EFFECT OF SWEET POTATO PECTIN ON MULTILAYER EMULSION STABILITY

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To my beloved parents, family and friends.

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ABSTRACT

Sweet potato (*Ipomoea batatas*) is a good alternative source of pectin which has the potential for stabilization of emulsion. This study aimed to optimize the extraction of pectin from sweet potato peels, and to investigate the effect of sweet potato pectin on multilayer emulsion stability. Response surface methodology was employed to optimize pectin extraction from sweet potato peels using citric acid. The factors included in the experimental design were temperature (60 − 100 °C), time (40 – 100 min) and pH (1 – 2). The yield of pectin was calculated on the basis of the sweet potato cell wall material. The degree of esterification (DE) of the pectin was analyzed using Fourier transform infrared spectroscopy. The extracted sweet potato pectin was incorporated in oil-in-water emulsion after homogenization using microfluidizer producing whey protein isolate-pectin multilayer emulsion. The effects of environmental conditions on the multilayer emulsion were studied by subjecting the emulsions to various conditions namely: 0 – 1.2 wt % pectin, pH 3 − 7 and 0 – 400 mM salt solution. The emulsion stability was characterized by determination of particle size, zeta-potential and creaming index (CI). The optimum conditions to extract the maximum yield of pectin of 65.8% were at extraction temperature 76°C, time 64 min and pH 1.2. The pectin was categorized as high-methoxyl pectin with DE of 58.5%. Decreasing of zeta-potential (+2.38 to –14.8 mV) showed the adsorption of pectin layer around the emulsion droplets. CI was the lowest (3.64%) at the highest pectin concentration (1.2 wt%) which showed great stability against creaming. Multilayer emulsion showed better stability at high pectin concentration and wide range of salt concentration compared to primary emulsion. Pectin addition largely improved the stability of emulsion especially at pH 4 and 5. Multilayer emulsion prepared using sweet potato pectin can be utilized to produce food emulsion with good stability against environmental pressure (pH and salt content) and can be further applied for controlled release of active ingredients in functional food and beverages.

ABSTRAK

Ubi keledek (*Ipomoea batatas*) adalah sumber alternatif bagi pektin yang berpotensi untuk menstabilkan emulsi. Kajian ini bertujuan untuk mengoptimumkan pengekstrakan pektin dari kulit ubi keledek dan mengkaji kesan pektin pada kestabilan emulsi dwi-lapis. Metodologi tindak balas permukaan digunakan untuk mengoptimumkan pengekstrakan pektin dari kulit ubi keledek menggunakan asid sitrik. Faktor yang termasuk dalam reka bentuk eksperimen adalah suhu (60 - 100 °C), masa (40 - 100 min) dan pH (1 - 2). Hasil pektin dihitung berdasarkan bahan dinding sel ubi keledek. Darjah pengesteran (DE) pektin dianalisa menggunakan spektroskopi inframerah transformasi Fourier. Pektin ubi keledek yang telah diekstrak dimasukkan ke dalam emulsi minyak dalam air selepas proses penghomogenan menggunakan mikropembendalir untuk menghasilkan emulsi dwi-lapis protein dadih susu terasing-pektin. Kesan persekitaran terhadap emulsi dwi-lapis telah dikaji dengan mengubah emulsi kepada pelbagai keadaan: 0 - 1.2 % berat pektin, pH 3 - 7 dan larutan garam 0 – 400 mM. Kestabilan emulsi dicirikan berdasarkan penentuan saiz zarah, potensi zeta dan pengukuran indeks pengkriman (CI). Keadaan optimum untuk mengekstrak hasil pektin yang maksimum iaitu 65.8% adalah pada suhu 76 °C, masa 64 minit dan pH 1.2. Pektin dikategorikan sebagai pektin metoksil tinggi dengan DE sebanyak 58.5%. Pengurangan potensi zeta (+2.38 hingga -14.8 mV) menunjukkan penjerapan lapisan pektin di sekitar titisan emulsi. CI adalah terendah (3.64%) pada kepekatan pektin yang tertinggi (1.2% berat) yang mana menunjukkan kestabilan yang baik terhadap pengkriman. Emulsi dwi-lapis lebih stabil pada kepekatan pektin tinggi dan pada julat kepekatan garam yang lebih luas berbanding emulsi asas. Penambahan pektin meningkatkan kestabilan emulsi terutama pada pH 4 dan pH 5. Emulsi dwi-lapis yang menggunakan pektin ubi keledek boleh digunakan untuk menghasilkan emulsi makanan dengan kestabilan yang baik terhadap tekanan persekitaran (pH dan kandungan garam) dan boleh digunakan untuk pembebasan terkawal bahan aktif dalam makanan dan minuman berfungsi.

