CHAPTER 1

Introduction

Aromatic rice from Thailand has famous among rice in the world because this cooked rice has soft, tender texture and flavor [1]. The unique flavor in aromatic rice is formed by incorporating of many aroma compounds [2]. The 2-acetyl-1-pyrroline (2AP) is the mainly aroma compound that gives the aromatic rice has flavor like pandan (*Pandanus amaryllifolius*) [3]. The valuable of Thai fragrant rice is due to its aroma. However, the reproducibility of aroma compounds especially 2AP from rice grain in each cultivation time and area is not reached. The fragrant rice grains from different planting areas have various content of 2AP. Until now, there have been many research studies about the important parameter effect on aroma compounds in aromatic rice such as genetic factor [4-6] and environmental factors [7-11]. For this work, the primarily research, the environmental factors are considered. Although some nutrients reported to effect 2AP in rice are nitrogen (N) and salt stress, no one simultaneously studied these factors. At this moment, soil factors are known to affect aroma of rice in ways not properly defined. The simultaneously studying of important factors effect on 2AP content in rice is interest.

1.1 An important aroma compound; 2-acetyl-1-pyrroline (2AP)

The volatile compounds that provide aroma and flavor in rice have been studied for decades. The 2-acetyl-1-pyrroline (Figure 1.1) in the term of a key odor-active compound to contribute flavor in rice and method to determine 2AP were reported by Buttery *et al*. in 1982 [12]. Later that, Laksanalamai *et al*. [2] and Bryant *et al*. [1] reported that the 2AP was not found in non-aromatic rice.

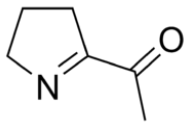

Figure 1.1 Chemical structure of 2AP

Many researchers have been tried to study the biological formation of 2AP in the fragrant rice [5, 6]. Yoshihashi *et al*. [5] reported that the nitrogen source of 2AP was proline, whereas the carbon source of acetyl group was not the carboxyl group of proline. Moreover, there have the reports about biosynthesis pathways due to the BADH2 enzyme as shown in Figure 1.2 [6]. However, the biosynthesis pathway of 2AP in fragrant rice was not properly defined.

