International Journal of Agricultural Technology 2024 Vol. 20(3):1017-1036
Available online http://www.ijat-aatsea.com
ISSN 2630-0192 (Online)

Differences in growing region affected grain yield, grain quality and 2-acetyl-1-pyrroline content in Thai jasmine rice (*Oryza sativa* L.)

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Dangthaisong, P., Sookgul, P., Suwannachin, K., Seiyot, A., Arikit, S., Wanchana, S. and Malumpong, C. (2024). Differences in growing region affected grain yield, grain quality and 2-acetyl-1-pyrroline content in Thai jasmine rice (*Oryza sativa* L.). International Journal of Agricultural Technology 20(3):1017-1036.

Abstract Thai jasmine rice var Khao Dawk Mali 105 (KDML105) rice traits in six provinces of Central, Northern, Western and Northeastern Thailand for two years was investigated. The results showed that the total variation in each trait of KDML105 was influenced by the location and year. In addition, the agronomic traits, grain yield and 2-acetyl-1-pyrroline (2AP) content among the six provinces including Pathum Thani, Nakhon Pathom, Chiang Rai, Chiang Mai, Surin and Nakhon Nayok were significantly different. In 2018, the grain yield in Chiang Rai (2506 kg/ha) was highest, while, in 2019, Chiang Mai (4063 kg/ha) and Chiang Rai (3950 kg/ha) had the highest in grain yield. When considering the 2AP content, it was found that Chiang Rai (4.12 ppm) and Surin (3.86 ppm) had the highest values, while, in 2019, Chiang Rai, Surin and Chiang Mai had the highest values at 5.83, 5.48 and 5.12 ppm, respectively. However, the amylose content and gelatinization temperature were not different. The reasons for the differences in grain yield and aroma levels among the provinces were due to air temperature, altitude, soil properties and salinity. However, mild drought stress and nitrogen from chemical fertilizers had no effect on either the grain yield or aroma levels of KDML105 rice. Thus, it can be concluded that the high levels of 2AP in KDML105 rice in the Northeastern region may be due to the natural low fertility of sandy soil, mildly saline soil, while the high 2AP content in the northern region resulted from the low temperature and high altitude.

Keywords: Aromatic rice, Topography, Microclimate, Soil properties

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Introduction

Aromatic rice varieties are very important in the Greater Mekong Subregion and South Asia and have gained wider acceptance in many countries around the world (Hori *et al*., 1992; Callingacion *et al*., 2014). Thai jasmine or Thai Hom Mali rice variety, also known as KDML105 rice, which was released to farmers in 1959, is the most popular fragrant rice from Thailand and is considered in the international market to have high cooking quality and unique aroma (Chinachanta *et al*., 2020). According to statistics on Thai jasmine rice exports from 2016 to 2019, Thailand's overall fragrant rice exports decreased by 32%, while Vietnam's decline was only 9%. In addition, the inability to maintain the top position as the best aromatic rice in the world in both 2018 and 2019 also caused the reputation of Thai fragrant rice to decline (Neo, 2023).

Among the numerous volatile compounds associated with the aroma in rice, the main aromatic compound has been identified as 2-acetyl-1-pyrroline (2AP) (Buttery *et al*., 1983), produced under the control of the recessive gene *badh2* (Bradbury *et al*., 2005). However, the 2AP content and aroma vary greatly depending on the environment (Vanavichit *et al*., 2018). The 2AP content also varies according to temperature, rainfall, day length, light intensity, humidity, soil type and fertility (Khush and Juliano, 1985). Yoshihashi *et al*. (2004) confirmed that the ecological conditions of an area, such as weather and soil conditions, were highly correlated with the quality of fragrant rice. Therefore, KDML105 rice grown in different parts of Thailand showed variation in aromatic content due to different climatic conditions, topography and cultivation management (Yoshihashi *et al*., 2004). In addition, the grain and cooking qualities of fragrant rice in Thailand vary greatly (Seanrungmueang *et al*., 2009).

KDML105 rice must be grown from traditional photoperiod-sensitive varieties that determine its flowering date around October, and farmers grow KDML105 rice in lowland rainfed areas during the rainy season only (Kumboonreang, 2011). KDML105 rice cultivated in different locations was subdivided into five areas as upper Central region, upper Northeast region, lower Northeast region, lower Central/Northeast region, lower Northern region and the upper Northeast region (Pitiphunpong and Suwannaporn, 2009). However, KDML105 rice is mainly grown in Northeastern (NE) Thailand, accounting for 80% of the total production (Lilavanichakul *et al*., 2021), followed by Northern (N) Thailand (Vanavichit *et al*., 2018). KDML105 rice is not successfully grown in the central plain due to the widespread occurrence of various diseases and insect pests in irrigated areas (Vanavichit *et al*., 2018). Yoshihashi *et al*. (2004) reported that the lowland rainfed area in the in Tung Kula Rong Hai (Surin, Yasothon, Maha Sarakham, Buriram and Roied) was the optimal environment for the accumulation of 2AP content in rice grain. In addition, the Tung Kula Rong Hai had the best rice quality in terms of physical appearance and cooking characteristics (Pitiphunpong and Suwannaporn, 2009).

KDML105 rice is not a well-known drought-tolerant variety, but it consistently expresses root growth adaptability to water stress via greater root branching in shallow soil layers (Kano-Nakata *et al*. 2013). However, KDML105 rice was adaptive to slightly to moderately salt-affected soils, with an electrical conductivity (EC) of 2–8 dS m^{-1} (Arunin and Pongwichian, 2015). The influence of these stresses on aroma quality has been reported, and drought (Yoshihashi *et al*., 2002; Dangthaisong *et al*., 2023) and salinity stress (Poonlaphdecha *et al*., 2012; Dangthaisong *et al*., 2023) have been shown to lead to higher 2AP contents in grains at the ripening stage. One prominent problem with KDML105 rice has been the reduction in 2AP content due to various production management, topography, microclimate and soil properties, causing the loss of competitive advantages and market share. Thus, the research aimed to evaluate the grain yield, grain quality and 2AP content of KDML105 which grown in different environments and cultivation practices in Central, Western, Northern and Northeastern regions in Thailand.

