สมบัติของเมือกจากผลพุงทะลาย *Scaphium scaphigerum* และผลต่อสมบัติและโครงสร้างจุลภาคของอิมัลชันเนื้อสัตว์ไขมันปกติและไขมันต่ำ

นางพร้อมลักษณ์ สมบูรณ์ปัญญากุล

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PROPERTIES OF MALVA NUT *Scaphium scaphigerum* MUCILAGE AND ITS EFFECTS ON PROPERTIES AND MICROSTRUCTURES OF NORMAL AND LOW FAT MEAT EMULSIONS

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พร้อมลักษณ์ สมบูรณ์ปัญญากุล: สมบัติของเมือกจากผลพุงทะลาย *Scaphium scaphigerum* และผลต่อสมบัติและโครงสร้างจุลภาคของอิมัลชันเนื้อสัตว์ไขมันปกติและไขมันต่ำ (PROPERTIES OF MALVA NUT *Scaphium scaphigerum* MUCILAGE AND ITS EFFECTS ON PROPERTIES AND MICROSTRUCTURES OF NORMAL AND LOW FAT MEAT EMULSIONS) อ. ที่ปรึกษา: รศ. ดร. พันธิพา จันทวัฒน์ อ. ที่ปรึกษาร่วม: รศ. ดร. นินนาท ชินประทีปฐ์, ศ. ดร. ชัย บานุท, 201 หน้า. ISBN 974-53-1863-9

กัมจากผลพุงทะลายที่สกัดด้วยด่างประกอบด้วยคาร์โบไฮเดรต 83.1%, เถ้า 8.4% และ โปรตีน 8.3% น้ำตาลโมเลกุลเดี่ยวที่เป็นองค์ประกอบ ได้แก่ น้ำตาลอะราบิโนส 17.1%, กาแลคโตส 15.1%, แรมโนส 15.0% ในอัตราส่วนโมล (molar ratio) 1.00 : 0.21 : 1.24 มีน้ำตาลกลูโคส,ไซโลสและแมนโนสในปริมาณเล็กน้อยและมีกรดยูโรนิก 6.4% กัมมีน้ำหนักโมเลกุลเฉลี่ยและ intrinsic viscosity (η) เป็น 6.65 x 10^6 Da และ 10.0 dl/g ตามลำดับ การทำให้บริสุทธิ์โดยวิธี dialysis หรือ dialysis และฟอกสีด้วยไฮโดรเจนเปอร์ออกไซด์ 30% ที่ความเป็นกรด-ด่าง 9 ส่งผลให้กัมที่สกัดด้วยด่างมีน้ำหนักโมเลกุลเฉลี่ยและ η ลดลง สเปกตรัมจาก FT-IR แสดงให้เห็นว่ากัมที่สกัดด้วยด่างไม่มีหมู่เมทธิลเนื่องจากด่างทำให้หมู่เมธิลที่เอสเทอริไฟด์ที่หมู่คาร์บอกซิลสูญเสียไป สารละลายจากกัมบริสุทธิ์ (PMG) เข้มข้น 0.1 ถึง 4.0% มีลักษณะการไหลในช่วงอัตราเฉือนระหว่าง 0.01 ถึง 1000 s^{-1} เป็นแบบ shear thinning PMG เข้มข้น 0.5 ถึง 4.0% มีโครงสร้างเจลที่ไม่แข็งแรง โดย G′ (storage modulus) มีค่ามากกว่า G″ (loss modulus) ตลอดช่วงความถี่จาก 0.01 ถึง 10.00 Hz นอกจากนี้ทั้ง G′ และ G″ ยังแปรตามความถี่โดยเฉพาะอย่างยิ่งที่ระดับความเข้มข้น 0.5% การเติมสารละลายแคลเซียมคลอไรด์ 5 ถึง 15 mM ใน PMG เข้มข้น 1.5% ไม่ทำให้ค่า G′ และ G″ เปลี่ยนแปลง PMG 1.5% ซึ่งมีแคลเซียมคลอไรด์ 5 ถึง 15 mM ไม่หลอมเหลวระหว่างช่วงอุณหภูมิ 10 ถึง 60 °C ในงานวิจัยนี้ยังได้ศึกษาถึงผลของกัมพุงทะลายที่มีความบริสุทธิ์น้อย (CMG) และ PMG ต่อคุณภาพ batter และอิมัลชันเนื้อไก่ สำหรับ batter เนื้อไก่ พบว่าการเพิ่มปริมาณแกลือมีผลในการเพิ่มน้ำหนักของ batter ภายหลังการทำให้สุก (cook yield) การเติม CMG 0.2% ช่วยเพิ่มน้ำหนักหลังทำให้สุกของ batter ที่มีเกลือผสมทุกระดับ แต่เป็นผลให้ค่า cohesiveness และ chewiness ลดลง การเติม CMG และ phosphate (STPP) ทำให้เสถียรภาพของ batter ดีขึ้น การเพิ่มปริมาณแกลือและเติม PMG 0.1% ใน batter ช่วยลดการสูญเสียน้ำหนักหลังทำให้สุก (cook loss) ได้ นอกจากนี้ batter ที่มี PMG และเกลือทุกระดับมีค่า springiness ต่ำกว่า batter ที่ปราศจาก PMG batter ที่มีส่วนผสมของเกลือ 2%, STPP 0.5% และ PMG 0.1% มีค่า fracture force, fracture distance, springiness และ chewiness ต่ำกว่า batter ที่มีเกลือ 2% และ STPP 0.5% สำหรับผลของ CMG และ PMG ต่อคุณภาพของอิมัลชันเนื้อไก่ 2 ชนิด พบว่าอิมัลชันเนื้อไก่บดแยกกระดูกด้วยเครื่อง (MDCM emulsion) ที่มีส่วนผสมของ CMG 0.2% หรือ 0.6% และ STPP 0.5% สูญเสียน้ำหนักจากการทำให้สุกต่ำกว่าอิมัลชันที่มีเฉพาะ CMG และอิมัลชันที่ไม่มี CMG และ STPP การเติม STPP 0.5% ช่วยลดการสูญเสียไขมันระหว่างการทำให้สุกและปรับปรุงเสถียรภาพของอิมัลชัน สำหรับอิมัลชันชนิดที่ 2 ซึ่งเตรียมจากเนื้ออกไก่ พบว่า PMG หรือ CMG ลดการสูญเสียน้ำหนักจากการทำให้สุก อิมัลชันที่มี PMG และ STPP สูญเสียไขมันจากการทำให้สุกน้อยกว่าอิมัลชันที่ไม่มีทั้งกัม และ STPP อิมัลชันที่มีส่วนผสมของ PMG 0.1% หรือ 0.3% และ STPP 0.4% มีค่า hardness, springiness, cohesiveness และ chewiness สูงกว่าอิมัลชันที่มีเฉพาะ PMG นอกจากนี้พบว่าการเติม PMG, CMG หรือ STPP มีผลให้ค่า G′ ในช่วงการทำให้สุกที่อุณหภูมิระหว่าง 30 °C ถึง 70 °C และการทำให้เย็นที่อุณหภูมิระหว่าง 70 °C ถึง 30 °C เพิ่มขึ้น ในส่วนของผลิตภัณฑ์แฟรงค์เฟอเตอร์ไก่ พบว่าแฟรงค์เฟอเตอร์ที่มี CMG 0.2% มีการสูญเสียน้ำหนักจากการทำให้สุกต่ำ มีลักษณะเนื้อสัมผัสดีกว่าแฟรงค์เฟอเตอร์ที่ไม่มี CMG และได้รับการยอมรับสูงสุด การเติม PMG หรือ CMG มีผลทำให้ค่าความสว่างของอิมัลชันและผลิตภัณฑ์ลดลง จากการศึกษาทั้งหมดจะเห็นได้ว่ากัมจากพุงทะลายมีศักยภาพในการปรับปรุงผลผลิตและเสถียรภาพของ batter และอิมัลชันจากเนื้อไก่

ภาควิชาเทคโนโลยีทางอาหาร
สาขาวิชาเทคโนโลยีทางอาหาร
ปีการศึกษา **2548**

ลายมือชื่อนิสิต.....
ลายมือชื่ออาจารย์ที่ปรึกษา.....
ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.....

###4473821423 : MAJOR FOOD TECHNOLOGY
KEY WORD : MALVA NUT/*Scaphium scaphigirum* /GUM/MUCILAGE/MEAT BATTER/ MEAT EMULSION/MICROSTRUCTURE

PROMLUCK SOMBOONPANYAKUL: PROPERTIES OF MALVA NUT *Scaphium scaphigerum* MUCILAGE AND ITS EFFECTS ON PROPERTIES AND MICROSTRUCTURES OF NORMAL AND LOW FAT MEAT EMULSIONS. THESIS ADVISOR: ASSOC. PROF. PANTIPA JANTAWAT, Ph. D. THESIS CO-ADVISOR: ASSOC. PROF. NINNART CHINPRAHAST, Ph. D. and PROF. SHAI BARBUT, Ph. D. 201 pp. ISBN : 974-53-1863-9

The alkaline extracted gum from malva nut (*Scaphium scaphigerum*) contained 83.1% carbohydrate, 8.4% ash, and 8.3% protein. The monosaccharide compositions of the gum were 17.1% arabinose, 15.1% galactose, 15.0% rhamnose with the molar ratio of 1.00 : 0.21 : 1.24 and small amounts of glucose, xylose and mannose. The gum also contained 6.4% uronic acid. The average molecular weight (Mw) and intrinsic viscosity (η) of the gum were 6.65×10^6 Da and 10.0 dl/g, respectively. Dialysis or dialysis plus decoloring with 30% H_2O_2 at pH 9 reduced Mw and η of the gum. The FT-IR spectrum of the alkaline extracted gum revealed the loss of methyl esterifed carboxyl group. The 0.1 to 4.0% purified malva nut gum (PMG) solutions had the shear thinning behavior over a range shear rates of 0.01 to 1000 s^{-1}. The 0.5 to 4.0% PMG solutions exhibited a weak gel structure with G′ (storage modulus) being higher than G″ (loss modulus) over the entire frequency ranges from 0.01 to 10.00 Hz. However, both G′ and G″ showed appreciable frequency dependence, especially for the 0.5% PMG. The addition of 5 to 15 mM $CaCl_2$ to 1.5% PMG solutions did not change the values of G′ and G″. All 1.5% PMG gels with 5 to 15 mM $CaCl_2$ did not melt within the temperature range studied (10-60 °C). Furthermore, effects of crude malva nut gum (CMG) and PMG on qualities of lean meat batters and meat emulsions were studied. In the lean poultry meat batters, increasing the salt level resulted in an overall increase of cook yield, and the addition of 0.2% CMG further improved yields at all salt levels. However cohesiveness and chewiness were reduced. Addition of CMG and STPP resulted in more stable batters. Furthermore, increasing salt level, along with the addition of 0.1% PMG, was beneficial in reducing the cook loss of lean chicken meat batter. At all salt levels, batters with PMG showed lower springiness comparing to batters without PMG. Batter with 2% salt, 0.5% STPP and 0.1% PMG showed lower fracture force, fracture distance, springiness and chewiness comparing to batter with only 2% salt and 0.5% STPP. In addition, the effects of CMG and PMG on qualities of the meat emulsions were investigated. Firstly, for the mechanically deboned chicken meat (MDCM) emulsions, the emulsions with 0.2% CMG or 0.6% CMG and 0.5% STPP had the lower cook loss when compared with the emulsions containing only CMG and the emulsion without CMG and STPP. Addition of 0.5% STPP decreased fat loss and improved stability of the MDCM emulsions. Secondly, for the emulsions from chicken breast meat, the addition of PMG or CMG reduced cook losses of the emulsions. The emulsions with PMG and STPP had lower fat losses when compared with the emulsion without malva nut gums and STPP. The addition of PMG and STPP resulted in more stable emulsions. The emulsions with 0.1% PMG or 0.3% PMG and 0.4% STPP showed higher values of hardness, springiness, cohesiveness and chewiness comparing to the emulsions with only PMG. Furthermore, G′ of the emulsions during cooking between 30 °C to 70 °C and cooling from 70 °C to 30 °C increased by the addition of PMG, CMG or STPP. For the chicken frankfurters, the frankfurters with 0.2% CMG had lower cook loss and better textural properties than the frankfurters without CMG. Sensory analysis results indicated that the frankfurters with 0.2% CMG were the most acceptable. Addition of CMG or PMG resulted in darker color of all poultry meat samples. Overall, this study indicated the potential use of malva nut gums for improving yield and stability of the lean meat batters and meat emulsions.

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CHAPTER I

INTRODUCTION

Sufficient binding of minced meat particles and high water retention are two important factors in marketing high quality processed meat products. The actual binding depends on factors such as the type and concentration of salt, temperature and pH of the meat. Binding of meat particles occurs during cooking as heat coagulation of the proteins takes place. This can be measured as an increase in the shear force value during cooking (Asghar *et al*., 1985; Saliba *et al*., 1987; Gordon and Barbut, 1992b). However, during cooking, water binding decreases due to protein denaturation (Schults and Wierbicki, 1973). Various non-meat proteins and carbohydrates especially hydrocolloid gums are often used to enhance the water binding and texture of meat products (Lanier, 1991). These meat binders are commonly used at low concentrations, ranging from 0.5% to 5%, and usually do not contribute much to the nutritional values, taste, or aroma of the final product (Krumel and Sarkar, 1975). Various researchers have reported on the applications of gums as meat binders, texture stabilizers and fat replacers. Fox *et al.* (1983) reported that xanthan gum and carrageenan stabilized the texture of pickled frankfurters while guar gum, gum arabic and locust bean gum had no such effect. Trudso (1985) showed that carrageenan improved water retention, consistency, sliceability and texture of poultry products especially with high levels of added brine. Foegeding and Ramsey (1986) revealed that ι-carrageenan, κ-carrageenan, guar gum,

locust bean gum, xanthan gum and κ-carrageenan/locust bean gum mixture could be used to make acceptable low-fat frankfurters. Bater *et al.* (1992) indicated that addition of 0.5% κ-carrageenan to oven-roasted turkey breasts improved sliceability, rigidity, and decreased expressible juice. Addition levels of salt, polyphosphates, and κ-carrageenan at approximately 2.7%, 0.17% and 2%, respectively, produced low fat emulsified meatballs which were more acceptable, and showed higher cooking yields (Hsu and Chung, 2001). Moreover, Morin *et al.* (2004) indicated that β-glucan gum held more water in cooked sausages than carboxymethyl cellulose due to its ability to form a tighter network within the protein matrix.

Malva nut fruit [*Scaphium scaphigerum* (G.Don) Guib & Planch] is known in Thailand as “Pungtalay” or “Sumrong”. The plant belongs to the *Sterculiaceae* family which includes other species such as *Scaphium macropodum* Beumee and *Sterculia lychnophora* Hance. *Scaphium scaphigerum* is growing in Vietnam, China, Malaysia, Indonesia as well as the eastern part of Thailand especially in the drier regions (Yamada *et al.*, 2000). Malva nuts are harvested from this native tall tree (20-40 m). The dry fruit is about 25 x 15 mm, ellipsoid in shape and glabrous. Large amount of mucilaginous substance can be extracted from the fruit by soaking it in water. The mucilage, when sweetened, can be consumed as a dessert, but its principal uses are in relief of canker sore and cough. It is also used, in China, as a traditional drug for the prevention of pharyngitis, treatment of tussis and constipation (Wang *et al*., 2003). Chemical analysis of an alkaline extracted malva nut gum (Somboonpanyakul *et al.*, 2004b) revealed that its carbohydrate content is 83.1%, ash 8.4%, and protein 8.3%. The major carbohydrates are the monosacharides arabinose (17.1%) and galactose

(15.1%). The gum also contains 6.4% uronic acid (as galacturonic acid equivalent) and small amounts of glucose, xylose and mannose; and the overall molecular weight is 3.3×10^{6} daltons.

Thailand imports a substantial volume of gums and mucilages every year. The most commonly imported gums are pectic substances, seed gums (locust bean and guar gums), with a 2004 annual value being $ 6,654,429 and $ 3,730,135, respectively (Thai Customs Department, 2004). Thus, the interest in finding a local source that can provide a less costly substitution for these imports is of a concern and there could be a substantial domestic market for a Thai-produced mucilage with appropriate properties and low cost to substitute some of the uses of these imported materials. Furthermore, malva nut gum is not commonly used as a stabilizer or a thickening agent by the food industry at present. The main reason appears to be the lack of reliable information on its functional properties and interactions with other food components such as meat proteins. Overall, a better understanding of an extraction procedure, physicochemical and rheological properties and the interactions between the gum and meat proteins is important in any new meat product development or even in improving the functionalities of existing meat and non-meat products on the market.

1.1 Objectives

The goal of this research was to develop an extraction procedure for malva nut gum and determine its physicochemical and rheological properties. Since a better understanding of interactions between gum and meat proteins is essential in meat product development, the more specific objectives of this research were established as follows:

1.1.1 to develop an extraction procedure to isolate the gum from dried malva nut fruits, partially purify and characterize its physicochemical properties,

1.1.2 to evaluate the rheological properties of the extracted gum,

1.1.3 to examine the effect of malva nut gums, either crude or purified, on cook yields, textural properties and microstructures of the lean poultry meat batters which include low and regular salt/phosphate levels, and

1.1.4 to determine the effect of malva nut gums, either crude or purified, on cook yields, color, textural and sensory properties, and microstructures of the emulsion type poultry meat systems.

CHAPTER II

LITERATURE REVIEW

2.1 Meat Binding

Some types of meat products have the ability to bind their constituent meat particles together to form a cohesive product. Factors that determine the efficiency of meat binding are protein extraction, mechanical treatment, ionic environment and temperature of heat processing. The binding abilities of meat proteins has been of interest to the meat processing industry for many years and has been the subject of many studies. Most research in this area has been directed towards studying the binding involved in meat patties and sausage products. Nowadays, interest in binding has turned to that involved in chunked and formed meat products. These products are made from meat pieces or chunks of meat that can vary in size from particles as small as 3 g to chunks, 250 g or greater. The meat pieces are processed in such a way as to form a protein matrix between them. This is achieved by applying mechanical work or energy to the meat, either fresh or cured, using methods such as mixing, churning, pounding, tumbling or massaging until the meat becomes soft and pliable (Schmidt and Trout, 1984).

2.2 Properties of Meat Proteins

Generally, lean meat is composed of 75% water, 20% protein, 3% fat and 2% non-nitrogenous soluble substances. The non-nitrogenous soluble substances consist carbohydrates (glycogen, glucose, glucose-6-phosphate and lactic acid) and minerals (phosphorus, magnesium, zinc, iron, potassium, sodium and trace metals). The water-soluble vitamins are also a part of this group (Schult, 1976; Forrest *et al*., 1975). Muscle proteins can be divided into three groups, i.e., the sarcoplasmic proteins or water soluble proteins (soluble in salt solutions of low ionic strength, $\leq$ 0.1), the myofibrillar or structural proteins (soluble in concentrated salt solutions of 0.5 to 0.6 ionic strength) and the connective tissue proteins or stroma protein (insoluble in both) (Szent-Gyorgyi, 1951; Perry, 1956).

2.2.1 Sarcoplasmic Proteins

Sarcoplasmic proteins are composed of globular proteins, which are soluble in solutions of low-salt concentration but not in water, and water-soluble albumins, such as myoglobin and the myogen fraction (to which most of the enzymes of glycolysis belong). The sarcoplasmic proteins have isoelectric points between pH 6.0 and 7.0, molecular weights in the range 30,000 to 100,000 (Bendall, 1964), and amount to about 30% of the total proteins (Lawrie, 1974).

2.2.2 Myofibrillar Proteins

The salt-soluble myofibrillar proteins comprised about 60% of the total muscle protein (Lawrie, 1974). The major protein groups are myosin-actin-actomyosin, tropomyosin-troponin, alpha actinin and minor myofibrillar components. Myosin, approximately 35% of muscle protein, is the most abundant of all proteins found in meat (Hansen and Lowy, 1964). It is a thread-like molecule with a high length-to-diameter ratio (40:1), having a molecular weight of 470,000 to 500,000 (Frederickson and Holtzer, 1968). Myosin contains a large amount of aspartic and glutamic acid residues and a fair amount of the basic residues histidine, lysine and arginine. From the amino acid composition, it is to be expected that myosin will be a negatively charged protein at physiological pH. The isoelectric point of myosin is approximately 5.4 (in KCl solution), but in the presence of magnesium or calcium ions it rises to 9.3, showing strong preferential bonding of the two divalent cations over the monovalent cations sodium and potassium (Bendall, 1964). Actin represents about 15% of the total muscle protein (Hansen and Lowy, 1964). When actin is extracted with water from muscle tissue, it is obtained in the globular form, having a molecular weight of from 44,000 to 49,000 (Hay *et al*., 1973). Tropomyosin and troponin together account for 9.5 to 12.0% of the muscle proteins and have molecular weights of 36,000 and 70,000, respectively (Forrest *et al*., 1975; Porzio and Pearson, 1977). Alpha actinin comprises 6% of the total muscle protein (Ebashi, 1966) and has a molecular weight of 90,000 (Offer *et al*., 1973).

2.2.3 Stroma Proteins

Stroma proteins include such constituents as collagen, elastin and recticulin. Collagen comprises of a triple helix that contains a higher hydroxy proline content than any other meat protein. At temperature of about 80 ^{o}C or higher, collagen is converted into gelatin (Schmidt and Trout, 1984). Elastin is a rather unique protein because it contains the uncommon amino acid residues desmosine and isodesmosine. These amino acids are involved in the crosslinking of the polypeptide chains and give elastin its characteristic elastic properties. Elastin is not decomposed by heat, contains very little swelling ability and is extremely resistant to acid and alkali (Bendall, 1964).

2.3 Factors Affecting the Binding Strength of a Meat Protein Matrix

The different types of meat products, such as emulsion, particulate and restructured meats, vary greatly in their method of preparation and meat particle size. In contrast, they do have one similar characteristic. This is their ability to bind their constituent meat particles together so as to form a cohesive product. The strength of this binding is important because it determines the quality of the products. The binding strength is defined as the force per unit cross-sectional area required to pull apart (either directly or indirectly) the bound pieces of meat. It includes a measure of both the cohesive force exerted between the binding matrix and the meat pieces and the strength of the binding matrix itself (Schmidt and Trout, 1984). The main factors that determine the efficacy of binding are protein extraction, mechanical treatment, presence

and concentration of added salts and temperature of heating (Schnell *et al*., 1970; Vadehra and Baker, 1970; Schmidt *et al*., 1981).

2.3.1 Protein Extraction

Bard (1965) reviewed some factors influencing the extractability of salt-soluble proteins from muscle tissue and concluded as follows: (1) temperatures in the range of –5 ^{o}C to 2 ^{o}C gave maximum protein extraction; (2) increasing extraction time increased protein extraction, up to an extraction time of 15 h; (3) prerigor meat is more extractable than postrigor meat; and (4) a sodium chloride concentration of 10% extracted the most proteins. The effect of vacuum and extraction time on the extractability of crude myosin from pre and postrigor meat was the object of a study by Solomon and Schmidt (1980). These authors concluded that: (1) the extraction of crude myosin increased linearly with extraction time; (2) vacuum increased the amount of crude myosin extracted by 20% over that of the non-vacuum treatment; and (3) 65% more crude myosin was extracted from prerigor meat than from postrigor meat. These results are in agreement with those of Saffle and Galbreath (1964) and Acton and Saffle (1969), who both found that the amount of salt-soluble proteins extracted from prerigor meat was 50% greater than that extracted from postrigor meat. Saffle and Galbreath (1964) studied the effect of increasing the pH of muscle from pH 5.5 to 6.5 in small increments on protein extraction. Increasing pH increased amounts of salt-souble proteins that were extracted. Both Awad *et al*. (1968) and Acton and Saffle (1969) found that freezing meat reduced the amount of total protein and salt-soluble protein extracted. The effect of

polyphosphates on the extractability of myofibrillar proteins and crude myosin was studied by Turner *et al.* (1979), using sodium tripolyphosphate, and Hamm and Grabowska (1979), using tetrasodium pyrophosphate. Both groups found that in the presence of salt and polyphosphate, the amount of proteins that could be extracted increased over that of salt alone. The role of solubilized meat proteins is binding to the insoluble components in the protein matrix to form a coherent stable combination with each other (Kotter and Fischer, 1975). Siegel and Schmidt (1979) found that increasing the amount of extracted myosin between meat surfaces produced a linear increase in binding strength in a model binding system. Randall and Voisey (1977) also found that when 2.5% of the total protein was replaced with salt-extracted proteins, the binding strength of the resultant product increased. Acton (1972) reported that reducing the particle size of poultry meat increased the amount of salt-soluble protein extracted and hence the binding in poultry rolls. Theno *et al.* (1978) found that increasing the massaging time of hams increased the amount of proteins extracted in a hyperbolic fashion, with the curve approaching an assymtote at 14% protein. The bind strength only increased with protein content up to 12%, after it remained fairly constant. This result may be explained by the fact that increased massaging time caused muscle fiber disruption and increased an additional protein extract which enhanced the binding among meat particles. When investigating the effect of storage time on the functionality of frozen meat (pork and beef), Miller *et al.* (1980) found that increased storage time reduced the total amount of protein that could be extracted from meat. The researchers found significant correlations between reduction in total extractable proteins and

reduction in binding strength of both beef and pork wieners, with the correlations for pork being greater than beef .

2.3.2 Mechanical Treatment

There are many types of mechanical treatments that can be applied to meat to improve its binding characteristics. Methods commonly used to increase binding in sectioned and formed meat products are mixing, massaging, tumbling and mechanical tenderization. Schnell *et al.* (1970) showed that the mixing causes cell disruption and breakage with subsequent release of the cell contents including the myofibrillar proteins. Siegel *et al.* (1978) reported that increased massaging time increased the amount of protein extracted. This result agreed with Schnell *et al*. (1970) who reported that increased protein extraction would be expected with increased cell disruption. Koo (1980) showed a similar effect occurred with tumbling. The author found that there was more myosin extracted from tumbled hams than from non-tumbled hams. Theno *et al.* (1978) showed that extended massaging time increased both fiber disruption and the amount of myofibrillar proteins solubilized as was evident by light microscopy. The increased myofibrillar protein solubilization may not be the only important contribution of cell disruption, as Schnell *et al.* (1970) observed that addition of RNA, which was also released by mixing, increased the binding strength of poultry rolls. Theno *et al.* (1978) showed that myofibrils and muscle fibers, which are both normally tightly packed, separate after massaging as were evident by the scanning electron microscope pictures. Once this structure is opened, the solubilized proteins of

the exudates are worked into loose fiber structure, allowing a more cohesive bond to form between the protein matrix and the meat surface. This finding is very similar to Kotter and Fischer (1975) who reported that the solubilized protein binds to the insoluble fibers which have been separated and dispersed by chopping. Maesso *et al*. (1970) found that mixing for 3 min increased the binding strength of poultry loaves when compared to non-mixed loaves. Later, Pepper and Schmidt (1975) also found that increasing the mixing time from 5 min to 30 min produced corresponding increases in binding strength of beef rolls. But when the mixing times exceeded 30 min, the binding strength did not increase with time and, in some cases, it actually decreased. The reduction in binding strength produced by extended mechanical treatment may be explained by the fact that at long periods of mechanical treatment the muscle fibers become excessively disrupted and the meat starts to lose its structural integrity. Krause *et al.* (1978) reported that total tumbling times of 95 min and 180 min improved the binding strength of canned hams over those hams not tumbled, and the intermittent tumbling had no advantage over continuous tumbling, as long as the total tumbling time was the same. Thus, the importance of a certain degree of meat fiber disruption for improved binding was well recognized and also indicated by McGowan (1970). The researcher reported that fraying at the meat surface, without disruption of the intregrity of the meat, allows a binding matrix to adhere more strongly to the meat surface and hence increase binding strength of the product.

2.3.3 The Presence and Concentration of Added Salts

Sodium chloride and the sodium salts of the polyphosphoric acids are widely used in the meat products on account of taste and toxological considerations (Schmidt and Trout, 1984). The important role of salts is to contribute to the ionic strength of the system, with an additional function being to alter the pH. Alkaline polyphosphates tend to increase the pH, usually by 0.1 to 0.4 units depending on type and concentration, while sodium chloride and other neutral salts tend to reduce the pH by 0.1 to 0.2 pH units (Mahon, 1961). The effect of the alkaline polyphosphates on pH is due to their alkaline nature, while that of sodium chloride has been theorized to be due to the displacement of hydrogen ions producing the drop in pH (Schmidt and Trout, 1984). These authors also reported the two mechanisms of salts to increase the binding ability of the protein matrix as follows: (1) by increasing the amount of proteins extracted; (2) by altering the ionic and pH environment so that the resultant heat-set protein matrix forms a coherent three-dimensional structure. Grabowska and Sikorski (1976) found that increasing the pH from 5 to 7.5 produced a corresponding increase in gel strength. When the salt concentration was increased from 0 to 5%, the gel strength increased up to a salt concentration of 3%, after which increasing salt concentration had little beneficial effect. Siegel and Schmidt (1979) showed that when myosin and actomyosin were heated in high ionic-strength salt solutions, the proteins formed a coherent three-dimensional network of fibers. In the absence of added salts, the same proteins formed a spongy structure with little strength as was evident by scanning electron microscopy. The researchers also investigated the effect of added polyphosphate (0.5%), change of pH

(6.0, 7.0 and 8.0) as well as added salt (0%, 2%, 4% and 6%) on binding strength of crude myosin. It was found that increasing the concentration of salt produced an increase in binding strength. In addition, the presence of 0.5% sodium tripolyphosphate produced a significant increase in binding strength but the pH changes had no significant effect. Swift and Ellis (1957) reported that the presence of 0.5% polyphosphate used in conjunction with 2% salt greatly improved the binding strength of the bologna when compared to the sample added with 2% salt alone.

2.3.4 The Temperature of Heating

As indicated by Schnell *et al*. (1970) and Vadehra and Baker (1970), the binding between meat pieces is a heat-initiated reaction. Kotter and Fisher (1975) found that heating caused the previously dissolved proteins to rearrange so that they could interact with the insoluble proteins on the meat surface and form a cohesive structure. Hamm (1966) indicated that the stabilizing bonds formed after heat denaturation were mainly hydrogen and ionic interactions. Yasui *et al.* (1980) reported that the gel strength of rabbit myosin and actomyosin gels started to increase at 40 ^{o}C and reached a maximum at 60 ^{o}C. A similar result was obtained by Grabowska and Sikorski (1976) using fish myofibrils, with the difference being that the increase in gel strength started at 30 ^{o}C and continued up to a temperature of 80 ^{o}C. Wright *et al.* (1977) showed that the temperature range of denaturation of the different protein components was characteristics of the species of animal from which the proteins came, the pH and the ionic strength. It can be concluded that there is an interaction between the temperature of

heating and the presence and concentration of different salts. The exact interaction has not been clearly clearified, but the implication is that the temperature at which maximum binding occurs is dependent on the presence of specific salts and hence the ionic strength and pH (Quinn *et al*., 1980).

2.3.5 Role of Specific Meat Proteins in Binding

From the three main groups of proteins found in muscle, sarcoplasmic, myofibrillar and stroma, the myofibrillar proteins have been implicated as being the most important in binding. The presence of salt-extractable myofibrillar proteins has been shown to be necessary for satisfactory binding in both emulsion and restructured meat products. Acton and Saffle (1969), Randall and Voisey (1977) and Miller *et al*. (1980) showed that increasing the proportion of salt-extractable myofibrillar proteins produced a concurrent increase in binding quality in the model emulsion systems. Yasui *et al.* (1980) showed that the addition of myosin to actomyosin produced a gel that was much stronger than the one with either myosin or actomyosin when used separately. The similar results were also obtained by Mcfarlane *et al*. (1977) which might be explained by the interaction of added myosin with the actomyosin at the surface of the meat to form a strong binding matrix. Although the myofibrillar proteins as a group are known to be the major contributors to binding in meat systems, it is difficult to verify the contribution of each individual proteins. This is due to the fact that the different proteins interact with each other when binding (Yasui *et al*., 1980) and that the method of extraction and purification of the proteins can have a profound influence on their binding ability

(Siegel and Schmidt, 1979). The contribution of sarcoplasmic proteins to binding has not been the subject of extensive studies and as a consequence very little quantitative information is available (Schmidt and Trout, 1984). The point to be considered in evaluating the importance of stroma proteins in the meat products is the temperature at which the product is assessed, which should correspond to the temperature at which the product will be consumed. This temperature can determine the textural characteristics of the products, as the gelatin formed on heat processing can liquefy if the product is reheated to above a certain critical temperature, which in the case of beef product is 49 ^{o}C. Once the product is heated above this critical temperature, the liquification of gelatin can result in collapse of product structure and loss of moisture and fat (Puolanne and Ruusunen, 1981).

2.4 Ingredients and Processing Factors Affecting Muscle Protein Functionality

Functional properties of proteins involve their ability to give the desired properties, whether defined in terms of biochemical interactions, analytical methods, or sensory characteristics. Proteins have an important role in meat product texture and the processing steps can be used to achieve the optimum functionality necessary for each particular product (Whiting, 1988).

2.4.1 Functional Properties of Meat Proteins

Muscle proteins can participate in three classes of interactions, namely, protein-water, protein-lipid and protein-protein (Acton and Dick, 1984). These basic interactions are characterized by the basic functional properties of water binding, fat binding and gelation. These functions are measured by many kinds of tests such as water-holding capacity, extract release volume, fat binding, emulsifying capacity, and shear force and are described by industry terms such as drip, yield, fat caps and bite. None of these industry terms represent simple or independent properties. For example, increased protein-protein interactions in a frankfurter, may improve firmness, cook yield and fat binding (Whiting, 1984). Different functional properties are important to different meat products. Bone-in hams and corned beef retain the muscular structure, and water binding is of primary importance. Increased water binding is usually desirable, but not in a dry-cured ham or fermented dried sausage. Poultry rolls and restructured products require adhesion of meat chunks as well as water binding. Liver sausages require water and fat binding and a spreadable texture, whereas the proteins in frankfurters must bind water and fat and form a firm, elastic gel. Meat proteins are usually gelled by heat; however, in sausages, such as salami, the proteins are gelled by the combination of salt, lactic acid and dehydration (Whiting, 1988).

2.4.2 Processing Steps

The general order of processing steps to make meat products is choosing and handling the lean meats and fatty tissues, grinding, adding spices, cutting, salting, curing (nitrite, nitrate, and/or ascorbate), fermenting, drying, smoking, cooking, shaping, and packaging. Individual steps are selected and controlled by the processor to create each meat product. Myosin (actomyosin in the postrigor state) is the muscle protein responsible for most of the textural properties of meat products. Myosin exists in a highly ordered and aggregated state *in vivo*. An ionic strength of about 0.6 is necessary to swell, hydrate, extract, and solubilize myosin or actomyosin. When binding meat pieces together, tumbling and massaging effectively accelerate this process, especially under vacuum (Whiting, 1988). For frankfurters, chopping breaks the cellular structure of meat and produces the sol of actomyosin. Sarcoplasmic proteins are in salt solution, and connective tissue proteins are suspended in the meat batters (Whiting, 1988). Adipose tissue is then added and chopped until fat particles of less than 200 μ are suspended in the water-protein matrix. Eventhough myosin/actomyosin can emulsify oil in model systems, the term “meat emulsion” is being replaced by “meat batter” to indicate the more complex nature of the system and to shift emphasis to the gelation and heat-setting behavior of the meat proteins (Whiting, 1984). Scanning electron micrographs show protein encapsulating the fat droplets and forming the surrounding matrix (Jones and Mandigo, 1982). The protein layer surrounding the fat must be strong enough to retain the fat and flexible enough to withstand fat liquefication and expansion during cooking (Whiting, 1988). Cooking causes protein unfolding and formation of an ordered,

three-dimensional network stabilized by hydrophobic and hydrogen bonding. This formation of network has been hypothesized to involve first the aggregation of the globular head regions of myosin at 30-50 ^{o}C followed by formation of cross-linked gel by the tail section at temperatures above 50 ^{o}C (Acton and Dick, 1984). The processor have to control a sensitive balance of protein interactions, to form a good frankfurter. Too little myosin extraction or protein-protein interaction results in excessive exudation and a mushy texture, but too much interaction can result in protein aggregation and batter failure. This batter and gel must contain the fat and water and retain the elastic texture through several cycles of solid-liquid fat transitions during smokehouse cooking, storage, freezing and recooking in the home (Whiting, 1984). The other processing steps that influence the functional properties of proteins are meat ingredients, formulation, comminution, stuffing, and smoking and cooking.

2.4.2.1 Meat Ingredients

Most skeletal muscles can be made into processed meat products. In the United States, poultry products are strongly challenging those made from beef and pork (Whiting, 1988). Thermal transitions of fish proteins occur at lower temperatures than mammalian or avian proteins (Lanier, 1985). When processing of meat products is concerned, fat and connective tissue contents must be considered and the color depends on myoglobin content, and strong-flavored fats such as those from sheep should be avoided. Fresh meat with neither excessively low pH; including pale, soft and exudative (PSE) pork from a rapid pH decline, nor excessive sarcomere contraction from cold

shortening has good functionality. Water binding decreases with decreasing pH until the proteins' isoelectric point nears pH 5.2. Optimum gelation of meat protein is at pH 5.5-6.0 (Acton *et al*., 1982; Whiting, 1984). Dark, firm, and dry (DFD) meat with a high pH and water-holding capacity may be advantageous in some products. Proper pre-slaughter handling of the animals and prerigor treatment of the meat are important to achieve the maximum amount of meat with a normal pH (Whiting, 1988). Frozen meat is not as good as fresh meat, but the former still has adequate functionality. This functionality is best preserved by using fast freezing rates, which increase juiciness and tenderness in ground beef patties because of smaller ice crystals (Nusbaum *et al*., 1983). Constant storage temperatures (less than –20 ^{o}C) resulted in slow growth of the ice crystals. However, even under optimal conditions, freezing is not a permanent preservation. Miller *et al*. (1980) showed significant declines in the functional quality during extended frozen storage. It was frequently better to directly incorporate frozen meat into the product without thawing (Groninger *et al*., 1983). Mechanically separated or deboned meats are a major meat source in the poultry industry (Froning, 1981). In general, the processing properties of mechanically separated meats compare satisfactorily to hand deboned meats (Field, 1981), although products containing too much mechanically separated meat have been described as having "less texture". The bone marrow content may improve processing properties by raising the pH and increasing the water-holding capacity, but higher iron, copper and magnesium of bone marrow may reverse this profit (Whiting, 1988). Preblending refers to mixing salt into ground meat and then allowing time for protein extraction. When subsequently made into

sausages or frankfurters, preblended meat has improved water- and fat-binding properties over meat that only added with salt but not preblended. (Puolanne and Terrell, 1983a, b).

