nutrition*, 131(2): 276-286.
- Behall, K. M., Scholfield, D. J. and Hallfrisch, J. (2006). Whole-grain diets reduce blood pressure in mildly hypercholesterolemia men and women. *Journal of American Diet Association*, 106: 1445–1449.
- Bourne, M. C. (1978). Texture profile analysis. *Journal of Food Technology*, 32: 62–66.
- Brown, I. L., Yotsuzuka, M., Birkett, A. and Henriksson, A. (2006). Prebiotics, synbiotics and resistant starch. *Journal of Japanese Association for Dietary Fibre Research*, 10: 1-9.
- Canovas, A. and Perez-Alvarez, J. A. (2006). Dietary fibre: An ingredient for the development of functional foods. *Journal of Food*, 370: 65-68.
- Carmona-Garcia, R., Sanchez-Rivera, M. M., Méndez-Montealvo, G., Garza-Montoya, B., and Bello-Pérez, L. A. (2009). Effect of the cross-linked reagent type on some morphological, physicochemical and functional characteristics of banana starch (*Musa paradisiaca*). *Journal of Carbohydrate Polymers*, 76: 117-122.
- Chung, H. J., Woo, K. S., and Lim, S. T. 2004. Glass transition and enthalpy relaxation of cross-linked corn starches. *Journal of Carbohydrate Polymers*, 55: 9-15.
- CIP, The International Potato Center. (2008). The international year of the potato, IPC. Retrieved from <http://www.cipotato.org>.
- Conde-Petit, B., Nuessli, J., Arrigoni, E., Escher, F., and Amadó, R. (2001). Perspectives of starch in food science. *Chimia* 55(3):201-205.
- Constantin, G., Voicu, G., Rusănescu C. O. and Ştefan, E. M. (2011). Researches on rheological characteristics of dough of wheat flour and their changes during storage. *Bulletin UASVM Agriculture*, 68(2):212-219.
- Delcour, J. A. and Eerlingen, R. C. (1995). Formation, analysis, structure and properties of type III enzyme resistant starch. *Journal of Cereal Science*, 22(2):129-138.

- Ellis, R. P., Cochrane, M. P., Dale, M. F. B., Duffus, C. M., Lynn, A., Morrison, I. M., Prentice, R. D. M., Swanston, J. S., and Tiller, S. A. (1998). Starch production and industrial use. *Journal of Science Food Agriculture*, 77: 289–311.
- Englyst, H. N., Kingman, S. M. and Cummings, J. H. (1992). Classification and measurement of nutritionally important starch fractions. *European Journal of Clinical Nutrition*, 46(2): 33-50.
- Faridi, H. A., Rubenthaler, G. L. (1983). Laboratory method for producing Chinese steamed bread and effects of formula, steaming and storage on bread starch gelatinization and freshness. Proceeding: 6th *International Wheat Genetics Symposium, Kyoto, Japan*. Kyoto University, 863-867.
- Ferguson, L. R., Tasman-Jones, C., Englyst, H. and Harris, P. J. (2000). Comparative effects of three resistant starch preparations on transit time and short-chain fatty acid production in rats. *Journal Nutrition and Cancer*, 36(2): 230–237
- Filer, L. J. (1971). Modified food starches for use in infant foods. *Nutrition Reviews*, 29: 55-59.
- Foschia, M., Beraldo, P., Peressini, D. (2017). Evaluation of the physicochemical properties of gluten-free pasta enriched with resistant starch. *Journal of the Science of Food and Agriculture*, 97(2): 572-577.
- Fu, L., Tian, J.C., Sun, Z., (2010). Effect of resistant starch on processing quality of northern style Chinese steamed bread. *Journal of China Cereals and Oils Association*, 25: 53–56 (In Chinese, English abstract).
- Fu, L., Tian, J., Sun, C. and Li, C. (2008). RVA and farinograph properties study on blends of resistant starch and wheat flour. *Journal Agriculture Science China*, 7: 812–822
- FAO/WHO (Food and Agriculture Organisation/World Health Organization). (2002). Diet, nutrition and the prevention of chronic diseases. Report of a joint FAO/WHO expert consultation. Rome and Geneva: WHO Technical Report Series No. 916.
- FDA, Food and Drug Administration (U.S.A), (1984). Bacteriological Analytical Manual. 6th ed. *Association of Official Analytical Chemists*, Arlington, VA.
- Fuentes, Z. E., Riquelme, N. M. J., Sánchez, Z. E. and Pérez, Á. J. A. (2010). Resistant starch as functional ingredient: A review. *Journal of Food Research International*, 43: 931-942.
- Gaillard, T., Bowler, P. (1987). In: Gaillard T, editor. *Starch: Properties and Potential*. New York: John Wiley and Sons. Vol. 55.