Materials and methods

Experimental sites and field preparation

An experiment was conducted in the rainy season (August–November) in 2018 and 2019 in transplanted paddy fields in lowland rainfed areas. In 2018, the experiment was conducted in four provinces, Pathum Thani, Nakhon Pathom, Chiang Rai and Surin, which represented the Central, Western, Northern and Northeastern regions, respectively. In 2019, the experiment was conducted in six provinces, including the same provinces in 2018, and Chiang Mai (northern region) and Nakhon Nayok (central region) were added. Details on the topography, planting date and harvesting date in each province are shown in Figure 1. The experiment was laid out in a randomized complete block design with eight replications in both years.

KDML105 rice seeds were obtained from the Rice Department, Ministry of Agriculture and Cooperative of Thailand. Twenty-day-old seedlings of KDML105 rice were transplanted by farmers at a rate of 3–5 plants per hill at a spacing of 25 x 25 cm The uniform areas in the field were used to represent the subplots for data collection, with a size of 2 × 5 m The locations were divided into two types based on fertilization practice. Chiang Rai and Surin were experimental areas for growing rice using organic fertilizer (90% chicken manure and 10% bagasse) applied at 500 kg/ha, while other areas grew rice following the conventional method using chemical fertilizers. Chemical fertilizer was applied via the conventional method at a rate of 75 kg N/ha (diammonium phosphate), 37.5 kg of P_2O_5/ha, and 37.5 kg of K_2O/ha. At the tillering stage, fertilizer was applied again at a rate of 37.5 kg N/ha (urea). Rain was used to manage the field, with the water level not exceeding 20 cm from 30 d after planting until 15 d after the flowering stage.

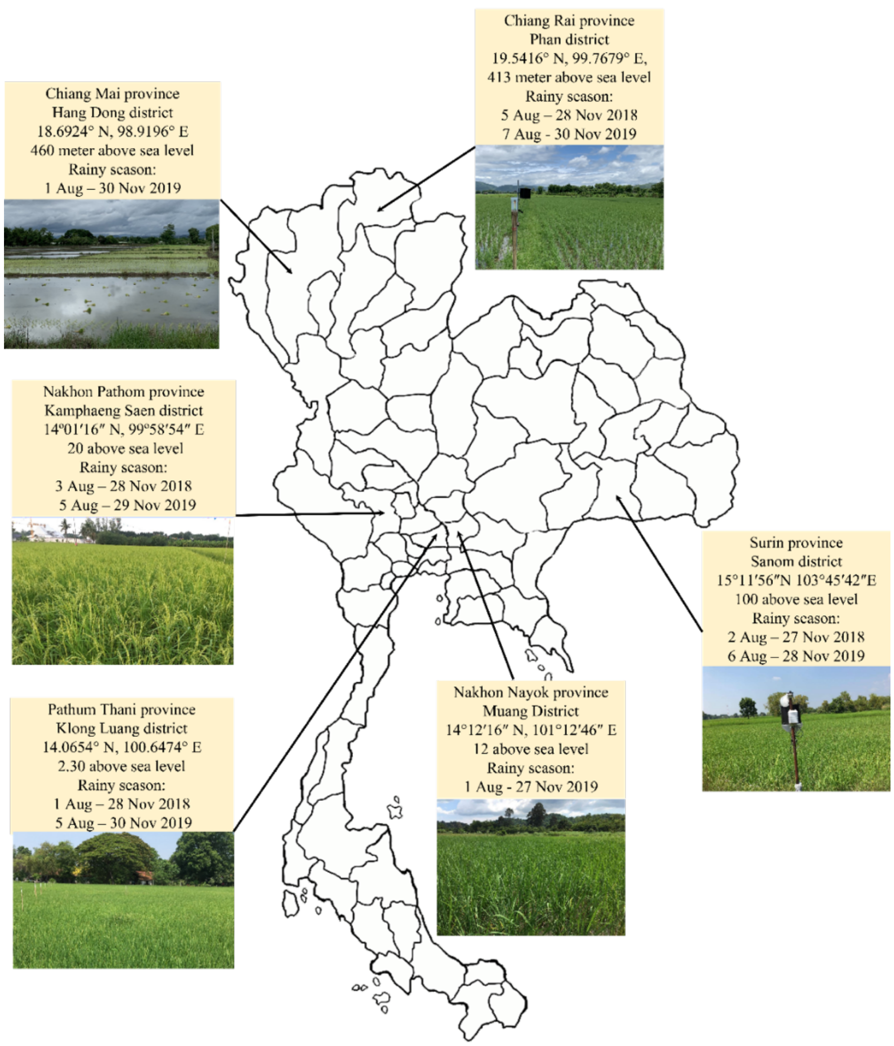

Figure 1. Six provinces representing the northern, northeastern, western and central regions where KDML105 rice was investigated in the rainy season in 2018 and 2019

Weather data and soil analysis

A data logger (WatchDog 2000 Series Micro Stations; Spectrum Technology Inc.; USA) was used to record daily air temperatures (minimum and maximum) and the amount of rain throughout the experimental period. The soil properties including organic matter, available phosphorus, available potassium, cation exchange capacity (CEC), soil pH and soil type) were analyzed. In addition, samples of organic fertilizers used by farmers in Chiang Rai and Surin provinces were analyzed to determine pH, electrical conductivity (EC), sodium, organic matter, carbon/nitrogen ratio, % moisture by weight, germination index, total nitrogen, total phosphorus (P_2O_5), total potassium (K_2O) and particle size. The analysis of soil and organic fertilizer was performed by the soil laboratory at the Department of Soil Science, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Thailand.

The agronomic traits were recorded as plant height, panicles per plant, filled grains per panicle, unfilled grains per panicle, percentage of seed set, 1000-grain weight and grain yield per ha. These traits were determined for rice plants grown during the field experiment. The grain yield in each subplot was determined per 10 m^2 of harvested area. The grain moisture was adjusted to 14% and then extrapolated to units of kg/ha.