2.4.2.2 Formulation

Meat batter requires a minimum amount of added water (10%) for myosin extraction (Morrison *et al*., 1971; Whiting, 1984). An increasing concentration of meat proteins increases firmness, but too much quantity increases rubberiness and dryness. Harder (more saturated) fats tend to increase the firmness of the frankfurter (Lee and Abdollahi, 1981; Whiting, 1987). Salt (NaCl) is essential for the manufacture of meat products (Sofos, 1986). Maximum water-holding capacity is with 0.8-1.0 M NaCl (4.6 to 5.8% salt) (Offer and Trinick, 1983), but 0.4-0.6 M is generally sufficient for good functionality (Trout and Schmidt, 1983). Frankfurters typically contain 2.5% salt (4.5% brine). Sofos (1983a, b) and Whiting (1984) showed that reducing sodium chloride levels below 2.0% (3.5% brine) resulted in increased water losses and a softer and eventually mealy texture. Only 0.50% to 0.75% salt is needed in fresh sausages and restructured products, but the salt is initially present in the thin extracted layer that binds the pieces together and, therefore, is at a much higher percent brine (Coon *et al*., 1983). Phosphates are widely used to improve or retain protein functionality. The most common forms for use in meat products are sodium acid pyrophosphate (SAPP), tetra sodium pyrophosphate (TSPP), and sodium tripolyphosphate (STPP). The first reduces the pH of the meat product, and the latter increases the pH about 0.2 units, which is frequently beneficial. Current regulations allow a total addition of 0.5% polyphosphates (Whiting, 1988).

Possible mechanisms for phosphates effects include higher pH and ionic strength, interaction with specific proteins including dissociation of actomyosin by pyrophosphate, and chelation of cations. Trout and Schmidt (1983, 1986b) showed that a pH of 6.0 and a total ionic strength of 0.6 were needed for satisfactory binding in a restructured product.

Furthermore, binders are added to improve qualities of meat products, primarily by retaining water. Non-meat proteins, modified proteins, and carbohydrates can be chosen depending on the specific product and process. Recent works demonstrated the improvement or better products' qualities with such proteins including soy protein in frankfurters (Terrell *et al*., 1979a), vital wheat gluten and egg white for binding meat pieces together (Siegel *et al*., 1979), blood plasma, egg albumin, soy and vital wheat gluten for binding (Terrell *et al*., 1982), bone and plasma proteins in sausages (Caldironi and Ockerman, 1982), gluten, calcium-reduced skim milk and soy protein in frankfurters (Keeton *et al*., 1984), and mechanically separated beef, soy protein and vital wheat gluten in restructured steaks (Parks and Carpenter, 1987). Blood fractions have excellent functional properties (Suter *et al*., 1976; Terrell *et al*., 1979b). Soy protein hydrolysates lower the Aw and can replace salt in extending the shelf life of meat products (Vallejo-Cordoba *et al*., 1986). Carbohydrates used in meat products include sugars, glycerol, cereals, starch, and gums. Means and Schmidt (1986) used calcium alginate for binding restructured meats and found that this product remained cohesive at room temperature in contrast to salt/phosphate restructured products which must remain frozen until cooked. Iota carrageenan and carboxymethyl cellulose improve water-holding capacity and texture of frozen minced cod (Da Ponte *et al*., 1985, 1986), and carrageenan is the most promising gum tested in low-salt frankfurters (Foegeding and Ramsey, 1986, 1987).

2.4.2.3 Comminution

In the typical American frankfurter, no cellular structure and few intact myofibrils remain in the batter after chopping. Temperatures between 5-7 ^{o}C are best for actomyosin extraction and dry ice can be added to prolong the chopping time. Vacuum chopping improves stability (Tantikarnjathep *et al*., 1983) and avoids small air pockets that decrease textural strength (Mawson *et al*., 1983). The completion of the comminution depends on temperature rather than time, although excessive chopping can destabilize the batter. Pork fat is chopped to 15.5 ^{o}C, yet the risk of catastrophic failure is high exceeding this temperature. Poultry frankfurters are chopped to 11 ^{o}C to 12 ^{o}C and all-beef frankfurters to 18 ^{o}C. The more saturated beef lipids are harder and have higher melting points. A batter chopped above these temperatures can be recooled and regain part of its functionality (Deng *et al*., 1981).

2.4.2.4 Stuffing

Good batters can be pumped from an emulsifier to a stuffer without causing their failure. Stuffing under vacuum produces a more dense product with better binding and texture than non-vacuum stuffing (Whiting, 1988).

2.4.2.5 Smoking and Cooking

The irreversible gelation of the myosin begins at 55 ^{o}C and peaks at 80 ^{o}C (Siegel and Schmidt, 1979). Water binding decreases as the cooking temperature increases from 60 ^{o}C to 80 ^{o}C (Puolanne and Kukkonen, 1983). Weight loss during thermal processing is proportional to the product temperature and to the protein-fat ratio (Mittal and Blaisdell, 1983). Low-salt batters, assuming no replacement with any ingredient, have increased water losses (Trout and Schmidt, 1986a). The initial low-temperature portion of the cooking cycle dries the surface and applies the smoke. Both natural wood smoke and liquid smoke contain organic acids that assist in forming the skin. An increase in relative humidity with the temperature rise results in a weak gel with some fat separation onto the surface and small fat pockets where the frankfurters touch the supports rods (Rust, 1976). Rapid contraction of the proteins with expanding and liquefying fat leads to rupture of the proteins barrier surrounding the fat droplets (Whiting, 1988).

2.5 Rheology of Meat Batters

Rheology is defined as the study of material deformation and flow (Scott-Blair, 1969) and includes what is termed “small strain” testing (deforming a small % of that required to break the sample) and “large-strain” testing (deforming to the point of permanent structural change). It is now well known that large-strain instrumental testing is required to consistently correlate with sensory texture (Hamann and Webb, 1979;

Montejano *et al*., 1985; Szczesniak, 1985), which is critical base in evaluating desirable gel forming functionality. Rheological parameters, such as stress and strain, can be used to predict sensory texture and muscle functionality of comminuted meat products. Stress can be defined as the response or internal reaction of a material to apply forces. It is a force intensity reaction dependent on the area on which forces are acting and is expressed as force per unit of area. Strain is the relative change in dimension or shape of a body subjected to stress. Strain is expressed as the ratio of the change in dimension to the original dimension and is therefore dimensionless (Rao and Steffe, 1992) .

2.5.1 Correlation of Instrumental and Sensory Evaluation

Cylindrical specimens compressed axially between flat plates always exhibited structural failure along planes of maximum shear stress and strain (these units can be independent of test method and specimen size and shape) (Hamann, 1988). Voisey *et al*. (1975) reported that the axial force required to produce failure (rupture force) correlated strongly (r = 0.89) with sensory ‘chewiness’ (desired bite) and inversely (r = -0.85) with permanent deformation resulting from a partial compression. The rupture force, called ‘initial break’ by Szczesniak and Hall (1975), was equivalent to ‘fracturability’ in the instrumental texture profile analysis (TPA) made with a universal testing machine (Bourne, 1978). If maximum force to compress to a specified post-rupture thickness is used instead of at rupture, the property is called TPA ‘hardness’ (Friedman *et al.*, 1963). A two compression sequence is used to determine TPA ‘cohesiveness’, which is expressed as the work of the second compression divided by

the work of the first. In sensory evaluations, hardness is the force required to bite through the specimen and first-bite cohesiveness is the deformation sensed before rupture by the teeth (Szczesniak, 1963). Sensory texture profile data on frankfurters generated by Hargett *et al*. (1980) were subjected to principal component analysis by Syarief *et al*. (1988). Results showed that sensory first-bite hardness, cohesiveness and springiness (elasticity) correlated strongly. Many of the sensory notes correlate with each other for the gel systems because they exhibit a high degree of elasticity (i.e., a rapid, high degree of shape recovery when deforming stress is removed). Elasticity represents a frankfurter or similar food with springy character.

2.5.2 Factors Affecting Rheological Parameters

Factors affecting rheological parameters are denaturation and dilution effects, influence of temperature history and effect of ingredients.

2.5.2.1 Denaturation and Dilution Effects

Stress and strain tend to increase or decrease in a similar fashion. Thus, the stress-to-strain ratio, also called shear modulus or rigidity, will change less than either of the parameters used in its determination; hence the lower correlations with sensory notes. Both stress and strain at rupture point of the texture are important parameters because their responses depend upon the ingredient and process variables. The protein quality is the important variable that affects stress and strain values.

For example, low–fat gels made from top-quality Alaskan Pollack surimi showed no significant differences in rigidity when subjected to 0, 3, 9, or 15 freeze-thaw cycles during storage. Even though the functional quality of the surimi was decreased through the freeze-thaw cycles, as indicated by consistent decreases in strain with the cycles, the ratio of stress-to-strain did not change since both parameters decreased proportionately. In contrast, significant changes in rigidity resulted when a fresh batch of top-grade Pollack surimi was subjected to higher water contents. Differences in strain were insignificant until 20% water was added. At 25% added water, the differences in strain were still small; however, stress dropped to about 1/3 of its value with no added water. This trend has been observed in numerous studies. Shear strain at failure is a fairly stable measure of protein functional quality whereas stress is strongly influenced by dilution and process variables. Similarly, in axial compression tests conducted on gels made with different concentrations of salt-soluble proteins extracted from turkey breast and thigh, stress and TPA hardness were influenced by concentration; strain was independent of concentration (Foegeding, 1987). The effect of changes in protein functionality on rupture rheology was also studied by Park *et al.* (1987), using gels made from beef. The gels were standardized with a 5:1 ratio of water-to-protein. The variables involved pre/post rigor muscle, cryoprotectants, muscle storage time, and time of ingredient additions. During 8 months' storage, strain values decreased with storage time for all treatments but the decreases was slow for the most advantageous treatments. For all treatments, stress changed proportionately with strain. However, because the changes in stress were greater, a plot of rigidity vs strain has a positive slope. A reduction in protein solubility correlated with the rheology parameters, and the cook loss changed inversely

with shear strain. In this study, stress was the most sensitive indicator of functionality loss because it changed proportionately more than strain. The greater sensitivity of stress is attributed to loss of protein solubility (dilution) and loss of other aspects of protein functionality (Hamann, 1988).

2.5.2.2 Influence of Temperature History

Stress is also more sensitive to thermal processing conditions than strain, as demonstrated in the study of Saliba *et al*. (1987) who found that heating at the rate of 0.9 ^{o}C/min produced 12% less stress than the 0.25 ^{o}C/min rate, but only 5% less strain. Kim *et al.* (1986) found that strain was a more consistent indicator of functional differences between gels made from two different sources and stress is an even stronger indicator of how temperature history influences the gels. The authors used three thermal treatments which were: (1) 40 ^{o}C for 30 min, (2) 90 ^{o}C for 15 min, and (3) a combination of treatments (1) and (2), to form surimi gels from Alaskan pollack and Atlantic croaker. Treatments (2) and (3) were used commercially. Strain values produced by the commercial treatments were consistently greater for Pollack, but the differences in strain between the two treatments were minor. In contrast, thermal treatment had major effects on stress. Stress values were doubled when the 90 ^{o}C treatment was preceded by the 40 ^{o}C treatment, which greatly strengthened the gels. Coaker gels were stronger than pollack gels with the 40/90 ^{o}C treatment and pollack gels were stronger than croaker gels with the 90 ^{o}C treatment.

2.5.2.3 Effect of Ingredients

The effect of added starch on stress and strain has been shown by Wu *et al*. (1985a) for surimi gels. The starches, which included modified and unmodified waxy maize starches, potato starch and pre-gelatinized tapioca starch, were used at 5% level. All starches with the exception of pre-gelatinized starch resulted in increased stress values but did not affect strain significantly. In gels cooked to 80 or 90 ^{o}C, the starches reduced the amount of free water separated by press method. Pre-gelatinized starch had a unique effect on the gels. It reduced strain by about 30% and reduced free water more than the amount obtained by ungelatinized starch. This could be a practical way of reducing sensory cohesiveness whenever desirable. Lanier *et al*. (1985) showed that gel cohesiveness is difficult to lower by simply reducing the concentration of functional protein. The authors evaluated the effect of adding up to 50% heat-denatured surimi to a standard gel preparation. The authors reported that strains were reduced by only 12% at 40% denatured protein but by 28% at 50% denatured protein. Corresponding stress reductions were 45% and 57%, respectively. The heat-denatured protein acts as a filler in this case and does not greatly influence strain at levels up to 40%.

2.5.3 Methods for Measurement of Rheological Changes During Thermally Induced Gelation of Proteins

The gelation of proteins is critical to the formation of desired texture in many foods. Structured of various meats are typical of foods in which protein gelation

produces various levels of hardness, cohesiveness, springiness, chewiness, etc. Generally, instrumental texture evaluation performed on fully cooked products, exhibits the effects of the study variables. A supplemental methodology is to study structure formation during thermal processing. Structure formation can also be studied using a rheological scan. A nondestructive (small-strain) test producing continuous or intermittent data during the processing schedule is required. Results are in terms of material rigidity (a modulus) and a measure of the proportion of input mechanical energy conserved (elasticity). Results depend on the functionality of proteins and other ingredients, as well as time-temperature history and test conditions (Hamann, 1987). Small-strain gel rigidity has been shown in several studies that it does not correlate well with sensory texture or rupture strength (Black and Smit, 1972; Hamann and Webb, 1979; Montejano *et al*., 1985). Thus, this type of test is not producing food texture data but is used to monitor physical properties changes in the gel that relate to molecular changes. Changes in gel rigidity or elasticity are circumstantial evidence that something occurred in the gel over a recorded time and/or temperature increment. This change may relate to protein unfolding, bonding of molecules, starch gelation (starch is often added to commercial protein gel-based foods) (Hamann, 1987). One of the simplest test fixtures is the small sample cup supplied by Brookfield Engineering Laboratories, Inc., (Stoughton, Mass.,) for their standard line of rotational viscometers. A cylindrical bob is suspended inside the water-jacketed cup from a 500-g load cell. Axial movement of cylinder shears the test fluid between it and cup wall because of relative motion and any flow of material from one end of the cylinder to the other. A fixture composed of the temperature-controlled cup with a cylindrical plunger to replace the suspended bob has been found to be a very

convenient device for temperature scanning rigidity measurements of pastes. This equipment is easy to fill, having a small sample surface exposed to the air, and good temperature control of the sample (Burgarella *et al*., 1985; Kim *et al*., 1986; Wu *et al*., 1985a,b). Graphs of force or apparent stress vs temperature show the slopes and transition temperatures but quantitative values of shear mondulus are only estimates since the influence of the material in compression at the end of the plunger is difficult to quantify, and the relative influence changes as the gel forms (Hamann, 1987). An accessory suspended bob that is submersible and produces a small annular gap for use with protein solutions is sometime used, when maximum sensitivity is needed. The bob and cup of this fixture have larger diameters. This provides two benefits: (1) sensitivity increase due to increased shear area, and (2) application of greater maximum shear stress during the downward movement of the bob. During the downward movement, support for the bob is transferred from the suspension cord to the gel. Force applied to shear the gel cannot exceed the weight of the bob. If the diameter of the bob increases, its volume (and weight) increases in proportion to the square of the diameters, while the shear area increases in proportion to the first power of the diameters. A larger sample is required due to the larger size of cup and bob (Hamann, 1987). Rheological monitoring has the potential to be extremely valuable and widely used instrumental method for helping to understand the chemistry of heat-set gel formation. In the case of commercial foods, the influence of ingredients and process changes can be studied for the alteration of the rheological properties of the products (Hamann, 1987).

2.6 Stabilization of Meat Batter

A meat batter can be described as a finely comminuted mixture of muscle proteins, fat, water, salt and other ingredients (Gordon and Barbut, 1992b). These mixtures are comminuted either in a bowl chopper or an emulsion mill in order to obtain a homogeneous mass. This mixture has a paste-like texture in the raw state but gradually changes into a rigid structure when the proteins start to denaturate and participate in protein-protein interactions. The emulsifying process is designed to reduce the lean meat and fat particle size. This helps to better extract the salt-soluble proteins (mainly myosin) and reduce the tendency of the fat particles to separate. These meat products include frankfurters, bologna, vienna sausages, some meat loaves and specialty items. Finely comminuted meat products have a complex structure and consist of numerous components that are held together by a variety of attractive forces (Barbut, 1995). In meat batters, fat globules constitute the dispersed phase (Swasdee *et al*., 1982), but are sometimes larger than the size required to form a true emulsion (dispersed particles not larger than 20 μ) (Lee, 1985).

In the preparation of meat batters, salt is added to lean meat and comminuted to extract the myofibrillar proteins (Barbut and Findlay, 1989). Water is then added and comminution creates a protein-rich slurry capable of binding moisture and fat. The proteins form a film or membrane around fat globules that helps to stabilize the fat. Gums and other non-meat proteins can also be used with salt to increase binding and reduce cooking losses (Comer and Allan-Wojtas, 1988; Lee *et al*., 1981). The non-meat proteins can either be added directly with all the ingredients or may be chopped with the

fat and water to form preemulsified fat system (Lin and Zayas, 1987). These non-meat ingredients mainly include dairy proteins (i.e., whey), plant proteins (i.e., soy bean), hydrocolloids, gums and starches (Comer and Allan-Wojtas, 1988). During cooking, coagulation of proteins takes place, thereby immobilizing the fat, water and other constituents (Sofos, 1986). This gives the characteristic texture to comminuted meat products. Therefore, the stabilization of fat and water within the system is important to the sensory acceptability of the product. The two theories that have been presented to explain meat emulsion stabilization are reviewed. The emulsion theory explains stabilization by the formation of an interfacial protein film around fat globules. The myofibrillar proteins, which have hydrophilic and hydrophobic sites, can position themselves in such a way that they reduce surface tension between fat droplets and water. Thus, the interfacial protein film could prevent fat coalescence and resulted in fat stabilization (Barbut, 1995). The physical entrapment theory emphasizes the role of the protein matrix in holding the fat in place during chopping and subsequent heating. The viscous protein matrix restricts the fat globule's movement and, hence, coalescence. (Gordon and Barbut, 1992a).

2.6.1 The Emulsion Theory

Fat is stabilized in the meat batter by the formation of an interfacial protein film (IPF) around the small fat globules. The film serves as an interfacial between the aqueous and the small fat globule phases and prevents their coalescence. Myosin is the major protein contributing to the mass of the IPF. It has been shown to be a good

emulsifier in an oil-in-water model system. Fat separation occurs where not enough myosin is extracted. (Galluzzo and Regenstein, 1978) The formation of a relatively thin, flexible protein film provides the best stability, whereas a thick inflexible film surrounding the fat globules results in large ruptured holes, in globule, during cooking (Jones and Mandigo, 1982). Thus, the three major factors contribute to fat stabilization in meat batters are (1) the biophysical properties of the IPF, (2) the gelation properties of the protein matrix, and (3) the physical characteristics and cell integrity of fat. The gelation properties of the protein matrix appear to play a major role in fat stabilization. However, interactions between the encapsulated fat droplets, matrix proteins and water also strongly influence the stability of the system (Gordon and Barbut, 1992b). Gordon and Barbut (1990) reported that the smooth fat globules were prevalent in the more stable batters and consisted of two subgroups: (1) thickly coated globules with a few evenly distributed and tiny pores and (2) globules with thin protein envelopes and larger pores. Lin and Zayas (1987) also reported that large, irregularly shape globules were thickly coated in frankfurters prepared with preemulsified fat. Gordon and Barbut (1990) observed that more stable batters had globules that exhibited several small, uniform pockets of exuding fat, while the unstable batters contained globules that showed large exudations at weak points in their protein coats. These large exudations have been shown to be more likely to form fat channels and facilitate coalescence (Gordon and Barbut, 1989). Fat coalescence can be distinguished from the exudation phenomenon in transmission electron microscopy (TEM) by the lack of a defined spherical shape of the fat within the matrix (Koolmees *et al*., 1989), and the existence of incomplete protein film residues within the fat (Gordon and Barbut, 1989; Comer and Allan-Wojtas, 1988).

Overall, the micrographic evidence very strongly supports the role of the IPF and the mechanism of fat emulsification in meat batter stabilization (Gordon and Barbut, 1992b).

2.6.2 The Physical Entrapment Theory

The physical entrapment theory emphasizes the fact that fat is entrapped within the protein matrix (before and during cooking). The important thing is that a protein structure has already been formed (prior to cooking) at a state in which the batter is still flowable (Barbut, 1995). According to the theory, the myofibrillar proteins in the continuous phase of the uncooked batter exist in a sol form (Acton *et al*., 1982), which aggregates during cooking to form a gel that physically traps the fat particles (Lee *et al*., 1981). This theory minimizes the contribution of the interfacial film that surrounds the fat globule and instead supports the importance of the high degree of fat cell integrity that along with physical entrapment, is thought to help to stabilize the fat in the uncooked emulsion (Gordon and Barbut, 1992b). Some studies have shown that the physical and chemical properties of fat are important in determining the stability of meat batters (Townsend *et al*., 1968). It was also shown that the size and distribution of fat particles as well as protein-lipid interactions were major factors affecting the stability of the system (Lee *et al*., 1981). Fat dispersion patterns and fat particle size were found to vary with hardness, density and melting properties of the fat, which therefore indirectly affect batter stability (Lee, 1985). Fat binding is also associated with water binding ability. Schmidt (1984) indicated that fat losses from a meat batter were associated with

initial moisture losses during cooking. It was further explained that this phenomenon was due to the formation of channels throughout the meat batter through which water and fat can migrate to the outside surface. Gordon and Barbut (1989) also showed this link between fat and water binding in meat batters. The formation of the protein matrix is also important to the stability of a meat batter system. The myofibrillar proteins undergo thermally induced gelation during cooking to form the three-dimensional network that comprises the protein matrix (Acton *et al*., 1982). Gelation occurs when a balance between protein-protein and protein-solute interactions is achieved and results in the induction of the unfolding and refolding of the protein molecules in different conformations to form a network (Oakenfull, 1987). The process is regulated by a combination of weak intermolecular forces, including hydrogen bonding, electrostatic forces, Van der Waals forces and hydrophobic interactions (Gordon and Barbut, 1992b). Gordon and Barbut (1991) observed that many fat globules were restricted physically by being bound to the protein matrix in both raw and cooked batters. Theno and Schmidt (1978) and Gordon and Barbut (1990) previously showed that physical binding of fat globules to the protein matrix took place in commercially produced frankfurters. This physical binding resulted from protein-protein interactions between the IPF and the matrix proteins. It was therefore logical to assume that protein aggregation during cooking increased the immobilization of protein-coated fat globules by binding them to the matrix, thereby further stabilizing these globules and preventing coalescence (Gordon and Barbut, 1992b). It appears that fat stabilization is a combination of the effectiveness of an IPF in localizing the fat and the physical restriction and binding provided by a cohesive protein matrix (Gordon and Barbut, 1992b). Thus, the physical properties of

the fat, the comminution, the end point chopping temperature history of the meat are among the factors that influence the extent to which either of the two mechanisms is involved in fat stabilization (Barbut, 1989; Koolmees *et al.*, 1989; Smith, 1988)

2.7 Hydrocolloids

Hydrocolloids, commonly referred to as "gums" are long-chain, high-molecular weight polymers that dissolve or disperse in water to give a thickening or gelling effect, and exhibit related secondary functional properties, such as emulsification, stabilization, and encapsulation. In a food emulsion system, hydrocolloids are generally used to stabilize the dispersed oil droplets against separation from a continuous or aqueous phase (Sharma, 1981).

2.7.1 Properties of Hydrocolloids

Hydrocolloids are used in processed foods as thickeners, stabilizers, gelling agents and, in some cases, emulsifiers. The meanings of these terms were defined by Sanderson (1981).

2.7.1.1 Thickening

Thickening, which is implied in the food technology terms " body", "mouth-feel", and "texture", refers to viscosity which is the resistance to flow of a liquid. Viscosity is defined as the ratio of shear stress to shear rate, where shear stress is the applied force and shear rate is the rate at which the liquid is being deformed. Only in a small number of liquid systems is the shear stress proportional to shear rate; these are referred to as Newtonian and are characterized by viscosities independent of shear rate. Most hydrocolloids solutions exhibit non-Newtonian flow and increasing shear rate can result in either a decrease or an increase in viscosity. In other words, the solutions are shear thinning or shear thickening, and are respectively described as being pseudoplastic or dilatant if, on removal of the shearing force, they immediately revert to their original states. Thixotropic and rheopectic solutions are also respectively shear thinning and shear thickening but require an interval of time to recover their original viscosity after shear removal. Clearly, for non-Newtonian solutions, viscosity is dependent on shear rate (Sanderson, 1996).

2.7.1.2 Stabilization

Stabilization with hydrocolloids applies to aqueous dispersions where the continuous phase is water and dispersed phase can either be solid, liquid or gas. Suspensions are solid dispersions, emulsions are liquid dispersions and foams are dispersions of gas. In all of these systems there is a tendency for the dispersed phase to

destabilize or separate out. Addition of the appropriate hydrocolloids to provide viscosity to the aqueous phase can minimize this tendency. Stabilization is not only favored by high solution viscosity but also by the existence of a solution yield value, defined as the shear stress or applied force below which the solution will not flow. If the suspended particles do not exert a force in excess of the yield value they can not separate and can remain effectively dispersed. A vast number of hydrocolloid applications in the food industry are concerned with the stabilization of suspensions, emulsions and foams (Sanderson, 1981).

2.7.1.3 Gel Formation

Although all water soluble hydrocolloids provide viscosity to a greater or lesser extent, only a few have the ability to form gels. Gelation results from intermolecular associations which give rise to a macroscopic three dimensional network within which the aqueous system is bound. Association may involve the same or different molecular species and is brought about physically by altering temperature or chemically adding an appropriate reagent (Rees and Welsh, 1977). Gels can be considered as the intermediate between solids and liquids since it possesses properties of both. For instance, the entrapped water retains many characteristics of liquid water. Pectins, alginates, and carrageenans are the most important gelling polysaccharides for food (Sanderson, 1996).

2.7.1.4 Emulsification

In the preparation of emulsions, an emulsifier is required to reduce interfacial surface tension so that extremely fine droplets of dispersed liquid can be formed. In most cases, hydrocolloids do not function as emulsifiers but are used to provide emulsion stability. There are, however, a few exceptions, for example propylene glycol, alginate and gum arabic, which act as both emulsifiers and stabilizers (Sanderson, 1981).

The ability of a hydrocolloid to function as a thickener, stabilizer, gelling agent, or emulsifier is governed by its functional properties in solution. Solution properties depend on molecular shape and since hydrocolloids can have greatly different molecular shapes they can provide vastly different solution properties (Sanderson, 1981).

2.7.2 Rheology of Hydrocolloids

The relation of mouthfeel or textural qualities imparted to rheological behavior is the key to the scientific application of gums to food products (Farkas and Glicksman, 1967). Natural and synthetic hydrocolloids or gums are mainly used in the food industry to control the rheological properties of many food products. They are normally used in low concentrations, ranging from 0.05% to about 5% and do not usually contribute to the nutritional value, taste or aroma of the finished product. They have been used individually or in combination for thickening and texture modification. They can also be used for gelation, water retention, gas entrainment, emulsification, film formation

and flavor fixation (Krumel and Sarkar, 1975). Rheology is defined as the science of deformation and flow behavior of matter. Hydrocolloids are complex polysaccharides that significantly modify the flow behavior of aqueous solutions. Some of the physical properties of a hydrocolloid that are responsible for modifying the rheology of a solution are molecular weight, molecular weight distribution, degree of hydration, extent of intra- and intermolecular interaction and ability to stabilize emulsions, foams and suspensions. The rheology of a solution is also affected by the measurement conditions, including temperature and concentration. Aqueous solutions of low-molecular-weight materials, most oils and syrups follow Newton's Law (Equation 1) in that their viscosities are not changed with changing shear rate:

$$\eta = \frac{\tau}{\gamma} \qquad (1)$$

In the expression, η = viscosity, τ = shear stress and γ = shear rate. In contrast, the viscosities of solutions of hydrocolloids usually change with changing rate of shear. The vast majority of gums display pseudoplastic flow properties-that is, the viscosity of the solution decreases as the rate of shear increases. The material behaves as a Newtonian fluid at very low and very high shear rates and is shear thinning (i.e., viscosity decreases with increasing shear) in the intermediate range. In many cases, the shear thinning portion of the flow curve plotted on log paper will yield a straight line over a certain region, the extent of which is dependent upon the gum and the measuring conditions. This behavior is described as following the power law expression shown in Equations 2 and 3 (Krumel and Sarkar, 1975):

$$\tau = K\gamma^{n} \qquad (2)$$

$$\eta = \frac{\tau}{\gamma} = K\gamma^{n-1} \qquad (3)$$

where K is the consistency index and n is a dimensionless number that is a measure of the deviation of flow properties of the fluid from Newtonian behavior. For pseudoplastic fluids, n is greater than 1.0 and the viscosity of the fluid would increase with increasing rate of shear. When n = 1.0, the fluid is Newtonian and Equation 3 reduces to Newton's law, Equation 1. For most polymer solutions, the power law is applicable only for a short range of shear rates.

Viscosity and concentration are usually interdependent, but not necessarily in a linear manner. At low concentrations, some gums will show Newtonian behavior, but as the concentration increases, the rheological properties tend to become non-Newtonian (Catacalos and Wood, 1964). The experimental way to recognize this behavior of gums is to measure and plot the viscosity against the shear rate. Farkas and Glicksman, (1967) reported that using of low-viscosity type carboxymethylcellulose (CMC) with concentrations of the gum in water up to 3.0%, showed fairly well-defined Newtonian behavior, but became distinctly thixotropic at higher concentrations. However, when using high-viscosity and high-molecular-weight CMC, the same thixotropic effect was achieved at the low concentration of 0.5%. Furthermore, the large majority of gums become less viscous as the temperature increases. This is similar to a reduction in effective concentration of gum. Since many gums are Newtonian at low concentrations, the effect of an increase in temperature can be to change the flow property of the gum from non-Newtonian to Newtonian (Farkas and Glicksman, 1967).

The molecular weight distribution of the polymer is also important. Two polymers having the same zero shear viscosity would show different flow properties at higher shears. The one having the broader molecular weight distribution would exhibit non-Newtonian flow at lower shear. Non-Newtonian behavior has been shown to occur only when the polymer chains are long enough to entangle, and when molecular weight is of a critical size (Farkas and Glickman, 1967). Other important factors are average molecular weight, chemical structure of the polymer and the presence of other active ingredients, all of which influence the rheological properties of gums (Farkas and Glicksman, 1967).

2.7.3 Isolation of Hydrocolloids from Plant Seeds

Plant hydrocolloids are polyhydroxylic and consequently largely hydrophilic compounds of high, variable molecular weights. Their molecular structures are made up of a selection of the less common monosaccharides units mutually linked as glycosides in a limited number of anomeric configuration and position of attachment. Incorporation of acidic sugars results in the ability to form gels from changes in pH of the dispersing medium (invariability water). Hydrocolloids have been isolated from complex pectic mixtures by fractional solubilization with chemicals or solvents of increasing strength (Ericson and Elbein, 1980; Odonmazig *et al*., 1985) or by fractional precipitation with solvents or metal complexes. Analyses have been based on hydrolysis with acid or enzymes and subsequent chromatographic separation of oligomers. Susheelamma (1987) isolated linseed mucilage by aqueous extraction and precipitation by acetone, ethanol and

isopropanol. The author found that ethanol, acetone or isopropanol gave similar yield of polysaccharide. The dried material easily redispersed in water and retained good viscosity. Mazza and Biliaderis (1989) prepared flax seed (*Linum usitatissimum* L.) by extraction of seeds with water followed by evaporation, precipitation with ethanol and freeze drying of extract. They found that the mucilage contained less carbohydrates, more minerals and more protein than commercial locust bean and guar gum. Cui *et al*. (1993) separated crude mucilage from yellow mustard (*Sinapis alba* L.) into a water-soluble fraction (52.0%) and a water-insoluble fraction (34.0%). Chemical composition of crude mucilage and its fractions were 80-94% carbohydrates, 1.7-15.0% ash and 2.2-4.4% protein. The monosaccharides composition were 22-35% glucose, 6.0-6.4% mannose, 1.6-4.0% rhamnose, 2.8-3.2% arabinose and 1.8-2.0% xylose. Later, Cui *et al*. (1994) found that temperature and pH have significant influences on both yield and quality of the extracted flaxseed crude gum while the water : seed ratio had only minor effects. Optimum conditions for the extraction were temperature of 85-90 ^{o}C; pH, 6.5-7.0 and a water : seed ratio of 13: 1. Balke and Diosady (2000) found that a two-stage extraction process at initial temperature of 45 ^{o}C and 8 : 1 water to seed ratio resulted in over 90% mucilage removal in approximately 3 h. An acidic polysaccharide gum was isolated from the fruit of *Cordia abyssinica* by extraction with alkali and precipitation with acid. This gum had 9.1% moisture, 0.7% ash and 8.7% uronic acid. The monosaccharides composition were 27% galactose, 21% rhamnose, 17% mannose, 11% xylose, 10% glucose and 9.5% arabinose (Benhura and Chidewe, 2002). Balyan *et al*. (2001) soaked fenugreek flour in water (1 : 10 w/v) for 20 min at 30 ^{o}C, 50 ^{o}C or 75 ^{o}C, followed by extraction with water (1 : 50 w/v) for 2 hours at pH 2, 5, 7, 9 or 11 with continuous

stirring. The researchers reported 30-32 % yield of crude mucilage. Mucilage extracted at pHs 2 and 11 had significantly higher carbohydrate contents, thereby indicating comparatively more purified mucilage. Brummer *et al*. (2003) extracted fenugreek gum with 10 ^{o}C water for 2 h and precipitated with 70% ethanol to give a 22% yield with only 2.36% protein contaminants.

2.7.4 Potential Use of Hydrocolloids in Meat Products

Hydrocolloids or gums have been used as water binders and texture modifiers in comminuted meat product. In addition, they have been incorporated into low fat meat product for their gel forming ability, water holding capacity improvement and textural characteristic contribution (Gordon and Barbut, 1992b). Various researchers reported on the applications of neutral seed gums (locust bean gum and guar gum), anionic gums (carrageenan, carboxymethylcellulose and xanthan gum) and cationic gum (chitosan) as meat binders and texture stabilizers. The proteins and polysaccharides interactions were occurred due to the general electrostatic attraction between the negatively charged polysaccharides and positively charged proteins. Other interactions such as hydrogen, hydrophobic or covalent bonds may also be affected through the stabilization of these complexes (Stainsby, 1980).

2.7.4.1 Application of neutral gums in meat products

Neutral gums such as locust bean gum, guar gum and other water soluble gums, give specific functional characteristics to the products. Because of their ability to form gels and retain water, neutral gums are used in the meat industry as texture modifiers, either for gelation and retention of the broth surrounding meat or the liquid contained in the meat. They may be used as a brine ingredient and introduced into the meat by massaging, injection, or simply by blending, as with meat batters (Whiting, 1988). Dziezak (1991) reported that the addition of 0.2 to 0.5% guar gum to sausages could bind the free water and retard shrinkage, while locust bean gum addition provided stability and imparted a smooth texture in ground meat products such as salami and bologna. Montero *et al*. (2000) found that the non-ionic hydrocolloids were dispersed throughout the fish (blue whiting) meat protein matrix but did not interact with it and were located simply by inclusion. The researchers also reported that when examined the fish protein gels with a scanning electron microscope (SEM), that locust bean gum (1%) is arranged as filaments inside the cavities of fish protein gels; there appear to be no areas of interaction or contact between the hydrocolloid and the protein matrix. The distribution of the locust bean gum caused an increase of breaking deformation and work of penetration when compared to the hydrocolloid-free gel. Moreover, the authors found that guar gum distributed in protein matrix in such the same way as locust bean gum, probably because both gums are polysaccharides comprising comprising mainly galactomannans. However, there is a small number of stained cavities, probably because the concentration was only half (0.5%) of that of locust bean gum. This gel structure was

significantly less firm (breaking force, work of penetration, hardness) than the control but had greater water holding capacity.

2.7.4.2 Application of anionic or cationic gums in meat products

Protein-hydrocolloid interactions are mainly electrostatic; between the anionic groups on the hydrocolloid and the positively charged groups on the protein, which are dependent upon hydrocolloid concentration and the proportion of the hydrocolloids used (Tolstoguzov, 1986). Fox *et al.* (1983) found that the addition of the anionic gums, xanthan and carrageenan, stabilized the texture of frankfurter emulsions against acid deterioration at 37 ^{o}C in vinegar pickle. The researchers also reported that the cationic gum, chitosan, formed an acid-stable gel but the gel would not hold the emulsion. The effect that the anionic gums stabilize frankfurter texture while cationic gums do not, is strong presumptive evidence that the frankfurter gel structure was also produced by a gum-protein interaction. When formed from a cationic gum, the gum-protein structure will not hold water. Naturally, pH of meat muscle proteins are on the alkaline side of their pKa, hence they have negatively charges. The resulting gel formed with a positively charged gum would be much less ionic than either component, and would not hold water as well, which account for the poor water-binding capacity of the frankfurters containing chitosan. Foegeding and Ramsey (1986) investigated the effects of adding xanthan gum and methylcellulose to low-fat, high moisture meat batters. The authors found that methylcellulose treatment showed an increase in weight loss between 60 and 70 ^{o}C. Increasing the concentration of xanthan gum decreased batter

hardness without affecting batter stability. Sensory evaluation indicated that low-fat frankfurters (11-12% fat) were as acceptable as control frankfurters (27% fat). Montero *et al.* (2000) reported that the addition of xanthan gum (0.5%) in dry state produced a decrease in the gel forming capacity of the fish (blue whiting) myofibrillar proteins, which was reflected by poorer resistance to the folding test, breaking force, work of penetration, hardness and adhesiveness of fish protein gels. This is because xanthan gum is in a dry state or has little available water and it tends to aggregate and coil upon itself, thus occupying a large volume which would distort the protein matrix. These authors also reported that the addition of the addition of 0.5% carboxymethylcellulose (CMC) powder negatively modified the fish protein gel rheology, probably through interaction of its carboxyl groups with the fish protein matrix, and because the matrix was distorted due to the high Mw of CMC. CMC also required a lot of water in order to disperse. The addition of the polysaccharides is therefore not a favorable condition in dry state.