- Gao, H., Cai, J., Han, W., Huai, H., Chen, Y., and Wei, C. (2014). Comparison of starches isolated from three different *Trapa* species. *Food Hydrocolloids*, 37: 174-181.
- Han, J. A., BeMiller, J. N. (2007). Preparation and physical characteristics of slowly digesting modified food starches. *Journal of Carbohydrate Polymer*, 67: 366-374.

- Haralampu, S. G. (2000). Resistant starch: A review of the physical properties and biological impact of RS3. *Journal of Carbohydrate Polymer*, 41: 285-292.
- Harkema, J. (1996). Starch derived fat mimetics from potato. In: *Handbook of Fat Replacers*, edited by S Roller & SA Jones. (pp. 119-129) USA: CRC Press LLC.
- He, J., Liu, J., Zhang, G. (2008). Slowly digestible waxy maize starch prepared by octenyl succinic anhydride esterification and heat-moisture treatment: glycemic response and mechanism. *Journal of Biomacromolecules*, 9: 175–84.
- Higgins, J. A. (2014). Resistant starch and energy balance: impact on weight loss and maintenance. *Journal Critical Review of Food Science and Nutrition* 54(9): 1158-1166.
- Hizukuri, S., Tabata, S., Nikuni, Z. (1970). Studies on starch phosphate: 1. Estimation of glucose-6-phosphate residues in starch and presence of other bound phosphate(s). *Journal of Starch*, 22: 338–343.
- Hoseney, R. C. (1994) Minor constituents of cereals. *Principles of Cereal Science and Technology* 2: 81-101. American Association of Cereal Chemists St. Paul, MN.
- Hou, G. G. and Popper, L. (2007). Chinese steamed bread. In: Popper, L., Schäfer, S. and Freund, W. (Eds.). *Future of flour-A compendium of flour improvement* (pp.309-318).
- Huber, K. C. and BeMiller, J. N. (2000). Channels of maize and sorghum starch granules. *Journal of Carbohydrate Polymers*, 41: 269–276.
- Huang, S., Miskelly, D. (2016). *Steamed Breads: Ingredients, Process and Quality*. Woodhead Publishing Series in Food Science, Technology, and Nutrition: 303
- Huang, S., Quail, K., Moss, R. and Best, J. (1995). Objective methods for the quality assessment of northern-style Chinese steamed bread. *Journal of Cereal Science*, 21: 49- 55.
- Huang, S. D., Miskelly, D. M. and Moss, R. (1991). Recent advances in research on Chinese steamed bread. In: OJ. Martin & e.W. Wrigley (eds.). *Proceeding of a Cereal International Conference*, Brisbane, Australia.
- Hung, P.V., Morita, N. (2005). Physicochemical properties and enzymatic digestability of starch from edible canna (*Canna edulis*) grown in Vietnam. *Journal of Carbohydrate Polymer*, 61: 314–321.
- Jane, J., Radosavljevic, M., and Seib, P. A. (1992). Location of amylose in normal starch granules. I. Susceptibility of amylose and amylopectin to cross-linking reagents. *Journal of Cereal Chemistry*, 69: 406–409.
- Kiew, C. F., and Chong, C. P., (2013). A Community Health Screening Conducted at Sungai Pinang Township, State of Pulau Pinang, Malaysia. *Malaysian Journal of Pharmaceutical Sciences*, 11(1): 59–68.

- Kim, M. J., Choi, S. J., Shin, S. I. and Sohn, M. R. (2008). Resistant glutarate starch from adlay: Preparation and properties. *Carbohydrate Polymers*, 74: 787–796.
- Lei, F. U., Tian, J-C., Sun, C-L. and Chun, L. I. (2008). RVA and Farinograph Properties Study on Blends of Resistant Starch and Wheat Flour. *Journal of Agricultural Sciences in China*, 7(7): 812-822.
- Lattimer, J. M., Haub, M. D. (2010). Effects of dietary fibre and its components on metabolic health. *Journal of Nutrients*, 2(12): 1266–1289.
- López, O. V., Zaritzky, N. E., and García, M. A. (2010). Physicochemical characterization of chemically modified corn starches related to rheological behavior, retrogradation and film forming capacity. *Journal of Food Engineering*, 100(1):160-168.
- Miller, R. A., Jeong, J. and Maningat, C. C. (2011). Effect of RS4 Resistant Starch on Extruded Ready-to-Eat (RTE) Breakfast Cereal Quality. *Journal of Cereal Chemistry*, 88(6): 584–588.
- Mir, J. A., Srikaeo, K., García, J. (2013). Effects of amylose and resistant starch on starch digestibility of rice flours and starches. *International Food Research Journal*, 20: 1329–1335.