Grain quality was evaluated using grains harvested from each location. The dried grains were stored at room temperature for one month prior to grain quality trait evaluation. The paddy grains were dehulled and polished using a minipolisher. Two chemical grain qualities, gelatinization temperature and amylose content, were evaluated following the procedures described by Juliano (1985). In addition, the samples of paddy rice grain from each subplot were evaluated for the percentage of head rice derived from milled rice with lengths longer than or equal to three-quarters of the average length of the entire milled rice.

Analysis of 2AP content in rice grains

The 2AP content in rice grains was evaluated over two seasons in each location and determined following Pitija *et al*. (2021). After harvest, the paddy grains were dehulled for brown grains and stored at -80 °C before analysis. Then, the brown grains were finely ground in liquid nitrogen, and the samples (0.50 g) were separately placed in headspace vials. Static HS-GC analyses were carried out using chromatography/NPD (Clarus 680 model) and GC/MS (SQ8 model, PerkinElmer Ltd., USA), a headspace autosampler, a flame ionization detector (FID), and a nitrogen-phosphorus detector (NPD). A sample headspace was collected through a 3-mL sample loop and automatically transferred to the GC instrument via a heated transfer line. A split/splitless GC injector equipped with a direct 0.2-mm i.d. glass liner was used. The system operation, data acquisition, collection including system evaluation were accomplished using Agilent Chem Station software versions A.01.04 and B.01.03 (Agilent Technologies, Waldbronn, Germany). The content of 2AP was recorded in ppm.

Statistical analysis

Data were analyzed using R version 3.6.1 to test the significance of the differences in agronomic traits, grain quality, and 2AP content. As the error variances for the two years were homogenous, a combined analysis of variance of data for the two years was performed. The means were separated using Duncan's test at an alpha level of 0.05. In addition, multiple correspondence analysis (MCA) was conducted in R version 3.6.1 to describe the relationship of variables with grain yield and 2AP content and to visualize the distances between the categories of the qualitative variables.

Results

Weather and soil properties

The microclimatic data for the four sites in the rainy season in 2018 (August to December) and six sites in the rainy season in 2019 (August to December) are shown in Table 1. The duration of the growing period in each year was approximately 120 d The flowering date of KDML105 rice in all regions in both years was approximately 16 to 20 October. The range of maximum temperatures in the rainy season in 2018 in the four locations was 31.4–34.9 °C, while the range of minimum temperatures was 22.3–25.2 °C. In the rainy season of 2019, the range of maximum temperatures was 31.5–33.8 °C, and the range of minimum temperatures was 23.2–25.8 °C. Thus, the range of maximum and minimum temperatures between the two seasons was not different. When comparing the temperature among locations, it was found that Chiang Rai had the lowest temperature, while Pathum Thani had the highest temperature in both years. The maximum temperature differences between Chiang Rai and Pathum Thani were 3.5 °C (2018) and 2.3 °C (2019), respectively. In addition, the minimum temperature differences between Chiang Rai and Pathum Thani were 2.9 °C (2018) and 2.6 °C (2019), respectively.

The total rainfall amounts during the growing period in 2018 and 2019 were 291–601 mm and 217–1489 mm, respectively. The highest total rainfall amount in 2018 was recorded in Nakhon Pathom (601 mm) and Chiang Rai (600 mm), while the lowest was recorded in Surin (291 mm). In 2019, Nakhon Nayok had the highest total rainfall of 1489 mm, while Nakhon Pathom Province had the lowest total rainfall of 217 mm. In addition, the mean relative humidity in all locations in the two years ranged from 78–80% RH (Table 1).

The soil at the six locations was classified into four soil textures, namely, clay soil in Chiang Rai, Pathum Thani and Nakhon Nayok, loam soil in Nakhon Pathom, clay loam soil in Chiang Mai and sandy loam in Surin. The soil chemicals differed among the six locations in the two years. The organic matter was in the range 0.24–2.93%, available P in the range 8.07–28.92 mg/kg, exchangeable K in the range 10.43–419.73 mg/kg, exchangeable Ca in the range 216.28-3457.47 mg/kg, exchangeable Mg in the range 19.55–616.74 mg/kg, electrical conductivity (EC) in the range 0.22–3.62 ds/m and soil pH in the range 4.93–6.69 (Table 1). Thus, the soils in every location were identified as neutral to slightly acidic. When considering EC, it was found that the soil in Surin was slightly salt affected, while the other sites were not found to have saline soil. In addition, organic matter in Surin, especially in 2019, was lower than recommended, while Pathum Thani had the highest organic matter. The available P and exchangeable K in Surin in 2018 were lower than the recommended soil values, while the average values of available P and exchangeable K in other sites were in the range or higher than the soil recommendation.

Fertilization properties

In this experiment, organic fertilizer was applied in Chiang Rai and Surin, while other provinces used chemical fertilizers. The results of organic fertilizer components compared to those of chemical fertilizer are shown in Table 2. The organic matter in the granular organic fertilizers applied in Chiang Rai and Surin was 39.48% and 42.21%, respectively. However, the pH (8.06 and 7.93) and EC (4.33 and 5.32 ds/m) of the granular organic fertilizers were high, while the macronutrients, including N (1.72 and 2.15%), P_2O_5 (0.26 and 2.33%) and K_2O (1.92 and 3.62%), were low in Chiang Rai and Surin, respectively. In contrast, the chemical fertilizer applied in other locations had only macronutrients in high amounts, namely, N (12.0%+46.0%), P_2O_5 (6.0%) and K_2O (6.0%).

Agronomic traits and grain yield

KDML105 rice has a photoperiod sensitivity that determines its flowering date around October. In this study, the flowering dates in the six locations ranged from 17 to 21 October, and the day length ranged from 11:45 to 11:42 h (Figure

2). KDML105 rice grown in Chiang Rai and Chiang Mai had an earlier flowering date than the other locations.