CHAPTER III

METHODOLOGY

3.1 Extraction and Physicochemical Characterization of Malva Nut Gum

3.1.1 Material

Dried malva nut fruits *Scaphium scaphigerum* were used in this experiment. Dried fruits which were collected between March and April 2003, in Eastern Thailand, were purchased from a local traditional herb store in Bangkok. The fruits were packed in polyethylene bags, transported to the laboratory, and stored at room temperature for up to 2 months.

3.1.2 Extraction of Malva Nut Gum

Endogeneous enzymes were inactivated by boiling the fruits in 80% ethanol for 1 h. The gum extraction followed a procedure described by Rombouts and Thibault (1986) with a few modification (Fig. 2). Briefly, the fruits were cooked in water at 90 ^{o}C for 2 h to extract water soluble gum (fraction 1). Insoluble solids were separated by filtration through a silk-screen cloth, while the water soluble fraction (fraction 1) was centrifuged to discard the non-soluble residues. The remaining solids were extracted with

0.05 M HCl at 85 ^{o}C for 30 min, filtered through a silk-screen cloth. The extract was centrifuged and the supernatant provided an acid soluble fraction (fraction 2). The remaining solids were further extracted by 0.05 M sodium hydroxide at 85 ^{o}C for 30 min, filtered through a silk-screen cloth. The alkaline soluble fraction (fraction 3) was centrifuged and supernatant collected to yield the alkaline soluble gum. The pHs of the acid soluble and alkaline soluble extracts were adjusted to 4.5 with 2 M NaOH or HCl. Finally, all three extracts were precipitated with 95% ethanol (1:3 v/v) and washed 3 times with 100% ethanol followed by air-drying.

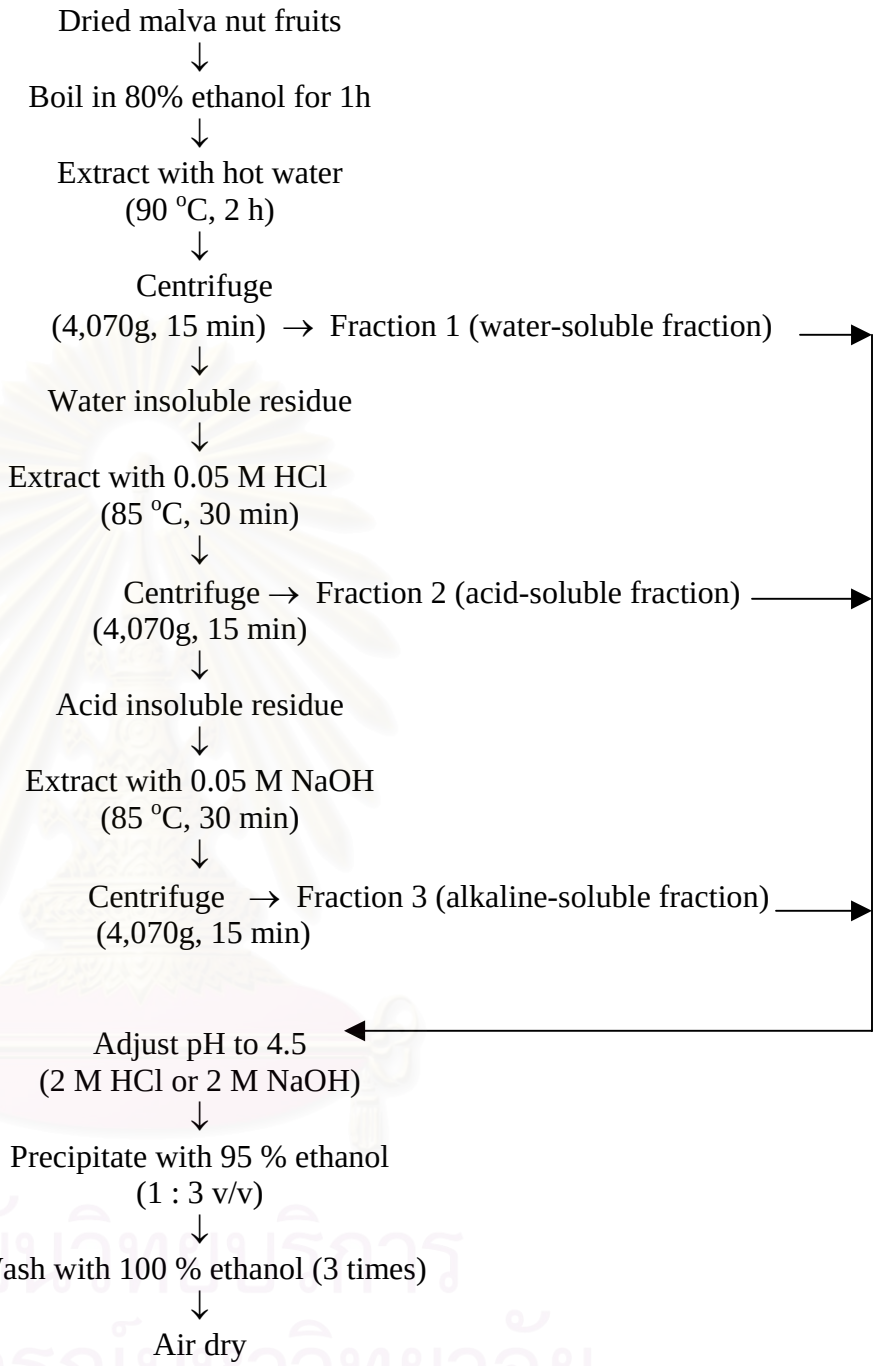

Fig. 3.1 Schematic flow diagram of extraction of the different fractions of malva nut gum.

3.1.3 Purification of Malva Nut Gum

3.1.3.1 Dialysis

The alkaline-soluble fraction was further purified by dialysis against deionized water for 48 h at 22 ^{o}C, using 3500 molecular weight cut off membrane (Spectra/Por® RC membrane, Spectrum Laboratories Inc., Rancho Dominguez, CA, U.S.A.). The purified gum was precipitated by 95% ethanol (1:3 v/v) and washed 3 times with 100% ethanol, followed by air-drying.

3.1.3.2 Decoloring

The gum decoloring followed a procedure described by Abdel-Aal *et al*. (1996) with a slight modification. The alkaline-soluble malva nut gum (2 g) was heated in water (800 ml) at 60 ^{o}C for 1 h. The gum solution (0.25% w/v) was cooled to the room temperature (22 ^{o}C) and the pH was adjusted to 9 with 2M NaOH. Then the gum solution was decolored with 30% H_2O_2 (1: 62.5 v/v) for 3 h and neutralized to pH 7 with 2 M HCl. The decolored gum solution was further purified by dialysis against deionized water for 48 h at 22 ^{o}C, using 3500 molecular weight cut off membrane (Spectra/Por® RC membrane, Spectrum Laboratories Inc., Rancho Dominguez, CA, U.S.A.). The decolored gum solution was vacuum evaporated (Buchi Vacuum Controller V-805, Brinkmann Instruments (Canada), Ltd., Ontario, Canada) at 40 ^{o}C (50 mbar, 35 rpm) until the volume of the solution decreased to 200 ml. After that the pH of

concentrated solution was adjusted to 4.5 with 2 M HCl and precipitated with 100% ethanol (1:3 v/v). The precipitate was separated by centrifugation (9020 g, 15 min), washed 3 times with 100% ethanol and air dried.

3.1.4 Composition of Malva Nut Gum

Ash and moisture contents were determined according to the AOAC methods (1996). Protein content was determined from the nitrogen content (N x 6.25) using an Automatic Elemental Analyzer (S.P.A. EA/NA 1110, ThermoQuest Italia, Strada Rivoltana, Milan, Italy). Total sugar content was determined according to the method of Dubois *et al*. (1956) and was measured as glucose equivalents. Uronic acid was determined according to Blumenkrantz and Asboe-Hansen (1973) and was measured as galacturonic acid equivalents. Monosaccharide content was determined using a Dionex HPLC system (Dionex Canada Ltd., Oakville, Ontario, Canada) with pulsed amperometric detection as previously described by Wood *et al.* (1994). In this method, the polysaccharide (2% w/v) was hydrolyzed in 1 M H_2SO_4 for 2 h at 100 ^oC to give constituent monosaccharides. The hydrolysate was cooled, diluted 1:10 with water, filtered through a 0.45 μm filter and injected into the HPLC column.

3.1.5 Methylation and GC-MS of Partially Methylated Alditol Acetates (PMAA)

3.1.5.1 Reduction of Uronic Acids

The reduction of the uronic acid was performed following a procedure described by Taylor and Conrad (1972) and York *et al.* (1986) with a slight modification. Alkaline extracted malva nut gum (5 mg) was dissolved in deuterium oxide (2 ml) and 50 mg of 1-cyclohexyl-3-(2-morpholino-ethyl)-carbodiimidemethyl-p-toluenesulfonate (CMC, Sigma, St. Louis, MO, U.S.A) was added. The pH was adjusted and maintained at 7.0, using 2.0 M HCl in deuterium oxide during the reduction reaction. After the addition of sodium borodeuteride, the reaction was carried on with constant stirring for 0.5 h at pH 7.0. After titration of the solution to pH 4.0, the salts were separated from the reduced polysaccharide by dialysis against distilled water overnight at 22 ^{o}C, using 3500 molecular weight cut off membrane (Spectra/Por® RC membrane, Spectrum Laboratories Inc., Rancho Dominguez, CA, U.S.A.), followed by lyophilizing the solution. The polysaccharide was dissolved in deionized water, 10% acetic acid in methanol was added and then dried with a nitrogen stream to remove boric acid. This process was repeated 3-4 times to make sure that most of the boric acid was removed. Finally, a few drops of methanol were added and the solution evaporated twice with a nitrogen stream to remove any boric acid remained.

3.1.5.2 Methylation Analysis

The methylation of the alkaline extracted malva nut gum with and without uronic acid reduction were analysed according to the method of Ciucanu and Kerek (1984) with a few modifications. The samples with and without uronic acid reduction (2-3 mg) were dried at 80 ^{o}C for 4-5 h and stored over P_2O_5 in a glass desiccator under vacuum. Dimethyl sulfoxide (DMSO) was added to the dried gum and the mixture sonicated for another 3 h followed by stirring at 85 ^{o}C for 2 h and another sonication for 3 h to increase sample solubility. Dry powder sodium hydroxide (20 mg) was added and the mixture was stirred at room temperature for 3 h. After adding methyl iodide (0.3 ml),the mixture was mixed for additional 2.5 h. The partially methylated polysaccharides were extracted with methylene chloride, passed through a sodium sulphate column and dried under a stream of nitrogen. The dried, partially methylated, polysaccharides were hydrolysed in a 4 M TFA (0.5 ml) at 100 ^{o}C for 6 h and dried under a stream of nitrogen. The acid hydrolysate was acetylated with acetic anhydride (0.5 ml). Aliquots of PMAA solution (0.5-1 μl) were injected onto a GC-MS system (ThermoQuest Finnigan, San Diego, CA, U.S.A.) fitted with a SP-2330 (Supelco, Bellefonte, PA) column (30m x 0.25 mm, 0.2 μm film thickness, 160 to 210 ^{o}C at 2 ^{o}C/min, then 210 to 240 ^{o}C at 5 ^{o}C/min) equipped with an ion trap MS detector.

3.1.6 Molecular Characterization

The molecular weight (Mw) and intrinsic viscosity of the alkaline extracted malva nut gum before and after dialysis were determined using a high performance size exclusion chromatography (HPSEC, Shimadzu SCL-10Avp, Shimadzu Scientific Instruments Inc., Columbia, MA) according to Wang *et al.* (2003). The column set consisted of two columns in series, a Shodex Ohpak KB-806 M (Showa Denko K.K. Tokyo, Japan) and an Ultrahydrogel linear (Waters, Milford, CT, U.S.A.). A Shimadzu SCL-10Avp pump unit (Shimadzu Scientific Instruments Inc., Columbia, MA) were kept at 40 ^{o}C during measurements. Triple detectors, a dual detector, refractive index and viscosity detectors (Model 250, Viscotek, Houston, TX, U.S.A.), and a right angle laser light-scattering system (RALLS, Viscotek, Houston, TX, U.S.A.) were used. The mobile phase was consisted of 50 mM $NaNO_3$ with a flow rate of 0.6 ml/min. The injection volume of sample was 100 μl. Pullulan of known Mw and intrinsic viscosity was used as a standard and dn/dc of 0.146 ml/g was used for malva nut gum fractions. The fractions were dissolved in water (60 ^{o}C, 3 h), cooled and filtered through a 0.45 μm filter prior to injection onto the column. The weight average molecular weight and intrinsic viscosity [η] were calculated using the software provided by Viscotek.

3.1.7 FT-IR Spectroscopy

Malva nut gum fractions, commercial pectin standards with known DE (26% and 59% DE) and gum arabic (Sigma, Steinheim, Germany) were dried in a vacuum oven at 80 ^{o}C for 4 h and desiccated in a vacuum jar prior to FT-IR analysis. FT-IR spectra of the gum fractions were obtained using a Golden-gate Diamond single reflectance ATR in a FTS 7000 FT-IR spectrometer equipped with a DTGS detector (Digilab, Randolph, TX, U.S.A.). The spectra were recorded at the absorbance mode from 4000 to 400 cm^{-1} (mid infrared region) at a resolution of 4 cm^{-1} with 128 co-added scans (Singthong *et al*., 2004). Triplicate spectra were recorded for each sample. The wave numbers corresponding to chemical species of interest included the ester region (1740-1720 cm^{-1}), carboxylate ion stretches (1600-1414 cm^{-1}) and amide peaks (1650 and 1550 cm^{-1}).

3.2 Rheological Properties of the Purified Malva Nut Gum

3.2.1 Preparation of Purified Malva Nut Gum (PMG)

Endogenous enzymes in the dried malva nut fruits were inactivated by boiling the fruits in 80% ethanol for 1 h. PMG extraction was conducted following a procedure described by Rombouts and Thibault (1986) with a few modification. Briefly, the fruits were boiled in 90 ^{o}C water for 2 h, Then the seeds were separated by filtration through a silk-screen cloth and centrifuged to discard the non water soluble gum. The non water soluble gum was treated with 0.05 M sodium hydroxide at 85 ^{o}C for 30 min, filtered through a silk-screen cloth, centrifuged to separate the non alkaline soluble gum, and then the pH of the alkaline soluble gum was adjusted to 4.5 with 2 M hydrochloric acid. The alkaline extract was further purified by dialysis against deionized water for 48 h at room temperature (22 ^{o}C). Finally, the purified gum was precipitated by adding 3 volumes of 95% ethanol and washed with 99.8 % ethanol for 3 times followed by air-drying. Proximate analyses of PMG (AOAC, 1996) were determined in duplicate. Average carbohydrate, ash, moisture, protein and fat contents were 88.86%, 5.15%, 3.57%, 2.42% and 0.00%, respectively.

3.2.2 Rheological Properties of Purified Malva Nut Gum

All rheological measurements were performed using a stress controlled rheometer (Bohlin CVO, Bohlin Instruments, NJ, U.S.A). Steady shear and oscillatory tests were performed at 25 ^{o}C using a parallel plate geometry (40 mm diameter, 1.0 mm gap).

3.2.2.1 Steady Shear Viscosity Test

Steady shear viscosity tests were determined for 0.1%, 0.3%, 0.5%, 0.8%, 1.0%, 2.0%, 3.0% and 4.0% (w/v) PMG solutions. Zero-shear-rate viscosity (η_o) and shear rate ($\dot{\gamma}$) value of PMG solutions at 0.1% and 0.3% (25 ^{o}C) were observed. The flow index (*n)* and consistency index (*K)* values of PMG at difference concentrations (25 ^{o}C) were calculated. Data presented are means of triplicate measurements.

3.2.2.2 Oscillatory Test

Viscoelastic properties (storage modulus - G′, loss modulus - G″,complex viscosity -η*) were determined through small amplitude oscillatory test at frequencies from 0.01 to 10 Hz. Prior to any dynamic experiments, a strain sweep test at a constant frequency of 1 Hz was used to determine the linear viscoelastic region (2% strain). All oscillatory tests were performed at a 2 % strain. PMG solutions were prepared by solubilizing the gum in deionized water at 60 ^{o}C for 3 h with stirring.

For temperature sweep, solutions of 1.5 % PMG were heated to 60 ^{o}C and rapidly mixed with 50 mM calcium chloride solutions (at 60 ^{o}C) to give gum solutions with 5 to 15 mM calcium chloride. After cooling, the malva nut gels were loaded onto the rheometer set at 10 ^{o}C. Temperature sweeps were performed between 10 and 60 ^{o}C; heating rate employed was 1 ^{o}C/min. The loss modulus (G″) and storage modulus (G′) were monitored at 1 Hz. Data presented are means of triplicate measurements.

3.3 Effect of Crude Malva Nut Gum (CMG), Purified Malva Nut Gum (PMG), Sodium Chloride and Phosphate on Cook Loss, Textural Properties and Microstructure of Lean Chicken Meat Batters

3.3.1 Materials

3.3.1.1 Hand Deboned Chicken Breast Meat

Hand deboned chicken breast meat (12 kg), obtained from an Ontario local processing plant was trimmed of all visible fat and connective tissue. The meat was chopped in a bowl chopper (SMK 40, Schneidmeister, Berlin, Germany), in three batches, at the low speed setting for 1 min at < 8 ^{o}C. All the meat was then mixed to obtain a homogeneous mass, then vacuum packed in 500 g bags (Multivac D-8941, Multivac GmbH, Wolfertschwerden, Germany) and kept frozen (-20 ^{o}C) for up to 4 weeks prior to use. Proximate analyses of the raw meat (AOAC, 1996) were determined in duplicate. Average moisture, protein, fat and ash contents were 74.78%, 22.46%, 0.62% and 1.02%, respectively.

3.3.1.2 Crude Malva Nut Gum (CMG)

CMG was extracted by soaking the nuts in water (1 : 80 w/v) at pH 7 for 15 h at room temperature to completely hydrate and swell the fruit. Excessive water was then removed by filtering through a 40-mesh screen. The left over fibrous

debris was removed by a pneumatic press. The crude mucilage was precipitated with 3 volumes of 95% ethanol and freeze dried (Heto Drywinner DW. 8-85, Heto-Holten A/S, Allerød, Denmark). The precipitate was then sieved through 50 mesh screen. Proximate analyses of the crude malva nut gum were determined in triplicate (AOAC, 1996). Average carbohydrate, ash, moisture, protein and fat contents were 86.62%, 5.87%, 5.34%, 2.17% and 0.00%, respectively.

3.3.1.3 Purified Malva Nut Gum (PMG)

PMG was prepared as described in 3.2.1.

3.3.2 Preparation of Lean Meat Batters

A 500 g bag of meat was thawed at 4 ^{o}C for 24 h. The meat was divided into 10 lots and each mixed, by hand, with all the dry ingredients (Table 3.1) for 3 min. All visible pieces of connective tissue were removed. The treatments were stored in a refrigerator for 1 h to allow adequate equilibration, and later three 30 g aliquots were placed into polypropylene centrifuge tubes, and centrifuged (Model 225, Fisher Scientific, Pittsburgh, PA, U.S.A) at the slow speed setting to remove all air bubbles. The chicken meat batters were cooked in a water bath (Haake W-26, Haake, Berlin, Germany) from 10 ^{o}C to 75 ^{o}C.

Table 3.1 Randomized complete block design with 10 treatments of chicken meat batters with 4 salt levels (0, 1, 2, 3%), 2 crude malva nut gum (CMG) levels (0, 0.2%) or 2 purified malva nut gum (PMG) levels (0, 0.1%) and 2 sodium tripolyphosphate (STPP) levels (0, 0.5%).

Treatment*	Salt (%)	CMG or (PMG) (%)	STPP (%)
1	0	0.0 (0.0)	0.0
2	0	0.2 (0.1)	0.0
3	1	0.0 (0.0)	0.0
4	1	0.2 (0.1)	0.0
5	2	0.0 (0.0)	0.0
6	2	0.2 (0.1)	0.0
7	3	0.0 (0.0)	0.0
8	3	0.2 (0.1)	0.0
9	2	0.0 (0.0)	0.5
10	2	0.2 (0.1)	0.5

* Treatments were composed of chicken breast meat and 33 % added water.

3.3.3 Cook Loss

Fluid separated from the batters was measured after cooling for 15 min at room temperature and expressed as:

$$\% \text{ Cook loss} = \frac{\text{g fluid expelled during cooking}}{\text{g sample before cooking}} \times 100$$

3.3.4 Texture Profile Analysis (TPA)

TPA was determined using 9 cooked cores (each 1 cm high, 1.5 cm diameter) per treatment (Bourne, 1978). The samples were compressed twice to 75% of their original height, using a Texture Analyzer (TA-XT2, Texture Techniques Corp, Scarsdale, NY, U.S.A). Hardness (maximum force required for 75% compression; indication of first bite hardness; Newton), springiness (height that sample recovers during time lapse between end of first bite and start of second ; mm), cohesiveness (ratio of area of second curve to area of first curve; dimensionless) and chewiness (gumminess x springiness) were determined. In addition, fracture force (force required to fracture; first significant break in curve; indication of fracturability; Newton) and fracture distance (deformation required to fracture sample, mm) were determined. See appendix A for additional definition of each texture profile.

3.3.5 Microscopical Evaluation

For light microscopy, small cooked sections (10x10x2 mm) were fixed in 10% neutral buffered formalin for 1.5 h followed by dehydrating in 70, 95 and 100% isopropanol each for 2 h, xylene for 2 h and later embedded in paraffin for 3 h. Samples preparation was done in an automated vacuum infiltration unit (Sakura Tissue-Tek VIP, Sakura Finetek, Torrance, U.S.A). The embedded sample were sectioned (Microtome HM 200, Ergostar, Walldoft, Germany) into 4 micron sections, dried for 40 min and stained with Periodic Acid Schiff's reagent (Elbert, 1992). A computerized image

analysis system attached to a microscope (Mode BX60F5, Olympus Optical Co, Ltd., Japan) was used to view (x 20 magnification) and record different areas from treatments 1, 2, 5, and 6.

3.3.6 Statistical Analysis

The experiment was designed as a randomized complete block design with 10 treatments in three independent trials. The treatments consisted of salt (0, 1, 2, 3%), CMG (0, 0.2%) and 0.5% STPP (see Table 3.1). Differences among treatment means were determined by the Duncan's multiple range test. SAS version 6.12 was used to perform the statistical analysis (SAS, 1997).

3.4 Effect of Crude Malva Nut Gum (CMG) and Phosphate on Cook Loss, Fat Loss, Color, Textural Properties and Microstructure of the Mechanically Deboned Chicken Meat Emulsions

3.4.1 Materials

3.4.1.1 Mechanically Deboned Chicken Meat (MDCM)

MDCM was obtained from an Ontario local processing plant. Chicken frames without skin were deboned by a Beehive machine. The MDCM was kept frozen up to 2 weeks at -20 ^{o}C prior to use. Proximate analysis of the MDCM, carried out according to AOAC (1996) was 72.74% moisture, 16.99% protein, 10.24% fat and 0.82% ash.

3.4.1.2 Beef Back-Fat

Beef back-fat was obtained from the University of Guelph abattoir. The beef fat was ground through a 9 mm plate to obtain a homogeneous mass and kept frozen at –20 ^{o} C for 15 min prior to use.

3.4.1.3 Crude Malva Nut Gum

CMG was prepared as described in 3.3.1.2.

3.4.2 Preparation of MDCM Emulsions

Six different MDCM batter formulations (Table 3.2) were prepared in three separate trials. For each treatment 1,500 g of MDCM was thawed (4 ^{o}C for 12 h). The MDCM was chopped (Mainca model J908110, Equipmentos Carnicos, S.L., Barcelona, Spain) at low speed for 30 sec followed by adding 1.5 g/100g salt (based on meat weight) and chopping at the high speed setting for 30 sec. This was followed by a 1.5 min break, i.e., allow time for protein extraction. Next, 23 g/100g beef fat was added and chopped at high speed for 1 min, followed by adding 20 g/100g ice, 3.6 g curing salt (150 mg/kg sodium nitrite by meat weight) and 0.5 g/100g sodium tripolyphosphate (STPP) while chopping at high speed for 1 min. In the treatment with CMG, a gum powder was mixed and added with the ice. The batters were chopped for an additional 2 min; batter temperature did not exceed 12 ^{o}C in any of the treatments. The batters were vacuum packed (Model D-8941, Multivac GmbH, Wolfertschwerden, Germany) to remove air bubbles and later three 30g batters were placed into polypropylene centrifuge tubes, which were centrifuged (Model 225, Fisher Scientific, Pittsburgh, U.S.A.) at the slow speed setting to remove any trapped air. The batters were cooked in a water bath (Haake W-26, Haake, Berlin, Germany) from 10 ^{o}C to 75 ^{o}C within 1.5 h.

Table 3.2 Factorial experiment design of chicken breast meat with 3 crude malva nut gum (CMG) levels (0, 0.2, 0.6%) and 2 sodium tripolyphosphate (STPP) levels (0, 0.5%).

Treatment*	CMG (%)	STPP (%)
1	0.0	0.0
2	0.2	0.0
3	0.6	0.0
4	0.0	0.5
5	0.2	0.5
6	0.6	0.5

* Treatments were composed of MDCM, 1.5% salt, 20% water, 23% beef fat and 150 ppm curing salt.

3.4.3 Cook Loss

Cook loss was measured as described in 3.3.3

3.4.4 Fat loss

Fat separated from the batters was weighed after pouring the separated moisture from batters and kept in the refrigerator at 5 ^{o}C overnight. Fat loss expressed as :

$$\% \text{ Fat loss} = \frac{\text{g fat expelled during cooking}}{\text{g sample before cooking}} \times 100$$

3.4.5 Color

Color of the cross-section of cooked batter (nine pieces per treatment) was measured using a colorimeter (Minolta, Co., Ltd., model CR-200, Osaka, Japan) and expressed as Hunter L (lightness), a (redness) and b (yellowness) values. Then, color difference (ΔE) and hue angle (θ) were calculated as follows:

$$\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]^{1/2}$$

$$\text{Hue angle } (\theta) = \arctan \frac{b}{a}$$

The color difference (ΔE) of higher than one indicates that the color between the 2 samples are different. The higher values of hue angle indicate less red color. The results are average values from three trials.

3.4.6 Texture Profile Analysis (TPA)

TPA was determined as described in 3.3.4

3.4.7 Microscopical Evaluation

Light microscopy of small cooked sections (10x10x2 mm) from treatments 1, 2, 3, 4, 5 and 6 were prepared as described in 3.3.5.

3.4.8 Statistical Analysis

A 2x3 factorial experiment was conducted with 3 separate trials. The principal effects were the presence or absence of 0.5% STPP (see Table 3.2) at 3 different CMG levels (0, 0.2, 0.6%). Differences among treatment means were determined by the Duncan's multiple range test. SAS version 6.12 was used to perform the statistical analysis (SAS, 1997).

3.5 Effect of Crude Malva Nut Gum (CMG), Purified Malva Nut Gum (PMG) and Phosphate on Cook loss, Fat loss, Color, Textural Properties , Rheological Properties and Microstructure of the Chicken Meat Emulsions

3.5.1 Materials

3.5.1.1 Hand Deboned Chicken Breast Meat

Hand deboned chicken breast meat (12 kg), obtained from an Ontario local processing plant was prepared as described in 3.3.1.1

3.5.1.2 Pork Back-Fat

Pork back-fat was obtained from the University of Guelph abattoir. The pork fat was cut to 2x2x2 cm^3 and kept frozen at –20 ^oC for 15 min prior to use.

3.5.1.3 Purified Malva Nut Gum

PMG was prepared as described in 3.2.1.

3.5.2 Preparation of Chicken Meat Emulsions

Seven different chicken batter formulations (Table 3.3) were prepared in three separate trials. For each treatment the chicken meat (375 g) was thawed (4 ^{o}C for 12 h) before used. The meat was chopped (SMK 40, Schneidmeister, Berlin, Germany) at low speed for 30 sec followed by adding 0.75 g/100g salt (based on meat weight) and chopping at the high speed setting for 30 sec. This was followed by a 1.5 min break, i.e., allow time for protein extraction. Next, 30 g/100g pork fat was added and chopped at high speed for 1 min, followed by adding 20 g/100g ice, 1.8 g curing salt (150 mg/kg sodium nitrite) and 0.4 g/100g sodium tripolyphosphate (STPP) while chopping at high speed for 1 min. All of the ingredients are based on batch size of the chicken batters (750 g). In the treatment with CMG, a gum solution was mixed and added with the ice. The batters were chopped for an additional 2 min; batter temperature did not exceed 12 ^{o}C in any of the treatments. The batters were vacuum packed (Model D-8941, Multivac GmbH, Wolfertschwerden, Germany) and later three 30g batters were placed into polypropylene centrifuge tubes which were centrifuged (Model 225, Fisher Scientific, Pittsburgh, U.S.A.) at the slow speed setting to remove any trapped air. The batters were cooked in a water bath (Haake W-26, Haake, Berlin, Germany) from 10 ^{o}C to 75 ^{o}C within 1.5 h.

Table 3.3 Randomized complete block design of chicken breast meat with 3 purified malva nut gum (PMG) levels (0, 0.1, 0.3%), 2 crude malva nut gum (CMG) levels (0, 0.3%) and 2 sodium tripolyphosphate (STPP) levels (0, 0.4%).

Treatment*	PMG (%)	CMG (%)	STPP (%)
1	0.0	0.0	0.0
2	0.1	0.0	0.0
3	0.3	0.0	0.0
4	0.0	0.0	0.4
5	0.1	0.0	0.4
6	0.3	0.0	0.4
7	0.0	0.3	0.0

* Treatments were composed of chicken breast meat, 0.75% salt, 20% water, 30% pork fat and 150 ppm curing salt.

3.5.3 Cook Loss

Cook loss was measured as described in 3.3.3

3.5.4 Fat loss

Fat separated from the batters was measured as described in 3.4.4.

3.5.5 Color

Color of cross-section cut cooked meat emulsion surfaces (nine pieces per treatment) was measured using a Hunter Lab Miniscan Spectrocolorimeter (Model MS/S-4500L, Hunter Associates Laboratory, Inc., Reston, VA, U.S.A.) and expressed as CIE L* (lightness), a* (redness) and b* (yellowness) values. Then, color difference (ΔE*) and hue angle (θ) were calculated as follows:

$$\Delta E^* = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$$

$$\text{Hue angle } (\theta) = \arctan \frac{b^*}{a^*}$$

If the color difference (ΔE) is higher than one, it indicates that the color between the 2 samples are different. The higher values of hue angle indicate less red color. The results are average values from three trials.

3.5.6 Texture Profile Analysis (TPA)

TPA was determined as described in 3.3.4

3.5.7 Rheological Properties

Continuous evaluations of the storage modulus (G′) during thermal processing of meat batters were performed by a Bohlin rheometer (Model CS 50, Bohlin Instruments Ltd, Gloucestershire, England). Measurements were carried out using the cup and bob geometry (C25 DIN53019) with a gap setting of 150 μm, frequency of 1 Hz and

amplitude oscillation of 0.0012 strain units. A small amount of mineral oil was put on the top to prevent it from drying out during the cooking process. The chicken batters were heated from 30 to 70 ^{o}C and cooled back down to 30 ^{o}C at a rate of 1.5 ^{o}C/min. Rheological data are presented as G′ value measured (Pa).

3.5.8 Microscopical Evaluations

Light microscopy of small cooked sections (10x10x2 mm) from treatments 1, 2, 3, 4, 5 and 6 were prepared as described in 3.3.5.

3.5.9 Statistical Analysis

The experiment was designed as a randomized complete block design with 7 treatments in three independent trials. The treatments consisted of PMG (0, 0.1, 0.3%), CMG (0, 0.3%) and 0.4% STPP (see Table 3.3). Differences among treatment means were determined by Duncan's multiple range test. SAS version 6.12 was used to perform the statistical analysis (SAS, 1997).

3.6 Effect of Crude Malva Nut Gum (CMG) Level on Physical and Sensory Properties of the Commercial Type Frankfurters

3.6.1 Materials

3.6.1.1 Mechanically Deboned Chicken Meat

The fresh MDCM was obtained from a local Thai processing plant. Chickens necks and frames without skin were deboned by a deboner. The meat was frozen and kept frozen in polyethylene bags at –20 ^{o}C for 2 day prior to use. Proximate analysis of the MDCM, carried out according to AOAC (1996), was 62.97% moisture, 12.64% protein, 22.98% fat and 1.02% ash.

3.6.1.2 Beef Back-Fat

Beef back-fat was obtained from a Thai slaughterhouse. The beef fat was ground through a 9 mm plate to obtain a homogeneous mass and kept frozen in polyethylene bags at –20 ^{o}C for up to 2 weeks prior to use.

3.6.1.3 Crude Malva Nut Gum (CMG)

CMG was prepared as described in 3.3.1.2

3.6.2 Preparation of Frankfurters

Three different MDCM frankfurter samples were prepared in three separate trials. The MDCM (15 kg/batch) was chopped in a silent cutter (Model K 41 RAS 661234, Maschinenfrabrik Seydelmann KG, Stuttgart, Germany) at low speed for 30 sec. 1.5% g/100g salt (based on the meat weight) was added and the chopping continued at high speed setting for 30 sec. After setting in the bowl chopper for 1.5 min to allow time for protein extraction, 2 g/100g beef fat was added and chopped at high speed for another 1 min. Then 2.5 g/100g ice, 2.5 g/100g water, 0.80 g/100g dextrose, 0.27 g/100g white pepper, 0.052 g/100g garlic powder, 0.052 g/100g nutmeg, 0.052 g/100g coriander root powder, 150 mg/kg sodium nitrite and 0.045 g/100g sodium erythorbate were added and chopped at high speed for 1 min. In the treatment with malva nut gum, the gum solution was prepared with 2.5 g/100g cold water and then added at the same time together with the ice. The batters were chopped for an additional 2 min and the batter temperature kept below 12 ^{o}C for all cases. The batters were then stuffed (Model F25-TOP-1124, Albert Handtmann Maschinenfabrik GmbH & Co. KG, Biberach, Germany) into 21 mm collagen casings (Nippi casing, Tokyo, Japan) and after 1 h in the cooler, they were smoked and cooked (Model UK 1-0/50-70 kg, GmbH & Co. KG, Waxweiler, Germany). The procedure included drying at 50 ^{o}C for 25 min, heating to 55^{o}C for 12 min, applying the natural smoke for 22 min and continuing heating in steps, up to 74 ^{o}C internal temperature. Relative humidity was gradually increasing up to 99% at the end. The frankfurters were then cold showered for 30 min, stored overnight at 3 ^{o}C, vacuum packed and stored at 5 ^{o}C.

3.6.3 Cook Loss

Weight differences of the sausages before and after cooking were measured and expressed as :

$$\% \text{ Cook loss} = \frac{\text{g sausage after cooking}}{\text{g sausage before cooking}} \times 100$$

3.6.4 Color

Color of cross-section cut cooked emulsion surfaces (nine pieces per treatment) was measured using a colorimeter (Model CR-300, Minolta, Co., Ltd., Osaka, Japan) and expressed as hunter L (lightness), a (redness) and b (yellowness) values. Then, color difference (ΔE) and hue angle (θ) were calculated as described in 3.4.5. The results are expressed as the average values from three different trials.

3.6.5 Texture Profile Analysis (TPA)

TPA was determined as described in 3.3.4.

3.6.6 Sensory Analysis

Sensory analysis was performed by graduate students and staff members of the Department of Food Technology, Chulalongkorn University. The six panelists who had experience in evaluating emulsion type meat products were trained by a texture profile panel training method (Civille and Szczesniak, 1973). The sensory notes were established by the trained panel. The cooked samples were identified by a 3-digit random number, were placed on a round plate. The samples were reheated in a microwave oven for 30 sec and served at 45 $\pm$ 4 ^{o}C. Descriptive sensory analysis evaluations were recorded by the panelists (Stone *et al*., 1974). The structured linear scales on ballots were coded on a 1-10 scale. Attributes included color (1 = brown, 10 = brownish pink), texture (1 = very soft, 10 = very firm), elasticity (1 = low elasticity, 10 = high elasticity), juciness (1 = very dry, 10 = very juicy), flavour (1 = unpleasant, 10 = extremely pleasant), and overall acceptability (1 = unacceptable, 10 = extremely acceptable).

3.6.7 Statistical Analysis

The experiment was designed as a randomized complete block with three treatments in three independent trials for sensory analysis and a completely randomized with three treatments in three independent trials for cook loss, color and textural properties. Differences among treatment means were determined by the Duncan's multiple range test. SAS version 6.12 was used to perform the statistical analysis (SAS, 1997).

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Extraction and Physicochemical Characterization of Malva Nut Gum

4.1.1 Extraction of Malva Nut Gum

The sequential extraction procedure indicated that malva nut polysaccharides could not be solubilized effectively neither by hot water nor by a dilute acid solution. The yields by hot water extraction and dilute acid extraction were only approximately 1.00±0.01% and 6.00±0.01% (dry basis), respectively. Extraction with 0.05 M NaOH provided the highest yield of approximately 20±0.02 % (dry basis). Clearly, most of the polysaccharides are covalently linked to other components or themselves in the plant cell so that they can be effectively released under alkaline condition.

