CHAPTER I

INTRODUCTION

1.1 Background

Food industries are expected to produce foods that are healthier, more nutritious, enjoyable, and attractive in their sensory properties to increase their worldwide demand. Color is one of the important attributes of foods as it is highlighted as sensorial attractiveness. Color directly affects the consumers' perceptions, desires, and considerations regarding food selection. The use of colorants has been applied for centuries in food products, namely as food colorants or color additives, to regain color losses after exposure to air, temperature, and light, enhance weak colors, and give color to colorless products. Both natural and synthetic food colorants are applied as food additives. Synthetic food colorants are more beneficial in terms of their production costs, stability, and usability, however, may produce adverse health problems. Therefore, the use of natural colorants has been increasing due to consumer awareness of their nutrition habits (Gençdag, *et al.*, 2022; Martins *et al.*, 2016).

Natural food colorants may be derived from plants, animals, minerals, and insects, providing environment-friendly and low or non-toxic colorants. However, there are several limitations on the utilization of natural food colorants, such as the lack of flexibility during processing and long-lasting effect because of their sensitivity towards pH, temperature, light, ascorbic acid, metals, and oxygen that may destruct the structure of the pigments (Jadhav and Bhujbal, 2020). Several main groups of natural food colorants include anthocyanins, carotenoids, carmine, riboflavin, betacyanins, and curcuminoids. Anthocyanin in acidic conditions appears in red, while in neutral pH appears in a purple hue, and in alkaline conditions appears in blue hue (Gençdag *et al.*, 2022). Anthocyanin represents one of the largest groups of phenolic compounds called flavonoids and is polar in nature. Temperature, pH, and light may affect the stability and cause the degradation of anthocyanins. Anthocyanins are also a good source of antioxidants, making them a good potential to substitute synthetic colorants due to their health benefits. Extraction using polar solvents by maceration method is done to obtain anthocyanin extract efficiently (Alappat, 2020; Oliveira and Antelo, 2020).

An example of a plant containing anthocyanin pigments is the butterfly pea (*Clitoria ternatea* L.) or *bunga telang*, which gives the plant an attractive blue color. It contains a specific anthocyanin, which is ternatins or also known as 3,5',5'-triglucoside (Suarna and Wijaya, 2021). Butterfly peas have been used worldwide as an edible natural colorant that can replace synthetic colorants. However, due to the instability of the anthocyanins, the application of substituting butterfly peas as a food colorant has been limited to short-shelf life products. The structure of anthocyanin may fade in color during processing causing economic losses. Therefore, various methods have been researched and developed to improve the stability of anthocyanins. The structural factors of anthocyanins can be modified using copigmentation, biosynthesis, and acylation method. To reduce the instability, encapsulation techniques that include microencapsulation, nanoparticles, and liposomes method are applied (Gençdag *et al.*, 2022).

As copigmentation produces stability on anthocyanins upon adverse environmental conditions, it has been widely studied for years, initially in winemaking. Copigmentation method is the non-covalent interaction between copigment and anthocyanins that can happen through self-association, intramolecular or intermolecular interaction, and metal complexes. Copigmentation process using copigment undergoes intermolecular interaction using the cofactors, such as, phenolic compounds, alkaloids, organic acids, amino acids, polysaccharides, etc. In addition, this process also induces variation in color, and therefore, is widely applied in many food products (Gordillo *et al.*, 2012).

Recently, relevant studies had found that polysaccharides are good potential for their stabilizing effect on anthocyanins. Macromolecules, as copigment agents, including gum Arabic, modified starch, kappa-carrageenan, pectin, alginate, chitosan, xanthan gum, and many more, have been investigated to enhance the stability of anthocyanins (Gençdag *et al.*, 2022). Research by Pinheiro *et al.* (2021) found that chitosan improved the thermal resistance of anthocyanins from *Pinot Noir* grape skins, therefore enhancing the stability. Another research by Zhao *et al.* (2020) found that xanthan gum, through intermolecular interactions, improves the color stability of black rice anthocyanins at a concentration of 0.25% (w/v). Also, xanthan gum improved the thermal stability of the black rice anthocyanins when exposed to high temperatures (80ºC, 90ºC, and 100ºC). The combination of various hydrocolloids had also been utilized for copigmentation of pomegranate juice in the research by Erkan-Koç *et al.* (2015). The combination of chitosan and xanthan gum with the concentration of 1% (w/v) obtained the best result by showing a higher copigmentation effect compared to other combinations, as both hydrocolloids prevented both anthocyanin and phenolic losses.

The effectiveness of copigmentation process relies on the type of copigment used, the molar ratio of copigment to anthocyanin, pH, and temperature (Rodriguez-Amaya and Carle, 2021). An optimum molar ratio is required because too high or too low copigment ratio may cause an inefficient copigmentation due to the disruption of the electron transfer process (Gordillo *et al.*, 2012). The utilization of xanthan gum and chitosan as copigments on butterfly pea extracts has not been researched yet, therefore, further analyses are needed. The effectiveness of chitosan and xanthan gum, or the combination of both, was tested. Therefore, this research aimed to find the most optimum molar ratio of copigment to anthocyanins to stabilize the anthocyanin extract of butterfly peas and also its antioxidant activity treated with different pH and temperatures.

1.2 Research Problem

Color is one of the important sensorial attributes of foods as it directly affects the consumers' perceptions, desires, and considerations regarding their food selection. The utilization of food colorants has been widely applied in food products, both synthetic and natural colorants. Synthetic colorants may cause adverse health problems, however, they are more beneficial than natural colorants in terms of their stability and production cost. Therefore, the use of natural colorants to substitute synthetic colorant is good potential in producing a safer food product.

Natural colorants may be derived from plants containing anthocyanin pigments, such as butterfly peas that produce an attractive blue color. Due to the instability of the anthocyanin pigments, the use of butterfly peas is still limited. The stability of anthocyanins and color intensity can be improved by copigmentation. Copigments that are derived from polysaccharides, such as chitosan and xanthan gum, have been proven to increase the stability of anthocyanins, however, have not been applied in butterfly pea anthocyanin extract. For that reason, this research was carried out to evaluate the effectiveness and find the best molar ratio of chitosan and xanthan gum as copigments in stabilizing the anthocyanin extract of butterfly peas, in terms of their color intensity, anthocyanin content, and antioxidant activity.

1.3 Objectives

1.3.1 General Objective

The general objective of this research was to utilize chitosan and xanthan gum as copigments of butterfly pea anthocyanin extract.

1.3.2 Specific Objective

The specific objectives of this research were:

1. To determine the best type of copigments (chitosan/xanthan gum/chitosan and xanthan gum) and the best ratio of pigment-copigment based on the analyses of thin layer chromatography, anthocyanin content, antioxidant activity, color intensity, and hyperchromic and bathochromic effect.
2. To determine the effect of different temperatures on the stability of butterfly pea anthocyanin extract based on the analyses of anthocyanin content, color intensity, and antioxidant activity.

3. To determine the effect of different pH on the stability of butterfly pea anthocyanin extract based on the analyses of anthocyanin content, color intensity, and antioxidant activity.