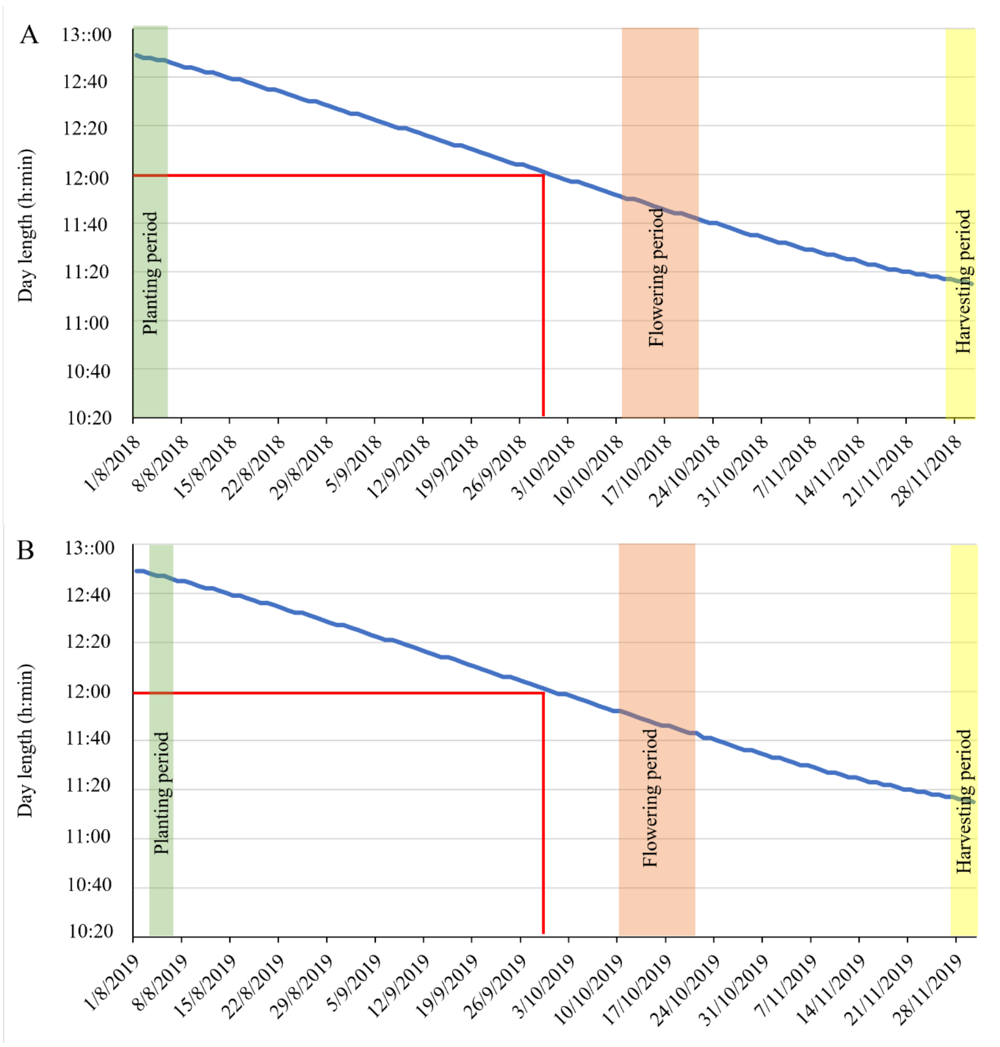

Figure 2. Day length (h:min) during KDML105 rice planting from August to November 2018 and 2019. The green box is the planting period, the orange box is the flowering period, and the yellow box is the harvesting period

A combined analysis of variance of agronomic traits was conducted for four sites in two years and is presented in Table 3. Regarding main effects, year effects were significant ($P<0.01$ and $P<0.05$) for most traits except filled grains/panicle, seed set and 1000-grain weight, while the location effect was significant for all traits ($P<0.01$) except filled grains/panicle. In addition, the effect of year × location was also significant ($P<0.01$ and $P<0.05$) for all traits except unfilled grains/panicle and seed set.

When considering the total variation in each trait, it was found that all traits were influenced by location. However, year (51.98%) and location (31.36%) both had a great effect on the 2AP content. In addition, the effects of interactions also made major and moderate contributions to total variations in plant height (46.28%), panicles/hill (21.73%) and grain yield (25.92%) (Table 3).

Some agronomic traits of KDML105 rice varied among the six sites and two years, as shown in Table 4. The plant height and panicles/hill ratio were significantly different among sites and years. In 2018, the plant height in Pathum Thani was shortest (127 cm), while the plant height in Chiang Rai was highest (163 cm). However, the rice plants that grew in Nakhon Pathom and Nakhon Nayok in 2019 were the shortest at 108 and 109 cm, respectively, while rice plants in Pathum Thani were the tallest (153 cm). High tillering ability and effectiveness of panicles were found in Chiang Rai both in 2018 and 2019 (14 and 18 panicles), but the panicles/hill in Surin was very low in both years (7 and 10 panicles). When considering the other traits, it was found that filled grains/panicle, seed set and 1000-grain weight were slightly different among sites and were not different between years.

The paddy grain yields varied among the locations and years. The mean grain yield in 2018 (2098 kg/ha) was lower than that in 2019 (2726 kg/ha). In 2018, KDML105 rice grown in Chiang Rai and Nakhon Pathom produced the highest grain yields of 2506 kg/ha and 2306 kg/ha, respectively, while the grain yield in Surin was the lowest (1469 kg/ha). In 2019, the grain yield in Chiang Rai (3950) was still the highest together with that in Chiang Mai (4063 kg/ha), but the grain yield in Surin (1669 kg/ha) was still the lowest together with that in Nakhon Pathom (1531 kg/ha) and Nakhon Nayok (1756 kg/ha) (Figure 3A).