4.1.2 Composition of Alkaline Extracted Malva Nut Gum

According to the highest yield obtained from extraction, the alkaline extracted malva nut gum was selected for further characterization. Chemical analyses showed the gum contained 83.10 $\pm$ 0.01% total carbohydrate, 8.30 $\pm$ 0.01% protein and 8.40 $\pm$ 0.13 % ash. The monosaccharide compositions of the alkaline extracted malva nut gum fraction are presented in Table 4.1. It contained mainly arabinose, galactose, and rhamnose in the ratio of galactose : arabinose : rhamnose = 0.21 : 1.00 : 1.24. This component is slightly different from that of the aqueous extract of *Sterculia lychnohora* (Hance) seeds (Chen *et al*., 1996) in which the major monosaccharide components reported were galactose : arabinose : rhamnose = 1.01 : 1.67 : 1.00. There are a number of other plant polysaccharide gums that have monosaccharide compositions similar to those found in malva nut gum. BeMiller (1973) reported that flaxseed mucilage was composed primarily of polysaccharides which, on acid-catalyzed hydrolysis, yielded L-galactose, L-arabinose, L-rhamnose, D-galacturonic acid and traces of D-glucose. The major monosaccharides in gum arabic (*Acacia senegal*) were 13% rhamnose, 27% arabinose, 44% galactose and 15% glucuronic acid (Williams and Phillips, 2000). The level of 6.40 $\pm$ 0.31% uronic acid (as galacturonic acid equivalent) in malva nut gums reflects the relative amount of acidic polysaccharides in the gum. The lower amounts of detected monosaccharides comparing to the total carbohydrate content was likely caused by the incomplete hydrolysis of the acidic fraction of the polysaccharides. In summary, the result from this study indicated that arabinose, galactose and rhamnose are the major monosaccharides present in malva nut gum.

Table 4.1 Monosaccharide content of the alkaline extracted malva nut gum.

Monosaccharide	(%, w/w)
Arabinose	17.10 ± 0.15
Galactose	15.10 ± 0.22
Rhamnose	15.00 ± 0.12
Glucose	0.52 ± 0.23
Xylose	0.32 ± 0.01

4.1.3 Molecular Weight Characterization

4.1.3.1 Effect of Dialysis

Molecular weights of malva nut gums, both before and after dialysis against de-ionized water were determined by high performance size exclusion chromatography (HPSEC). The HPSEC chromatograms of both samples (before and after dialysis) are shown in Fig. 4.1. The gum solutions were eluted from the column at similar retention volumes. HPSEC coupled with three detectors permitted the determination of average molecular weights (M_w) and intrinsic viscosity ([η]). The results are summarized in Table 4.2. The weight average molecular weight of the gum was 6.65 x 10^6 Daltons before dialysis. After dialysis, it was reduced to 3.30 x 10^6 Daltons. Chemical analysis showed that the ash content of the gums was reduced from 8.40% to 5.34% (dry basis) after dialysis. It is likely that the reduction in molecular weight is associated with the disruption of some structural associations among gum molecules, due to ion removal

during dialysis. Cui *et al.* (1994), also reported that dialyzed flaxseed gum exhibited lower apparent viscosity and much weaker viscoelastic responses when compared to a crude flaxseed gum. The M_w of malva nut gum obtained in the present study is much higher than the M_w of water extracted polysaccharides gum obtained from *Sterculia lychnophora* (Hance) with a value of 162,200 Daltons (Chen *et al*., 1996). The reason might be associated with the different species of malva nut used in the separated studies. As mentioned previously, the monosaccharide composition of both polysaccharides are also different. The intrinsic viscosity of the malva nut gum after dialysis was also lower than that before dialysis. This is consistent with the molecular weight result.

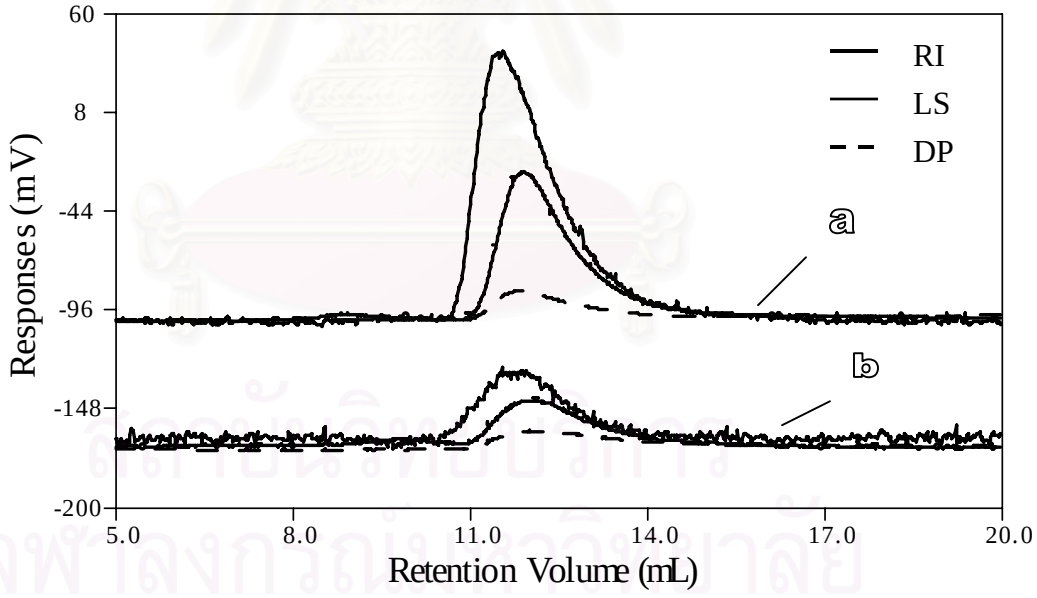

Fig. 4.1 High performance size exclusion chromatograms of the alkaline extracted malva nut gum before dialysis (a) and after dialysis (b).

Table 4.2 Molecular weights and intrinsic viscosities of the alkaline extracted malva nut gum solutions before and after dialysis.

Alkaline extracted malva nut gum	Mw (Daltons)	Intrinsic viscosity (dl/g)
Before dialysis	$6.65 \times 10^6 \pm 1.70 \times 10^6$	10.0 ± 1.51
After dialysis	$3.30 \times 10^6 \pm 7.41 \times 10^5$	7.70 ± 1.24

4.1.3.2 Effect of Decoloring by Hydrogen Peroxide

Molecular weights of malva nut gums, both before and after decoloring by hydrogen peroxide were determined once again by size exclusion chromatography. HPSEC coupled with three detectors permitted the determination of average molecular weights (M_w) and intrinsic viscosity [η]. The results are summarized in Table 4.3. The weight average molecular weight of the gum was 6.65×10^6 Daltons before decoloring. After decoloring, it was reduced to 2.27×10^6 Daltons. This is probably because hydrogen peroxide is a very strong oxidizing agent that might cause some structural degradation and resulted in the molecular weight reduction. Furthermore, the alkaline condition in the decoloring process probably enhanced the degradation of the gum. Rombouts and Thibault (1986) also reported that the average molecular weight (36,400) and intrinsic viscosity (181 ml/g) of alkaline-soluble pectin from sugar beet were lower than the average molecular weight (47,700) and intrinsic viscosity (259 ml/g) of the water-soluble pectin. The intrinsic viscosity of the malva nut gum after decoloring

was also lower than that before decoloring. This is in accordance with the molecular weight result.

Table 4.3 Molecular weights and intrinsic viscosities of the alkaline extracted malva nut gum solutions before and after decolorization by hydrogen peroxide.

Alkaline extracted malva nut gum	Mw (Dalton)	Intrinsic viscosity (dl/g)
Before decoloring	$6.65 \times 10^6 \pm 1.70 \times 10^6$	10.0 ± 1.51
After decoloring	$2.27 \times 10^6 \pm 1.63 \times 10^5$	5.43 ± 0.22

4.1.4 Methylation Analysis

Carbodiimide-activated reduction of the carboxyl groups of glycosyluronic acids with sodium borodeuteride ($NaBD_4$) resulted in easily identified sugars (deuterized). There were three major peaks detected from the GC-MS analysis of the partially methylated alditol acetate (PMAA) derived from the carboxyl reduced alkaline extracted malva nut gum (Fig. 1, Appendix B) and their corresponding mass spectra are showed in Figures 2A, 2B and 3 (Appendix B). Methylation analysis of PMAA, derived from the alkaline extracted malva nut gum revealed that it is primarily composed of 1,4-D-Gal*p*, 1,3-linked-L-Ara*f* and 1,2 and 1,3-linked Rham*p* in the molar ratios of 0.21 : 1.00 : 1.24, respectively (Table 4.4). It is worth noting that the amount of 1,4-D-Gal*p* was much lower than that of the galactose detected by the monosaccharide analysis before uronic acid reduction (Table 4.4) and it increased dramatically after uronic acid reduction. Since the total uronic acid content was only about 6.4%, it is unlikely that all the increase of 1,4-D-Gal*p* was caused by the reduction of galacturonic acid. This indicated that the 1,4-D-Gal*p* was not completely converted during the methylation analysis possibly due to its presence near the galacturonic acid which prohibited the hydrolysis and complete methylation of the 1,4-linked-D-Gal*p*. This finding is similar to Chen *et al*. (1996) who reported that there were rhamnoses in the main chain of isolated and purified malva nut polysaccharides which were linked by α-(1→3) glycosidic linkages but the molar ratio of galactose, arabinose and rhamnose (1.01 : 1.67 : 1.00), were remarkably different from the values found in the present

study. This might be due to the fact that different species of malva nut gums possess different amounts of monosaccharides.

Table 4.4 Chemical names and deduced linkage of partially methylated alditol acetates (PMAA) of the alkaline extracted malva nut gum with and without uronic acid reduction.

Chemical name	Deduced linkage	Molar ratio* (a)	Molar ratio* (b)
1,4,5-tri-*O*-acetyl-(1-deuterio)-2,3,6-tri-*O*-methyl galactol	1,4-D-Gal*p*	0.21	1.56
1,3,4-tri-*O*-acetyl-(1-deuterio)-2,5-di-*O*-methyl-arabinotol	1,3-L-Ara*f*	1.00	1.00
1,3,5-tri-*O*-acetyl-(1-deuterio)-2,4-di-*O*-methyl-6-deoxy-rhamnotol 1,2,5-tri-*O*-acetyl-(1-deuterio)-3,4-di-*O*-methyl-6-rhamnotol	1,3-Rham*p* and 1,2-Rham*p*	1.24	1.97

* Relative molar ratio calculated from the ratio of peak area.

(a) Without uronic acid reduction.

(b) With uronic acid reduction.

4.1.5 FT-IR Spectroscopy

The FT-IR spectrum of alkaline extracted malva nut gum (Fig. 4.2) is very noticeably similar to that of gum arabic. This observation confirmed the result of monosaccharide composition and methylation analysis. In contrast, the spectra of water and acid extracted gums exhibited characteristic absorbances of low DE pectins. It is worth noting that the gum arabic-like portion of malva nut gum was not soluble in hot water and weak acid but it was only extractable by alkaline solution. The poor solubility in hot water and acidic conditions are possibly due to their extremely large molecular weight (> 6 millions Dalton comparing to 0.6 millions Dalton for gum arabic) and to the possibility of interpolymeric cross links via covalent bonds. It appeared that the alkaline extraction caused saponification of the methyl esterified carboxyl group. Bands in the 1,000-2,000 cm^{-1} region are independent of pectin source and may be used to identify the presence of galacturonic acid (Filippov and Shamshurina, 1972; Wellner *et al.*, 1998). The carbonyl absorption bands at 1,600 and 1,740 cm^{-1} were from free (COO-) and esterified (COO-R) carboxyl groups, respectively. It was observed that the total carbonyl absorption band area increased as the polygalacturonic acid content increased. All spectra showed carboxylate ion peaks at 1,600 and 1,414 cm^{-1} and carbohydrate peaks at 1,140, 1,100, 1,060 and 990 cm^{-1}. The carbohydrate peaks of the alkaline extracted malva nut gum were similar to those of gum arabic because they had a similar monosaccharides composition. According to Glicksman (1969), the complete acid hydrolysis of gum arabic resulted in D-galactose, L-arabinose,

L-rhamnose, and D-glucuronic acid. It was later reported that the neutral sugar composition and uronic acid content of gum arabic were 34% rhamnose, 24% arabinose, 45% galactose and 15% glucuronic acid (Cui and Mazza, 1996; Anderson and Morrison, 1990).

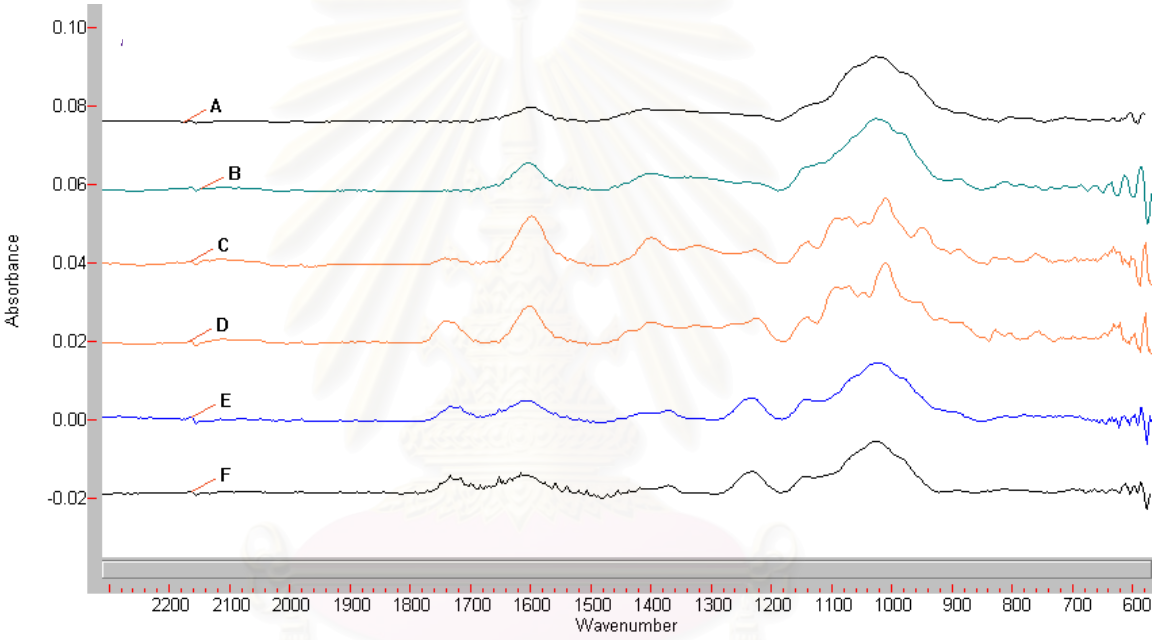

Fig. 4.2 FT-IR spectra of gum arabic (A), malva nut gum (alkaline extraction) (B), 26% DE citrus pectin (C), 59% DE citrus pectin (D), malva nut gum (acid extraction) (E), and malva nut gum (water extraction) (F) .

4.2 Rheological Properties of the Purified Malva Nut Gum (PMG)

4.2.1 Steady Shear Viscosity of PMG

The effect of shear rate on viscosity of purified malva nut gum (Fig. 4.3) revealed that PMG solutions are highly shear thinning, i.e. decreasing apparent viscosities being greatly noticeable with increases in shear rate. This behavior is similar to that observed with xanthan solutions, a stiff polysaccharides giving high viscosity solution. Morris *et al.* (1981) found that shear thinning was much more dramatic for 1.50, 2.75 and 5.00% (w/v) of λ-carrageenan solutions. Graessley (1974) indicated that the interpenetration of polymer coils in concentrated solutions gaves rise to a dynamic 'entangled' network structure. Morris *et al.* (1981) also reported that at low rates of shear, those entanglements which were disrupted by the imposed deformation were replaced by new interactions between different molecules, with no net change in the extent of entanglement, and hence no reduction in viscosity. This agrees with the present study that Newtonian plateaus were observed at concentrations of 0.1% and 0.3%. Both zero-shear-rate viscosity (η_o) and shear rate value ($\dot{\gamma}$) at the onset of shear thinning behavior could be determined, as shown in Table 4.5. It was previously reported that the weight average molecular weight and intrinsic viscosity ([η]) of the PMG were found to be 3.3×10^6 Daltons and 7.7 dl/g, respectively. The overlap concentration c*, which is the concentration at which molecules start to contact and penetrate each other, is then estimated to be $1/[\eta] = 0.13\%$. It can be seen from Figure 4.3 that, even at a dilute solution of 0.1%; the value being lower than the overlap concentration, PMG exhibited

appreciable shear thinning behavior and the shear rate of onset shear thinning was very low (approximately 0.3 s^{-1}). In contrast, for many linear polysaccharides such as guar gum and xyloglucans of similar molecular weight, the shear thinning behavior is only evident at such low shear rate when concentration is higher than approximately 1% (Wang *et al*., 1997). It has been shown that PMG has a highly branched structure similar to gum arabic (Somboonpanyakul *et al*., 2004b), although the latter has a much lower molecular weight. However, solutions of gum arabic at concentrations as high as 30% show essentially Newtonian flow behavior (Williams and Phillips, 2000). The present data suggests that there are strong intermolecular interactions among PMG molecules in water solution. Both high molecular weight nature and intermolecular interactions contribute to the properties of high viscosity solution and strong shear thinning flow behavior of PMG solution.

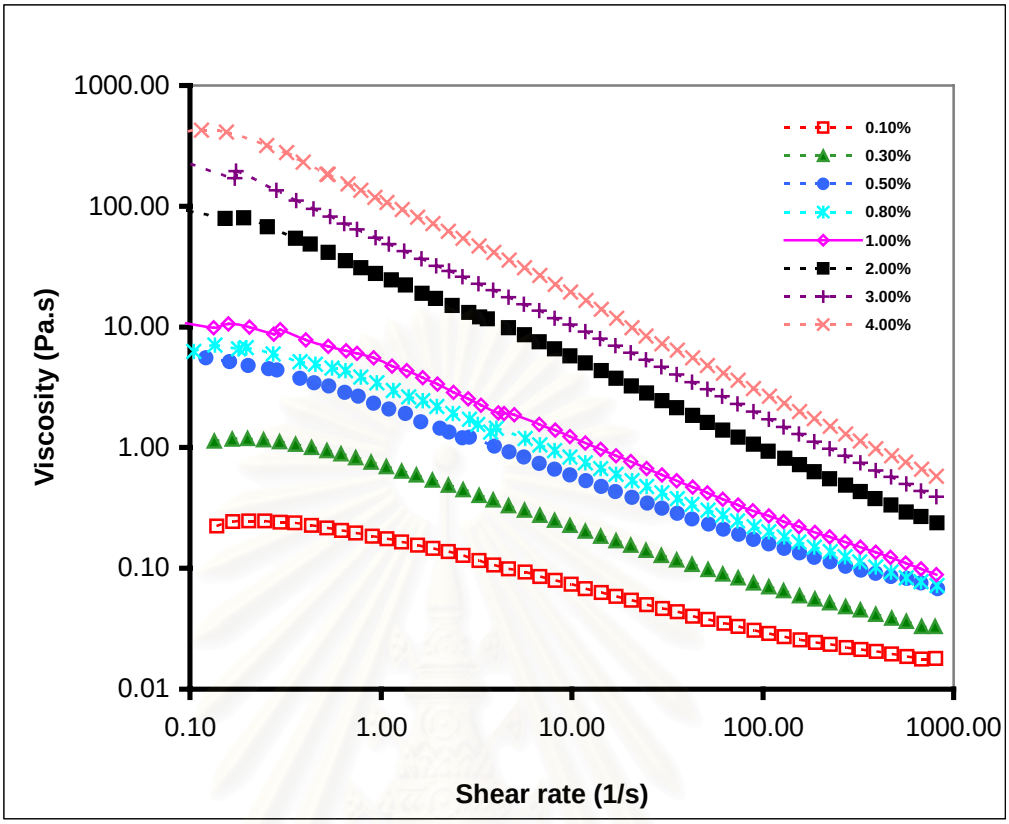

Fig. 4.3 Effect of shear rate on viscosity of purified malva nut gum at concentration between 0.1 to 4.0%, 25 ^{o}C.

The zero-shear-rate viscosity value of 0.3% purified malva nut gum solution was higher than that value for 0.1 % gum solution (Table 4.5). The $\dot{\gamma}$ value of the 0.3 % PMG solution was lower than a 0.1 % gum. It showed that at the higher gum concentration, there was more elastic behavior than at the lower concentration. Cui *et al.* (1993) also recorded Newtonian plateaus for 0.3 % of yellow mustard mucilage solution.

Table 4.5 Observed zero-shear-rate viscosity (η_o) and shear rate ($\dot{\gamma}$) value at which onset of shear thinning occurred for 0.1% and 0.3% purified malva nut gum solutions (25 ^oC).

Concentration (% w/v)	η_o (mPa.s)	$\dot{\gamma}$ value (s^{-1})
0.1	237	0.355
0.3	1120	0.294

The most widely used mathematical expression for pseudoplastic rheological behavior of hydrocolloid solutions/dispersions is the power law model described by Ostwald (Whitcomb *et al*., 1980):

$$\eta = K \dot{\gamma}^{n-1} \qquad (1)$$

where η is the apparent viscosity (Pa.s), $\dot{\gamma}$ is the shear rate, *K* is the consistency index (Pa.s), and *n* is the flow index which measures the pseudoplasticity of the system. A comparison of *n* and *K* values of PMG solutions is given in Table 4.6.

The consistency index (*K*) and flow index (*n*) (Table 4.6), were obtained from the power-law model (Whitcomb *et al*., 1980). As gum concentration increased, the *K* values increased but the *n* values decreased. The increase in *K* with increasing concentration suggests that a more viscous system is obtained at higher concentrations. In contrast, the decrease in *n* with increasing concentration implies a more prominent shear thinning behavior of the system. A linear plot of n versus log c has been obtained

for a number of polysaccharide solutions (Launay *et al*., 1986). However, a power law relationship between n and log c was found for PMG solutions as illustrated by Figure 4.4. The value of n decreased with increased log concentration more rapidly and especially at log concentrations lower than 1%, then gradually leveled off to a value of approximately 0.25.

Table 4.6 Comparison of *n* and *K* values of the purified malva nut gum solutions at different concentrations (25 ^{o}C).

Concentration (% w/v)	*n*	*K* (Pa.s)
0.1	0.653	0.141
0.3	0.539	0.645
0.5	0.465	2.090
0.8	0.442	2.904
1.0	0.420	4.377
2.0	0.308	25.221
3.0	0.284	50.104
4.0	0.224	105.880

Parameters *n* and *K* were calculated using the power-law model: $\eta = K \cdot \gamma^{n-1}$ (Whitcomb *et al*., 1980).

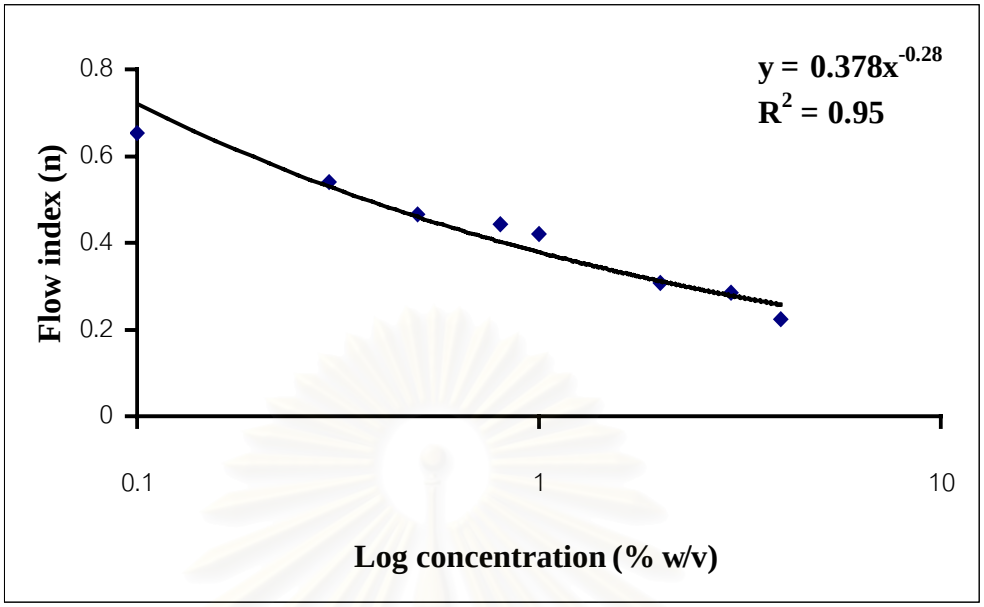

Fig. 4.4 A power law relationship between flow index (n) and log concentration (% w/v) of PMG solutions.

4.2.2 **Oscillatory Testing of PMG**

Small deformation oscillatory tests are useful in examining the molecular origin of the rheological properties of hydrocolloid solutions or dispersions (Cui *et al*., 1994). The storage modulus (G′) reflects the solid-like properties of a viscoelastic material, while the loss modulus reflects its liquid-like character (Whitcomb *et al*., 1980). The mechanical spectra of the PMG solutions showed G′ > G″ over the entire frequency range examined (Fig. 4.5). However, both G′ and G″ showed appreciable frequency dependence, especially for the 0.5 % PMG dispersion. The frequency dependency became less evident when the polysaccharide concentration was increased. In addition, G′ was only slightly higher than G″, i.e. less than 10 folds. Complex viscosity (η*), a parameter related to the viscoelastic response of the PMG solutions, also decreased as a function of frequency. Overall, the current data indicates a typical weak gel structure which corresponds to a solution of entangled macromolecules (Ross-Murphy, 1995; Steffe, 1996).

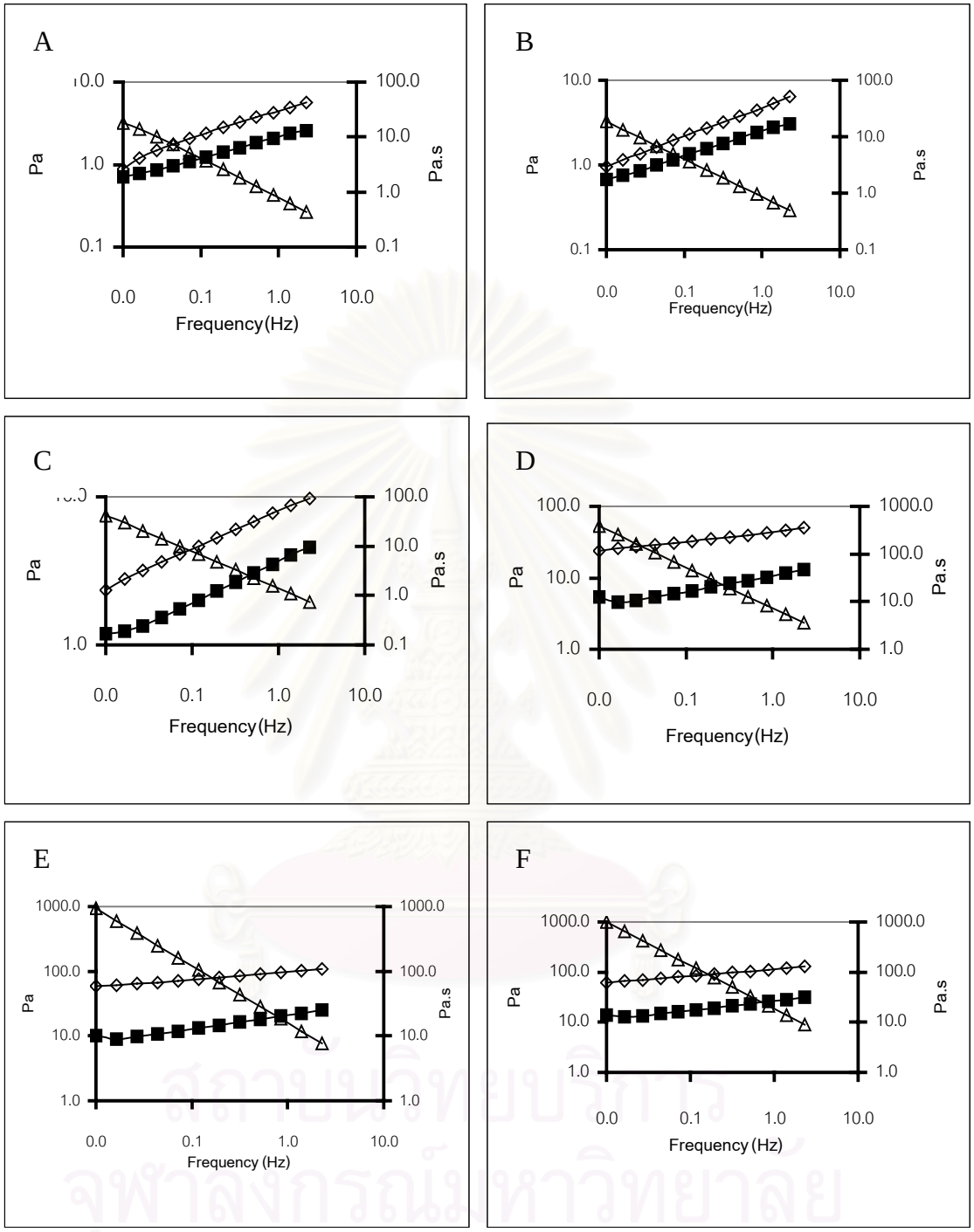

Fig. 4.5 Frequency dependence of storage (G′, —◇—) and loss (G′, —■—) moduli, and complex viscosity (η*,—△—) of the purified malva nut gum solutions at different concentrations. (A, 0.5%), (B, 0.8%), (C, 1.0%), (D, 2.0%), (E, 3.0%) and (F, 4.0 %).

4.2.3 **Effects of Ca^{2+} and Temperature on PMG Solutions' Property.**

Since the PMG contains approximately 6.40% uronic acid (as galacturonic acid equivalent), it is interesting to investigate if an addition of Ca^{2+} and heat treatment affect the solution's properties. The addition of 5 to 15 mM calcium chloride to 1.5% PMG solutions did not change the values of G′ and G″ (Fig. 4.6). Moreover, the changes in G′ and G″ during heating (1 ^{o}C/min) the PMG gels with three different Ca^{2+} ion concentrations could be observed. All samples did not show a melting point within the temperature range studied. G′ and G″decreased slightly with increasing temperature; however, there was no apparent difference between the three samples with different Ca^{2+} concentration. The rate of decrease of G″ was more evident than that of G′. From these data, it is clear that the origin of G′ did not result from the formation of crosslinks by complexation between Ca^{2+} ions and PMG. The weak gel structures of PMG solutions were more likely attributed to the formation of entangled macromolecules networks.

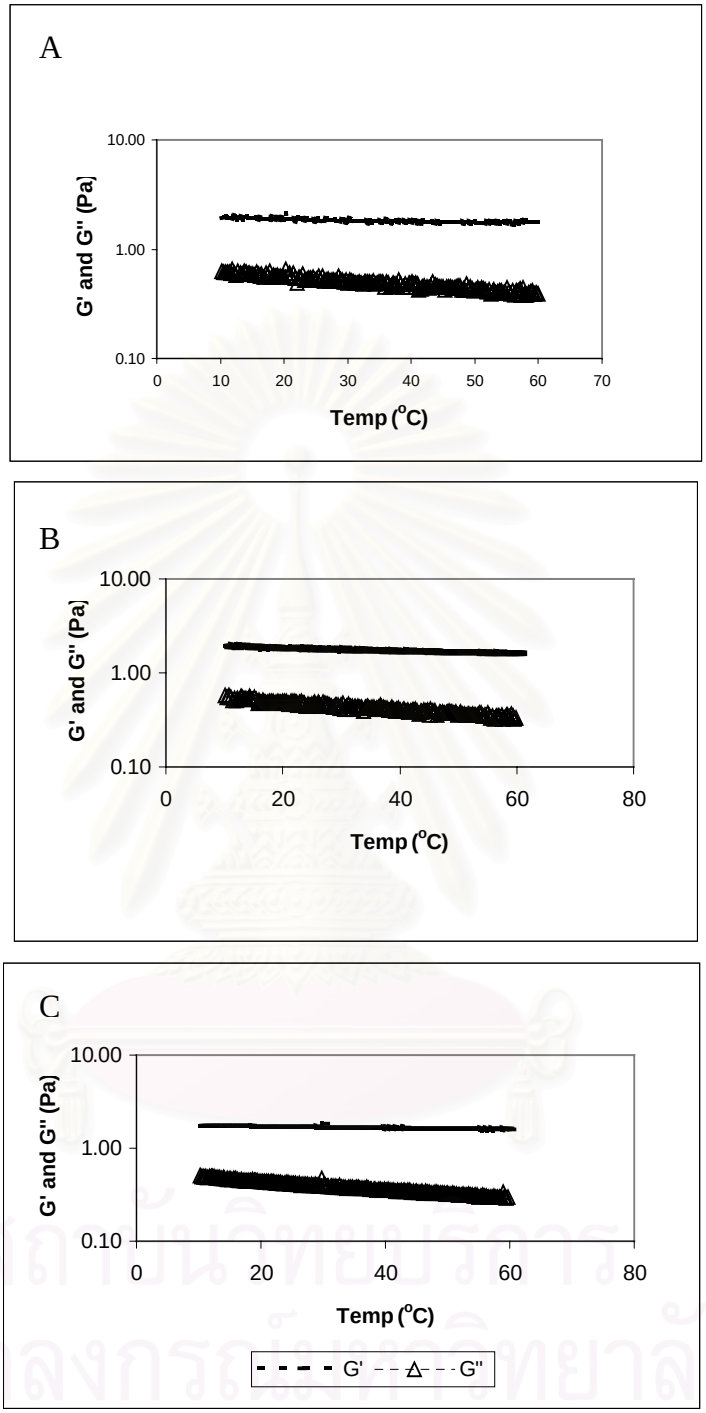

Fig. 4.6 Temperature dependence of storage (G′) and loss (G″) moduli during heating from 10 to 60 ^{o}C at a rate of 1 ^{o}C/min for 1.5% (w/v) of the purified malva nut gum gels with (A) 5 mM $CaCl_2$, (B) 10 mM $CaCl_2$ and (C) 15 mM $CaCl_2$.

In conclusion, PMG was an acidic polysaccharide that contained anionic carboxyl groups from galacturonic acid. PMG was composed of D-galactopyranosyluronic acid units. They also contained L-arabinose residues and other neutral units (galactose, rhamnose, glucose and xylose). D-galacturonic acid residues were joined by (1→4) linkage to form the main chain. The structure of PMG was similar to low methoxyl pectin (LMP) that is primarily composed of (1→4)-D-galactopyranosyluronic acid units except that the carboxyl groups are partially esterified with methanol. Pectin also contained in a lesser extent L-rhamnose residues and other neutral sugars (galactose, arabinose, and xylose) (May, 1997). Both PMG and LMP were soluble in cold water although the molecular weight of PMG (6.65×10^6 Daltons) was much higher than pectins (0.09×10^6 to 0.20×10^6 Daltons) (Corredig *et al*., 2000). Solution of PMG exhibits pseudoplastic behavior, similar to that of LMP (Krumel and Sarkar, 1975). Gelation behavior of PMG can be established for a dilute solution (1.5%), heating for 3 h at 60 ^{o}C and cool to room temperature (25 ^{o}C) to form a weak gel while gelation of LMP can take place across a wide pH range (from 2.9 to 5.5), a soluble solids content from 10 % to 80% and a controlled amount of calcium ions (Dziezak, 1991). Due to a very high water absorption and gelling property, PMG can be used as gelling and thickening agent in the food products, as well as LMP.

4.3 Effect of Crude Malva Nut Gum (CMG), Purified Malva Nut Gum (PMG), Sodium Chloride and Phosphate on Cook Loss, Textural Properties and Microstructure of Lean Chicken Meat Batters

4.3.1 Effect of CMG, Sodium Chloride and Phosphate on Cook Loss, Textural Properties and Microstructure of Lean Chicken Meat Batters

Chicken breast meat used in this experiment was composed of 74.78% moisture, 22.46% protein , 0.62% fat and 1.02% ash. Lean chicken meat batter was prepared as described in 3.3.2.

4.3.1.1 Cook Loss from Lean Chicken Meat Batters

The statistical analysis (Table 1, Appendix C) revealed that treatment (varying amounts of functional ingredients) had a highly significant ($P < 0.01$) effect on cook loss of the chicken batters. There was an overall decrease in cook loss as salt level was increased from 0 to 3% (Fig. 4.7A). Higher salt content allowed more meat protein extraction and thus more moisture binding, Schults and Wierbicki (1973) also showed that increasing salt from 0 to 1% reduced shrinkage of chicken meat after cooking from 35% to 18%, but using 2 or 5% salt resulted in shrinkages with a marginal difference at approximately 16%. Gordon and Barbut (1992a) reported that increasing NaCl from 1.5 to 2.5% enhanced protein extraction (i.e. determined by gel electrophoresis) in chicken breast meat batters. This implies that the higher level of salt extracted more salt soluble meat proteins and further enhanced network formation.

Treatment 10 showed the low cook loss due to the combination of 2% salt, 0.5% STPP and 0.2% CMG. Added STPP and CMG helped to further reduce the cook loss (15.92% to 9.97%; Trt. 5 vs. 10; Fig. 4.7A). This is because STPP enhanced extraction and solubilization of myofibrillar proteins contributing to meat particle binding while CMG help improve water-holding capacity by absorption of water. Thus decreased cook losses and improved texture of the meat batters were obtained. Many researchers reported that NaCl levels of 1.0 to 1.5% resulted in unstable emulsified meat products, while levels of 1.5 to 2.5% NaCl were needed for formation of acceptable products (Seman *et al*., 1980; Puolanne and Terrell, 1983a; Sofos, 1983b; Whiting, 1984). Moreover, polyphosphates can attach to a positively charged group, while the rest of the chain can attract water molecules and increase water-holding capacity by acting as a polyanion (Steinhauer, 1983). The influence of phosphates in improving water-holding capacity is greatly enhanced when used in combination with NaCl (Swift and Ellis, 1956; Hamm, 1960; Hellendoorn, 1962; Shults and Wierbicki, 1973). Pepper and Schmidt (1975) reported that a 2.0% NaCl and 0.5% of STPP and hexametaphosphate was an optimal combination for obtaining the highest cook yield in beef rolls. CMG helped to decrease cook losses of meat batters because the interactions between their anionic carboxyl groups and positively charge meat proteins were much stronger at pH 6 (pH of the meat) (Samant *et al*., 1993). Barbut and Mittal (1996) reported that moisture loss, during cooking, was reduced from 10% to 6% when 0.35% carboxymethylcellulose was added to low fat pork/beef frankfurters. Berry and Bigner (1996) also reported that adding 1.5% salt with 0.38% i-carrageenan improved cooking yield, juiciness and tenderness scores of partially

cooked low-fat nuggets compared with an all-pork nugget produced without the gum and salt.

