CHAPTER 1

INTRODUCTION

Roselle (*Hibiscus sabdariffa* L) is commonly known as "kra-jeab" in Thai, calyx of roselle contains various antioxidants, such as anthocyanin, quercetin, L-ascorbic acid and proto-catechuic acid (Rice-Evans et al., 1996). Roselle anthocyanins are natural food colorant (E163) that has been approved on positive lists issued by Food and Drug Administration in the US, EU and Japan (Socaciu, 2008). Many biological activities of anthocyanins have been extensively reported, such as antihypertensive and cardioprotective agents, hepatoprotector, inhibitor against porcine pancreatic α-amylase and antioxidant capacity (Rice-Evans et al., 1996).

Anthocyanin pigments, in a model system, are highly affected by pH and solution system. They are in equilibrium of color (cationic) and/or colorless (pseudobase) structure. Moreover, other factors such as temperature, light, oxygen and hydrogen are also important in the stability of anthocyanins. In foods system, at high temperature, high sugar concentration, pH or ascorbic acid and others additives may affect the rate of anthocyanins destruction. Another factor that can give negative effect on anthocyanin stability is metal reactivity as anthocyanins form stable complex with tin (Sn), copper (Cu) and iron (Fe) (Patras et al., 2010).

Over the past decade, the use of a small particle has been studied extensively in biomedical and pharmaceutical application (Chen et al., 2006; Salata, 2004). By reducing particle size, the nanotechnology can contribute to improve the properties of bioactive compounds, such as delivery properties, solubility, prolonged residence time in the gastrointestinal tract and efficient absorption through cells (Sozer and Kokini, 2009). In recent report, Morris (2007) has shown that nanoparticle of carotenoids can be dispersed in water, allowing them to be added to fruit drinks providing improved bioavailability. However, no scientific finding has been reported on physicochemical properties of anthocyanains extracted from nano-particle plant materials compared to those prepared from traditional ground particles.

Nowadays, consumers' demand for natural products, as well as their concern over commonly use of synthetic antioxidants, suggests that it is important to identify functional natural antioxidants to use in meat and meat products. Lipid and protein oxidation is important to the meat industry because it is one of the major causes of quality deterioration which undergo auto-oxidation before meat is cooked. Lipid and protein oxidation can impart negative effects on

sensory attributes such as color, texture, odor and flavor, as well as negatively impacting the nutritional quality of the products (Nunez de Gonzalez et al., 2008; Raghavan and Richards, 2007). In the food industry, the most common approach to control oxidative rancidity is through the use of synthetic antioxidants such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), tert-butylhydroquinone (TBHQ) and propylgallate (PG) (Jayathilakan et al., 2007). However, the uses of these synthetic antioxidants at high concentration are undesirables in terms of toxicological effects (Sebranek and Bacus, 2007). Moreover, a list of some synthetic antioxidant seems incompatible with functional claims. Based on these reasons, food processors are seeking for natural antioxidants to replace the synthetic ones.

Roselle anthocyanins have been reported to be a good antioxidant (in terms of antilipoperoxidant activity) in ethnic meat products including Chinese-style sausage and pork chips with sucrose content lower than 10 %. However, high concentrations of sucrose (> 10 %) have been shown to negatively affect the antilipoperoxidant capacity of roselle anthocyanins when incorporated at 0.3 % (w/w) in Chinese-style sausage (Pinsirodom, 2008). In addition, when the xylitol was used to replace sucrose in Chinese-style sausage, the roselle anthocyanins efficiently exhibited antilipoperoxidant capacity. It is, therefore, of interest to further investigate whether different types of sugar alcohols will affect the antioxidant activity of roselle anthocyanins in the same manner.

The use of sugar alcohols as an alternative sweetener in meat products has not yet been reported elsewhere. The interest in application of sugar alcohols in food products is not only due to their low calorie and ability to improve blood sugar control, but also their excellent hydroxyl radical ($HO^{\bullet}$) scavenging capacity and *in vitro* inhibition of diazocompound-induced erythrocyte damage (den Hartog et al., 2010). Thus, sugar alcohols would be a promising sweetener for meat products especially when roselle anthocyanins will be incorporated as anti-lipid oxidation agent (Pinsirodom, 2008).

In the case of processed meat such as ham, bacon, frankfurter, bologna and others that are typical cured by addition of sodium/potassium nitrite or nitrate, which are used to control the growth of food born pathogens, provide cured meat flavor and color and extent shelf-life of products (Sebranek and Bacus, 2007). However, the concern about the potentially adverse health implications of the breakdown products of nitrate, i.e. nitrites and nitrosamines, have led the health authorities to set upper limits for nitrate in meat products and regulate the use of nitrate and nitrite as food additives. Thus, the reduction of the residual nitrite could be an acceptable

alternative to reduce the intake of nitrite in processed meats. The use of nitrate and/or nitrite has fallen under scrutiny due to their potential toxicological effects (Nunez de Gonzalez et al., 2008; Naveena et al., 2008). Plant extracts high in phenolic contents such as tomato and citrus co-products have been reported to be effective nitrite reduction agent (Østerlie and Lerfall, 2005; Viuda-Martos et al., 2009). It is possible to use the roselle anthocyanins extract to retard lipid and protein oxidation and at the same time reduce residual nitrite of meat products during storage. Successful application of roselle anthocyanins in meat products may enhance the value of roselle, thus expanding the utilization of the roselle as an inhibitor against lipid and protein oxidation and nitrite reduction agent.

The aim of this dissertation is to assess factors (the particle size of roselle extract, pH, sucrose and temperature) that can influence the physicochemical properties and antioxidant capacities of roselle extracts preparing from different extraction of dried roselle calyxes in model solution. The application of roselle extracts as natural antioxidant and nitrite residual reduction agent in meat products will also studied.

The objectives of this study were:

1 To determine physicochemical and antioxidant properties of roselle extracts prepared by original-grinding and nano-grinding method.

2 To evaluate antioxidant capacity of roselle anthocyanin extracts on lipid and protein oxidation as affected by sucrose and different sugar alcohols in Chinese-style sausage.

3 To investigate the ability of roselle anthocyanin extracts in scavenging of reactive nitrogen species in model system and evaluates their effect on nitrite reduction in meat products.