2AP content and grain quality

In 2018, the brown rice of KDML105 rice in Chiang Rai and Surin had the highest 2AP content in grains of 4.12 and 3.86 ppm, respectively, whereas Nakhon Pathom and Pathum Thani had 2AP contents of 2.46 and 2.37 ppm, respectively (Figure 3B). However, the 2AP content in 2019 was higher than that in 2018 at every location. The 2AP contents of KDML105 brown rice in Chiang Rai, Surin and Chiang Mai were not significantly different and were the highest, 5.83, 5.48 and 5.12 ppm, respectively, whereas the brown rice grown in Nakhon Pathom, Nakhon Nayok, and Pathum Thani had the lowest 2AP contents of 3.06, 3.20 and 3.81 ppm, respectively.

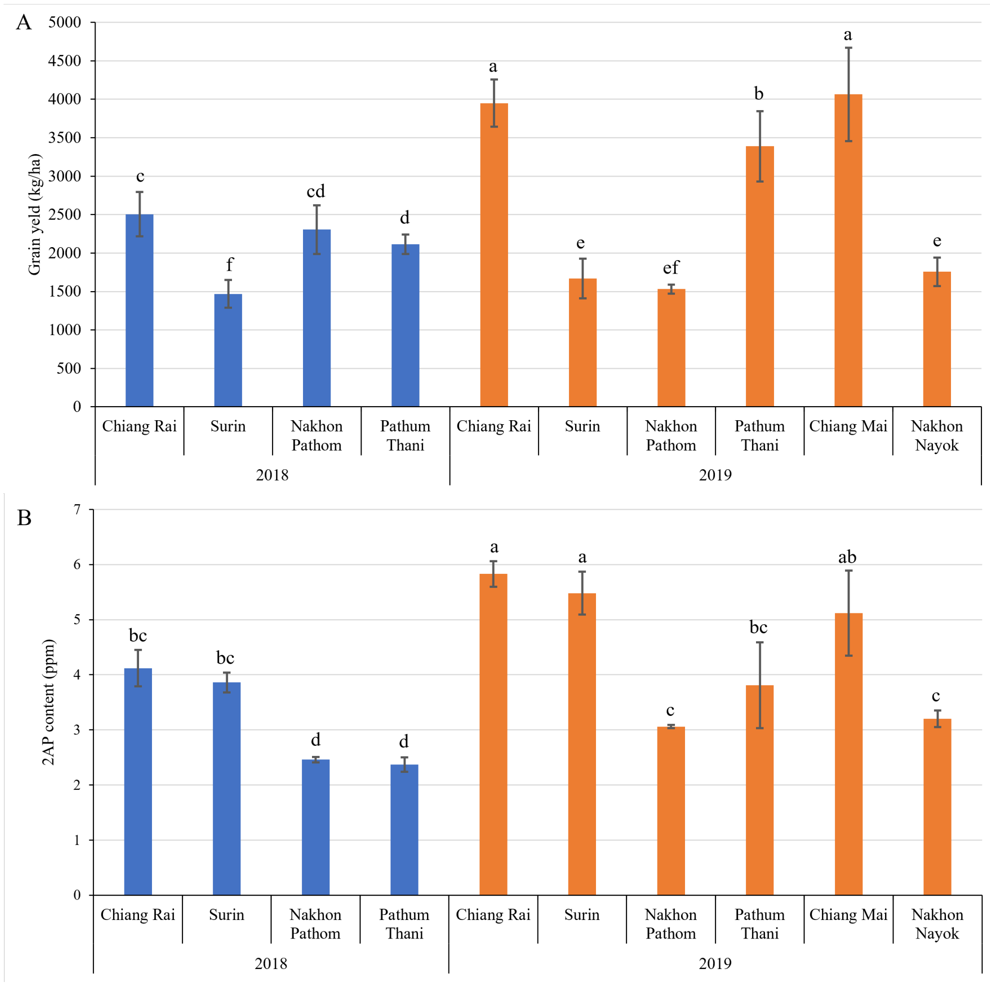

Figure 3. Grain yield and 2AP content of KDML105 rice compared among six provinces in the rainy seasons of 2018 and 2019

The percentage of head rice compared among locations in each year was significantly different. However, the percentage of head rice across years in each location was not significantly different, except for Pathum Thani. In 2018, the highest head rice yield was found in Pathum Thani (52%), followed by Chiang Rai (50%), Nakhon Pathom (49%), and Surin (43%). In 2019, Chiang Mai had the highest head rice yield (52%), while Nakhon Nayok had the lowest (44%) (Table 4).

The chemical grain properties were not significantly different among locations or between the two years. The mean amylose content of KDML105 rice in 2018 was 17.6%, while in 2019, it was 17.7%. In addition, the alkaline value in the two years was 7, denoting a low gelatinization temperature (Table 4).

Multiple correspondence analysis

Multiple correspondence analysis (MCA) classified locations into four groups based on grain yield and 2AP content (Figure 4). Group I included Chiang Rai and Chiang Mai, which were classified as having a high grain yield and high 2AP content, and Group II included Surin, which was classified as having a low grain yield and high 2AP content. Group III was classified as having moderate grain yield and 2AP content, and Pathum Thani was classified into this group. Finally, Group IV was classified as having low grain yield and moderate 2AP content, and Nakhon Pathom and Nakhon Nayok were classified into this group. In addition, the grain yield and 2AP content in the 2018 season were lower than those in the 2019 season.