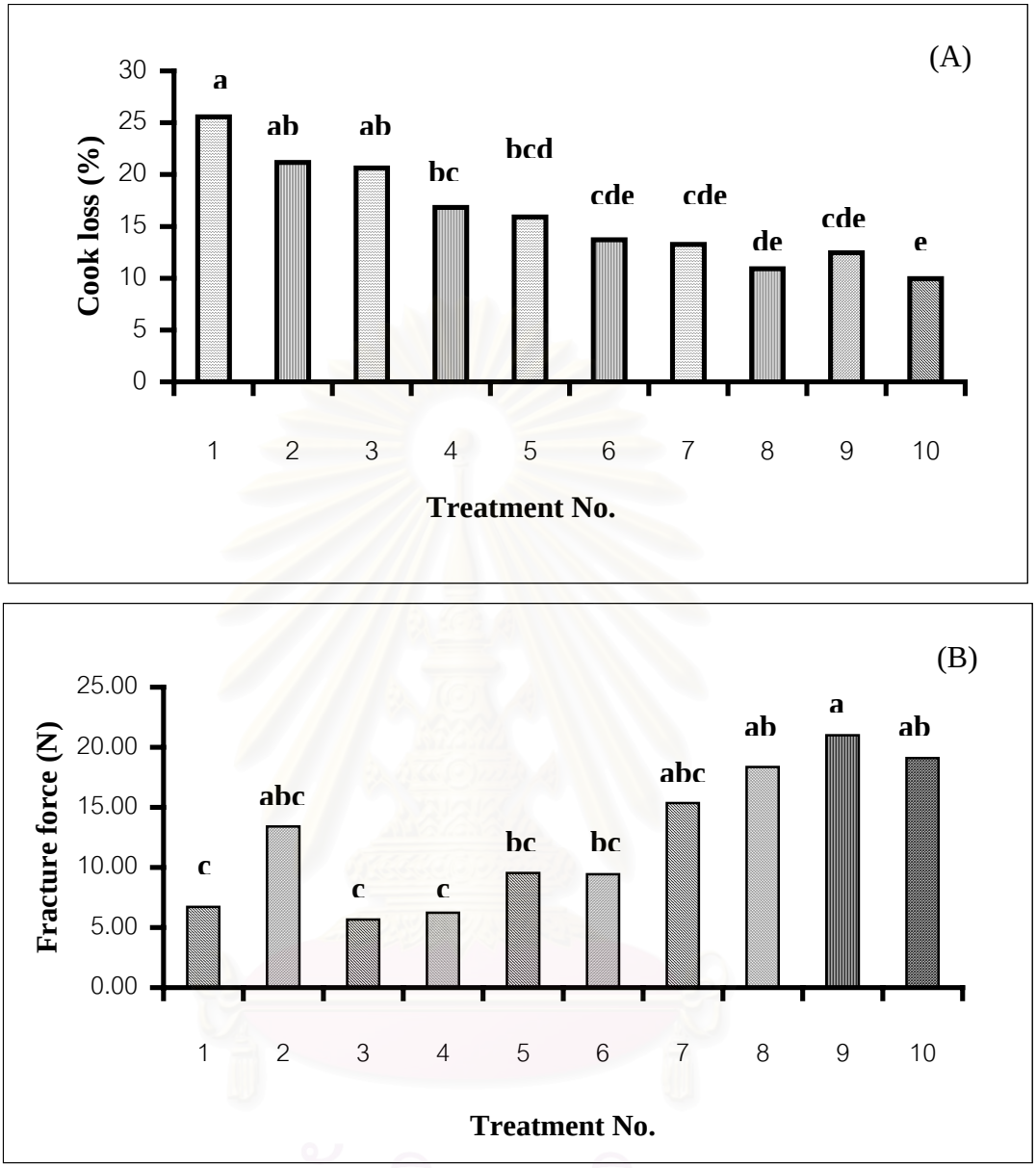

Fig. 4.7 Means for cook loss (A) and fracture force (B) of lean chicken meat batters containing different levels of salt, sodium tripolyphosphate (STPP) and crude malva nut gum (CMG). Bars with different letters are different ($P < 0.01$). Treatment numbers are: 1 = no salt, 2 = 0.2 % CMG, 3 = 1 % salt, 4 = 1 % salt and 0.2 % CMG, 5 = 2 % salt, 6 = 2 % salt and 0.2 % CMG, 7 = 3 % salt, 8 = 3 % salt and 0.2 % CMG, 9 = 2 % salt and 0.5 % STPP and 10 = 2 % salt, 0.5 % STPP and 0.2 % CMG.

4.3.1.2 Textural Properties of Lean Chicken Meat batters

The statistical analysis results (Table 1, Appendix C) revealed that treatment (varying amounts of functional ingredients) had a significant ($P < 0.01$) effect on fracture force, fracture distance, springiness, chewiness and hardness of the lean chicken meat batters. Fracture force and springiness showed an overall increase when salt level was raised (Fig. 4.7B, Table 4.7). The high salt level (3%) in treatments 7 and 8, helped to extract more proteins, which improved the binding of meat particles. This agrees with Barbut and Mittal (1989), who indicated that cooked poultry meat batters became more rigid when a higher salt level was used since more proteins were extracted. Treatment 9 showed high numerical fracture force, fracture distance, springiness, cohesiveness, chewiness and hardness (Fig. 4.7B, Table 4.7). This is because STPP helped to enhance further solubilization of myofibrillar proteins, which during heating, increased binding among meat particles. Addition of salt and/or polyphosphates to meat products has been reported to improve water-binding ability (Hellendoorn, 1962; Schults and Wierbicki, 1973). Theno *et al.* (1978) reported that sectioned and formed hams showed good binding characteristics, with a high degree of alignment, when ≥ 2 % salt and phosphate were added. The main mechanisms associated with phosphate activity in meat are believed to be: increasing ionic strength, acting as polyanion and shifting the pH from its isoelectric range (Sofos, 1986). However, cohesiveness and chewiness of the cooked chicken batter with 2% salt and 0.5% STPP (Trt. 9) were reduced when 0.2% CMG was added into the batter (Trt. 10, Table 4.7). This might be explained by a possible interference with the binding caused by the gum, without affecting meat batter

stability as reflected by no significant difference in cook loss. Cooking loss and water-holding capacity are important indicators of the strength of the protein gel matrix, as well as the eating quality of the final product (Foegeding and Ramsey, 1986). The high molecular weight of CMG (3.3×10^6 Daltons) probably obstructed the formation of the protein matrix in the chicken batter. Foegeding and Ramsey (1986) also reported that increasing xanthan gum (anionic gum) concentration, decreased beef meat batter hardness without affecting batter stability. Later, Montero *et al.* (2000) found that the presence of xanthan gum produced a decrease in gel forming capacity of the myofibrillar proteins from fish (blue whiting), which was reflected by poorer resistance to their folding test and lower values of breaking force, penetration work and hardness. The authors further explained that the high molecular weight of xanthan gum might have hindered protein network formation and resulted in large cavities in the matrix.

Table 4.7 Textural parameters of the corresponding lean meat batters prepared with different levels of sodium chloride, sodium tripolyphosphate (STPP) and crude malva nut gum (CMG).

	Treatment									
	1	2	3	4	5	6	7	8	9	10
Salt (%)	0	0	1	1	2	2	3	3	2	2
STPP (%)	0	0	0	0	0	0	0	0	0.5	0.5
CMG (%)	0	0.2	0	0.2	0	0.2	0	0.2	0	0.2
Textural Parameter										
Springiness (mm)	0.28^{f}	0.39^{ef}	0.34^{f}	0.46^{de}	0.48^{de}	0.50^{cde}	0.59^{bc}	0.56^{bcd}	0.71^{a}	0.62^{ab}
Fracture distance (mm)	3.93^{cde}	3.41^{ef}	3.08^{f}	4.76^{ab}	3.57^{def}	4.00^{bcde}	4.32^{bcd}	4.58^{abc}	5.37^{a}	4.76^{ab}
Cohesivenes	0.26^{b}	0.26^{b}	0.30^{b}	0.29^{b}	0.29^{b}	0.29^{b}	0.33^{b}	0.32^{b}	0.57^{a}	0.32^{b}
Chewiness (N.mm)	2.96^{de}	3.03^{de}	2.57^{e}	4.34^{cde}	5.91^{c}	5.14^{cd}	10.41^{b}	8.84^{b}	13.87^{a}	9.77^{b}
Hardness (N)	40.68^{cde}	29.77^{e}	30.16^{e}	32.89^{e}	44.08^{bcd}	36.68^{de}	52.24^{ab}	49.88^{abc}	55.88^{a}	48.25^{abc}

$^{a-f}$ Means followed by different letters within the same row are highly significant different ($P < 0.01$).

4.3.1.3 Light Microscopy of Lean Chicken Meat Batters

The micrographs (Fig. 4.8) show differences among the treatments. Overall, the no salt treatment (A) showed a much more open structure comparing to the 2% salt treatment (B). This is because salt extracted more proteins and resulted in a denser protein matrix formation. Increasing NaCl from 1% to 2% improved the stability of the meat batters, as was evident by the lower cook loss (Fig. 4.7A). Trout and Schmidt (1986a) indicated that the ionic environment of a whole muscle system affected its water binding ability. The addition of salt to a meat batter influences the amount of protein extracted (Gordon and Barbut, 1992a) and hence the structure, density and stability of the meat product. Structural changes are effected by alterations in the electrostatic and hydrophobic interactions among proteins (Trout and Schmidt, 1986a and b) or by effects dependent on the specific properties of the ions involved (Asghar *et al*., 1985). The 0.2% CMG without salt treatment (C) showed a fairly similar structure to the no salt treatment but with a residual malva nut gum particle. It is possible that the gum interacted with the meat protein matrix; e.g., occupying some of the interstitial spaces within the matrix structure and resulted in increasing gel strength of the batter as reflected by various textural parameters (Table 4.7). The treatment with 2% salt and 0.2% CMG (Trt. 6; D) showed a consolidation of the protein matrix as well as residual CMG particles in between some muscle fibers (see the arrow in Fig. 4.8D). This resultant microstructure is associated with both the gum present and the more salt-extracted myofibrillar proteins which resulted in more close structure and within the system.

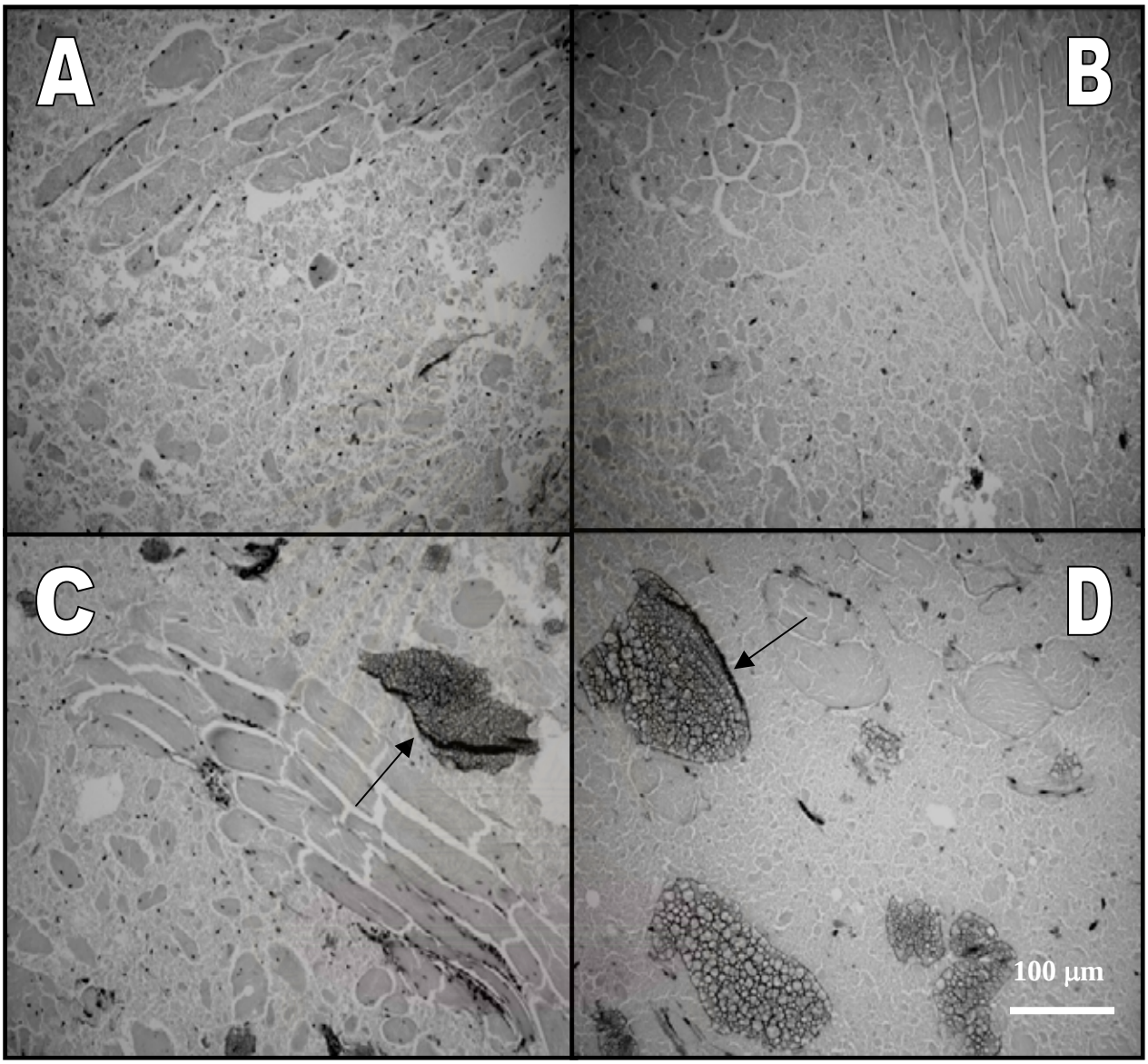

Fig. 4.8 Light micrographs of lean chicken meat batter: (A) with no salt (Trt. 1), (B) with 2 % salt (Trt. 5), (C) with 0.2 % crude malva nut gum (CMG) (Trt. 2), and (D) with 2 % salt and 0.2 % CMG (Trt. 6), Arrowheads are pointed to CMG particles. Bar = 100 μ.

4.3.2 Effect of PMG, Sodium Chloride and Phosphate on Cook Loss and Textural Properties of Lean Chicken Meat Batters

4.3.2.1 Cook Loss from Lean Chicken Meat Batters

The statistical analysis (Table 2, Appendix C) showed that treatment (varying amounts of functional ingredients) had a highly significant ($P < 0.01$) effect on cook loss of the chicken batters. Increasing the salt level reduced ($P < 0.01$) cook loss (Fig. 4.9A). More salt content increases protein extraction and water-binding properties of the chicken batters as found and explained in 4.3.1.1. In this experiment, cook loss decreased ($P < 0.01$) from 18.54% (Trt. 5) to 14.60% (Trt. 9) when phosphate was added. STPP addition further assisted in extracting proteins which during heating increased binding among meat particles. Addition of salt and/or polyphosphates to meat products increases water-binding ability (Hellendoorn, 1962; Schults and Wierbicki, 1973). Several researchers have reported that some phosphates, in the presence of salt, have a synergistic effect to increase water holding capacity of meat proteins (Schnell *et al*., 1970; Pepper and Schmit, 1975; McMahon and Dawson, 1976; Neer and Mandigo, 1977). Keeton (1983) reported that a combination of 1% NaCl and 0.25% phosphate reduced cooking losses of the pork patties. This implies that higher amounts of salt soluble meat proteins were also extracted in this study as salt level was increased and later involved in network formation. The addition of PMG further decreased cook loss (Trt. 2, 4, 6 and 8, Fig. 4.9A). This is basically due to PMG particles bind water during the thermal processing of meat batters and gel on cooling. Treatment 10 showed the low cooking loss (8.51%, Fig. 4.9A) because in addition to the 2% salt, 0.5% STPP and 0.1%

PMG were also added. Trius and Sebranek (1996) also reported that addition of 0.5% STPP to pork sausages formulated with or without carrageenan (κ, i, and λ) reduced cooking loss. This result agrees with a previous study by Somboonpanyakul *et al*. (2004a) who reported that a chicken meat batter that contained 2% salt, 0.5% STPP and 0.2% CMG showed the low cooking loss. However, the cook loss of the chicken meat batter with 2% salt, 0.5% STPP and 0.1% PMG (8.51%, Trt. 10) was not significantly different from the one with 3% salt and 0.1% PMG (9.75%, Trt. 8). This is probably because the same water-binding properties possibly being lessened by salt reduction ,in treatment 10, may be compensated by adding STPP.

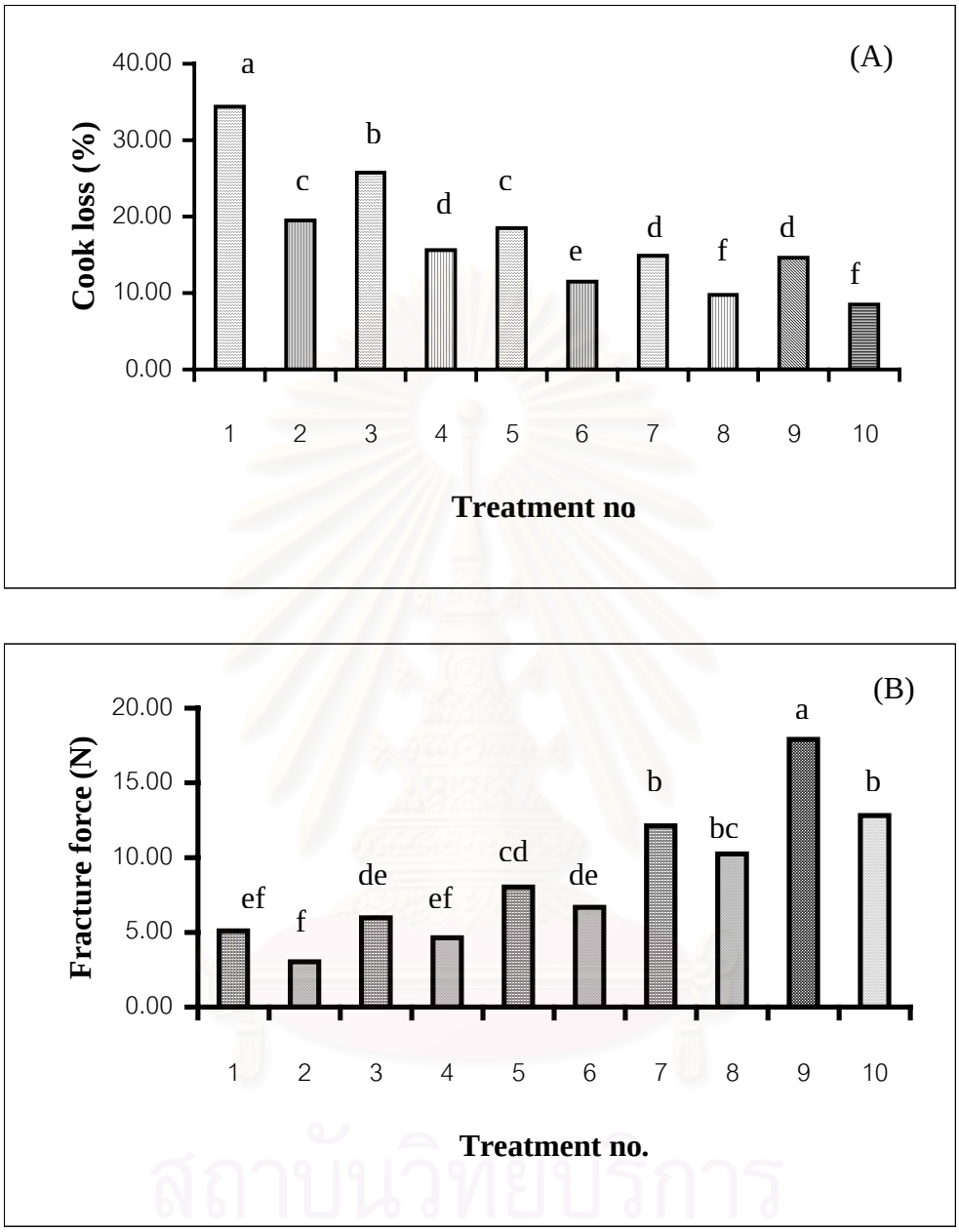

Fig. 4.9 Means for cook loss (A) and fracture force (B) of lean chicken meat batters containing different levels of salt, sodium tripolyphosphate (STPP) and purified malva nut gum (PMG). Bars with different superscripts are different ($P < 0.01$). Treatment numbers are: 1 = no salt, 2 = 0.1% PMG, 3 = 1% salt, 4 = 1% salt and 0.1% PMG, 5 = 2% salt, 6 = 2% salt and 0.1% PMG, 7 = 3% salt, 8 = 3% salt and 0.1% PMG, 9 = 2% salt and 0.5% STPP and 10 = 2% salt, 0.5% STPP and 0.1% PMG.

4.3.2.2 Textural Properties of Lean Chicken Meat Batters

The statistical analysis showed that treatment had highly significant ($P < 0.01$) effects on fracture force, fracture distance, springiness, cohesiveness, chewiness and hardness (Table 2, Appendix C). The treatment with 2% salt and 0.5% STPP (Trt. 9) showed the highest fracture force (17.92 N, Fig. 4.9B) because it also contained STPP which helped to extract more proteins and improved binding among meat particles. Vadehra and Baker (1970) concluded that the binding between chunks of meat is a phenomenon involving structural rearrangement of the solubilized meat proteins. However, when 0.1% PMG was added into the batter with 2% salt and 0.5% STPP (Trt. 10, Fig. 4.9B), significant reductions in fracture force, fracture distance, springiness and chewiness were observed. This might be explained by the reduction of binding among meat particles caused by the interfering effect of the gum. The strength of the bonds between myofibrillar proteins should be stronger than those between PMG and proteins. This is because PMG is anionic gum that contains only 6.40% of uronic acid (as galacturonic acid equivalent). The negatively charged carboxyl groups of the PMG are expected to be lower than the highly charged residual amino acids on polypeptide chain of myofibrillar proteins. Also in a system with limited level of water such as in meat batter, water might be retained mainly by the myofibrillar proteins and hence PMG woud have little available water in comparison with aqueous system. In this condition, PMG might aggregate and coil upon itself, thus occupying a large volume which could distort the protein matrix.

Springiness values of the treatments with PMG (Trt. 2, 4, 6 and 8) were lower ($P < 0.01$) than the corresponding treatments without PMG (Trt. 1, 3, 5 and 7, Table 4.8). This might be explained by certain gum interference with the binding of meat particles, without increasing cook loss of the batters. The present study also confirmed Somboonpanyakul *et al.* (2004a) results which showed that cohesiveness and chewiness of lean chicken batter with 0.5% STPP were decreased ($P < 0.01$) when 0.2% CMG was added.

Table 4.8 Textural parameters of the corresponding meat batters prepared with different levels of sodium chloride, sodium tripolyphosphate (STPP) and purified malva nut gum (PMG).

	Treatment									
	1	2	3	4	5	6	7	8	9	10
Salt (%)	0	0	1	1	2	2	3	3	2	2
STPP (%)	0	0	0	0	0	0	0	0	0.5	0.5
PMG (%)	0	0.1	0	0.1	0	0.1	0	0.1	0	0.1
Textural Parameter										
Springiness (mm)	0.34^{e}	0.26^{f}	0.41^{d}	0.36^{e}	0.48^{c}	0.43^{d}	0.61^{b}	0.52^{c}	0.71^{a}	0.61^{b}
Fracture distance (mm)	3.60^{bcd}	3.44^{bcd}	3.34^{d}	3.36^{cd}	3.66^{bcd}	3.54^{bcd}	4.26^{ab}	3.90^{bcd}	4.96^{a}	4.18^{bc}
Cohesiveness	0.27^{de}	0.25^{e}	0.29^{d}	0.28^{d}	0.30^{cd}	0.28^{d}	0.34^{a}	0.32^{bc}	0.36^{a}	0.34^{a}
Chewiness (N.mm)	2.93^{ef}	1.38^{f}	4.13^{cde}	2.60^{ef}	4.88^{cd}	3.50^{de}	8.62^{b}	5.85^{c}	10.63^{a}	8.25^{b}
Hardness (N)	32.00^{bcd}	20.91^{e}	36.19^{abc}	25.98^{de}	34.25^{abcd}	28.39^{cde}	41.10^{a}	35.83^{abc}	41.80^{a}	39.96^{ab}

[a-f] Means followed by different letters within the same row are significantly different ($P < 0.01$).

In conclusion, addition of 0.2% CMG into the lean chicken meat batter with 2% salt and 0.5% STPP resulted in lower cohesiveness and chewiness when compared to the batter with only 2% salt and 0.5% STPP. For the lean chicken meat batter with PMG, addition of 0.1% PMG and 0.5% STPP also resulted in lower fracture force, fracture distance, springiness and chewiness of the batters. This is probably because both crude and purified malva nut gums had similar chemical compositions as shown in 3.3.1.2 and 3.2.1. Thus, both of them could similarly affect some textural properties of the meat batters. The level of PMG (0.1%) that could effectively improve cook yield of the lean meat batters was lower than the CMG (0.2%) due to the lower ash (5.15%) and protein (2.42%) contents of PMG. Moreover, PMG had more portions of carbohydrate (88.86%) and uronic acid (6.40% as galacturonic acid equivalent) to enhance its gelation property than CMG (86.62% carbohydrate, 5.87% ash, 5.34% moisture, 2.17% protein).

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4.4 Effect of Crude Malva Nut Gum (CMG) and Phosphate on Cook Loss, Fat Loss, Color, Textural Properties, and Microstructure of the Mechanically Deboned Chicken Meat (MDCM) Emulsions

4.4.1 Cook Loss from MDCM Emulsions

The statistical analysis (Table 3, Appendix C) revealed that STPP and CMG had a highly significant ($P < 0.01$) overall effect on cook loss of the MDCM emulsions. Figure 4.10A shows the effect of CMG and STPP levels on the cook loss of the MDCM emulsions. The 0.2% or 0.6% CMG emulsions with 0.5% STPP (Trts. 5 and 6) and the emulsion with only 0.5% STPP (Trt. 4) showed the lower cook losses when compared to the emulsion with only CMG (Trts 2 and 3) and the emulsion without CMG and STPP (Trt. 1). This is probably because the addition of STPP increased the pH of the meat emulsions and when pH is high, meat proteins are more highly charged and repel each other, creating larger spaces for water molecules. The mechanism of CMG in improving water holding properties of meat emulsions, is proposed as holding water in the interstitial spaces of the protein gel network rather than true interactions with the proteins in the formation of network. Trius *et al.* (1994) investigated effect of pork meat pH or STPP on performance of carrageenans (κ, i and λ) in low-fat pork sausage model systems. Adding STPP or using high-pH meat (pH > 6.0) affected performance of the meat batter in similar ways. High-pH meat batters or those containing STPP had lower force values when extruded, lower cooking losses, and were firmer when cooked. The addition of λ-carrageenan produced the softest texture. Only the κ- and i-carrageenan

improved water retention of low-pH meat batters. Increasing level of CMG and the addition of STPP enhanced the binding among the meat particles and improved stability of the meat emulsions as accordingly supported by the fat loss results (Table 4.9 and 4.10).

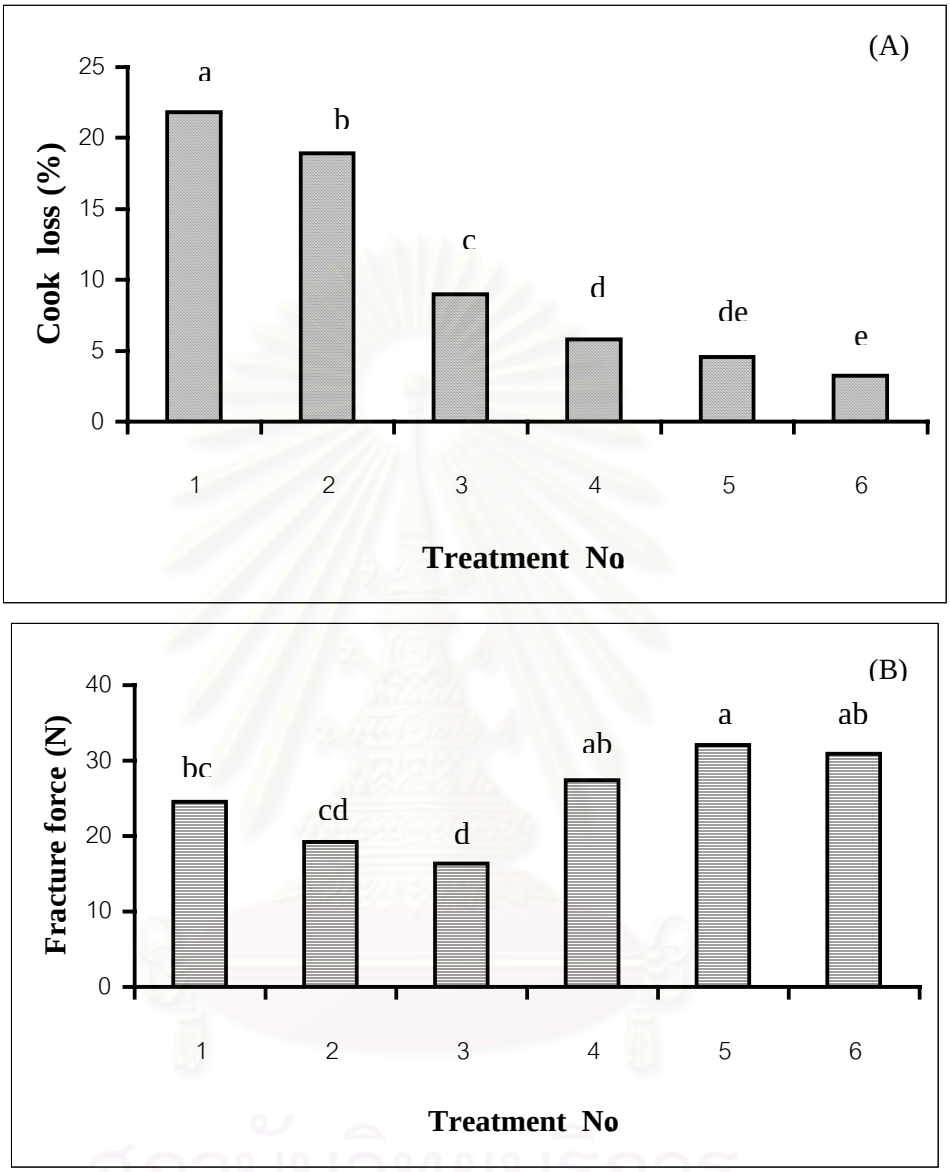

Fig. 4.10 Effect of crude malva nut gum (CMG) level and sodium tripolyphosphate (STPP) (1 = no CMG and STPP, 2 = 0.2% CMG, 3 = 0.6% CMG, 4 = 0.5% STPP, 5 = 0.2% CMG and 0.5% STPP, 6 = 0.6% CMG and 0.5% STPP) on the cook loss (A) and fracture force (B) of mechanically deboned chicken meat emulsions. Bars with different letters are highly significant different ($P < 0.01$).

4.4.2 Fat Loss from MDCM Emulsions

The main factors influencing fat loss from MDCM emulsions were CMG and STPP contents (Table 3, Appendix C). Tables 4.9 and 4.10 show the effect of CMG and STPP levels on the fat loss of cooked MDCM emulsions, respectively. In general, the presence of CMG, provided the MDCM emulsion with lower fat loss. This is probably because CMG is also capable of binding water and forming gel and hence further enhance the stability of the meat emulsions. Schmidt (1984) indicated that fat losses from the meat batters were mutually associated with initial moisture losses during cooking. Hughes *et al*. (1997) also reported that an addition of carrageenan or oat fiber reduced cook loss and increased both water holding capacity and batter stability of pork/beef frankfurters. Addition of STPP decreased ($P < 0.01$) fat losses of the emulsions (Table 4.10). This is basically because STPP was reported to be able to increase the extracted salt soluble proteins from meat and hence provided the meat emulsion with a more stable structure. Whiting (1984) also reported that the fat exudates in reduced salt beef frankfurters (1.5% salt) were prevented by the addition of 0.12% pyrophosphate.

4.4.3 Color of MDCM Emulsions

The main factor influencing lightness (L) and redness (a) of MDCM emulsions was CMG and the main factor influencing redness (a) and yelloness (b) was STPP (Table 3, Appendix C). Table 4.9 shows the effects of CMG levels on color of the cooked MDCM emulsions. Increasing CMG level decreased ($P < 0.01$) L and (a) values

of the MDCM emulsions indicating that the addition of CMG resulted in darker and redder cooked emulsions. This might be due to the proportionally higher amount of the dark-colored CMG in the emulsions. The color difference (ΔE) results also confirmed that the overall color of emulsions with CMG was more pronounced when the CMG level was increased. The MDCM emulsion with 0.6% CMG also had higher hue angle value indicating less red than other treatments. Addition of STPP decreased ($P < 0.05$) hue angle values of the emulsions (Table 4.11), indicating that the MDCM emulsions with STPP were more red than the MDCM emulsions without STPP. This is probably because STPP increases the pH of chicken meat from the isoelectric range of 5.4 (Sofos, 1986), which in turn alters proteins solubility and/or water holding capacity, it may affect the color reflectance of meat pigments in the batter. Lin and Huang (2003) found that reduced-fat (18%) frankfurters incorporated with mixed gels of konjac (1%, 2%) and gellan gum (0.25%, 0.5%) had the highest ($P < 0.05$) lightness (L*), yellowness (b*) values but the lowest redness (a*) and also had the highest hue angle value indicating more yellow (but less red) color. The color (ΔE) difference also confirmed that the overall color of the MDCM emulsions with STPP were different from the MDCM emulsions without STPP.

Table 4.9 Effect of crude malva nut gum (CMG) on the fat loss and color of mechanically deboned chicken meat emulsions.

Treatment		Color				
CMG (%)	Fat loss (%)	L	a	b	ΔE	Hue angle (θ)
0.0	0.14	57.36[a*]	14.76[a*]	8.71[a]	0.00	30.55
0.2	0.10	56.56[b*]	13.74[b*]	8.42[a]	1.33	31.50
0.6	0.09	54.48[c*]	13.03[c*]	8.53[a]	3.36	33.21

[a-b] Means followed by different letters within the same column are significantly different ($P < 0.05$).

[a-c*] Means followed by different letters and * within the same column are highly significant different ($P < 0.01$).

Table 4.10 Effect of sodium tripolyphosphate (STPP) on the fat loss and textural properties of mechanically deboned chicken meat emulsions.

Treatment		Textural properties		
STPP (%)	Fat loss (%)	Springiness (mm)	Cohesiveness	Chewiness (N.mm)
0.0	0.17	0.55^{b*}	0.22^{b}	3.78^{b*}
0.5	0.05	0.69^{a*}	0.26^{a}	6.53^{a*}

$^{a-b}$ Means followed by different letters within the same column are significantly different ($P < 0.05$).

$^{a-b*}$ Means followed by different letters and * within the same column are highly significant different ($P < 0.01$).

Table 4.11 Effect of sodium tripolyphosphate (STPP) on color of mechanically deboned chicken meat emulsions.

Treatment	Color				
STPP (%)	L	a	b	ΔE	Hue angle (θ)
0.00	56.27^{a}	14.16^{a}	9.02^{a*}	0.00	32.50
0.50	55.99^{a}	13.53^{b}	8.09^{b*}	1.16	30.88

$^{a-b}$ Means followed by different letters within the same column are significantly different ($P < 0.05$).

$^{a-b*}$ Means followed by different letters and * within the same column are highly significant different ($P < 0.01$).

4.4.4 Textural Properties of MDCM Emulsions

The statistical analysis (Table 3, Appendix C) revealed that STPP and CMG had a highly significant ($P < 0.01$) overall effect on fracture force of MDCM emulsions. Figure 4.10B shows the effect of CMG and STPP levels on fracture force of cooked MDCM emulsions. High level of CMG tended to decrease fracture force of the product containing CMG (Trt. 3). This might be due to the possibility that the high amount of CMG powder could be more evenly distributed around the emulsion mass and hence hindered the binding among the meat particles and decreased gel strength of the emulsion. Similar findings have been reported by Whiting (1984) who found that xanthan gum (at 0.1 or 0.3%) decreased cooking losses and gel strength of reduced-salt beef frankfurters. Montero *et al.* (2000) also reported that the presence of xanthan gum produced a decrease in the gel forming capacity of the myofibrillar proteins. These researchers explained that the high molecular weight of xanthan gum probably hindered formation of the protein network, thus occupying a large volume which would distort the protein matrix. The samples with 0.2% or 0.6% CMG and 0.5% STPP (Trt. 5 and 6) had higher fracture force when compared to the batters with only 0.2% or 0.6% CMG (Trt. 2 and 3). This is probably because STPP improve water-holding capacity (WHC) of the meat protein. The higher amount of the water in the meat system could enhance the gelation of gum and increase gel strength of the meat emulsion.

The main factor influencing springiness, cohesiveness and chewiness of MDCM emulsions was STPP. Table 4.10 shows that addition of 0.5% STPP increased springiness, cohesiveness and chewiness of the MDCM emulsions. This is basically

because STPP increases the pH of meat from the isoelectric range (pH 5.4) and thus improve water-holding capacity and binding of the emulsion (Hamm, 1970; Hellendoorn, 1962). Trout and Schmidt (1984) also indicated that tetrasodium pyrophosphate and sodium tripolyphosphate were the most effective in increasing water-binding capacity in beef rolls. Moreover, the researchers concluded that pH and ionic strength are probably the most important influences of polyphosphates in improving meat particle-to-particle binding and yield.