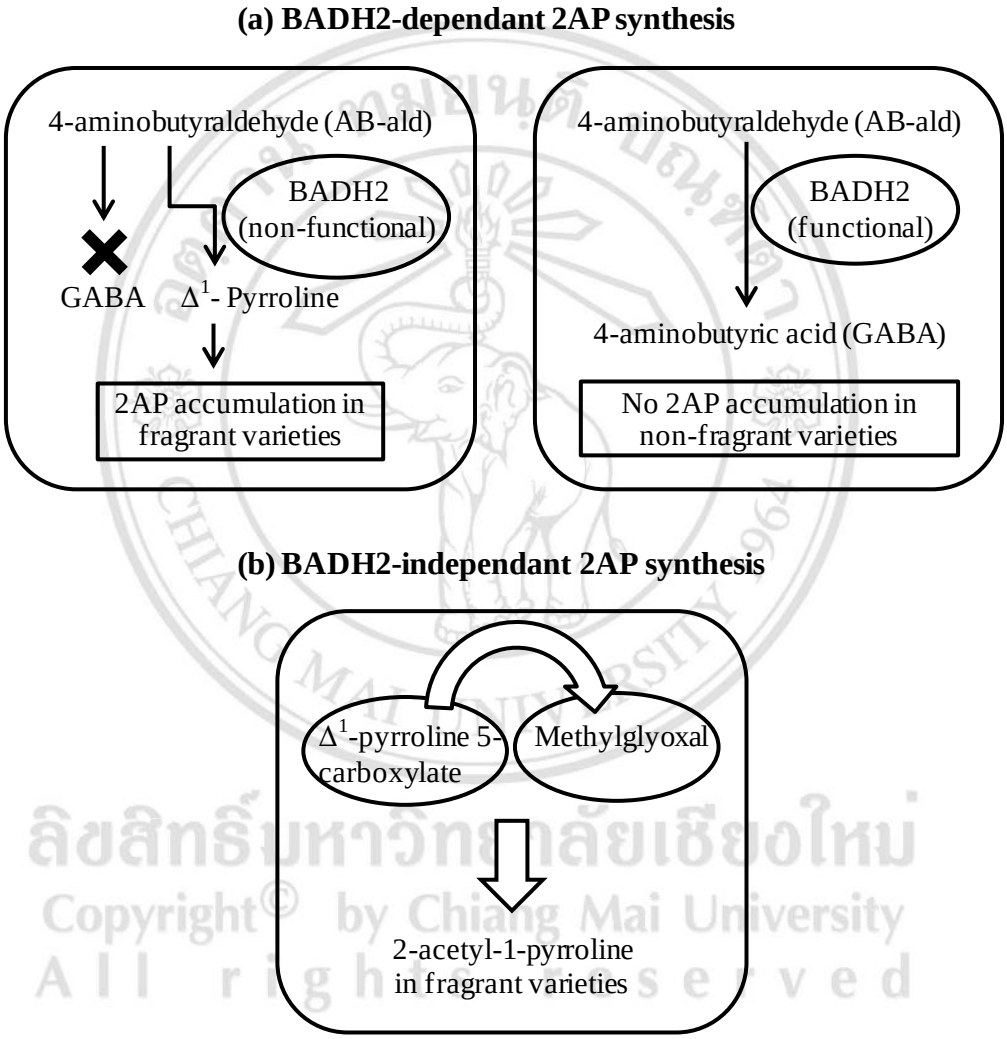

Figure 1.2 Pathway of 2AP biosynthesis in rice (a) BADH2-dependant 2AP synthesis [13, 14], (b) BADH2-independant 2AP synthesis [15].

1.2 Environmental factors effecting on aromatic compounds in rice grain

Some reports claimed that N content in soil and soil salinity were among the key factors in producing of the aroma in aromatic rice [2, 6]. An amount of N fertilizer applied could boost the growth of the rice plant but this might reduce the aromatic quality of the grain. On the other hand, soil salinity may have contributed to the concentration of this aromatic flavor. So far, the effects of N fertilization and salt salinity were investigated separately. The effect of N rate applied was not taken into account the consequence of the salinity treatments, and vice versa, however these factors in fact may not be a sole factor.

1.3 Hydroponics

Hydroponics is a method for growing plant in other material, without soil. The hydroponic plants were feed from mineral nutrient solutions. Hydroponics is suitable for people who live in city that do not have enough area for cultivation. Moreover, it is the way to prevent the contamination of pesticide residue in food from soil. From this advantage, we hydroponically planted the rice due to the convenient controlling of amount of nutrient in each treatment and convincing that did not have interferes from the remained nutrients in soil. In this research, the water culture, the simplest of hydroponic systems was used as show in Figure 1.3.

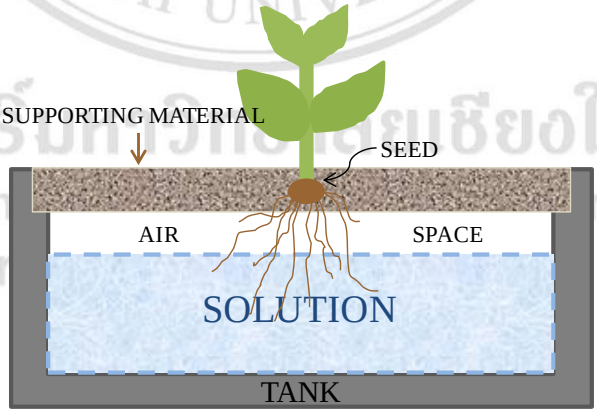

Figure 1.3 Water culture system for growing the rice plant

1.4 Chemometrics

Chemometrics is the combination of applied mathematics, multivariate statistics and computer science to interpret the data or solve the problem in chemistry. Chemometrics may have categorized in design of experiments (DOEs), multivariate calibration, classification, exploratory data analysis and process monitoring. In this research, design of experiment and multivariate calibration were used. The methods were described in Chapter 2.

1.5 Literature review

Oryza sativa L. ssp.indica cv. Pathum Thani 1 or Pathum Thani1 (PT1) has been well known among Thai jasmine rice. Although the aromatic quality of PT1 somehow may not compete with Khao Dawk Mali 105 (KDML 105), the most recognized Thai jasmine rice, PT1 has very much the same delicious flavor and the texture properties of the cooked rice. PT1 plays an important role in Thai economics because it is a short-day rice or photoperiod-insensitive rice which means that the rice cultivation could be this rice can be planted at any time of a year whereas KDM 105 can be grown only once a year particularly during the rainy season. Also, PT1 has the reputation for its resistance to some important diseases such as leaf blight and pest such as brown plant hopper [16].