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LIST OF ABBREVIATIONS

O/W	-	Oil-in-water
W/O	-	Water-in-oil
M-O/W	-	Multilayer oil-in-water
DE	-	Degree of esterification
HM	-	High methoxyl
LM	-	Low methoxyl
CI	-	Creaming index
GalA	-	Galacturonic acid
Rha	-	Rhamnose
O	-	Oxygen
C	-	Carbon
H	-	Hydrogen
Na^+	-	Sodium ion
Cl^-	-	Chlorine ion
NaCl	-	Sodium chloride
NaOH	-	Sodium hydroxide
HCl	-	Hydrochloric acid
SDS	-	Sodium dodecyl sulphate
pKa	-	Acid dissociation constant
pH	-	Measure of acidity/basicity
pI	-	Isoelectric point
WPI	-	Whey protein isolate
GC	-	Gas chromatography
SD	-	Standard deviation
n	-	Number of analysis
RSM	-	Response surface methodology

LIST OF SYMBOLS

~	-	Approximately
>	-	More than
<	-	Less than
±	-	Plus minus
α	-	Alpha
β	-	Beta
%	-	Percentage
wt	-	Weight
v/v	-	Volume per volume
M	-	Molar
mM	-	Milimolar
min	-	Minute
s	-	Second
°C	-	Degree celcius
:	-	Ratio
m	-	Meter
mm	-	Milimeter
µm	-	Micrometer
g	-	Gram
mg	-	Miligram
kg	-	Kilogram
L	-	Liter
mL	-	Mililiter
mg/L	-	Miligram per liter
rpm	-	Revolutions per minute
MPa	-	Megapascal
i.d	-	Inside diameter

CHAPTER 1

INTRODUCTION

1.1 Background of Study

Products such as milk, cream, salad dressings, soups, sauces, mayonnaise, butter and margarine are example of emulsions in the food industry. Conventionally, oil-in-water (O/W) emulsion is produced by homogenizing oil and aqueous phase together in addition of one or more emulsifiers (Friberg *et al.*, 2004; McClements, 2004). There are several limitations that can be attained by using current food emulsifier and traditional method of producing emulsion such as restricted stability to pH, salt, dehydration, heating and freezing. These limitations have driven researchers to carry out extended researches to come out with another method to improve the stability of emulsion. One of the methods is creating an interfacial layer around oil droplets that consists of several layers of emulsifiers and/or polyelectrolytes using a layer-by-layer electrostatic deposition technique that produces a multilayer emulsion (Guzey & McClements, 2006). The multiple layers provide strong steric repulsive and electrostatic forces which stabilize the emulsion against variety of environmental conditions such as temperature, pH and salt content (Guzey *et al*., 2004; Moreau *et al.*, 2003).

According to a study by Djordjevic *et al*. (2007), multilayer emulsion improved the stability of encapsulated limonene against oxidation. Previous studies on multilayer emulsion have been reported for encapsulation of lemon/orange oil, fish oil and β-carotene (Yang *et al.,* 2011; Klinkesorn *et al*., 2005; Hou *et al.,* 2011) which were found to improve the stability of the emulsion. In a multilayer emulsion, by manipulating the pH, salt concentration or temperature, the outer layer can simply be detached from the oil droplets. Therefore, provides flexible encapsulation abilities against different environmental conditions (Ogawa *et al*., 2003; Gu *et al*., 2005). In food industry, the highly-charged multilayer interface is useful to produce food emulsion that is stable against droplet aggregation and lipid oxidation. Besides, multilayer emulsion can be used as delivery system for controlled or triggered release of active ingredients in functional food (Guzey & McClements, 2006).

Polysaccharides are widely used as emulsifier for the production of food emulsions which include gum arabic, soy soluble polysaccharides (SSP), some galactomannans, modified starches, and cellulose, as well as several types of pectin (Chanamai & McClements 2001; Dickinson, 2003; Nakamura *et al*., 2004). Pectin is a polysaccharide consists of galacturonic acid that occurs in the cell walls of most plant. Pectin was widely used in the food industry due to its gelling, thickening and stabilizing properties (Vriesmann *et al.*, 2003; Yapo *et al.*, 2007; Takamine *et al.*, 2007). Commercial pectin is often extracted from citrus and apple pomace with yield of 15 to 30% and 10 to 15% respectively (Yapo & Koffi, 2013). Recent study has reported that industrial potato waste contains appreciable amount of rhamnogalacturonan I (hairy region of pectin) (Byg *et al.*, 2012). This opens the possibility to investigate the potential use of other crop residue material such as sweet potato peels, as an alternative pectin source. Several studies have been carried out to investigate pectin extraction from sweet potato using hydrochloric acid (Nurdjanah, 2008); Zhang *et al.,* 2013; Abang Zaidel *et al*., 2015, Foo *et al.*, 2016), sodium hydroxide (Nurdjanah, 2008; Abang Zaidel *et al.,* 2017) and disodium phosphate (Takamine *et al*., 2007; Nurdjanah, 2008).