4.4.5 Microstructure of MDCM Emulsions

Light microscopy (Fig. 4.11) revealed differences among the treatments. The control emulsion with no CMG and STPP (A) showed a less dense structure. The resulting fat globules were large and angular with a number of small globules interspersed. Gordon and Barbut (1990) observed that more stable batters had fat globules that exhibited several small, uniform pockets of exuding fat, while the unstable batters contained fat globules that showed large exudations at weak points in their protein coats and thus facilitating coalescence. Increasing CMG from 0.2% (B) to 0.6% (C) showed more dense protein matrix and the higher stability of the meat emulsions, as was evident by the lower cook loss (Fig. 4.10A) result. The resulting fat globules of the samples containing only CMG (B and C) were still in angular shapes but smaller and more uniformly distributed in the emulsions when compared to the control emulsion (A). This might be explained by the rationale that CMG was incorporated and dispersed in a discrete form in the matrix without affecting the fat dispersion at a low level (0.2%) and even up to a high level (0.6%). The emulsions with 0.2% or 0.6% CMG and 0.5% STPP

(E and F) showed very dense and organized protein matrix as well as some residual CMG particles distributed in the matrix. Moreover, the emulsion stability was increased, as was evident by less cook loss of the cooked emulsions (Fig. 4.10A). The fat globules were more round and consistently distributed in the protein matrix. This was due to the fact that phosphate addition improved the emulsion stability of the MDCM emulsions. Moreover, CMG particles could hold moisture and also contributed to matrix formation. However, the emulsion with only 0.5% STPP (Trt. 4) showed the most dense protein matrix and the fat globules were spherical in shapes and separated rather evenly within the protein matrix as shown in D. This is in agreement with Schmidt (1984) who investigated the structural differences in the protein matrix of the three meat batters that contained different salt level and STPP. The researcher found that the protein matrix of batter with 0.58% salt had larger aggregates and capillary areas than the batter with only 2.82% salt and the batter with 2.82% salt and 0.37% STPP. Moreover, the higher salt treatments showed smaller capillary size in the matrix structure which resulted in higher water-binding ability as indirectly measured by cooking losses.

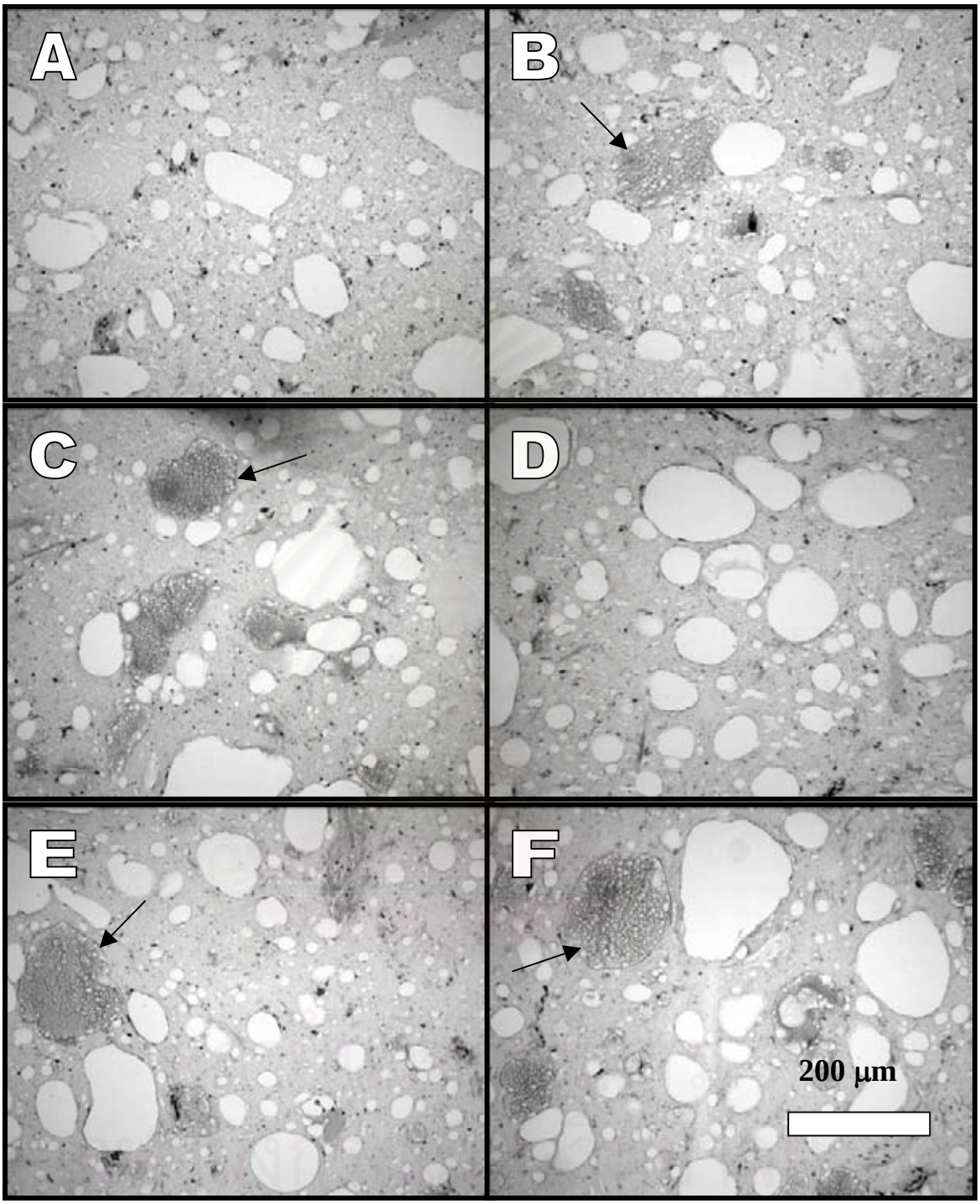

Fig. 4.11 Light micrographs of the mechanically deboned chicken meat emulsion: (A) without sodium tripolyphosphate (STPP) and crude malva nut gum (CMG) (Trt. 1), (B) with 0.2 % CMG (Trt. 2), (C) with 0.6 % CMG (Trt. 3), (D) with 0.5 % STPP (Trt. 4), (E) with 0.5 % STPP and 0.2% CMG (Trt. 5) and (F) with 0.5 % STPP and 0.6 % CMG (Trt. 6). Arrowheads are pointed to CMG particles. Bar = 200μm.

4.5 Effect of Crude Malva Nut Gum (CMG), Purified Malva Nut Gum (PMG) and Phosphate on Cook Loss, Fat Loss, Color, Textural Properties, Rheological Properties and Microstructure of the Chicken Meat Emulsions

4.5.1 Cook Loss from Chicken Meat Emulsions

The statistical analysis (Table 4, Appendix C) revealed that treatment (the contents of functional ingredients used) had a highly significant ($P < 0.01$) effect on cook loss of the chicken meat emulsions. Figure 4.12A shows the effect of PMG level, CMG and STPP on the cook loss of chicken meat emulsions. Cook losses between 13.22% and 0.58% observed for the various treatments showed a trend for decreasing cook loss. The addition of 0.1% PMG, 0.3% PMG or 0.3% CMG significantly decreased cook losses of the control meat emulsions (Trt.1) from 13.22% to 6.66%, 1.70% and 2.98%, respectively. This might have been due to the ability of both PMG and CMG to form gel and retain water of the meat emulsions. Similar results were reported by several researchers, as already cited in 4.3.1.1 and 4.3.2.1. However, when compared at the same level of malva nut gums (0.3%), the chicken emulsion with 0.3% PMG (Trt. 3) had lower cook loss than the one with 0.3% CMG (Trt. 7) as shown in Figure 4.12A. This is probably because PMG had higher proportion of carbohydrate content (88.86% dry basis) than CMG (86.62% dry basis). Thus, PMG was more effective to bind with meat proteins than CMG. In the meat emulsion with STPP, increasing the level of PMG from 0.1% to 0.3% did not help to decrease cook loss of the emulsions. The chicken emulsions with 0.4% STPP and with and without PMG had reduced cooking losses when compared to

some other treatments. STPP which is polyanionic substance could attach to positively charged groups of the meat proteins, while the rest of the chain attracted water molecules and increase water-holding capacity of the emulsions (Steinhauer, 1983). Similar result was reported by Trius *et al.* (1994) who reported that addition of 0.5% STPP to pork sausages formulated with or without carrageenans (κ, i and λ) increased hardness and reduced cooking losses. DeFreitas *et al.* (1997) investigated the effects of κ-, i- or λ-carrageenan, STPP (0 or 0.5%), chloride salts (KCl or NaCl) and meat pH (5.47 or 6.18) on the freeze/thaw stability of cooked pork sausages. The authors found that STPP decreased thaw drip (TD) and increased hardness for all treatments regardless of type of salt or carrageenan. Increased meat pH increased the hardness and decreased thaw drip for all carrageenan treatments, except for λ-carrageenan, which remained unchanged. Kappa-, and iota-carrageenan may be useful for increasing freeze/thaw stability of cooked meat products, particulary those made with low-pH meat such as pale soft and exudative (PSE) pork. These carrageenans improved moisture retention of cooked pork sausage both with and without addition of STPP. Carrageenans and the phosphate salt with NaCl provided the most effective combination for moisture retention in cooked pork sausage. Hsu and Chung (2001) also reported that addition levels of salt, polyphosphates and κ-carrageenan at around 2.7%, 0.17% and 2.0%, respectively, produced low fat emulsified meatballs that had higher cook yields and acceptability.

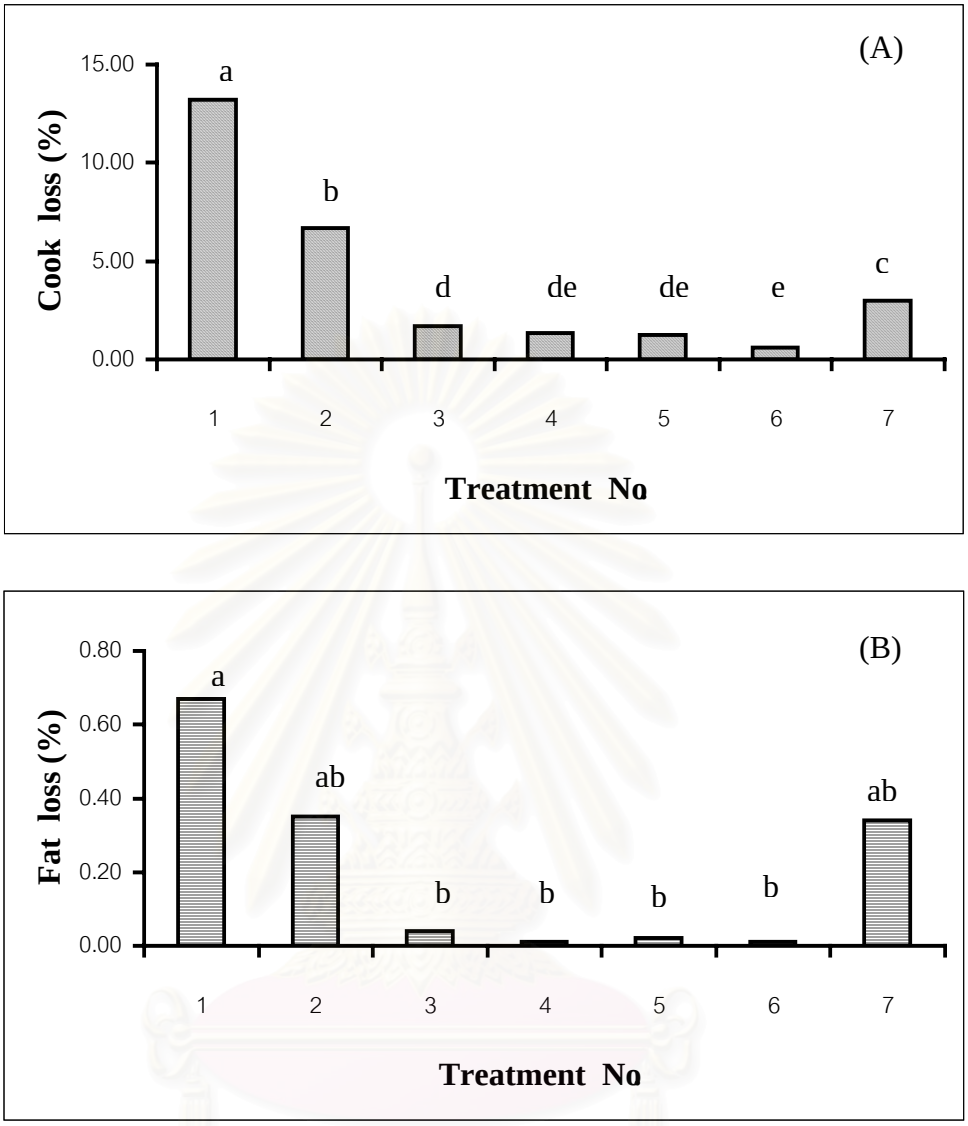

Fig. 4.12 Effect of crude malva nut gum (CMG) level, purified malva nut gum (PMG) and sodium tripolyphosphate (STPP) (1 = no PMG and STPP, 2 = 0.1% PMG, 3 = 0.3% PMG, 4 = 0.4% STPP, 5 = 0.1% PMG and 0.4% STPP, 6 = 0.3% PMG and 0.4% STPP, 7 = 0.3% crude malva nut gum (CMG) on the cook loss (A) and fat loss (B) of chicken meat emulsions.
Bars with different letters are highly significant different ($P < 0.01$).

4.5.2 Fat Loss from Chicken Meat Emulsions

The statistical analysis (Table 4, Appendix C) revealed that treatment (the contents of functional ingredients used) had a highly significant ($P < 0.01$) effect on fat loss of the chicken emulsions. Figure 4.12B shows the effect of PMG level, CMG and STPP on the fat losses of chicken meat emulsions. The chicken emulsions, with 0.1% or 0.3% PMG and 0.4% STPP (Trts. 5 and 6), with 0.3% PMG (Trt. 3) and with only 0.4% STPP (Trt. 4) had lower fat losses when compared to the control emulsion (Trt. 1). These results were supported by the low cook loss results (Fig. 4.12A). This is in agreement with Schmidt (1984) who indicated that the fat losses from a meat batter are always associated with initial moisture losses during cooking. The addition of 0.1% PMG (Trt. 2) or 0.3% CMG (Trt. 7) did not affect fat losses of the batters.

4.5.3 Color of Chicken Meat Emulsions

The statistical analysis (Table 4, Appendix C) revealed that treatment (the contents of functional ingredients used) had highly significant ($P < 0.01$) effects on lightness (L*), redness (a*) and yellowness (b*) of the chicken emulsions. Table 4.12 shows the effect of PMG, CMG and STPP levels on the color values of chicken meat emulsions. The lightness (L*) values of the control emulsion (Trt. 1) and of the with only 0.4% STPP (Trt. 4) were higher than those of the other emulsions. This was expected since treatments without malva nut gums would produce the lighter chicken emulsions. Increasing PMG level from 0.1 to 0.3% reduced L* value of the emulsion (Trt. 2 and

Trt. 3). This is because the higher proportion of the dark-colored PMG would produce the darker emulsion. Redness (a*) and yellowness (b*) values of the control emulsion was increased by the addition of 0.3% PMG (Trt. 3) or 0.3% CMG (Trt. 7). This is probably due to the reddish brown and dark-colored PMG or CMG. The color difference (ΔE*) results confirmed that the overall color was more pronounced when the level of PMG was increased. The emulsions with 0.3% PMG also had lower hue angle values indicating more reddish brown than the emulsions with 0.1% PMG. The emulsions with 0.1% or 0.3% PMG and 0.4% STPP (Trts. 5 and 6) would also produce the darker emulsions comparing to the emulsion with only STPP (Trt. 4). This is also due to the presence of PMG. Moreover, the L* value of the 0.3% PMG emulsion (Trt. 3) was lower than that value of the emulsion with 0.3% CMG (Trt. 7). This can be concluded that the PMG affected the lightness of the emulsions more than CMG. The color (ΔE*) difference also showed that the overall color of the 0.3% PMG emulsion (Trt. 3) was more different from the control emulsion (Trt. 1) than the 0.3% CMG emulsion (Trt. 7). The 0.3% PMG emulsion also had lower hue angle values indicating more reddish brown than the 0.3% CMG emulsion.

4.5.4 Textural Properties of Chicken Meat Emulsions

The statistical analysis (Table 4, Appendix C) revealed that treatment (the contents of functional ingredients used) had highly significant ($P < 0.01$) effects on springiness, cohesiveness, chewiness and hardness of the chicken emulsions. Table 4.12 summarizes the effect of PMG, CMG and STPP levels on the textural

properties of chicken meat emulsions. Increasing PMG level from 0.1% to 0.3% did not affect all of textural parameters of the chicken emulsion (Trt. 3) when compared to the control (Trt. 1). The emulsions with 0.1 or 0.3% PMG and 0.4% STPP (Trts. 5 and 6) showed higher values of hardness, springiness, cohesiveness and chewiness comparing to the emulsions with only 0.1% or 0.3% PMG (Trts. 2 and 3). This was probably because the combined effect between PMG and STPP that increased binding among the meat particles. The ability of phosphates to increase pH, enhance water-holding capacity, induce solubilization of actomyosin and improve texture has been documented (Molins, 1991). However, the emulsion with 0.3% PMG and 0.4% STPP (Trt. 6) showed lower values of hardness, springiness and chewiness when compared to the one with only 0.4% STPP (Trt. 4). This is probably because PMG was easy to aggregate and it was difficult to be homogeneously mixed within the emulsion. The aggregated gum interfered the binding among meat particles thus reducing the numerical values of some textural parameters of the cooked product. Similar findings have been reported by Foegeding and Ramsey (1986) and Mittal and Barbut (1994). The addition of 0.3% PMG (Trt. 3) or 0.3 % CMG (Trt. 7) resulted in the emulsions with no differences in textural profiles when compared to the control (Trt. 1). This can be concluded that the addition of 0.3% PMG or 0.3% CMG did not improved textural properties of the chicken emulsions without STPP.

Table 4.12 Textural properties and color (L*, a*, b*) of the chicken meat emulsions prepared with different levels of purified malva nut gum (PMG) and sodium tripolyphosphate (STPP).

	Treatment						
	1	2	3	4	5	6	7
STPP (%)	0.0	0.0	0.0	0.4	0.4	0.4	0.0
PMG (%)	0.0	0.1	0.3	0.0	0.1	0.3	0.0
CMG (%)	0.0	0.0	0.0	0.0	0.0	0.0	0.3
Textural Parameter							
Hardness (N)	18.54^{bc}	17.92^{bc}	15.80^{c}	34.51^{a}	35.52^{a}	24.94^{b}	15.40^{c}
Springiness (mm)	0.37^{c}	0.39^{c}	0.35^{c}	0.76^{a}	0.68^{ab}	0.67^{b}	0.33^{c}
Cohesiveness	0.16^{b}	0.16^{b}	0.16^{b}	0.27^{a}	0.27^{a}	0.23^{a}	0.17^{b}
Chewiness (N.mm)	1.06^{c}	1.12^{c}	0.93^{c}	7.13^{a}	6.82^{a}	3.83^{b}	0.85^{c}
Color							
L*	80.72^{a}	77.39^{b}	72.45^{c}	80.60^{a}	75.90^{b}	70.28^{d}	76.48^{b}
a*	1.41^{d}	3.79^{c}	5.92^{a}	1.63^{d}	3.44^{c}	5.33^{b}	3.61^{c}
b*	9.84^{bc}	10.46^{b}	11.88^{a}	9.52^{c}	9.48^{c}	10.59^{b}	12.16^{a}
ΔE*	0.00	4.14	9.64	0.41	5.24	11.18	5.31
Hue angle (θ)	81.85	70.08	63.51	80.28	70.06	63.28	73.47

$^{a-c}$ Means followed by a different letter within the same row are highly significant different ($P < 0.01$).

4.5.5 Rheological Properties of Chicken Meat Emulsions

The statistical analysis (Table 5, Appendix C) revealed that treatment (the contents of functional ingredients used) had a significant ($P < 0.05$) effect on G′ at 30 ^{o}C (cooking) and highly significant ($P < 0.01$) effects on G′ at 70 ^{o}C (cooking) and G′ at 30 ^{o}C (cooling). Plots of storage modulus (G′) versus the internal temperature of the chicken emulsions containing different levels of PMG, CMG and STPP and heated from 30 to 70 ^{o}C are shown in Fig. 4.13. Each point on the curve is the average of three trials. During heating, the emulsions with 0.1% or 0.3% PMG and 0.4% STPP (Trts. 5 and 6) and the emulsion with only 0.4% STPP (Trt. 4) showed a slightly increase in G′ upto 55 ^{o}C indicating that a stiff protein matrix was continuously developing. In the temperature range of 58 ^{o}C to 63 ^{o}C, there was a rapid linear increase in G′ exhibiting the maximum G′ values at 70 ^{o}C. The transition at 55 ^{o}C has been attributed to myosin and the one at 63 ^{o}C to collagen and sarcoplasmic proteins (Wright *et al*., 1977). The decreases in G′ at 55 ^{o}C for treatments 1, 2, 3 and 7 and 57 ^{o}C for treatments 4, 5 and 6 were due to the melting of fat (Acton *et al.*, 1982). According to Patana-anake and Foegeding (1985), shear modulus sharply decreased from 20-35 ^{o}C due to the melting of pork fat; and G′ remained relatively constant over the 40-50 ^{o}C range. The major rigidity transition occurred around 54-57 ^{o}C and continued to increase for the remainder of the heating period. Wu *et al.* (1985b) studied transitions occurring during the gelation of meat batter by the thermal scanning rigidity monitor (TSRM) using a constant heating rate of 1 ^{o}C/min. The authors reported three transition temperatures at 38, 46 and 60 ^{o}C.

The first transition was contributed to fat melting and the increased rigidity beyond 46 ^{o}C was related to the formulation of a stable network and a strong gel structure.

From figure 4.13, two patterns of the storage modulus (G′) were observed. The first group with same G′ pattern comprises treatments 4, 5 and 6. Within this group, there was a transition in G′ at 58 ^{o}C. This transition indicated the melting of fat followed by the gelation of the meat proteins at temperature between 58 ^{o}C to 70 ^{o}C (Wright *et al*., 1977). Schweid and Toledo (1981) also reported the transition points in meat batter to occur at 33-36 ^{o}C and 57-67 ^{o}C. These researchers indicated that these temperatures represent the points where insolubilization and solubilization of collagen occur, respectively. Saliba *et al.* (1987) reported that the major modulus of rigidity of beef frankfurter batters started to increase at 58 ^{o}C and the major decrease in energy loss occurred in the 40-60 ^{o}C range. Foegeding and Ramsey (1987) determined the rigidity changes during heating of meat batters containing various carrageenans and xanthan gum. There were slight variations of G′ values from 34 to 58 ^{o}C but major variations in rigidity developed from 58 to 70 ^{o}C due to the functionalities of the gums. The plots of G′ values for the emulsions with 0.1% or 0.3% PMG and 0.4% STPP (Trts. 5 and 6) and batter with only 0.4% STPP (Trt. 4) showed very different profiles to those obtained for the emulsions with only 0.1% or 0.3% PMG (Trts. 2 and 3), the emulsion with 0.3% CMG (Trt. 7) and the control emulsion (Trt. 1). The differences in G′ values on addition of PMG and STPP are probably because STPP increased the extracted myofibrillar proteins content and PMG was increasing the viscosity of the system. Thus, a change in the thermally induced rigidity transition at 57 to 58 ^{o}C indicated that STPP enhanced a PMG-protein interaction and affected protein stability of the emulsions. Foegeding and

Ramsey (1987) reported that beef batters with xanthan gum exhibited decreased rigidity during the entire heating process but the gum did not affect the transition temperature of 58-60 ^{o}C. The authors also explained that meat batters are viscoelastic systems and the viscous component was greatest at the temperatures of < 50 ^{o}C. If the xanthan gum was increasing the viscosity of the system an increase in the initial rigidity was expected and the opposite occurred. A change in thermally induced rigidity transition at 58 to 60 ^{o}C would implicate a xanthan gum-protein interaction affecting protein stability; however, the transition was unaltered. The mechanism by which xanthan gum interferes with rigidity could be related to its surface properties or an unknown role in the protein aggregation process. The emulsion with only 0.4% STPP (Trt. 4) showed the highest G′ value at 70 ^{o}C (8,418 Pa) followed by the emulsion with 0.3% PMG and 0.4% STPP (7,274 Pa, Trt. 6) and the emulsion with 0.1% PMG and 0.4% STPP (7,035 Pa, Trt. 5). These results agree with the texture profile analysis (TPA) in that the emulsions with 0.1 or 0.3% PMG and STPP (Trts. 5 and 6) had higher values of hardness, springiness, cohesiveness and chewiness when compared to the chicken emulsions with only PMG (Trts. 2 and 3, Table 4.12). This also confirmed that STPP was an important factor that involved with the gel strength of the meat emulsions. Foegeding and Ramsey (1987) reported that an addition of i-carrageenan increased water holding capacity, rigidity at 70 ^{o}C, force-to-fracture and true shear strain. Addition of κ-carrageenan increased rigidity at 70 ^{o}C and it was the most effective for increasing hardness.

Another group with the same G′ pattern was composed of treatments 1, 2, 3 and 7. For this group, there was a linear decrease in G′ during 30 to 55 ^{o}C followed by rapid increased up to 56-70 ^{o}C where the maximum value obtained. This indicated that

the gel strength of the protein matrix in these emulsions were lower than those with PMG and STPP (Trts. 4, 5 and 6). The change in the thermally induced rigidity transition at 54 to 55 ^{o}C implicated the malva nut gums and protein interaction affected protein stability. The rapid increase in G′ which started at 58 ^{o}C, indicated the myosin denaturation. The general pattern observed here for the denaturation of the control batter (0.75% salt) is similar to the pattern reported for the white poultry meat batters with 1% salt (Mittal and Barbut, 1989). The lowest G′ was observed in the control emulsion (Trt. 1) and its G′ value (5,792 Pa) was not significantly different from the G′ value of the 0.3% CMG emulsion (5814 Pa, Trt. 7, Table 4.13). It can be concluded that addition of PMG or CMG without STPP might interfere rigidity of the emulsions. These results agree with the TPA results (Table 4.12).

Table 4.13 summarizes the statistics of the G′ values for different treatments. Generally, G′ increases with the increase of temperature. The average values of G′ (at 70 ^{o}C) in increasing order for the various emulsions, were: the control emulsion (5,792 Pa, Trt. 1); the emulsion with 0.3% CMG (5,814 Pa, Trt. 7); the emulsion with 0.1% PMG (6,204 Pa, Trt. 2); the emulsion with 0.1% PMG and 0.4% STPP (7,035 Pa, Trt. 5); the emulsion with 0.3% PMG and 0.4% STPP (7,274 Pa, Trt. 6); the emulsion with 0.3% PMG (7,884 Pa, Trt. 3); and the emulsion with only 0.4% STPP (8,418 Pa, Trt. 4). During the cooling step to 30 ^{o}C, the emulsions with 0.1% PMG and 0.4% STPP (Trt. 5), with only 0.4% STPP (Trt. 4) and with 0.3% PMG and 0.4% STPP (Trt. 6) showed the high G′ values of 18,916 Pa, 18,275 Pa and 17,716 Pa, respectively. These agree with the TPA results (Table 4.12) that the 0.1% PMG and 0.4% STPP emulsion

(Trt. 5) and the 0.4% STPP emulsion (Trt. 4) showed higher values of all textural parameters than the emulsions without STPP (Trts. 1, 2, 3 and 7).

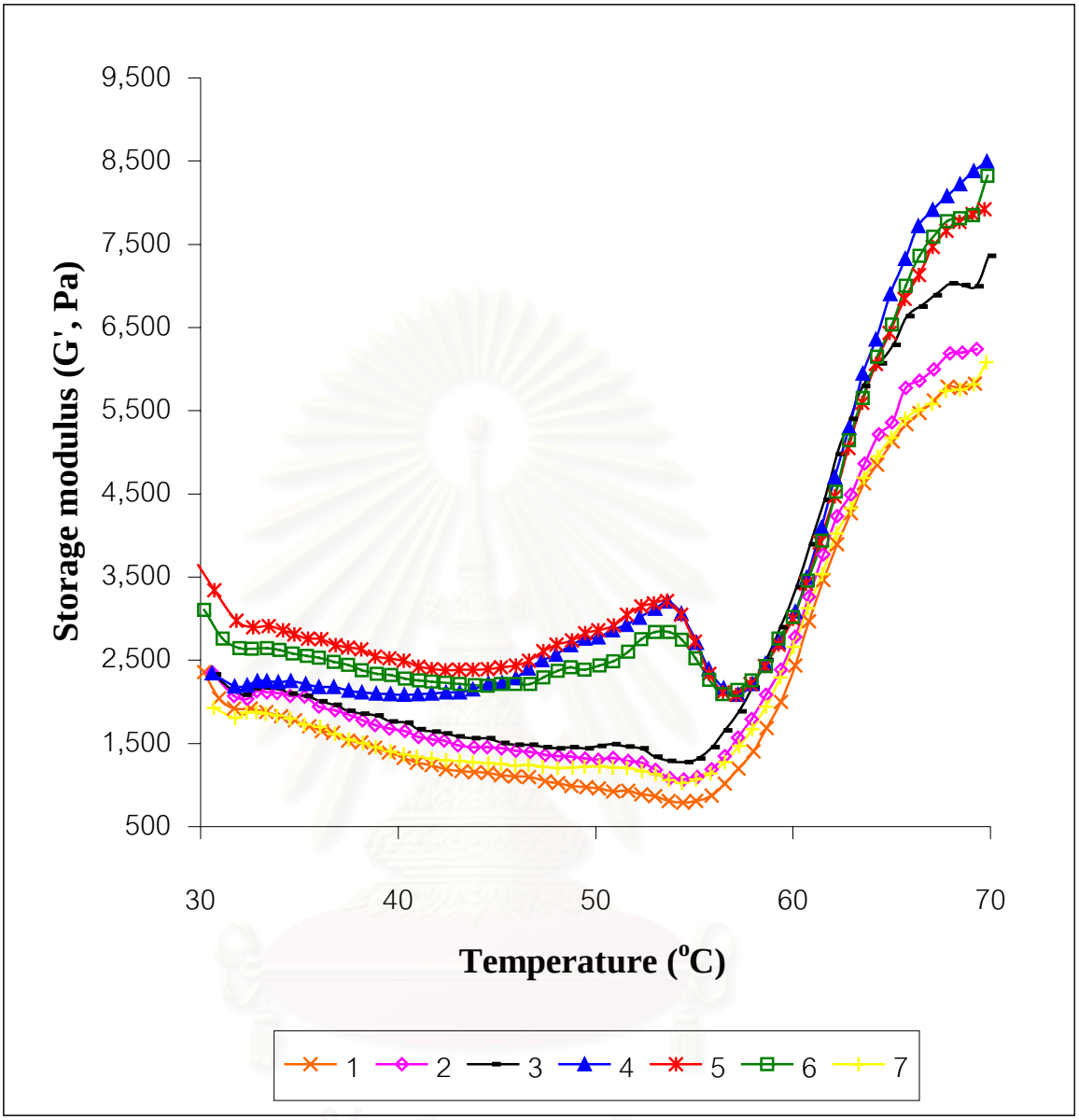

Fig. 4.13 Representative rheograms illustrating the storage moduli (G′) of chicken meat emulsions prepared with different levels of crude malva nut gum (CMG), purified malva nut gum (PMG) and sodium tripolyphosphate (STPP) during cooking (30 to 70 °C). Treatment numbers are: 1 = no STPP and PMG, 2 = 0.1 % PMG, 3 = 0.3 % PMG, 4 = 0.4 % STPP, 5 = 0.4 % STPP and 0.1 % PMG, 6 = 0.4 % STPP and 0.3 % PMG, 7 = 0.3 % CMG.

Table 4.13 Storage moduli (G′) of chicken meat emulsions before cooking (30 ^{o}C), after cooking (70 ^{o}C) and after cooling to 30 ^{o}C. Emulsions were prepared with different levels of crude malva nut gum (CMG), purified malva nut gum (PMG) and sodium tripolyphosphate (STPP).

Treatment				Storage modulus (G′, Pa) (cooking)		Storage modulus (G′, Pa) (cooling)
No.	STPP (%)	PMG (%)	CMG (%)	30 ^{o}C	70 ^{o}C	30 ^{o}C
1	0.0	0.0	0.0	2,200^{bc}	5,792^{d*}	11,716^{c*}
2	0.0	0.1	0.0	2,205^{bc}	6,204^{cd*}	13,775^{bc*}
3	0.0	0.3	0.0	2,268^{bc}	7,884^{ab*}	14,941^{b*}
4	0.4	0.0	0.0	2,933^{ab}	8,418^{a*}	18,275^{a*}
5	0.4	0.1	0.0	2,169^{bc}	7,035^{bc*}	18,916^{a*}
6	0.4	0.3	0.0	3,490^{a}	7,274^{b*}	17,716^{a*}
7	0.0	0.0	0.3	1,880^{c}	5,814^{d*}	12,866^{bc*}

$^{a-c}$ Means followed by different letters within the same column are significantly different ($P < 0.05$).

$^{a-d\ *}$ Means followed by different letters and * within the same column are highly significant different ($P < 0.01$).

4.5.6 Microstructure of Chicken Meat Emulsions

The light micrographs of fat dispersion of chicken emulsions with different levels of PMG, STPP and CMG are shown in Figure 4.14. The control emulsion (A), the emulsion with 0.1% PMG (B) and the emulsion with 0.3% PMG (C) showed less uniform fat dispersion and the fat was partly coalesced into larger fat particles.

The control emulsion (A) and the emulsion with 0.1% PMG (B) showed more coarse protein matrix structure containing larger fat globules when compared with the 0.3% PMG emulsion (C). Moreover, the fat globules of the control emulsion and the 0.1% PMG emulsion were larger and more elongate than the ones of 0.3% PMG emulsion. These were in agreement with the cook and fat losses results (A and B), which indicated that the 0.3% PMG emulsion (Trt. 3) showed lower cook and fat losses than control and 0.1% PMG emulsions. This is probably because the higher level of PMG decreased cook and fat losses and improved stability of the batters. Koolmees *et al*. (1989) also indicated that the predominant way of fat stabilization in the absence of polyphosphate was developed through physical entrapment of the larger fat particles in a coarser matrix.

The reverse results were found in the emulsions with 0.1% or 0.3% PMG and 0.4% STPP (Trts. 5 and 6) and the emulsion with only 0.4% STPP (Trt. 4). All these three batters showed dense protein matrix and the large number of finely distributed small fat particles, especially in the emulsion with only 0.4% STPP (D). Most of fat particles of the emulsions that contained STPP (D, E and F) were more round, smaller and better distributed in the protein matrix when compared to the emulsions without STPP (A, B and C). Moreover, most of fat particles in the emulsions with STPP did not coalesce. This might be explained by the fact that the addition of STPP resulted in a higher density of the protein matrix and stabilized the fat droplets. The similar finding was reported by Koolmees *et al.* (1989) who found that the addition of polyphosphate into beef batters resulted in a dense protein matrix which stabilized numerous small fat droplets, even after extended chopping time and increased fat content. Without polyphosphates, the meat batters became less stable with increased chopping time and fat

content. Furthermore, the PMG particles that distributed in the protein matrix, as the arrowheads pointed in D, E and F, bound some of the water and prevent it from mobilizing which allowed the meat proteins to form a firmer gel structure and improved stability of the batters. Comer and Allan-Wojtas (1988) also reported that the principal functional fillers are polysaccharides, especially starches, and proteins, i.e. ingredients which had the ability to hold moisture and/or fat in a gel structure after the heat treatment used in processing. The fillers added to a comminuted meat system generally increased both stability and textural firmness of the comminuted meat product. Gordon and Barbut (1990) observed that more stable batters had globules that exhibited several small, uniform pockets of exuding fat, while the unstable batters contained globules that showed large exudations at weak points in their protein coats. These large exudations have been shown to be more likely to form fat channels and facilitate coalescence (Gordon and Barbut, 1989).

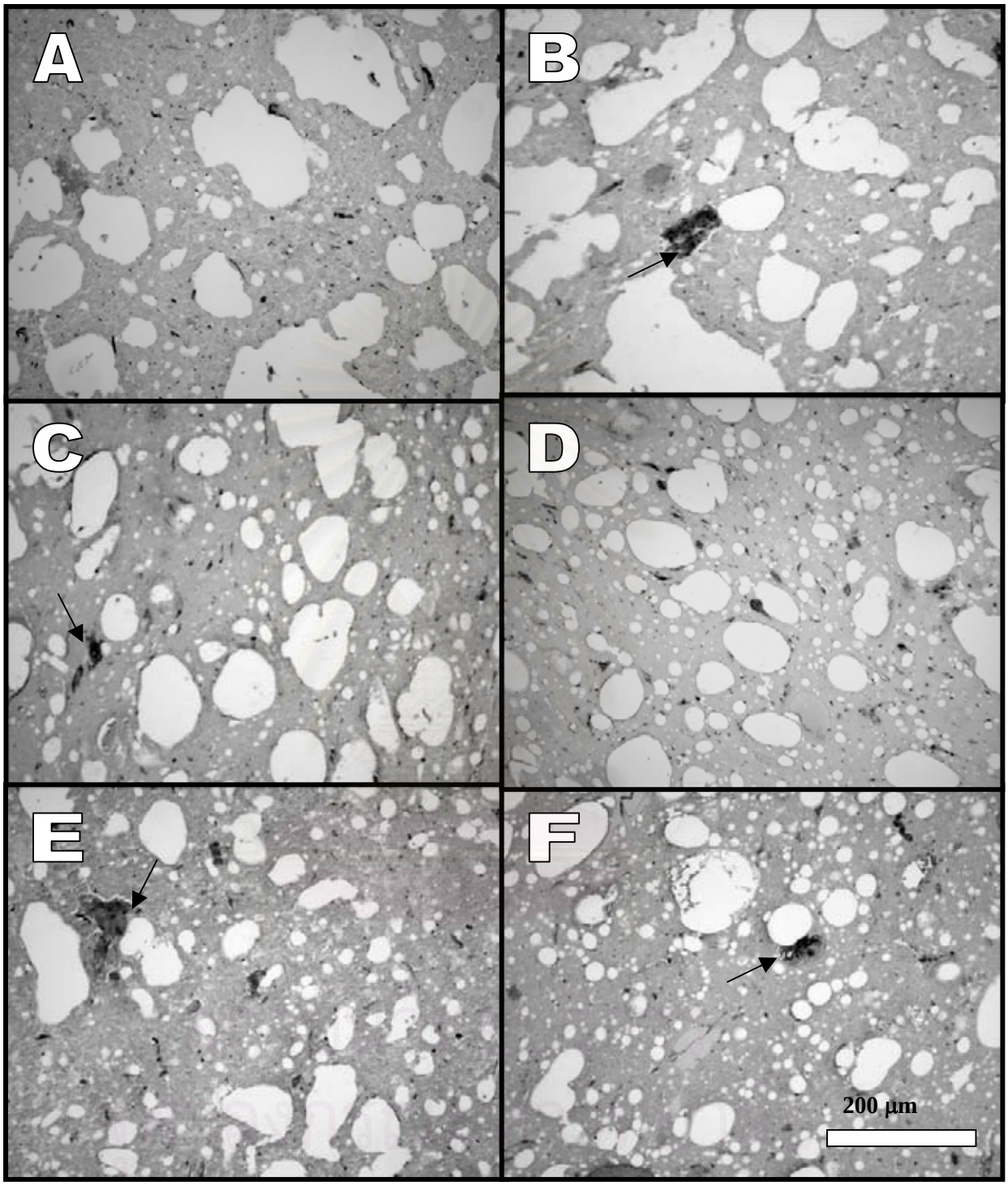

Fig. 4.14 Light micrographs of the chicken emulsion: (A) without sodium tripolyphosphate (STPP) and purified malva nut gum (PMG) (Trt. 1), (B) 0.1% PMG (Trt. 2), (C) 0.3% PMG (Trt. 3), (D) with 0.4% STPP (Trt. 4), (E) with 0.4% STPP and 0.1 % PMG (Trt. 5) and (F) with 0.4% STPP and 0.3% PMG (Trt. 6). Arrowheads are pointed to the PMG particles. Bar = 200 µm.