A number of studies have investigated on rice flavor chemistry and it was found that 2-acetyl-1-pyrroline (2AP) was identified as a key odor-active component in rice grains [2]. The content of this aromatic component in grains of rice heavily contributed to the satisfaction of consumers [4]. Two main factors that contribute to the aromatic flavor in fragrant rice are genetic and environmental factors. As a matter of fact that it would be not easy to modify the rice genetics accordingly, this research will put the attention on the latter factor.

For fragrant rice cultivation, it is believed that growing conditions such as fertilizer rate applied and soil salinity have been shown to affect the aromatic quality of the grains, however only few studies have done in order to confirm this investigation. This could be due to the fact that most of the studies in the previous time were intended for the optimum yield not for the scent. Some studies have investigated effect of varied N fertilization on yield and 2AP contents of some the aromatic cultivars. Still, the

reports showed contradictory evidences on whether or not the fertilizer content was key factors in producing the strong aroma. For example, Srivastava *et al*. [17] reported that aroma strength of a Basmati rice (Taraori variety) was inversely related to the nitrogen content in soil. On the contrary, Yang *et al*. [11] reports that the higher the total N content of the soil, the stronger the aroma of the grain. However, this study did not focus on the content of 2AP and the aroma quality instead was monitored using some other chemical compounds and a sensory test.

Besides the rate of N fertilizer, soil salinity was also identified as another issue for the quality of the aromatic rice production. Gay *et al*. [8] reported that salinity may have contributed to the concentration of this aromatic flavor but the consequence of applying the salt stress is the lower in grain yield. Performance of aroma and yield of KDML 105 in some soil series in Tung kula rong hai was investigated. Although, the results revealed that sodium and soluble sodium could be related with the aroma of KDML 105, the rigid interpretation of results could not be provided due to the complexity of data collected. Up to now, the effects of N rate and salt salinity were not clearly investigated. Also, it is not clear what the relationship between the N rate applied is and soil salinity that affects to the content of this aromatic flavor and also the grain yield.

Chemometrics is the application of mathematical and statistical methods to data that is chemical in nature in order to obtain relevant information [18]. There is a variety of applications that chemometrics can be useful such as experimental design [19], quality control [20], pattern recognition/classification [21], multivariate calibration [22], variable selection [23] and data exploratory [24]. Although chemometrics is more closely related to analytical chemistry since there is the need to deal with a huge amount of data, which provides a great opportunity for chemometrics. In fact, the need for chemometrics and its tools can be observed in many life sciences where data interpretation is of a great interest such as physical chemistry [25], biology [26], environment [27], agriculture [4] and engineering [28].

In this research, chemometrics will be used as a tool to ease the analysis of data collected from a greenhouse-scale experiment. PT1 rice will be cultivated in pots using a modified Hoagland's nutrient solution applying different rates of N fertilization and

different salinity treatments (NaCl). The experiment will be designed using a central composite design (CCD) in order to investigate the relationship between the studied parameters and their effects. The 2AP contents in rice grains will be determined using gas chromatography-nitrogen phosphorous detector (GC-NPD). Based on design data, growth parameters such as number of tillers, length of root and height of plant and some yield parameters will be used to establish a multivariate prediction model called partial least square regression (PLS). This model will be used to estimate the content of 2AP in grains of the cultivated rice. Using PLS coefficient, it is possible to investigate the important parameters effecting on the 2AP content and the behaviors of these selected parameters will be confirmed and visualized using supervised self-organizing map (SSOM). It is important to note that ideally this experiment should be done using KDML 105; however, as a pilot study PT1 will be used since it is photoperiod-insensitive rice and its disease resistances.

1.6 Research objectives

1) To predict the 2AP content in PT1 rice grains based on the design data and some growth and yield parameters using chemometric technique such as PLS.
2) To investigate the effects of N and Na on the 2AP content in grains of PT1 rice.
3) To evaluate the important parameters effecting on the 2AP content in PT1 rice grains using PLS and SSOM.