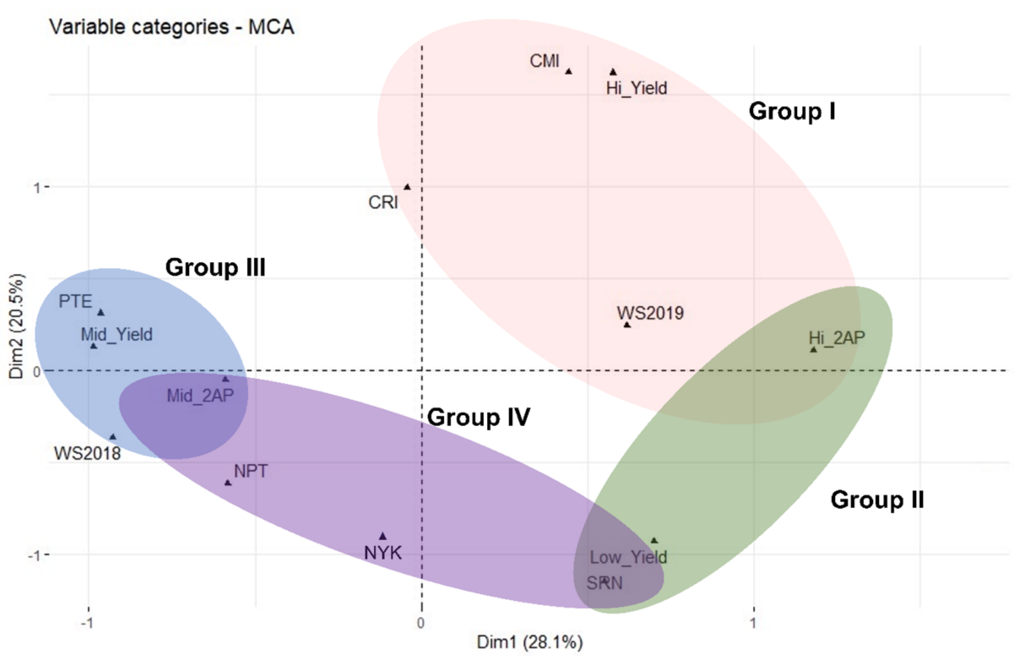

Figure 4. MCA of grain yield and 2AP content among six provinces in the rainy seasons of 2018 and 2019 that were divided into four groups. Group I: high yield and high 2AP content, Group II: low yield and high 2AP content, Group III: low yield and moderate 2AP content; and Group IV: moderate yield and moderate 2AP content

Table 1. Soil properties and weather conditions in six provinces compared with the suitable conditions for paddy rice in the rainy seasons of 2018 and 2019

Soil properties	Chiang Rai		Surin		Nakhon Pathom		Pathum Thani		Chiang Mai[1]	Nakhon Nayok[1]	Suitable condition for rice
	2018	2019	2018	2019	2018	2019	2018	2019	2019	2019	
Soil properties:											
pH	5.40	5.42	4.93	6.05	5.80	6.69	5.68	6.69	6.60	6.19	5.5-6.0[1/]
EC (ds/m)	0.22	0.36	2.92	3.62	0.48	0.58	0.47	1.09	0.66	0.47	<2.0[1/]
Organic Matter (%)	1.56	1.40	1.08	0.24	1.56	2.93	2.78	2.93	2.43	1.47	>1.5[1/]
Available P (mg/kg)	26.70	20.01	8.07	10.07	29.45	20.10	18.16	20.10	11.83	28.92	>15[1/]
Exchangeable K (mg/kg)	73.54	88.19	10.43	11.46	59.30	419.73	72.18	419.73	142.14	189.32	>20[1/]
Exchangeable Ca (mg/kg)	629.90	533.66	216.28	809.50	1120.20	3457.47	3113.48	3457.47	2115.92	722.97	-
Exchangeable Mg (mg/kg)	76.50	54.13	50.83	35.00	69.73	554.75	616.74	554.75	19.55	53.95	-
Soil texture	Clay	Clay	Sandy loam	Sandy loam	Loam	Loam	Clay	Clay	Clay loam	Clay	Clay, Silt clay[1/]
Weather conditions:											
Amount of rainfall (mm)	600	557	292	643	601	217	514	414	693	1489	700-1500[2/]
Amount of evaporation (mm)	383	450	480	472	496	481	524	420	401	410	-
Maximum temp (°C)	31.4	31.5	33.0	31.6	32.9	32.7	34.9	33.8	32.0	31.9	24-36[3/]
Minimum temp (°C)	22.3	23.2	23.8	24.2	24.0	24.9	25.2	25.8	23.7	25.4	>15[3/]
Mean temp at seedling (°C)	26.2	27.5	28.3	28.2	28.1	28.5	29.0	29.4	27.5	29.5	25-30[3/]
Mean temp at tillering (°C)	26.2	27.1	28.7	27.6	28.2	28.4	29.4	29.1	27.7	29.9	25-31[3/]
Mean temp at flowering (°C)	25.7	27.1	28.4	28.2	27.5	29.6	28.8	31.1	28.5	31.2	30-33[3/]
Mean temp at ripening (°C)	23.2	24.3	27.4	26.2	28.0	28.0	29.0	28.8	25.2	29.0	20-29[3/]
Mean of relative humidity (%RH)	80.5	78.0	76.8	78.0	79.1	78.0	80.1	78.0	79.0	79.0	50-90[3/]

[1] Sys *et al*. (1993), [2] Bhuiyan (1992), [3] Yoshida (1978)

Table 2. Fertilization properties compared between granule organic and chemical fertilizers

Locations/Fertilizer properties	Chiang Rai	Surin	Nakhon Pathom	Pathum Thani	Chiang Mai	Nakhon Nayok
Fertilizer	Organic	Organic	Chemical	Chemical	Chemical	Chemical
	-	-	12-6-6 46-0-0	12-6-6 46-0-0	12-6-6 46-0-0	12-6-6 46-0-0
pH, 1:2	8.06	7.93	-	-	-	-
EC (ds/m)	4.33	5.32	-	-	-	-
Na (%)	0.23	0.20	-	-	-	-
Organic Matter (%)	39.48	42.21	-	-	-	-
Organic carbon (%)	22.90	24.48	-	-	-	-
C/N Ratio	13.31	11.39	-	-	-	-
Moisture by weight (%)	16.29	12.21	-	-	-	-
Germination Index (%)	25.52	30.71	-	-	-	-
Total N (%)	1.72	2.15	12.00 46.00	12.00 46.00	12.00 46.00	12.00 46.00
Total P_2O_5 (%)	0.26	2.33	6.00 -	6.00 -	6.00 -	6.00 -
Total K_2O (%)	1.92	3.62	6.00 -	6.00 -	6.00 -	6.00 -
Particle size (mm)	100	100	-	-	-	-
Gravel size > 5 mm, (%)	None	None	-	-	-	-
Plastic, Glass, etc, (%)	None	None	-	-	-	-