4.6 Effect of Crude Malva Nut Gum (CMG) Level on Physical and Sensory Properties of Commercial Type Frankfurters.

4.6.1 Cook Loss from Frankfurters

The statistical analysis (Table 6, Appendix C) revealed that treatment (the contents of functional ingredients used) had a highly significant ($P < 0.01$) effect on cook loss of the frankfurters. Cook loss was significantly ($P < 0.01$) affected by CMG level. Treatment 3 showed the lowest cook loss (2.24%, Table 4.14). This seems to be due to the highest level of CMG (0.6%), which enabled more moisture binding. Whiting (1984) concluded that 0.1 to 0.3% xanthan gum could improve water binding in beef frankfurters. Barbut and Mittal (1996) found that moisture loss during cooking of low fat pork/beef frankfurter was reduced from 10 to 6% due to the addition of carboxymethylcellulose. Lurueña-Martínez *et al.* (2004) also reported that addition of locust bean gum/xanthan gum into pork frankfurters significantly increased cooking yield, improved batter stability and decreased jelly and fat separation. This experiment demonstrated that CMG was more effective in retaining moisture in cooked poultry frankfurters than the control frankfurter without CMG.

4.6.2 Textural Properties of Frankfurters

The statistical analysis (Table 6, Appendix C) revealed that treatment (the contents of functional ingredients used) had highly significant ($P < 0.01$) effects on springiness, chewiness and hardness of the frankfurters. Table 4.14 shows the effect of CMG levels on textural properties of the frankfurters. Adding 0.2 and 0.6% of CMG (Trts. 2 and 3) resulted in higher hardness, springiness and chewiness values when compared to the control frankfurters (Trt. 1); however CMG level did not affect cohesiveness. The increases of some textural parameters might be explained by the interactions of CMG with the chopped meat particles and hence the gel strength of the frankfurters. The frankfurters with the highest level of CMG (0.6%) was the hardest sample (22.81 N). This might have been due to the high level of gum that increased the binding among meat particles more than the low CMG treatment. A similar trend was also observed for springiness when the gum level was increased. The increase was mainly because CMG helped improve the gel structure and increased binding within the product. Moreover, chewiness of the frankfurters were also increased with an increase of CMG level. The maximum chewiness values were obtained for the 0.6% CMG product (Trt. 3, Table 4.14) with the values of 5.30 N and 4.28 N.mm, respectively.

Table 4.14 Effect of crude malva nut gum (CMG) level on cook loss and textural properties of commercial type frankfurters.

Treatment			Texture Profile Analysis			
No.	CMG (%)	Cook loss (%)	Hardness (N)	Springiness (mm)	Cohesiveness	Chewiness (N.mm)
1	0.0	2.92[a]	16.37[c]	0.53[b]	0.24[a]	2.79[b]
2	0.2	2.55[b]	19.28[b]	0.80[a]	0.24[a]	3.67[a]
3	0.6	2.24[c]	22.81[a]	0.81[a]	0.23[a]	4.28[a]

[a-c] Means followed by different letters within the same column are significantly different ($P < 0.01$).

4.6.3 Color of Frankfurters

The statistical analysis (Table 6, Appendix C) revealed that treatment (the contents of functional ingredients used) had highly significant ($P < 0.01$) effects on lightness (L) and redness (a) of the frankfurters. The addition of CMG reduced ($P < 0.01$) L and a values of the frankfurters (Table 4.15). This could be explained by the higher amount of the reddish brown and dark-colored CMG in the frankfurters. The color difference (ΔE) results also confirmed that the overall color was more pronounced when the level of CMG was increased. The frankfurters with 0.6% CMG also had higher hue angle value indicating less red than other treatments.

Mittal and Barbut (1993) also found that the lightness values of low-fat pork breakfast sausages were reduced due to the addition of carboxymethylcellulose and microcrystalline cellulose. No effect of CMG was noticed on yellowness (b) value of the frankfurters.

Table 4.15 Effect of crude malva nut gum (CMG) level on color of commercial type frankfurters.

Treatment		Color				
No.	CMG (%)	L	a	b	ΔE	Hue angle (θ)
1	0.00	62.12[a]	16.91[a]	12.78[a]	0.00	37.08
2	0.20	60.70[b]	15.96[b]	12.68[a]	1.71	38.47
3	0.60	58.69[c]	15.39[c]	12.81[a]	3.75	39.77

[a-c] Means followed by different letters within the same column are significantly different ($P < 0.01$).

4.6.4 Sensory Properties of Frankfurters

The statistical analysis (Table 7, Appendix C) revealed that treatment (the contents of functional ingredients used) had highly significant ($P < 0.01$) effects on sensory color, firmness, elasticity, juiciness and overall acceptability of the frankfurters. Table 4.16 shows the effect of CMG level on sensory properties of the frankfurters. The panel's scores for color revealed major differences in the perceived color of the frankfurters, being in agreement with the instrumental color evaluation (Table 4.15). Overall, the frankfurters without CMG were evaluated by the panelists as more pink than the ones with 0.6% CMG. This agrees with the objective color measurement that the sample without CMG had higher L and a values than the one with 0.6% CMG (Table 4.15). The panelists found that firmness and elasticity significantly increased by increasing the CMG content. This also agrees with the objective texture analysis that showed higher hardness, springiness and chewiness values for the sample added with CMG. The addition of CMG resulted in the products perceived to be less juicy than the control (without CMG). The CMG added frankfurters (Trts. 2 and 3) were perceived to be more firm and elastic than the control (Trt. 1). No effect of CMG on the flavor ($P > 0.01$) of frankfurters with 0.2 and 0.6% CMG content (Trts. 2 and 3) was detected. Moreover, the frankfurters with 0.2% CMG had the higher flavor score (8.41, Table 4.16) than the control (without CMG). The overall acceptability results indicated that the panelists preferred the frankfurters with 0.2% CMG (Trt. 2). This could have been due to the more firm and elastic product as well as the color of this product which was not too

dark when compared to the frankfurters with 0.6% CMG (Trt. 3). In summary, the 0.2% CMG frankfurters (Trt. 2) can be produced with a high degree of consumer acceptance.

Table 4.16 Effect of crude malva nut gum (CMG) level on sensory properties of commercial type frankfurters.

Treatment		Sensory Attributes*					
No.	CMG (%)	Color	Firmness	Elasticity	Juiciness	Flavor	Overall acceptability
1	0.0	8.27[a]	6.74[b]	7.25[b]	8.43[a]	7.86[b]	7.69[b]
2	0.2	7.68[b]	7.71[a]	8.01[a]	7.95[b]	8.41[a]	8.26[a]
3	0.6	6.65[c]	8.19[a]	8.28[a]	7.63[b]	7.97[ab]	7.68[b]

* Scales: Color (1 = brown, 10 = brownish pink), texture (1 = very soft, 10 = very firm), elasticity (1 = low elasticity, 10 = high elasticity), juiciness (1 = very dry, 10 = very juicy), flavor (1 = unpleasant, 10 = extremely pleasant), (1 = very dry, 10 = very juicy) and overall acceptability (1 = unacceptable, 10 = extremely acceptable).

[a-c] Means followed by different letters within the same column are significantly different ($P < 0.01$).

CHAPTER V

CONCLUSIONS

5.1 Extraction and Physicochemical Characterization of Malva Nut Gum

- The sequential extraction procedure indicated that malva nut polysaccharides could not be solubilized effectively by hot water nor by a diluted acid solution.
- Extraction malva nut fruits with 0.05 M NaOH provided the highest yield of 20% dried malva nut gum.
- The alkaline extracted malva nut gum contained 83.10% carbohydrate, 8.30% protein and 8.40% ash.
- The level of 6.40% uronic acid (as galacturonic acid equivalent) in alkaline extracted malva nut gum reflects the relative amount of acidic polysaccharides in the gum.
- The weight average molecular weight of the alkaline extracted malva nut gum was 6.65×10^6 Daltons before dialysis. After dialysis, it was reduced to 3.30×10^6 Daltons.
- The weight average molecular weight of the alkaline extracted malva nut gum was 6.65×10^6 Daltons before decoloring. After decoloring, it was also reduced to 2.27×10^6 Daltons.

- Methylation analysis of PMAA, derived from the alkaline extracted malva nut gum revealed that it is primarily composed of 1,4-D-Gal*p*, 1,3-linked-L-Ara*f* and 1,2 and 1,3-linked Rham*p* in the molar ratios of 0.21 : 1.00 : 1.24.
- The FT-IR spectrum of alkaline extracted malva nut gum showed that the alkaline extraction caused saponification of the methyl esterified carboxyl group. In contrast, the spectra of water and acid extracted malva nut gums exhibited characteristic absorbances of low DE pectins. It was observed that the total carbonyl absorption band area increased as the polygalacturonic acid content increased. All spectra showed carboxylate ion peaks at 1,600 and 1,414 cm^{-1} and carbohydrate peaks at 1,140, 1,100, 1,060 and 990 cm^{-1}. The carbohydrate peaks of the alkaline extracted malva nut gum were similar to gum arabic because they had a similar monosaccharides composition.

5.2 Rheological Properties of the Purified Malva Nut Gum (PMG)

- Both high molecular weight nature and intermolecular interactions contribute to the properties of high viscosity solution and strong shear thinning flow behavior of PMG solution.
- The Newtonian plateaus of PMG solutions were observed at concentrations of 0.1% and 0.3%.
- The dilute PMG solution of 0.1%, which is lower than the overlap concentration, exhibited appreciable shear thinning behavior and the shear rate of onset shear thinning was very low (approximately 0.3 s^{-1}).
- The zero-shear-rate viscosity and the $\dot{\gamma}$ value of 0.3% PMG was higher than at 0.1% gum.
- As PMG concentration increased, The consistency index (*K)* increased and flow index (*n)* decreased. The increase in *K* of PMG solutions with increasing concentration suggests that a more viscous system is obtained at higher concentions. Moreover, the value of n decreased with concentration more rapidly at concentrations lower than 1%, then gradually leveled off to a value of approximately 0.25.
- The PMG solutions showed $G' > G''$ over the entire frequency range examined. However, both G' and G'' showed appreciable frequency dependence, especially for the 0.5% PMG dispersions. The frequency dependency became less evident when the polysaccharide concentration was

increased. Complex viscosity (η*), related to the viscoelastic response of the PMG solutions, also decreased as a function of frequency.

- The addition of 5 to 15 mM calcium to 1.5% PMG solutions did not change the values of G′ and G″. The PMG gels with 5 to 15 mM calcium did not show a melting point within the temperature range studied. Futhermore, G′ and G″ slightly decreased with increasing temperature; however, there was no apparent difference between all PMG solutions with different Ca^{2+} concentration.
- The weak gel structure of PMG solutions were more likely attributed to the formation of entangled macromolecules networks.

5.3 Effect of Crude Malva Nut Gum (CMG), Purified Malva Nut Gum (PMG), Sodium Chloride and Phosphate on Cook Loss, Textural Properties and Microstructure of Lean Chicken Meat Batters.

5.3.1 Effect of CMG, Sodium Chloride and Phosphate on Cook Loss, Textural Properties and Microstructure of Lean Chicken Meat Batters

- There was an overall decrease in cook loss of lean chicken meat batters as salt level was increased from 0 to 3%.
- The combination of 2% salt, 0.5% STPP and 0.2% CMG showed the low cook loss of lean chicken batter (9.97%).
- Fracture force and springiness showed an overall increase when salt level was raised from 0 to 3%.
- The combination of 2% salt and 0.5% STPP showed the high numerical fracture force, fracture distance, springiness, cohesiveness, chewiness and hardness. However, cohesiveness and chewiness of the chicken batter with 2% salt and 0.5% STPP was reduced ($p < 0.01$) when 0.2% CMG was added into the batter.
- The micrographs of lean chicken batters show differences among the treatments. Overall, the no salt treatment showed a much more open structure compared to the 2% salt treatment. Increasing NaCl from 1% to

2% improved the stability of the meat batters, as was evident by the lower cook loss.

- The microstructure of the chicken batter with no salt and 0.2% CMG showed a fairly similar structure to the batter with no salt but with a residual CMG particle.
- The microstructure of the chicken batter with 2% salt and 0.2% CMG showed a consolidation of the protein matrix as well as residual CMG particles in between some muscle fibers.

5.3.1 Effect of PMG, Sodium Chloride and Phosphate on Cook Loss and Textural Properties of Lean Chicken Meat Batters.

- Increasing the salt level reduced ($p < 0.01$) cook loss of the chicken batters. Cook loss of the batters with 2% salt decreased ($p < 0.01$) from 18.54% to 14.60% when STPP was added. The addition of PMG further decreased cook loss of chicken batters. Thus, the treatment with 2% salt, 0.5% STPP and 0.1% PMG showed low cook loss (8.51%).
- The chicken batters with 2% salt and 0.5% STPP showed the highest fracture force (17.92 N). The fracture force, fracture distance, springiness and chewiness values of the chicken batter with 2% salt and 0.5% STPP were higher than the chicken batter with 2% salt, 0.5% STPP and 0.1% PMG.

5.4 Effect of Crude Malva Nut Gum (CMG) and Phosphate on Cook Loss, Fat Loss, Color, Textural Properties and Microstructure of the Mechanically Deboned Chicken Meat (MDCM) Emulsions.

- STPP and CMG had a highly significant ($p < 0.01$) overall effect on cook loss of the MDCM emulsions. The 0.2% or 0.6% CMG emulsions with 0.5% STPP and the emulsion with only 0.5% STPP showed the lower ($p < 0.01$) cook losses when compared to the emulsions with only CMG and the emulsion without CMG and STPP.
- Fat loss was affected by CMG or STPP levels. The higher level of CMG showed an overall decreased fat loss of the MDCM emulsions. Addition of STPP decreased ($p < 0.01$) fat losses of the emulsions.
- Increasing CMG level decreased ($p < 0.01$) L and a values but increased color difference (ΔE) and hue angle values of the MDCM emulsions. The addition of CMG resulted in darker and redder cooked emulsions.
- High level of CMG showed an overall decrease fracture force of MDCM emulsions with CMG. In the 0.2% or 0.6% CMG emulsions with 0.5% STPP addition resulted in higher fracture force when compared to the emulsion with only 0.2% or 0.6% CMG. STPP also increased the values of springiness, cohesiveness and chewiness of the emulsions.
- Light microscopy revealed differences among the treatments of MDCM emulsions. The control emulsion with no CMG and STPP showed a less dense structure when compared to the other treaments. The resulting fat

globules were large and angular with a number of small globules interspersed.

- Increasing CMG from 0.2% to 0.6% showed more dense protein matrix The resulting fat globules were still angular shape but smaller and more uniformly distributed in the emulsions than the control emulsion.

- The emulsions with 0.2% and 0.6% CMG and 0.5% STPP showed very dense and organized protein matrix as well as some residual CMG particles distributed in the matrix. Moreover, the fat globules were more round and consistently distributed in the protein matrix. However, the emulsion with only 0.5% STPP showed the most dense protein matrix and the fat globules were spherical shape and separated within the protein matrix.

5.5 Effect of Crude Malva Nut Gum (CMG), Purified Malva Nut Gum (PMG) and Phosphate on Cook loss, Fat Loss, Color, Textural Properties, Rheological Properties and Microstructure of the Chicken Meat Emulsions.

- The addition of 0.1% PMG, 0.3% PMG or 0.3% CMG decreased ($p < 0.01$) cook losses of the control meat emulsions.
- When compared at the same level of malva nut gums (0.3%), the chicken emulsion with 0.3% PMG had lower cook loss than the chicken emulsion with 0.3% CMG.
- The chicken emulsions with 0.4% STPP and with and without PMG had reduced cooking losses when compared to some other treatments.
- The chicken emulsions with 0.1% PMG or 0.3% and 0.4% STPP, the chicken emulsion with 0.3% PMG and the chicken emulsion with only 0.4% STPP had lower fat losses when compared to the control chicken emulsion. In contrast, the chicken emulsions with only 0.1% PMG or 0.3% CMG did not affect fat losses of the chicken emulsions.
- Increasing PMG level from 0.1 to 0.3% also reduced ($p < 0.01$) L* and hue angle values but increased color difference (ΔE^*) of the emulsions.
- Redness (a*) and yellowness (b*) values of the control chicken emulsion was increased ($p < 0.01$) by the addition of 0.3% PMG or 0.3% CMG.

- The emulsions with PMG and STPP would produce the darker emulsions compared to the emulsion with only STPP. Moreover, the L* and a* values of the 0.3% PMG emulsion was lower ($p < 0.05$) than the L* and a* values of emulsion with 0.3% CMG. Thus, PMG affected the color of the emulsions more than CMG.

- Increasing PMG level from 0.1% to 0.3% did not affect all of textural parameters of the chicken emulsion. However, the emulsions with 0.1% or 0.3% PMG and 0.4% STPP showed higher values of hardness, springiness, cohesiveness and chewiness when compared to the emulsions with only PMG.

- The emulsion with 0.3% PMG and 0.4% STPP showed lower values of hardness, springiness and chewiness when compared to the emulsion with only 0.4% STPP.

- The addition of 0.3% PMG or 0.3% CMG did not improved textural properties of the chicken emulsions without STPP.

- The average values of G′ (at 70 ^{o}C) in increasing order for the various chicken emulsions, were: the control chicken emulsion (5,792 Pa); the chicken emulsion with 0.3% CMG (5,814 Pa); the chicken emulsion with 0.1% PMG (6,204 Pa); the chicken emulsion with 0.1% PMG and 0.4% STPP (7,035 Pa); the chicken emulsion with 0.3% PMG and 0.4% STPP (7,224 Pa); the chicken emulsion with 0.3% PMG (7,884 Pa); and the chicken emulsion with only 0.4% STPP (8,418 Pa).

- At the cooling step to 30 ^{o}C, the chicken emulsions with 0.1% PMG and 0.4% STPP, the chicken emulsion with 0.4% STPP and the chicken emulsion with 0.3% PMG and 0.4% STPP showed the high G′ values of 18,916 Pa, 18,275 Pa and 17,716 Pa, respectively.
- The light micrographs of the control chicken emulsion and the chicken emulsions with 0.1% or 0.3% PMG showed less uniform fat dispersion and the fat was partly coalesced into larger fat particles.
- The control chicken emulsion and the chicken emulsion with 0.1% PMG showed more coarse protein matrix structure containing larger fat globules when compared to 0.3% PMG emulsion. Moreover, the fat globules of control emulsion and the 0.1% PMG emulsion were larger and more elongate than 0.3% PMG emulsion.
- The chicken emulsions with PMG and STPP and the emulsion with only STPP showed dense protein matrix and the large number of finely distributed small fat particles, especially in the emulsion with only STPP. Most of fat particles of the emulsions that contained STPP were more round, smaller and better distributed in the protein matrix when compared to the emulsions without STPP.

5.6 Effect of Crude Malva Nut Gum (CMG) Level on Physical and Sensory Properties of Commercial Type Frankfurters.

- Cook loss was affected ($p < 0.01$) by CMG level. The frankfurters with 0.6% CMG showed the lowest cook loss (2.24%). Thus, the frankfurters with CMG were more effective in retaining moisture in the frankfurters than the control frankfurters without CMG.
- The addition of CMG reduced ($p < 0.01$) L and a values but increased color difference (ΔE) and hue angle values of the frankfurters. In contrast, no effect of CMG was noticed on yellowness (b) value of the frankfurters.
- Adding 0.2 and 0.6% of CMG resulted in higher hardness, springiness and chewiness values when compared to the no CMG frankfurters; however CMG level had not affected cohesiveness.
- The frankfurters with the highest level of CMG (0.6%) resulted in the hardest product (22.82 N). Springiness values also showed an increasing trend as the gum level was increased. Moreover, chewiness behaved similarly to hardness. The maximum and chewiness values were obtained for the 0.6% CMG frankfurters.
- The no CMG frankfurters were evaluated by the panelists as more pink than the frankfurters with 0.6% CMG.

- The panelists found that firmness and elasticity increased ($p < 0.01$) by increasing the CMG content. The CMG added frankfurters were perceived to be more firm and elastic than the control frankfurters. The addition of CMG resulted in products perceived to be less juicy than the control (without CMG).
- No effect of CMG on the flavor of frankfurters with 0.2 and 0.6% CMG content was detected. Moreover, the frankfurters with 0.2% CMG showed higher flavor scores than the control (without CMG). The overall acceptability results indicated that the panelists preferred the frankfurters with 0.2% CMG. Overall, the 0.2% CMG frankfurters can be produced with a high degree of consumer acceptance.

REFERENCES

Abdel-Aal, E. S. M., Sosulski, F. W., and Sokhansanj, S. 1996. Bleaching of wheat distillers' grains and its fibre and protein fractions with alkaline hydrogen peroxide. Lebensmittel-Wissenschaft und- Technologie. 29: 210-216.

Acton, J. C. 1972. The effect of meat particle size on extractable protein, cooking loss and binding strength in chicken loaves. Journal of Food Science. 37: 240-243.

Acton, J. C., and Dick, R. L. 1984. Protein-protein interaction in processed meats. Proceeding of the 37th Annual Reciprocal Meat Conference. 37: 36-43.

Acton, J. C., and Saffle, R. L. 1969. Preblended and prerigor meat in sausage emulsions. Food Technology. 23: 367-371.

Acton, J. C., Ziegler, J., and Burge, G. R. 1982. Functionality of muscle constituents in the processing of comminuted meat products. Critical Reviews in Food Science and Nutrition. 18: 99-121.

Anderson, D. M. W., and Morrison, N. A. 1990. The identification of *Combretum* gums which are not permitted food additives, II. Food Additives and Contaminants. 7: 181-188.

AOAC. 1996. Official Methods of Analysis. 16th ed. Arlington, VA: Association of Official Analytical Chemists.

Asghar, A., Samejima, K., and Yasui, T. 1985. Functionality of muscle proteins in gelation mechanisms of structured meat products. Critical Reviews in Food Science and Nutrition. 22: 27-106.

Awad, A., Powrie, W. D., and Fennema, O. 1968. Chemical deterioration of frozen bovine muscle at –4 ^{o}C. Journal of Food Science. 33: 227-235.

Balke, D. T., and Diosady, L. L. 2000. Rapid aqueous extraction of mucilage from whole white mustard seed. Food Research International. 33: 347-356.

Balyan, D. K., Tyagi, S. M., Singh, D., and Tanwar, V. K. 2001. Effect of extraction parameters on the properties of fenugreek mucilage and its use in ice cream as stabilizer. Journal of Food Science and Technology. 38: 171-174.

Barbut, S. 1989. Effect of three chloride salts and chopping time on the microstructure and texture of meat batters. Canadian Institute of Food Science and Technology Journal. 22: 284-289.

Barbut, S. 1995. Importance of fat emulsification and protein matrix characteristic in meat batter stability. Journal of Muscle Foods. 6: 161-177.

Barbut, S., and Findlay, C. J. 1989. Sodium reduction in poultry products-A review. Critical Reviews in Poultry Biology. 2: 59-75.

Barbut, S., and Mittal, G. S. 1989. Effect of salt reduction on the rheological properties of beef, pork and poultry meat batters. Meat Science. 26: 177-191.

Barbut, S., and Mittal, G. S. 1996. Effects of three cellulose gums on the texture profile and sensory properties of low fat frankfurters. International Journal of Food Science and Technology. 31: 241-247.

Bard, J. C. 1965. Some factors influencing extractability of salt-soluble proteins. Proceedings of the Meat Industry Research Conference, pp. 96-98. Arlington, VA.

Bater, B., Descamps, O., and Maurer, A. J. 1992. Quality characteristics of hydrocolloid-added oven-roasted turkey breasts. Journal of Food Science. 57: 1068-1070.

BeMiller, J. N. 1973. Quince seed, psyllium seed, flaxseed and okra gums. In R. S. Whistler, and J. N. BeMiller (eds.), Industrial Gums, pp. 331-337, New York: Academic Press.

Bendall, J. R. 1964. Meat proteins. In H. W. Schultz; and A. F. Anglemier (eds.), Symposium on foods: Proteins and their reactions, p. 225. Westport, CT: AVI Publishing Company.

Benhura, M. A. N., and Chidewe, C. 2002. Some properties of polysaccharide preparation that is isolated from the fruit of *Cordia abyssinica*. Food Chemistry. 76: 343-347.

Bernal, V. M., Smajda, C. H. Smith, J. L., and Stanley, D. W. 1987. Interactions in protein/polysaccharide/calcium gels. Journal of Food Science. 52: 1121-1125, 1136.

Berry, B.W. and Bigner, M. E. 1996. Use of carrageenan and konjac flour gel in low-fat restructured pork nuggets. Food Research International. 29: 355-362.

Black, S. A., and Smit, G. J. B. 1972. The effect of demethylation procedures on the quality of low-ester pectins used in dessert gels. Journal of Food Science. 37: 730-735.

Blumenkrantz, N., and Asboe-Hansen, G. 1973. New method for quantitative determination of uronic acids. Analytical Biochemistry. 54: 484-489.

Bourne, M. C. 1978. Texture profile analysis. Food Technology. 32: 62-72.

Brummer, Y., Cui, W., and Wang, Q. 2003. Extraction, purification and physicochemical characterization of fenugreek gum. Food Hydrocolloids. 17: 229-236.

Burgarella, J. C., Lanier, T. C., and Hamann, D. D. 1985. Effects of added egg white or whey protein concentrate on thermal transitions in rigidity of croaker surimi. Journal of Food Science. 50: 1588-1595, 1606.

Caldironi, H. A., and Ockerman, H. W. 1982. Bone and plasma protein extracts in sausages. Journal of Food Science. 47: 1622-1625.

Catacalos, G., and Wood, J. H. 1964. Suspending potential of gum systems and their general flow properties. Journal of Pharmaceutical Science. 53: 1089-1093.

Chen, J., Cao, P., and Song, H. 1996. Purification and properties of polysaccharide PP III from *Sterculia lychnophora* Hance. China Journal of Chinese Materia Medica. 21: 39-41.

Ciucanu, I., and Kerek, F. 1984. A simple and rapid method for the permethylation of carbohydrates. Carbohydrate Research. 131: 209-217.

Civille, G. V., and Szczesniak, A. S. 1973. Guidelines to training a texture profile panel. Journal of Texture Studies. 4: 204-223.

Comer, F. W., and Allan-Wojtas. 1988. Functional and microstructural effects of fillers in comminuted meat products. Food Microstructure. 7: 25-46.

Coon, F. P., Calkins, C. R., and Mandigo, R. W. 1983. Pre- and post-rigor sectioned and formed beef steaks manufactured with different salt levels, mixing times and tempering times. Journal of Food Science. 48: 1731-1735.

Corredig, M., Kerr, W., and Wicker, L. 2000. Molecular characterization of commercial pectins by separation with linear mix gel permeation columns in-line with multi-angle light scattering detection. Food Hydrocolloids. 14: 41-47.

Cui, W., Eskin, N. A. M., and Biliaderis, C. G. 1993. Chemical and physical properties of yellow mustard (*Sinapis alba* L.) mucilage. Food Chemistry. 46: 169-176.

Cui, W., and Mazza, G. 1996. Physicochemical characteristics of flaxseed gum. Food Research International. 29: 397-402.

Cui, W., Mazza, G., Oomah, B. D., and Biliaderis, C. G. 1994. Optimization of an aqueous extraction process for flaxseed gum by response surface methodology. Lebensmittel-Wissenschaft und- Technologie. 27: 363-369.

Da Ponte, D. J. B., Boozen, J. P., and Pihrik, W. 1986. Effect of irradiation on the stability of minced cod, with and without hydrocolloids during frozen storage. Lebensmittel-Wissenschaft und- Technologie. 19: 167-171.

Da Ponte, D. J. B., Herfst, J. M., Boozen, J. P., and Pihrik, W. 1985. Effects of different types of carrageenans and carboxymethylcelluloses on the stability of frozen stored minced fillets of cod. Journal of Food Technology. 20: 587-590.

DeFreitas, Z., Sebranek, J. G., Olson, D. G., and Carr, J. M. 1997. Freeze/thaw stability of cooked pork sausages as affected by salt, phosphate, pH, and carrageenan. Journal of Food Science. 62: 539-543.

Deng, J. C., Toledo, R. T., and Lillard, D. A. 1981. Protein-protein interaction and fat and water binding in comminuted flesh products. Journal of Food Science. 46: 1117-1980.

Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. A., and Smith, F. 1956. Colorimetric method for determination of sugars and related substances. Analytical Chemistry. 28: 350-356.

Dziezak, J. D. 1991. A focus on gums. Food Technology. 45: 116-130, 132.

Ebashi, S. 1966. Structural proteins controlling the interaction between actin and myosin. In E. Kuhn (ed.), Progressive muskeldystrophie-myatonic-myasthenic, p. 506. Berlin: Springer-Verlag.

Elbert, G. 1992. Carbohydrate. In B. P. Edna; Bobmills; B. A. Jacquelyn; and H. S. Leslie (eds.), Armed forces institute of pathology laboratory methods in histotechnology, pp. 149-150. Washington, DC: Armed Forces Institute of Pathology.

Ericson, M. C., and Elbein, A. D. 1980. Biosynthesis of cell wall polysaccharides and glycoproteins. In P. K. Stumpf; and E. E. Conn (eds.), The biochemistry of plants, pp 589-616. New York: Academic Press.

Farkas, E., and Glicksman, M. 1967. Hydrocolloid rheology in the formulation of convenience foods. Food Technology. 21: 49-52.

Field, R. A. 1981. Mechanically deboned red meat. Advances in Food Research. 27: 23-27.

Filippov, M. P., and Shamshurina, S. A. 1972. Comparative study of pectin substances by IR spectroscopy. Food Science and Technology Abstract. 6: 7A2601974.

Foegeding, E. A. 1987. Functional properties of turkey salt-soluble proteins. Journal of Food Science. 52: 1495-1499.

Foegeding, E. A., and Ramsey, S. R. 1986. Effect of gums on low-fat meat batters. Journal of Food Science. 51: 33-36, 46.

Foegeding, E. A., and Ramsey, S. R. 1987. Rheological an water-holding properties of gelled meat emulsions containing iota carrageenan, kappa carrageenan or xanthan gum. Journal of Food Science. 52: 549-553.

Forrest, J. C., Aberle, E. D., Hedrick, H. B., Judge, M. D., and Merkel, R. A. 1975. Principles of meat science. San Francisco, CA: W. H. Freeman and Company, p. 288.

Fox Jr., J. B., Ackerman, S. A., and Jenkins, R. K. 1983. Effect of anionic gums on the texture of pickled frankfurters. Journal of Food Science. 48: 1031-1035.

Frederickson, D. W., and Holtzer, A. 1968. The substructure of the myosin molecule: Production and properties of the alkali subunits. Biochemistry. 7: 3935-3950.

Friedman, H. H., Whiting, J. E., and Szczesniak, A. S. 1963. The texturometer – A new instrument for objective texture measurement. Journal of Food Science. 28: 390-395.

Froning, G. W. 1981. Mechanical deboning of poultry and fish. Advances in Food Research. 27: 109-147.

Galluzzo, S. J., and Regenstein, J. M. 1978. Emulsion capacity and timed emulsification of chicken breast muscle myosin. Journal of Food Science. 43: 1757-1760.

Glicksman, M. 1969. Gum Technology in the Food Industry. New York: Academic Press, p. 590.

Gordon, A., and Barbut, S. 1989. The effect of chloride salts on the texture, microstructure and stability of meat batters. Food Microstructure. 8: 271-283.

Gordon, A., and Barbut, S. 1990. The role of the interfacial protein film in meat batter stabilization. Food Structure. 9: 77-90.

Gordon, A., and Barbut, S. 1991. Raw meat batter stabilization: Morphological study of the role of the interfacial protein film. Canadian Institute of Food Science and Technology Journal. 24: 136-142.

Gordon, A., and Barbut, S. 1992a. Effect of chloride salts on protein extraction and interfacial protein film formation in meat batters. Journal of the Science of Food and Agriculture. 58: 227-238.

Gordon, A., and Barbut, S. 1992b. Mechanisms of meat batter stabilization: A review. Critical Reviews in Food Science and Nutrition. 4: 299-332.

Grabowska, E. J., and Sikorski, Z. E. 1976. The gel-forming capacity of fish myofibrillar proteins. Lebensmittel-Wissenschaft und- Technologie. 9: 33-35.

Graessley, W. W. 1974. The entanglement concept in polymer rheology. Advanced Polymer Chemistry. 17: 19-151.

Groninger, H., Hawkes, J. W., and Babbit, J. K. 1983. Functional and morphological changes in processed frozen fish muscle. Journal of Food Science. 48: 1388-1392.

Hamann, D. D. 1987. Methods for measurement of rheological changes during thermally induced gelation of proteins. Food Technology. 41: 100-108.

Hamann, D. D. 1988. Rheology as a means of evaluating muscle functionality of processed foods. Food Technology. 42: 66-71.

Hamann, D. D., and Webb, N. B. 1979. Sensory and instrumental evaluation of material properties of fish gels. Journal of Texture Studies. 10: 117-122.

Hamm, R. 1960. Biochemistry of meat hydration. Advances in Food Research. 10: 355-463.

Hamm, R. 1966. Heating of muscle systems. In E. J. Briskey; R. G. Cassens; and J. C. Trautman (eds.), The physiology and biochemistry of muscle as a food, p. 363. Madison, WI: The University of Wisconsin Press.

Hamm, R. 1970. Biochemistry of meat hydration. Advances in Food Research. 10: 355-392.

Hamm, R., and Grabowska, E. J. 1979. Protein solubility and water binding under conditions obtained in frankfurter mixtures. Communication II. the effects of heat on dissolved proteins. Die Fleischwirtschaft. 58: 1345- 1349.

Hansen, J., and Lowy, J. 1964. The structure of actin. In J. Gergely (ed.), Biochemistry of muscle contraction, p. 141. Boston: Brown and Co.

Hargett, S. M., Blumer, T. N., Hamann, D. D., Keeton, J. T., and Monroe, R. J. 1980. Effect of sodium pyrophosphate on sensory, chemical, and physical properties of frankfurters. Journal of Food Science. 45: 905-909.

Hay, J. D., Currie, R. W., and Wolfe, F. H. 1973. Effect of postmortem aging on chicken muscle fibers. Journal of Food Science. 38: 981-986.

Hellendoorn, E. W. 1962. Water-binding capacity of meat as affected by phosphates. I. Influence of sodium chloride and phosphates on the water retention of comminuted meat at various pH values. Food Technology. 16: 119-124.

Hsu, S. Y., and Chung, H. Y. 2001. Effects of κ-carrageenan, salt, phosphates and fat on qualities of low fat emulsified meatballs. Journal of Food Engineering. 47: 115-121.

Hughes, E., Cofrades, S., and Troy, D. J. 1997. Effects of fat level, oat fibre and carrageenan on frankfurters formulated with 5, 12 and 30% fat. Meat Science. 45: 273-281.

Jones, K. W., and Mandigo, R. W. 1982. Effects of chopping temperature on the microstructures of meat emulsions. Journal of Food Science. 47: 1930-1935.

Keeton, J. T. 1983. Effects of fat and NaCl/phosphate levels on the sensory properties of pork patties. Journal of Food Science. 48: 878-881.

Keeton, J. T., Foegeding, E. A., and Patana-Anake, C. 1984. A comparison of nonmeat proteins, sodium tripolyphosphate and processing temperature effects on physical and sensory properties of frankfurters. Journal of Food Science. 49: 1462-1465.

Kim, B. Y., Hamann, D. D., Lanier, T. C., and Wu, M. C. 1986. Effects of freeze-thaw abuse on the viscosity and gel-forming ability of surimi from two species. Journal of Food Science. 51: 951-956, 1004.

Koo, K. H. 1980. Determination of myofibrillar proteins binding strength, yield and TBA effected by mechanical treatment, salt and micro-organisms in a pre-cooked (uncured) chunked and formed beef product. Dissertation Abstracts International B40(10) 4726.

Koolmees, P. A., Moerman, P. C., and Zijderveld, M. H. G. 1989. Image analysis of the fat dispersion in a comminuted meat system. Food Microstructure. 8: 81-90.

Kotter, L., and Fischer, A. 1975. The influence of phosphates or polyphosphates on the stability of foams and emulsions in meat technology. Die Fleischwirtschaft. 3: 360-362, 365-368.