1.2 Problem Statement

Since emulsions are thermodynamically unstable systems, ittend to be stabilized (coalescence, creaming) during food handling and processing. Emulsion breakdown is usually retarded by using emulsifiers, which are surface-active ingredients that adsorb to the surface of freshly formed lipid droplets during homogenization (Moreau *et al.,* 2003). Once adsorbed, itfacilitates further droplet disruption by lowering the interfacial tension, thereby reducing the size of the droplets produced during homogenization. There are various kinds of synthetic and natural emulsifiers that can be legally used in food emulsions, including small-molecule surfactants, phospholipids, proteins, and polysaccharides (Stauffer, 1999; Charlambous & Doxastakis, 1989; Krog, 1997). Each type of emulsifier has its own particular advantages and disadvantages. For example, some emulsifiers are effective at generating small emulsion droplets during homogenization, but are poor at providing long-term stability against droplet aggregation and vice versa (Stang *et al.*, 1994).

Consequently, there is no single emulsifier that is ideal for use in every food product. A number of studies have shown it is possible to improve emulsion stability by combining the beneficial attributes of different types of emulsifiers by forming multilayer emulsion (Ogawa *et al.*, 2003; Mao *et al.,* 2013, Gu *et al.,* 2004; Gu *et al.*, 2005). Multilayer emulsion can be produced by firstly using an ionic emulsifier that facilitates the formation of small droplets during homogenization. Then, an emulsifier with the opposite charge is introduced as the second layer that adsorbed to the droplets surface. The resulting droplets are coated by a two-layer interfacial membrane that may provide improved emulsion stability.

Pectin is a good potential emulsifier for food emulsions because it is already widely utilized in the food industry as an ingredient (Ngouémazong *et al.,* 2015). Commercial pectins are commonly extracted from citrus and apple. However, these two sources are not easily available in Malaysia as compared to sweet potato. It has been reported that sweet potato residues contain about 15% pectin on a dry matter basis (Mei *et al.,* 2010). Sweet potatoes are grown widely in Malaysia and are used to produce starchy food products in large food industry as well as small food business selling sweet potato snacks. Therefore, it is generating a lot of waste that could in turn pollute the environment. By using sweet potato residues as an alternative source of pectin, the waste can be converted into a value-added product that is profitable while reducing pollution to the environment.

Several studies reported that citric acid which is an organic acid, is an effective solvent for extraction of pectin in terms of the yield and its physicochemical properties (Canteri-Schemin *et al*, 2015; Pinheiro *et al.*, 2008, Kliemann *et al.*, 2009). Besides, citric acid is a natural and safe food additive and is thus more attractive than commonly used strong mineral acids such as nitric, hydrochloric and sulfuric acid for the extraction of pectin (Yapo, 2009). Extraction of pectin from sweet potato peels using citric acid would not only manage the disposal of sweet potato waste, but also reduce the impact of the corrosive effluents to the environment that was caused by the use of conventional acids during pectin extraction.

A lot of study regarding multilayer emulsion using pectin as emulsifier only focused on the commercial apple and citrus pectin (Benjamin *et al*, 2012; Benjamin *et al*., 2013, Moreau *et al*., 2003; Ogawa *et al*., 2004). Therefore, since pectin derived from sweet potato has the potential to be commercialized, this study aims to optimize pectin extraction from sweet potato peels using citric acid and to investigate multilayer emulsion stability as affected by sweet potato pectin.

1.3 Objective of Study

The objectives of this study are;

1. To optimize pectin extraction from sweet potato peels using citric acid.
2. To study the effect of sweet potato pectin concentration, pH and salt on multilayer emulsion stability.

1.4 Scope of Study

The scopes of this study are;

1. Extraction of pectin from sweet potato peels using citric acid at different conditions (temperature 60 to 100°C, time 40 to 100 min, pH 1 to 2) to optimize the sweet potato pectin yield using Response Surface Methodology (RSM).
2. Determination of degree of esterification of sweet potato pectin using Fourier Transform Infrared (FTIR) Spectroscopy.
3. Investigation on the effect of sweet potato pectin concentration (0 - 1.2 wt %), pH (3 - 7) and salt (0 - 400mM NaCl) on the multilayer emulsion stability using Dynamic Light Scattering for particle size and zeta-potential and measurement of creaming index.

1.5 Significance of Study

By conducting this study, sweet potatoes peels can be proven to be an alternative source of pectin that is available in Malaysia. This research can be the first step to the commercialization of sweet potato pectin which may open more opportunities in the food industry. Optimization of pectin extraction carried out in this study may increase the efficiency of the extraction. Besides, this study can provide the method and formulation of multilayer emulsion that utilizes pectin as a food-grade emulsifier. The study of emulsion stability at various environmental conditions can give more understanding regarding the characteristic of WPI-sweet potato pectin interfacial layer. In the future, this knowledge can be applied to control the thickness, structure and composition of the interfacial membrane of multilayer emulsion for food application.

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