Table 3. Combined analysis of variance for agronomic traits, grain yield, grain quality and 2AP content of KDML105 rice grown in four locations in the rainy seasons of 2018 and 2019

Source of Variance	Year	Rep within year	Location	Year × Location	Error
df	1	14	3	3	42
Plant height	2150.6**	4.9ns	2375.2**	2920.5**	16.4
Respective SS (%)	11.36	1.09	37.64	46.28	3.63
Panicles/hill	16.0**	0.6ns	166.2**	54.5**	1.34
Respective SS (%)	2.13	2.33	66.31	21.73	7.51
Filled grains/panicle	9.0ns	74.0ns	2464.1ns	282.0**	62.4
Respective SS (%)	0.08	7.60	62.83	7.19	22.30
Unfilled grains/panicle	81.0*	10.9ns	215.7**	44.9ns	19.1
Respective SS (%)	4.23	13.15	33.79	7.03	41.80
Seed setting	21.4ns	6.95ns	134.4**	11.84ns	7.52
Respective SS (%)	2.43	12.72	45.82	4.04	34.98
1000 grain weight	0.009ns	0.021ns	13.58**	1.25*	0.25
Respective SS (%)	0.02	3.53	70.28	6.50	19.67
Grain yield	4599952**	50210ns	9248710**	4270006**	77019
Respective SS (%)	9.31	1.76	56.15	25.92	6.86
Head rice	17.02*	2.34ns	51.14**	11.05**	2.30
Respective SS (%)	2.93	9.88	60.72	13.12	13.35
2AP content	20.4**	0.22ns	4.10**	1.52**	0.11
Respective SS (%)	51.98	1.38	31.36	11.66	3.61

Mean square value; ns, nonsignificant, * and ** significant at $P< 0.05$ and $P< 0.01$, respectively. % Respective SS included the effects of Y + L+ Y×L

Table 4. Agronomic traits and grain quality of KDML105 rice compared among six provinces in the rainy seasons of 2018 and 2019

Agronomic and grain quality traits	Year	Chiang Rai	Surin	Nakhon Pathom	Pathum Thani	Chiang Mai	Nakhon Nayok	Mean	F-test	C.V. (%)
Plant height (cm)	2018	163a[1/]A[2/]	132cA	142bA	127dB	-	-	141	**	3.21
	2019	137cB	119dB	108eB	153aA	143b	109e	128	**	2.43
	Mean	150	126	125	140	-	-	135		
Panicles/hill	2018	14abA	7cB	13bA	15aA	-	-	12	**	9.67
	2019	18aB	10cA	9cB	14bA	15b	9c	13	**	9.19
	Mean	16	9	11	15	-	-			
Filled grain/panicle	2018	118a	127a	92c	102b	-	-	110	**	8.64
	2019	120b	114bc	98e	104de	139a	111cd	114	**	5.52
	Mean	119	121	95	103	-	-	112		
Unfilled grains/panicle	2018	27bA	28bA	33aA	22cA	-	-	28	**	17.52
	2019	26bcA	29bA	35aA	29bB	24c	27bc	28	**	12.62
	Mean	27	29	34	26	-	-	28		
Seed set (%)	2018	81a	82a	76b	81a	-	-	80	**	3.02
	2019	82b	81b	75d	77cd	85a	80bc	80	**	3.47
	Mean	82	82	76	79	-	-	80		
1000 Grain weight (g)	2018	26.6a	25.4b	25.5b	24.4c	-	-	25.5	**	2.00
	2019	26.9b	25.2c	24.8c	24.9c	27.6a	24.0d	25.6	**	2.05
	Mean	26.8	25.3	25.2	24.7	-	-	25.6		
Head rice (%)	2018	50bA	43cA	49bA	52aA	-	-	49	**	3.15
	2019	49bA	45dA	47cA	48cB	52a	44e	48	**	2.24
	Mean	50	44	48	50	-	-	49		
Amylose content (%)	2018	17.5	17.8	17.5	17.6	-	-	17.6	ns	2.15
	2019	17.7	17.9	18.1	17.4	17.6	17.7	17.7	ns	1.93
	Mean	17.6	17.9	17.8	17.5	-	-	17.7		
Alkaline test (score)	2018	7	7	7	7	-	-	7	ns	1.50
	2019	7	7	6	6	7	7	7	ns	1.50
	Mean	7	7	7	7	-	-	7		

[1/]Different lowercase letters in the same row indicate significant differences at the 0.05 level using Duncan's test.
[2/]Different uppercase letters for each trait in the same column indicate significant differences at the 0.05 level using Duncan's test

Discussion

The study hypothesized that weather, altitude, soil properties and fertilizer type may affect the grain yield, 2AP content and grain quality of KDML105 rice grains. An experiment was conducted in fields in Central, Northern, Western and Northeastern Thailand for two years to observe and investigate the production of KDML105 rice and its traits.

KDML105 rice grown in distinct locations displayed significant variations in grain yield and aromatic content (2AP), but the grain quality was uniform. This result is consistent with Sangwongchai *et al*. (2021) that the climatic conditions, soil type, soil fertility and cultivation practices play significant roles in controlling grain yield and 2AP content in KDML105 rice.

The 2AP content in KDML105 rice in Surin, which is located at Tung Kula Rong Hai, was high over two years, but the 2AP content between the two years was different. However, KDML105 rice grown in the Northern regions of Chiang Rai and Chiang Mai had the same high 2AP content as that grown in Surin. In addition, Central and Western Thailand (Pathum Thani, Nakhon Nayok, Nakhon Pathom) had the lowest 2AP content. Therefore, this experiment confirms the reported of the 2AP content from 7 locations in Thailand by Yoshihashi *et al*. (2004) that the 2AP content in Northeastern Thailand, especially in Tung Kula Rong Hai, was higher than that in other locations. In addition, the 2AP content in central Thailand was the lowest (Pathum Thani). However, the survey study by Changsri *et al*. (2015) reported that the range of 2AP content in KDML105 rice in Northeastern Thailand in 2012 was 1.04–10.69 ppm. In addition, Phuseerit and Siriamornpun (2020) reported that the 2AP content in KDML105 rice from three sites in Tung Kula Rong Hai ranged from 5.00–5.47 ppm. These reports confirm the results of this experiment that 2AP content is variable across locations and years.