Krause, R. J., Ockerman, H. W., Krol, B., Moerman, P. C., and Plimpton, R. F., Jr. 1978. Influence of tumbling, tumbling time, trim and sodium tripolyphosphate on quality and yield of cured hams. Journal of Food Science. 43: 853-855.

Krumel, K. F., and Sarkar, N. 1975. Flow properties of gums useful to the food industry. Food Technology. 29: 36-44.

Lanier, T. C. 1985. Fish protein in processed meats. Proceeding of the 38th Annual Reciprocol Meat Conference. 38: 129-139.

Lanier, T. C. 1991. Interactions of muscle and non-muscle protein affecting heat-set gel rheology. In N. Parris; and R. Barford (eds.), Interactions of food proteins, pp. 268-284. Washington, DC: American Chemical Society.

Lanier, T. C., Hamann, D. D., and Wu, M. C. 1985. Development of methods for quality and functionality assessment of surimi and minced fish to be used in gel-type food products. Final Report for Alaska Fisheries Development Foundation, pp. 10-25. Ancholage, Alaska.

Launay B., Doublier J. R., Cuvelier G. 1986. Dispersed system (starch pastes). In J. R. Mitcheil; and D. A. Ledward (eds.), Functional properties of food macromolecules, pp. 1-78. London: Elsevier Applied Science.

Lawrie, R. A. 1974. Meat Science. 2nd ed. New York: Pergamon Press, p. 419.

Lee, C. M. 1985. Microstructure of meat emulsions in relation to fat stabilization. Food Microstructure. 4: 63-72.

Lee, C. M., and Abdollahi, A. 1981. Effect of hardness of plastic fat on structure and material properties of fat-protein gel product. Journal of Food Science. 46: 1755-1759.

Lee, C. M., Carroll, R. J., and Abdollahi, A. 1981. A microscopical study of the structure of meat emulsions and its relationship to thermal stability. Journal of Food Science. 46: 1789-1793.

Lin, K. W., and Huang, H. Y. 2003. Konjac/gellan gum mixed gels improve the quality of reduced-fat frankfurters. Meat Science. 65: 749-755.

Lin, C. S., and Zayas, J. F. 1987. Microstructural comparisons of meat emulsions prepared with corn protein emulsified and unemulsified fat. Journal of Food Science. 52: 267-271.

Lurueña-Martínez, M. A. Vivar-Quintana, A. M., and Revilla, I. 2004. Effect of locust bean/xanthan gum addition and replacement of pork fat with olive oil on the quality characteristics of low-fat frankfurters. Meat Science. 68: 383-389.

Maesso, E. R., Baker, R. C., Bourne, M. C., and Vadehra, D. V. 1970. Effect of some physical and chemical treatments on the binding quality of poultry loaves. Journal of Food Science. 35: 440- 445.

Mahon, J. H. 1961. Tripolyphosphate-salt synergism and its effect on cured meat volume. Proceeding of the 13th Research Conference of American Meat Institute Foundation, pp. 20-35.

Mawson, R. F., Miller, B. F., and Schmidt, G. R. 1983. Studies on pasteurized and commercially sterilized poultry meat bologna. Effects of nitrite addition and vacuum cutting. Journal of Food Science. 48: 322-326.

May, C. D. 1997. Pectins. In A. Imeson, (ed.), Thickening and gelling agents for food, pp. 230-260. London, UK: Blackie Academic and Professional.

Mazza, G., and Biliaderis, C. G. 1989. Functional properties of flax seed mucilage. Journal of Food Science. 54 : 1302-1305.

Mcfarlane, J. J., Schmidt, G. R., and Turner, R. H. 1977. Binding of meat pieces: A comparison of myosin, actomyosin and sarcoplasmic proteins as binding agents. Journal of Food Science. 42: 1603-1605.

McGowan, R. G. 1970. Method of preparing a poultry product. U. S. Patent 3,503,755.

McMahon, E. F., and Dawson, L. E. 1976. Effects of salt and phosphates on some functional characteristics of hand and mechanically deboned turkey meat. Poultry Science. 55: 573-578.

Means, W. J., and Schmidt, G. R. 1986. Algin/calcium gel as a raw and cooked binder in structured beef steaks. Journal of Food Science. 51: 60-65.

Miller, A. J., Ackerman, S. A., and Palumbo, S. A. 1980. Effects of frozen storage on functionality of meat for processing. Journal of Food Science. 45: 1466-1471.

Mittal, G. S., and Barbut, S. 1989. Effects of salt reduction on the rheological and gelation properties of white and dark poultry meat emulsions. Journal of Texture Studies. 20: 209-222.

Mittal, G. S., and Barbut, S. 1993. Effects of various cellulose gums on the quality parameters of low-fat breakfast sausages. Meat Science. 35: 93-103.

Mittal, G. S., and Barbut, S. 1994. Effects of carrageenans and xanthan gum on the texture and acceptability of low fat frankfurters. Journal of Food Processing and Preservation. 18: 201-216.

Mittal, G. S., and Blaisdell, J. L. 1983. Weight loss in frankfurters during thermal processing. Meat Science. 9: 79-88.

Molins, R. A. 1991. Phosphates in Food. Boca Raton, FL: CRC Press, pp. 235-251.

Montejano, J. G., Hamann, D. D., and Lanier, T. C. 1985. Comparison of two instrumental methods with sensory texture of protein gels. Journal of Texture Studies. 16: 403-424.

Montero, P., Hurtado, J. L., and Pérez-Mateos, M. 2000. Microstructural behavior and gelling characteristics of myosystem protein gels interacting with hydrocolloids. Food Hydrocolloids. 14: 455-461.

Morin, L. A., Temelli, F., and McMullen, L. 2004. Interactions between meat proteins and barley (Hordeum spp.) β-glucan within a reduced-fat breakfast sausage system. Meat Science. 68: 419-430.

Morris, R., Culter, A. N., Ross-Murphy, S. B., and Rees, D. A. 1981. Concentration and shear rate dependence of viscosity in random coil polysaccharide solutions. Carbohydrate Polymers. 1: 5-21.

Morrison, G. S., Webb, N. B., Blumer, T. N., Ivey, F. J., and Haq, A. 1971. Relationship between composition and stability of sausage-type emulsions. Journal of Food Science. 36: 426-430.

Neer, K. L., and Mandigo, R. W. 1977. Effects of salt, sodium tripolyphosphate and frozen storage time on properties of flaked, cured pork product. Journal of Food Science. 42: 738-742.

Nusbaum, R. P., Sebranek, J. G., Topel, D. G., and Rust, R. E. 1983. Structural and palatability relationships in frozen ground beef patties as a function of freezing treatments and product formulation. Meat Science. 8: 135-140.

Oakenfull, D. 1987. Gelling agents. Critical Reviews in Food Science and Nutrition. 26: 1-25.

Odonmazig, P., Ebringerova, A., Badga, D., and Janecek, F. 1985. Carbohydrate components of Mongolian apricot fruit (*Armeniaca sibirica* L.) fractional extraction and general characteristics. Journal of the Science of Food and Agriculture. 36: 575-582.

Offer, G., Moos, C., and Starr, R. 1973. A new protein of the thick filaments of vertebrate skeletal myofibrils. Journal of Molecular Biology. 74- 653-676.

Offer, G., and Trinick, J. 1983. On the mechanism of water holding in meat: The swelling and shrinkage of myofibrils. Meat Science. 8: 245-281.

Parks, L. L., and Carpenter, J. A. 1987. Functionality of six nonmeat proteins in meat emulsion systems. Journal of Food Science. 52: 271-275.

Park, J. W., Lanier, T. C., Keeton, J. T., and Hamann, D. D. 1987. Use of cryoprotectants to stabilize functional properties of prerigor salted beef during frozen storage. Journal of Food Science. 52: 537-542.

Patana-anake, C., and Foegeding, E. A. 1985. Rheological and stability transitions in meat emulsions containing soy protein and vital wheat gluten. Journal of Food Science. 50: 160-164.

Pepper, F. H., and Schmidt, G. R. 1975. Effect of blending time, salt, phosphate and hot-boned beef on binding strength and cook yield of beef rolls. Journal of Food Science. 40: 227-230.

Perry, S. V. 1956. Relation between chemical and contractile function and structure of the skeletal muscle cell. Physical Review. 36: 3-67.

Porzio, M. A., and Pearson, A. M. 1977. Improved resolution of myofibrillar proteins with sodium dodecyl sulfate-polyacrylamide gel electrophoresis. Biochimica et Biophysica Acta. 490: 27-34.

Puolanne, E. J., and Kukkonen, E. 1983. Influence of core temperature at the end of cooking on the water binding capacity of the meat in bruhwurst. Fleischwirtschaft. 63: 1495-1500.

Puolanne, E. J., and Ruusunen, M. 1981. The properties of connective tissue membrane and pig skin as raw materials for cooked sausage. Meat Science. 5: 371-382.

Puolanne, E. J., and Terrell, R. N. 1983a. Effects of salt levels in prerigor blends and cooked sausages on the water binding, released fat and pH. Journal of Food Science. 48: 1022-1026.

Puolanne, E. J., and Terrell, R. N. 1983b. Effects of rigor-state, levels of salt and sodium tripolyphosphate on physical, chemical and sensory properties of frankfurter-type sausages. Journal of Food Science. 48: 1036-1041.

Quinn, J. R., Raymond, D. P., and Harwalker, V. R. 1980. Differential scanning calorimetry of meat proteins as affected by processing treatment. Journal of Food Science. 45: 1146-1149.

Randall, C. J., and Voisey. P. W. 1977. Effect of meat protein fractions on textural characteristics of meat emulsions. Canadian Institute of Food Science and Technology Journal. 10: 88-92.

Rao, M. A., and Steffe, J. F. 1992. Viscoelastic properties of foods. London, UK: Elsevier Applied Science, p. 441.

Rees, D. A., and Welsh, E. J. 1977. Secondary and tertiary structure of polysaccharides in solutions and in gels. Angewandte Chemie (International Ed.). 16: 214-224.

Rombouts, F. M., and Thibault J. F. 1986. Feruloylated pectic substances from sugar-beet pulp. Carbohydrate Research. 154: 177-187.

Rongey, E. H., and Bratzler, L. J. 1966. The effect of various binders and meats on the palatability and processing characteristics of bologna. Food Technology. 20: 1228-1231.

Ross-Murphy, S. B. 1995. Rheological characterisation of gels. Journal of Texture Studies. 26: 391-400.

Rust, R. E. 1976. Sausage and processed meats manufacturing. Chicago: American Meat Institute Center for Continuing Education, p. 123.

Saffle, R. L., and Galbreath, J. W. 1964. Quantitative determination of salt-soluble protein in various types of meat. Food Technology. 18: 1943-1953.

Saliba, D. A., Foegeding, E. A., and Hamann, D. D. 1987. Structural failure and nondestructive rheological analyses of frankfurter batters: Effects of heating rates and sugars. Journal of Texture Studies. 18: 241-259.

Samant, S. K., Singhal, R. S., Kulkarni, P. R., and Rege, D. V. 1993. Protein-polysaccharide interactions: A new approach in food formulations. International Journal of Food Science and Technology. 28: 547-562.

Sanderson, G. R. 1981. Polysaccharides in foods. Food Technology. 35: 50-57, 83.

Sanderson, G. R. 1996. Gums and their use in food systems. Food Technology. 50: 81-84.

SAS Institute. 1997. User's guide. Cary, NC: SAS Institute.

Schmidt, G. R. 1984. Processing effects on meat product microstructure. Food Microstructure. 3: 33-39.

Schmidt, G. R., Mawson, R. F., and Siegel, D. G. 1981. Functional properties of a protein matrix in comminuted meat products. Food Technology. 35: 235-252.

Schmidt, G. R., and Trout, G. R. 1984. Chemistry of meat binding. In A. J. Bailey, (ed), Recent advances in the chemistry of meat, pp. 231-145. London, UK: The Royal Society of Chemistry.

Schnell, P. G., Vadhehra, D. V., and Baker, R. C. 1970. Mechanism of binding chunks of meat. I. Effects of physical and chemical treatments. Canadian Institute of Food Technology Journal. 3: 44-48.

Schult, J. 1976. Meat emulsions. In S. Friberg, (ed), Food emulsions, pp. 50-55. New York: Marcel Dekker.

Schults, G. W., and Wierbicki, E. 1973. Effects of sodium chloride and condensed phosphates on the water holding capacity, pH and swelling of chicken muscle. Journal of Food Science. 38: 991-994.

Schweid, J. M., and Toledo, R. T. 1981. Changes in physical properties of meat batters during heating. Journal of Food Science. 46: 850-854.

Scott-Blair, G. W. 1969. Elementary rheology. New York: Academic Press, pp. 81-89.

Seman, D. L., Olson, D. G., and Mandigo, R. W. 1980. Effect of reduction and partial replacement of sodium on bologna characteristics and acceptability. Journal of Food Science. 45: 1116-1121.

Sharma, S. C. 1981. Gums and hydrocolloids in oil-water emulsions. Food Technology. 35: 59-64.

Siegel, D. G., Church, K. E., and Schmidt, G. R. 1979. Gel structure of nonmeat proteins as related to their ability to bind meat pieces. Journal of Food Science. 44: 1276-1279, 1284.

Siegel, D. G., and Schmidt, G. R. 1979. Ionic, pH and temperature effects on the binding quality of myosin. Journal of Food Science. 44: 1686-1689.

Siegel, D. G., Theno, D. M., Schmidt, G. R., and Norton, H. W. 1978. Meat massaging: The effects of salt, phosphate and massaging on cooking loss, binding strength and exudates composition in sectioned and formed ham. Journal of Food Science. 43: 331-333.

Singthong, J., Ningsanond, S, Cui, S.W. and Goff, H.D. 2004. Structural characterization, degree of esterification and some gelling properties of Krueo Ma Noy (*Cissampelos pareira*) pectin. Carbohydrate Polymers. 58: 391-400.

Smith, D. M. 1988. Meat proteins: Functional properties in comminuted meat products. Food Technology. 42: 116-121.

Sofos, J. N. 1983a. Effects of reduced salt (NaCl) levels on the stability of frankfurters. Journal of Food Science. 48: 1684-1691.

Sofos, J. N. 1983b. Effects of reduced salt (NaCl) levels on sensory and instrumental evaluation of frankfurters. Journal of Food Science. 48: 1692-1695, 1699.

Sofos, J. N. 1986. Use of phosphates in low sodium meat products. Food Technology. 40: 52-69.

Solomon, L. W., and Schmidt, G. R. 1980. Effect of vacuum and mixing time on the extractability and functionality of pre- and post-rigor beef. Journal of Food Science. 45: 283-287.

Somboonpanyakul, P., Barbut, S., Jantawat, P. and Chinprahast, N. 2004a. Effect of malva nut gum, sodium chloride and phosphate on textural properties of poultry meat batters. Abstracts of the Canadian Institute of Food Science and Technology and Agriculture and Agri-Food Canada Joint Conference, p. 110. Guelph, Ontario.

Somboonpanyakul, P., Wang, Q., Cui, W., Barbut, S., Jantawat, P. and Chinprahast, N. 2004b. Extraction and physicochemical characterization of malva nut gum. Abstracts of the Canadian Institute of Food Science and Technology and Agriculture and Agri-Food Canada Joint Conference, p. 83. Guelph, Ontario.

Stainsby, G. 1980. Proteinaceous gelling systems and their complexes with polysaccharides. Food Chemistry. 6: 3-14.

Steffe, J. 1996. Rheological methods in food process engineering. 2nd ed. East Lansing, MI: Freeman Press, pp. 294-348.

Steinhauer, J. E. 1983. Food phosphates for use in the meat, poultry and seafood industry. Dairy and Food Sanitation. 3: 244-249.

Stone, H., Sidel, J., and Oliver, S. 1974. Sensory evaluation by quantitative descriptive analysis. Food Technology. 28: 24-34.

Susheelamma, N. S. 1987. Isolation and properties of linseed mucilage. Journal of Food Science and Technology. 24: 103-106.

Suter, D. A., Sustek, E., Dill, C. W., Marshall, W. H., and Carpenter, Z. L. 1976. A method for measurement of the effect of blood protein concentrates on the binding forces in cooked ground beef patties. Journal of Food Science. 41: 1428-1432.

Swasdee, R. L., Terrell, R. N., Dutson, T. R., and Lewis, R. E. 1982. Ultrastructural changes during chopping and cooking of a frankfurter batter. Journal of Food Science. 47: 1011-1013.

Swift, C. E., and Ellis, R. 1956. The action of phosphates in sausage products. I. Factors affecting the water retention of phosphate-treated ground meat. Food Technology. 10: 546-552.

Swift, C. E., and Ellis, R. 1957. Action of phosphates in sausage products. II. Pilot plant studies of the effects of some phosphates on binding and color. Food Technology. 11: 450-456.

Syarief, H., Hamann, D. D., Giesbrecht, F. G., Young, C. T., and Monroe, R. J. 1988. Interdependency and underlying dimensions of sensory textural characteristics of selected foods. Journal of Texture Studies. 19: 29-35.

Szczesniak, A. S. 1963. Classification of textural characteristic. Journal of Food Science. 28: 385-389.

Szczesniak, A. S. 1985. Rheological basis for selecting hydrocolloids for specific applications. In G. O. Phillips; D. J. Wedlock; and P. A. Williams (eds.), Gums and stabilizers for the food industry 3, p. 311. London, UK: Elsevier Applied Science.

Szczesniak, A. S., and Hall, B. J. 1975. Application of the general foods texturometer to specific food products. Journal of Texture Studies. 6: 117-121.

Szent-Gyorgyi, A. G. 1951. Chemistry of muscular contraction. 2nd ed. New York: Academic Press, p. 178.

Tantikarnjathep, K., Sebranek, J. G., and Topel, D. G. 1983. Use of vacuum during formation of meat emulsions. Journal of Food Science. 48: 1039-1044.

Taylor, R. L., and Conrad, H. E. 1972. Stoichiometric depolymerization of polyuronides and glycosaminoglycuronans to monosaccharides following reduction of their carbodiimide-activated carboxyl groups. Biochemistry. 11: 1383-1388.

Terrell, R. N., Brown, J. A., Carpenter, Z. L., Mattil, K. F., and Monagle, C. W. 1979a. Effects of oilseed proteins, at two replacement levels, on chemical, sensory and physical properties of frankfurters. Journal of Food Science. 44: 865-869.

Terrell, R. N., Crenwelge, C. H., Dutson, T. R., and Smith, G. C. 1982. A technique to measure binding properties of nonmeat proteins in muscle-junction formation. Journal of Food Science. 47: 711-713.

Terrell, R. N., Weinblatt, P. J., Smith, G. C., Carpenter, Z. L., Dill, C. W., and Morgan, R. G. 1979b. Plasma protein isolate effects on physical characteristics of all-meat and extended frankfurters. Journal of Food Science. 44: 1041-1045.

Thai Customs Department. 2004. Import/Export statistics from Jan – Oct 2004 [online]. available from: http://www.customs.go.th/statistic [2004, Dec 15].

Theno, D. M., and Schmidt, G. R. 1978. Microstructural comparison of three commercial frankfurters. Journal of Food Science. 43: 845-848.

Theno, D. M., Siegel, D. G., and Schmidt, G. R. 1978. Meat massaging: Effects of salt and phosphate on the microstructure of binding junctions in sectioned and formed ham. Journal of Food Science. 43: 493-498.

Tolstoguzov, V. B. 1986. Functional properties of protein polysaccharide mixtures. In J. R. Mitchell; and D. A. Ledward (eds.), Functional properties of food macromolecules, pp. 385-415. London, UK: Elsevier Applied Science.

Townsend, W. E., Witnauer, L. P., Riloff, J. A., and Swift, C. E. 1968. Comminuted meat emulsions: Differential thermal analysis of fat transitions. Food Technology. 22: 319-325.

Trius, A., and Sebranek, J. G. 1996. Carrageenans and their use in meat products. Critical Reviews in Food Science and Nutrition. 36: 69-85.

Trius, A., Sebranek, J. G., Rust, R. E., and Carr, J. M. 1994. Low-fat bologna and beaker sausage: Effects of carrageenans and chloride salts. Journal of Food Science. 59: 941-945.

Trout, G. R., and Schmidt, G. R. 1983. Utilization of phosphates in meat products. Proceeding of the 36th Annual Reciprocal Meat Conference. 36: 24-47.

Trout, G. R. and Schmidt, G. R. 1984. Effect of phosphate type and concentration, salt level and method of preparation on binding in restructured beef rolls. Journal of Food Science. 49: 687-694.

Trout, G. R., and Schmidt, G. R. 1986a. Water binding ability of meat products: Effect of fat level, effective salt concentration and cooking temperature. Journal of Food Science. 51: 1061-1062.

Trout, G. R., and Schmidt, G. R. 1986b. Effect of phosphates on the functional properties of restructured beef rolls: The role of pH, ionic strength, and phosphate type. Journal of Food Science. 51: 1416-1423.

Trudso, J. E. 1985. Increasing yields with carrageenan. Meat Processing. 24: 37-38, 40-42.

Turner, R. H., Jones, P. N., and Macfarlane, J. J. 1979. Binding of meat pieces: An investigation of the use of myosin-containing extracts from pre- and post-rigor bovine muscle as meat binding agents. Journal of Food Science. 44: 1443-1446.

Vadehra, D. V., and Baker, R. C. 1970. The mechanism of heat-initiated binding of poultry meat. Food Technology. 24: 42-55.

Vallejo-Cordoba, B., Nakai, S., Powrie, W. D., and Beveridge, T. 1986. Protein hydrolysates for reducing water activity in meat products. Journal of Food Science. 51: 1156-1161.

Voisey, P. W., Randall, C. J., and Larmond, E. 1975. Selection of an objective test of weiner texture by sensory analysis. Canadian Institute of Food Science and Technology Journal. 8: 23-29.

Wang, Q., Ellis, P. R., Ross-Murphy, S. B., and Burchard, W. 1997. Solution characteristics of the xyloglucan extracted from *Detarium senegalense* Gmelin. Carbohydrate Polymers. 33: 115-124.

Wang, R. F., Yang, X. W., Ma, C. M., Shang, M. Y., Liang, J. Y., Wang, X., Cai, S. Q., and Shoyama, Y. 2003. Alkaloids from the seeds of *Sterculia lychnophora* (Pangdahai). Phytochemistry. 63: 475-478.

Wellner, N., Kacurakova, M., Malovikova, A., Wilson, R., and Belton, P. S. 1998. FTIR study of pectate and pectinate gels formed by divalent cations. Carbohydrate Research. 308: 123-131.

Whitcomb, P. J., Gutowski, J., and Howland, W. W. 1980. Rheology of guar solutions. Journal of Applied Polymer Science. 25: 2815-2827.

Whiting, R. C. 1984. Stability and gel strength of frankfurter batters made with reduced NaCl. Journal of Food Science. 49: 1350-1362.

Whiting, R. C. 1987. Influence of lipid composition of water and fat exudation and gel strength of meat batters. Journal of Food Science. 52: 1126-1130.

Whiting, R. C. 1988. Ingredients and processing factors that control muscle protein functionality. Food Technology. 42: 104-114, 210.

Williams, P. A., and Phillips, G. O. 2000. Gum Arabic. In G. O. Phillips; and P. A. Williams (eds.), Handbook of hydrocolloids, pp. 155-168. New York: CRC Press.

Wood, P. J., Weisz, J., and Blackwell, B. A. 1994. Structural studies of (1 --> 3), (1 --> 4)-b-D-glucans by 13C-nuclear magnetic resonance spectroscopy and by rapid analysis of cellulose-like regions using high-performance anion-exchange chromatography of oligosaccharides released by lichenase. Cereal Chemistry. 71: 301-307.

Wright, D. J., Leach, I. B., and Wilding, P. 1977. Differential scanning calorimetric studies of muscle and its constituent proteins. Journal of the Science of Food and Agriculture. 28: 557-564.

Wu, M. C., Hamann, D. D., and Lanier, T. C. 1985a. Rheological and calorimetric investigations of starch-fish protein systems during thermal processing. Journal of Texture Studies. 16: 53-74.

Wu, M. C., Lanier, T. C., and Hamann, D. D. 1985b. Rigidity and viscosity changes of croaker actomyosin during thermal gelation. Journal of Food Science. 50: 14-19,25.

Yamada, T., Itoh, A., Kanzaki, M., Yamakura, T., Suzuki, E. and Ashton, P. S. 2000. Local and geographical distributions for a tropical tree genus, *Scaphium* (Sterculiaceae) in the Far East. Plant Ecology. 148: 23-28.

Yasui, T., Ishioroshi, M., and Samejima, K. 1980. Heat-induced gelation of myosin in the presence of actin. Journal of Food Biochemistry. 4: 61-78.

York, W. S., Darvill, A. G., McNeil, M., Stevenson, T. T., and Albersheim, P. 1986. Isolation and characterization of plant cell wall components. Methods in Enzymology. 118: 3-40.

APPENDICES

APPENDIX A

According to Civille and Szczesniak (1973) defined the definitions of physical textural characteristics of hardness, springiness, cohesiveness, chewiness and fractureability as follows:

(a) Hardness is a force necessary to attain a given deformation.

(b) Springiness is a rate at which a deformed material goes back to its undeformed condition after the deforming force is removed.

(c) Cohesiveness is an extent to which a material can be deformed before it ruptures.

(d) Chewiness is an energy required to masticate a solid food to a state ready for swallowing.

(e) Fracturability is a force with which a material fractures; a product of high degree of hardness and low degree of cohesiveness.

APPENDIX B

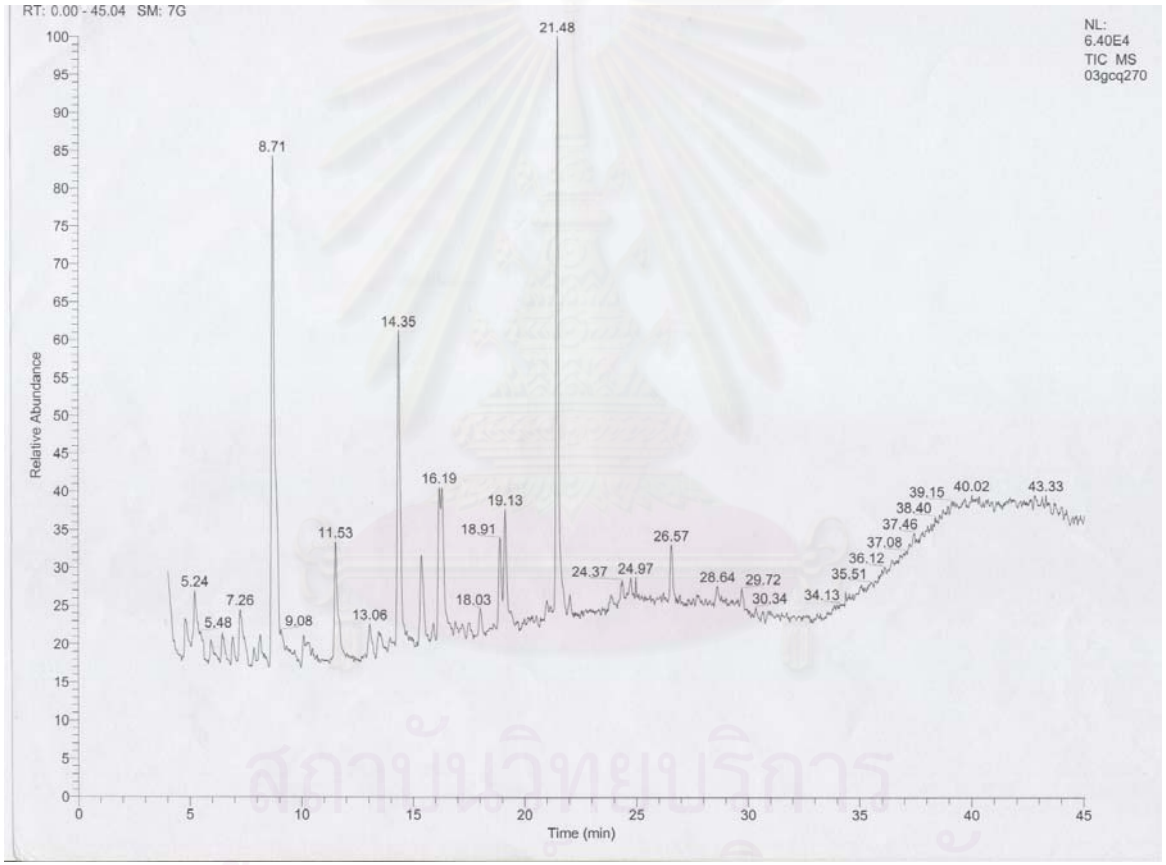

Fig. 1 Chromatogram of GC of alkaline extracted malva nut gum.

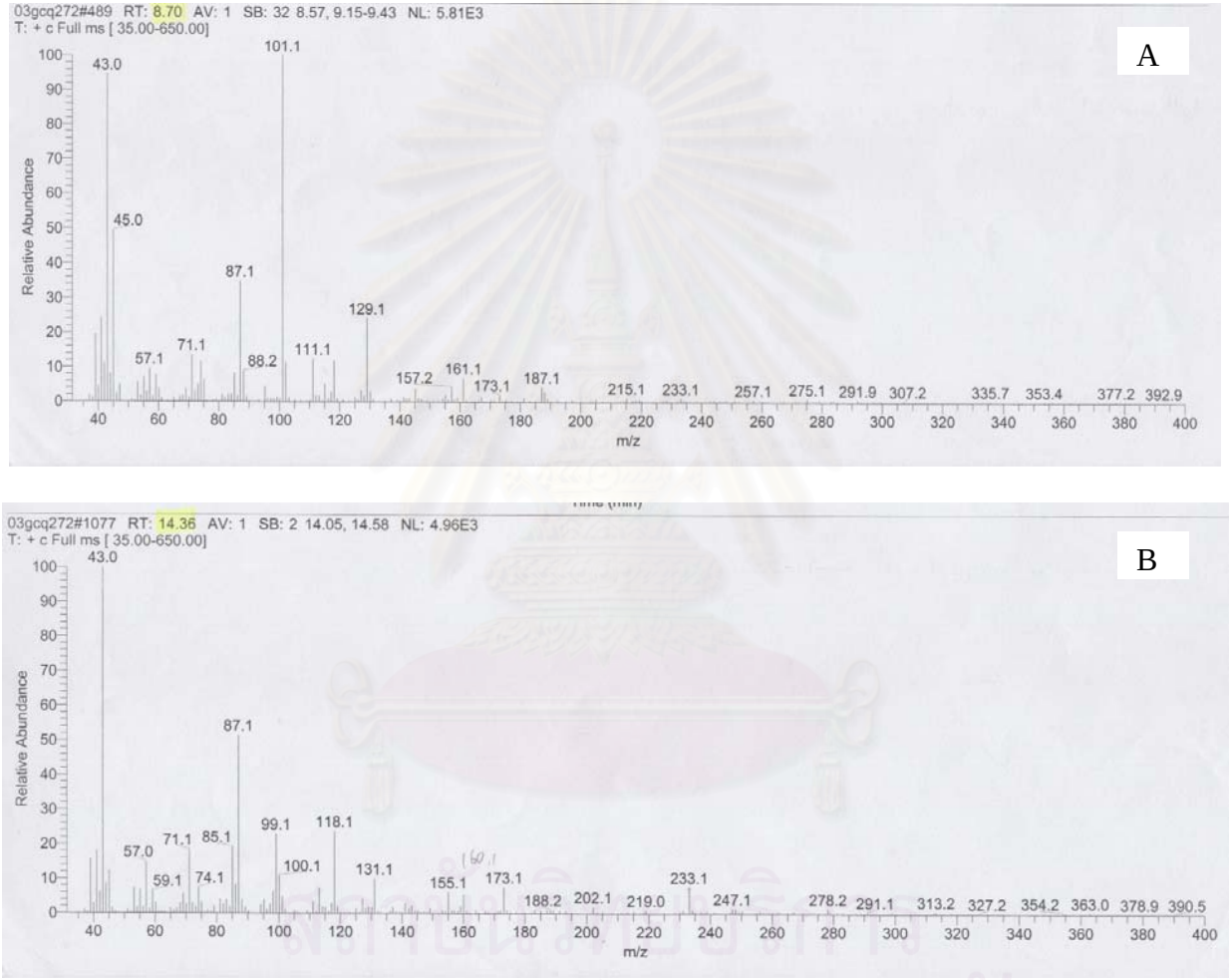

Fig. 2 Mass spectrum of alkaline extracted malva nut gum. (A) peak at 8.70 min and (B) peak at 14.36 min.

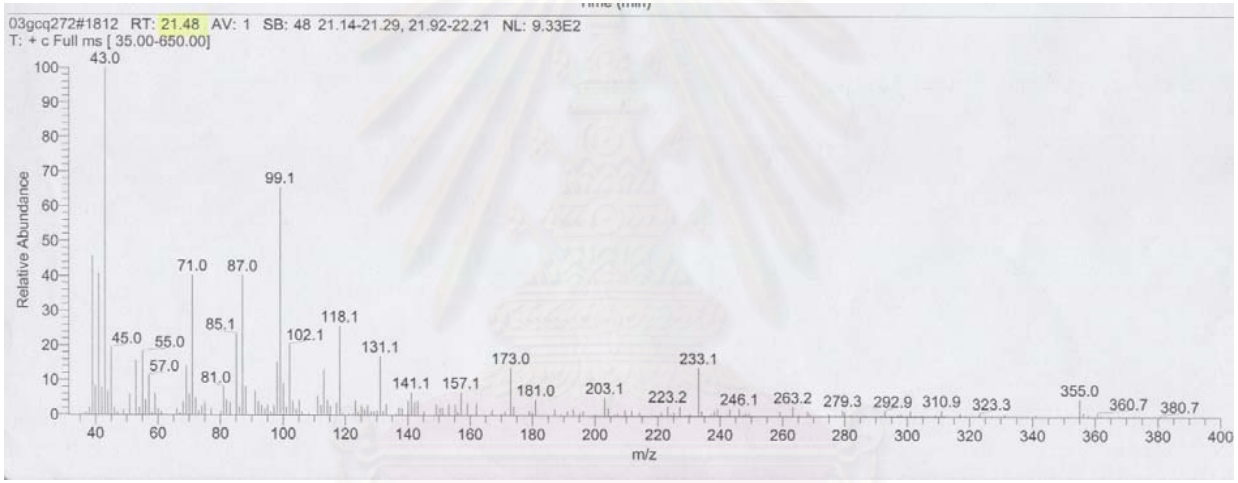

Fig. 3 Mass spectrum of peak at 21.48 min of alkaline extracted malva nut gum.

APPENDIX C

Table 1 ANOVA: overall effect of independent variables on response variable.

Independent variable	df	Sum of squares						
		Cook loss	Fracture force	Fracture distance	Springiness	Cohesiveness	Chewiness	Hardness
Treatment	9	683.29**	889.80**	13.59**	0.48**	0.22	396.96**	2,416.76**
Block	2	267.47**	15.56	1.63*	0.06**	0.04	15.51*	159.15
Error	18	161.19	524.07	3.32	0.07	0.26	27.77	622.00

* Significant at 5% level
** Significant at 1% level

Table 2 ANOVA: overall effect of independent variables on response variable.

Independent variable	df	Sum of squares						
		Cook loss	Fracture force	Fracture distance	Springiness	Cohesiveness	Chewiness	Hardness
Treatment	9	1,660.15**	569.73**	7.10**	0.53**	0.03**	244.36**	1,273.96**
Block	2	90.92**	51.67**	0.60	0.11**	0.00*	20.82**	77.22
Error	18	12.66	38.50	3.30	0.01	0.00	19.28	396.48

* Significant at 5% level
** Significant at 1% level

Table 3 ANOVA: overall effect of independent variables on response variable.

Independent Variable	df	Sum of squares							
		Cook loss	Springiness	Cohesiveness	Chewiness	Fracture force	L	a	b
STPP	1	649.20**	0.09**	0.01*	33.84**	459.95**	0.36	1.76*	3.91**
CMG	2	178.90**	0.01	0.01	10.76	19.67	26.61**	9.04**	0.26
STTP*CMG	2	102.83**	0.01	0.00	0.02	117.84*	2.56	0.99	0.69
Block	1	1.48	0.01	0.00	2.90	13.72	0.05	0.54	0.01
Error	11	18.89	0.03	0.01	23.77	130.62	3.55	3.22	0.91

* Significant at 5% level
** Significant at 1% level

Table 4 ANOVA: overall effect of independent variables on response variable.

Independent Variable	df	Sum of squares							
		Cook loss	Springiness	Cohesiveness	Chewiness	Hardness	L*	a*	b*
Treatment	6	373.75**	0.63**	0.05**	145.39**	1,343.48**	271.19**	51.39**	21.23**
Block	2	0.98	0.01	0.00	7.60	32.21	9.07*	1.24*	0.12
Error	12	3.63	0.03	0.01	20.53	224.39	10.95	1.13	2.00

* Significant at 5% level
** Significant at 1% level

Table 5 ANOVA: overall effect of independent variables on response variable.

Independent Variable	df	Sum of squares G′ (30 °C) (cooking)	G′ (70 °C) (cooking)	G′ (30 °C) (cooling)
Treatment	6	5,620,666.07*	18,959,329.17**	146,433,333.33**
Block	2	596,275.60	2,519,908.93*	14,616,488.10*
Error	12	2,234,653.57	3,291,261.91	16,296,845.24

* Significant at 5% level
** Significant at 1% level

Table 6 ANOVA: overall effect of independent variables on response variable.

Independent Variable	df	Sum of squares Cook loss	Springiness	Cohesiveness	Chewiness	Hardness	L	a	b
Treatment	2	2.06**	0.46**	0.00	10.01**	187.80**	53.45**	10.56**	0.08
Error	24	0.03	0.25	0.01	12.68	65.59	23.84	8.40	0.61

* Significant at 5% level
** Significant at 1% level

Table 7 ANOVA: overall effect of independent variables on response variable.

Independent Variable	df	Sum of squares					
		Color	Firmness	Elasticity	Juiciness	Flavor	Overall acceptability
Treatment	2	24.04**	19.66**	10.16**	5.88**	3.02*	3.94**
Block	5	14.37	15.94	10.13	16.58	14.28*	9.17*
Error	46	24.93	23.97	22.51	18.65	23.87	14.56

* Significant at 5% level
** Significant at 1% level

VITAE

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