- Mishra, S., and Rai, T. (2006). Morphology and functional properties of corn, potato and tapioca starches. *Food Hydrocolloids*, 20: 557–566.
- Morais, M. B., Feste, A., Miller, R. G. and Lifichitz, C. H. (1996). Effect of resistant starch and digestible starch on intestinal absorption of calcium, iron and zinc in infant pigs. *Paediatric Research*, 39(5): 872–876
- Morrison, W. R., and Azudin, M. N. (1987). Variation in the amylose and lipid contents and some physical properties of rice starches. *Journal of Cereal Science*, 5(1): 35-44.
- Nofrarías, M., Martínez-Puig, D., and Pérez J. F. (2009). Potential health benefits of potato starch. *Journal of Food*, 3(2).
- Östergård, K., Björck, I. and Gunnarsson, A. (1988). A study of native and chemically modified potato starch. Part I: Analysis and enzymic availability in vitro. *Journal of Starch*, 40: 58–66.
- Páramo-Calderón, D. E., Carrillo-Ahumada, J., Juárez-Arellano, E. A., Bello-Pérez, L. A., Aparicio-Saguilán, A. and Alvarez-Ramirez, J. (2016), Effect of cross-linking on the physicochemical, functional and digestibility properties of starch from Macho (*Musa paradisiaca L.*) and Roatan (*Musa sapientum L.*) banana varieties. *Journal of Starch*, 68: 584–592.
- Pateras, Irene M. C. (2007), Bread Spoilage and Staling. In: *Technology of Breadmaking* (pp. 275-298). Boston, MA, USA.
- Pérez, S. and Bertoft, E. (2010). The molecular structures of starch components and their contribution to the architecture of starch granules: A comprehensive review. *Journal of Starch*, 62(8): 389-420.

- Raben, A., Tagliabue, A., Christensen, N. J., Madsen, J., Holst, J. J. and Astrup, A. (1994). Resistant starch: The effect on postprandial glycemia, hormonal response, and obesity. *American Journal Clinical Nutrition*, 60: 544-550.
- Ratnayake, W.S. and Jackson, D.S. (2008), Thermal Behavior of Resistant Starches RS 2, RS 3, and RS 4. *Journal of Food Science*, 73: 356–366.
- Ren, C. and Shin, M. (2013). Effects of Cross-linked Resistant Rice Starch on the Quality of Korean Traditional Rice Cake. *Journal of Food Science and Biotechnology*, 22(3): 697-704.
- Rosell, C. M., Rojas, J. A. and Benedito de Barber, C. (2001). Influence of hydrocolloids on dough rheology and bread quality. *Journal of Food Hydrocolloids*, 15(1): 75–81.
- Rubenthaler, G. L., Huang, M. L., and Pomeranz, Y. (1990). Steamed Bread. I. Chinese Steamed Bread Formulation and Interactions. *Journal of Cereal Chemistry*, 67(5):471-475.
- Sajilata, M. G., Singhal R. S. and Kulkarni, P. R. (2006). Resistant Starch-A Review. *Comprehensive Reviews in Food Science and Food Safety*, 5(1): 1-17.
- Seneviratne, H. D., Biliaderis, C. G. 1991. Action of α -amylases on amylose-lipid complex superstructures. *Journal of Cereal Science*, 13: 129–143.
- Seyer, M. E. and Gélinasy, P. 2009. Bran characteristics and wheat performance in whole wheat bread. *International Journal of Food Science and Technology*, 44: 688–693.
- Shelton, D. (2008). Wheat and Flour Testing Methods: A Guide to Understanding Wheat and Flour Quality, Version 2. *Wheat Marketing Center*, Kansas State University, USA.
- Shukri, R., and Shi, Y. (2015). Physicochemical properties of highly cross-linked maize starches and their enzymatic digestibilities by three analytical methods. *Journal of Cereal Science*, 63: 72-80.
- Shukri, R., Zhu, L., Seib, P. A., Maningat, C., and Shi, Y. (2014). Direct in-vitro assay of resistant starch in phosphorylated cross-linked starch. *Journal of Bioactive Carbohydrates and Dietary Fibre*, 5(1): 1-9.
- Siegrist, M., Stampfli, N., Kastenholz, H., (2008). Consumers' willingness to buy functional foods: The influence of carrier, benefit and trust. *Appetite*, 51:526–9.
- Singh, J., Kaur, L., and McCarthy, O. J. (2007). Factors influencing the physico-chemical, morphological, thermal and rheological properties of some chemically modified starches for food applications-A review. *Journal of Food Hydrocolloids*, 21(1): 1-22.
- Singh, N., Kaur, L., Sandhu, K. S., Kaur, J., and Nishinari, K. (2006). Relationships between physicochemical, morphological, thermal, rheological properties of rice starches. *Journal of Food Hydrocolloids*, 20: 532–542.