When considering the air temperature during the growing period, it was found that the temperatures of all six locations were in the optimum range for rice growth (Yoshida, 1978). However, the mean temperature at the ripening stage, which is an important period for 2AP accumulation in grains was 2–5 °C lower in Chiang Rai than in other locations. Thus, this may have resulted in higher 2AP content than in the Central and Western regions. These results are consistent with those of Dela Cruz *et al.* (1989) and Itani *et al.* (2004), who reported that the 2AP content was higher in brown rice ripened at a low temperature (day: 25 °C/night: 20 °C) than in brown rice ripened at a high temperature (day: 35 °C/night: 30 °C).

KDML105 rice planted in Chiang Rai and Chiang Mai, which are located at high altitudes, showed higher 2AP contents than KDML105 rice planted in the Northeastern, Central and Western regions. This result is consistent with those of Lorieux *et al.* (1996), who noted that KDML105 rice was usually harvested at the beginning of winter (November) to obtain a strong aroma content. Moreover, Nakamura (1998) reported that aromatic rice cultivated at a higher altitude than usual had a higher 2AP content than that cultivated at a low altitude.

Yoshihashi *et al.* (2002) and Dangthaisong *et al.* (2023) found that drought stresses during cultivation have led to higher 2AP contents in rice grains. These reports were contrast with this study. The amount of rainfall in all locations was lower than the recommended condition, except for Nakhon Nayok. In addition, less rainfall was observed in Surin in 2018 and in Nakhon Pathom in 2019, and there was less evaporation. Thus, KDML105 rice grown in Surin in 2018 and in Nakhon Pathom in 2019 was affected by mild drought conditions. However, the 2AP content in Nakhon Pathom was the lowest, while 2AP content in Surin was the highest. Moreover, the soil physicochemical properties in Surin were identified as slightly acidic sandy loam, slightly to salt-affected with lower fertility (OM, P and K) than recommended. The reported of Singh *et al*. (2003) supported that lighter soils with low nitrogen were generally perceived to favor aroma formation in rice. In addition, the 2AP content in rice grown in sandy soils is higher than that in rice grown in loamy soils, followed by clayey soils (Saetung and Trelo-ges, 2017). Moreover, Poonlaphdecha *et al*. (2012) and Dangthaisong *et al*. (2023) supported that slightly saline soil has led to higher 2AP contents in rice grains.

The total nitrogen from chemical fertilizer in this study did not increase the 2AP content in rice grains. However, Chiang Rai and Surin, which used granule organic fertilizer, showed a high 2AP content in grains. This result contradicted with Yang *et al*. (2012), who reported that total nitrogen in the soil is one of the key factors increasing the 2AP content of Chinese aromatic rice. However, this result showed the same as Chinachanta *et al*. (2020), who suggested that chemical fertilizer addition is not very successful, as this practice exerts a negative effect on the 2AP content of KDML105 rice. In addition, several studies have shown that organic inputs can enhance the 2AP content compared to chemical fertilizer applications (Saha *et al*., 2007; Surekha *et al*., 2013; Chinachanta *et al*., 2020).

In terms of grain yield, the average yield of KDML105 rice is 2268 kg/ha (Rice Department, 2010). The grain yield in Surin and Pathum Thani in 2018 was lower than the average yield. The grain yield in Surin, Nakhon Pathom, and Nakhon Nayok in 2019 was lower than the average yield, and the grain yield in Surin was the lowest in both years.

The grain quality, including the percentage of head rice, amylose content and alkaline test to determine gelatinization temperature for all locations, was in accordance with the standard for Thai Hom Mali (Ministry of Agriculture and

Cooperative, 2018). This means that these characteristics were not affected by the different environments.

This study hypothesized that temperature, altitude, soil properties, stress condition and fertilizer type may affect the grain yield, 2AP content and grain quality of KDML105 rice grains. An experiment was conducted in fields in Central, Northern, Western and Northeastern of Thailand for two years to observe and investigate the production of KDML105 rice and its agronomic traits. In this study, KDML105 seeds were obtained from the same source; thus, the genotype had no effect on the variability in 2AP content and yield. Therefore, it was confirmed that the 2AP content was variable between the growing seasons and among locations. When consider the environment factors, temperature and altitude had affect to the increase 2AP content in grains, which can be confirmed the results from Chiang Rai and Chiang Mai. In addition, other factors also cause high 2AP contents in addition to temperature and altitude, as a high 2AP content was also observed in Surin. Moderate of saline soil and others soil properties may affect the 2AP content of aromatic rice that can be observed in Surin. However, moderate drought condition and the addition of chemical fertilizers cannot stimulate the generation of 2AP in the pathway. To explain the higher 2AP content during salt stress, Kavi Kishor *et al*. (2005) suggested that a high level of proline is biosynthesized via the intermediate Δ1-pyrroline-5-carboxylic acid by both glutamate and ornithine pathways during salinity stress. Thus, the increased proline content also resulted in higher 2AP synthesis. In terms of grain yield, drought, salinity (Dangthaisong *et al*., 2023) and infertile soil conditions (Chinachanta *et al*., 2020) were likely the reasons for the low yield, especially in Surin. In addition, it was confirmed that KDML105 rice was not suitable for planting in the Central region due to the low yield and low 2AP content (Vanavichit *et al*. 2018).

Acknowledgements

The authors would like to thank the Rice Department, the Ministry of Agriculture and Cooperatives, Thailand where supported their germplasm for the experiments. We thank to the Rice Science Center at Kasetsart University, Kamphaeng Saen Campus, Thailand for providing the facilities for this study.

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(Received: 6 April 2023, Revised: 11 March 2024, Accepted: 15 March 2024)