- Singh, N., Singh, J., Kaur, L., Sodhi, N. S., and Gill, B. S. (2003). Morphological, thermal and rheological properties of starches from different botanical sources. *Journal of Food Chemistry*, 81(2): 219-231.
- Tester, R. F., Karkalas, J. and Qi, X. (2004). Starch - Composition, fine structure and architecture. *Journal of Cereal Science*, 39(2): 151-165.
- Tester, R. F. and Debon, S. J. J. (2000). Annealing of starch - a review. *International Journal of Biological Macromolecules* 27(1):1-12.
- Topping, D. L. and Clifton, P. M. 2001. Short-chain fatty acids and human colonic function: Roles of resistant starch and non-starch polysaccharides. *Journal of Physiological Review*, 81(3): 1031-1064.
- Tsen, C. C., Reddy, P. R. K. and Gehrke, C. W. (2006). Effects of conventional baking, microwave baking, and steaming on the nutritive value of regular and fortified breads. *Journal of Food Science*, 42(2): 402-406.
- Udeme Joshua Josiah Ijah, Helen Shnada Auta, Mercy Oluwayemisi Aduloju, and Sesan Abiodun Aransiola. (2014). Microbiological, Nutritional, and Sensory Quality of Bread Produced from Wheat and Potato Flour Blends. *International Journal of Food Science*, 6.
- Vasanthan, T., Bergthaller, W., Driedger, D., Yeung, J., and Sporns, P. (1999). Starch from Alberta potatoes: Wet-isolation and some physicochemical properties. *Food Research International*, 32: 355–365.
- Vonk, R. J., Hagedoorn, R. E., Graaff, R. (2000). Digestion of so-called resistant starch sources in the human small intestine. *Journal of American Society for Clinical Nutrition*, 72(2):432-438.
- Wang, J., Rosell, C. M., Barber, C. B. (2002). Effect of the addition of different fibres on wheat dough performance and bread quality. *Journal of Food Chemistry*, 79: 221-226.
- Whistler, R. L., BeMiller, J. N. (1997). *Carbohydrate Chemistry for Food Scientists*, (pp. 117–151). Eagan Press: St. Paul, USA.
- Włodarczyk-Stasiak, M., Mazurek, A., Kowalski, R., Pankiewicz, U., Jamroz, J. (2017). Physicochemical Properties of Waxy Corn Starch after Three-Stage Modification. *Food Hydrocolloids*, 62: 182–190.
- Wolf, B. W., Bauer, L. L., and Fahey, G. C. (1999). Effects of chemical modification on in vitro rate and extent of food starch digestion: an attempt to discover a slowly digested starch. *Journal of Agriculture Food Chemistry*, 47: 4178–4183.
- Wongsagonsup, R., Shobsngob, S., Oonkhanond, B., Varavinit, S. (2005). Zeta Potential (ζ) and Pasting Properties of Phosphorylated or Crosslinked Rice Starches. *Starch-Stärke*, 57: 32–37.
- Woo, K. S. and Seib, P. A. (2002). Cross-linked Resistant Starch: Preparation and Properties. *Journal of Cereal Chemistry*, 79(6): 819-825.
- Yao, N., Paez, A. V., and White, P. J. (2009). Structure and function of starch and resistant starch from corn with different doses of mutant amylose-extender and floury-1 alleles. *Journal of Agriculture and Food Chemistry*, 57: 2040-2048.

- Yeo, L. L., and Seib, P. A. (2009). White pan bread and sugar-snap cookies containing wheat starch phosphate, a cross-linked resistant starch. *Journal of Cereal Chemistry*, 86: 210-220.
- Yue, P. and Waring, S. (1998). Resistant starch in food applications. *Cereal Foods World*, 43(9): 690–695.
- Yusuph, M., Tester, R. F., Ansell, R., and Snape, C. E. (2003). Composition and properties of starches extracted from tubers of different potato varieties grown under the same environmental conditions. *Journal of Food Chemistry*, 82: 283–289.
- Zaman S. A. and Sarbini S. R. (2015). The potential of resistant starch as a prebiotic. *Journal Critical Review in Biotechnology*, 36(3): 578-584.
- Ziobro, R., Korus, J., Witczak, M., and Juszczak, L. (2012). Influence of modified starches on properties of gluten-free dough and bread. Part II: Quality and staling of gluten-free bread. *Journal of Food Hydrocolloids*, 29(1):68–74.
- Zipkes, M. R., Gilchrist, J. E., and Peeler J. T. (1981). Comparison of yeast and mold counts by spiral, pour, and streak plate methods. *Journal of Association Official Analytical Chemistry*, 64: 1465-1469.