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Selection of Indigo Plant Varieties and Other Plants that Yield Indigo Dye

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Abstract

This study was to investigate the four indigo plant varieties and other plants that give indigo dye. The findings revealed that Nakhon Panom variety gave the greatest yield. The leaf produce was 5520 kg/Acre A followed by Kalasin variety, Mukdahan variety and Sakon Nakhon variety with leaf yield of 4560, 4320 and 3920 kg/Acre, respectively. Kalasin variety was proved to be the best. This variety gave the highest amount of indigo dye, 0.022 g per 100 g of indigo leaves. Mukdahan variety, Sakon Nakhon and Nakhon Phanom variety gave 0.017, 0.015 and 0.015 g of indigo dye respectively. The difference was not statistically different. From the study the plant that could be used to supplement the indigo plants was *Marsdenia tinctoria* R. Br. which gave 0.025 g per 100 g of indigo leaves at 40 °C, whereas *Memecylin edule* Roxb. had high acidity, it did not give the color of indigo dye.

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Keywords: selection; indigo plant varieties; other plants that yield indigo dye

1. Introduction

The dye from an indigo plant (true indigo) (*Indigofera tinctoria* L.) which human beings use to dye cloth has been known long for 2,000 years. Its blue color is distinctly attractive and impressive for those who touch. No wonder, it was named “the King of Dyes” [1]. In Thailand, the indigo color was brought to dye cloth long time ago, especially in the North and North-East Handicrafts [2] and natural color-dyed

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fabrics have been promoted and developed as OTOP (one tambon, one product) products in Sakon Nakhon province Indigo-dyed fabrics were selected as the Province's OTOP products. That resulted in gaining wider acceptance from consumers. In addition to such promotion, some group of weavers of indigo-dyed fabrics has developed such the products and did distributed the products throughout the country and abroad such as Italy, Japan and France which were interested in and ordered more and more indigo-dyed fabric products from Sakon Nakhon province [3]. The indigo dye, thus, was increasingly needed. Indigo plants comprised several varieties and other plants that yield the indigo color is needed to be done for development of high quality of indigo-dyed fabrics. It is a response to Sakon Nakhon province's policy and strategy for becoming "the City of Indigo".

2. Materials and Methods

2.1 A study of growth and production of the indigo plant

A experimental plan using the Randomized Complete Block Design (RCBD) was made comprising 4 treatments and 4 blocks: treatment 1 (Nakhon Phanom variety), treatment 2 (Sakon Nakhon variety), treatment 3 (Kalasin variety), and treatment 4 (Mukdahan variety); plot size of $1 \times 5 \text{ m}^2$, spacing $25 \times 60 \text{ cm}^2$, fertilizing Bogashi compost after planting for every 4, 5, 6, 7, 8, 9, and 10 weeks with an amount of 5 kg/plot. Rate of growth, plant height, number of branches and sub-branches were recorded. Its fresh indigo leaves begin to be harvested when the indigo plant is 4 months old. Indigo leaves are harvested for 2 times: the first time from observation of the dark-green colour of the lower surface of mature leaves, and the second time when the indigo leaves grow mature after 21 days following the first time of harvest. All the produce of fresh indigo leaves is weighted.

2.2 A study of a quantity of indigo color from leaves of each of the 4 varieties

A experiment plan using the Complete Block Design (CBD) of 4 treatments and 3 replications comprises treatment 1 (Nakhon Phanom variety), treatment 2 (Sakon Nakhon variety), treatment 3 (Kalasin variety) and treatment 4 (Mukdahan variety). Indigo leaves were selected and collected from each variety of indigo plants grown on the first experiment plot as determined by being dark-green and mature. Fresh mature leaves of each indigo plant variety weighting 100 g were put in 1000 ml beaker which was filled with 350 g of water. They were soaked for 24 hr. Then, the waste of indigo leaves was separated from the water. Indigo solution was weighted. It was stirred by a stirrer for 2 hr until the indigo solution became constant dark-blue. The dark-blue indigo solution weighting 10 g was filtered using dried filter paper through a vacuum pump. The residue was washed with water for 2 or 3 times. Then, the indigo content was heated to $105 \text{ }^\circ\text{C}$ for 2 hr in a desiccators. Then, it was brought to weight and heated 30 min. more. It was brought to weight again; repetitions were done until its weight was constant. After that, an amount of indigo powder of each indigo plant variety was calculated.

2.3 A study of other plants that yield the indigo color

From a survey of local plants which yield the indigo color through in-depth interviews with those local people whose prowess of using other plants that yield the indigo color and of using them in mixing with the indigo color was found outstanding, both varieties of plants named *Marsdenia tinctoria* R. Br. and *Memecylon edule* Roxb., each were investigated. The plants of both varieties which yield the indigo color were grown for a study of botany characteristics by analyses of solutions derived from soaking plant leaves. The leaves of *Marsdenia tinctoria* R. Br. and *Memecylon edule* Roxb., each were picked from the plants. The separated leaves of both plants weighting 100 g for 3 replications were put into the beakers of 1000 ml and filled with 350 g of water. Then, it was soaked in the room temperature and at the $40 \text{ }^\circ\text{C}$ for being as long as 24, 30, and 36 hr, respectively. The waste of both varieties plants' leaves was separated

from each beaker. The solutions were then filtered to make clean. The pH value levels of solutions of both varieties plants' leaves were measured. Solutions of *Marsdenia tinctoria* R. Br s' leaves, was stirred by a stirrer for 2 hr. until solution of *Marsdenia tinctoria* R. Br. s' leaves became constant dark-blue. One solutions of the *Marsdenia tinctoria* R. Br. s' leaves which was dark blue, weighting 10 g were filtered by dried filter paper through a vacuum pump. *Marsdenia tinctoria* R. Br. residue gained was washed with water for 2 or 3 times. Then, it was heated to the temperature of 105 °C for 2 hr. *Marsdenia tinctoria* R.Br. was weighted and heated for more 30 min. *Marsdenia tinctoria* R. Br. was weighted repeatedly until the weight was constant. The amounts of dye powder from *Marsdenia tinctoria* R. Br. leaves at each temperature level and the lengths of soaking leaves time were calculated. The *Memecylin edule* Roxb. leaves on the contrary cannot yield the indigo color.

3. Results and Discussion

3.1. A study of growth and produce of 4 indigo plant varieties: Kalasin, Mukdahan, Sakon Nakhon, and Nakhon Phanom

From the comparisons of 4 indigo plant varieties, their heights were found no significant difference. The heights average of Mukdahan variety was 109.26 cm. which was highest. The heights average of other varieties saying Sakon Nakhon variety, Nakhon Phanom variety and Kalasin variety were 107.96, 107.39 and 102.74 cm, respectively.

From the comparisons of 4 indigo plant varieties, each variety's main branches and sub branches of all the 4 indigo plant varieties were found a significant difference among them. The most branch-producing indigo plant variety was Sakon Nakhon variety producing as many as 35.19 branches, while Nakhon Phanom variety, Kalasin variety, and Mukdahan variety yielded 30.85, 22.89, and 22.83 branches, respectively.

As the indigo leaf produces for each of the 4 varieties was compared to one another, it was found a significant difference. The highest produce of fresh indigo leaves belonged to Nakhon Phanom variety, yielding 5520 kg/Acre; while the Kalasin variety, the Mukdahan variety and the Sakon Nakhon variety yielded fresh indigo leaves of 4560, 4320 and 3920 kg/Acre, respectively (Table 1).

Table 1. To show plant height, Number of leaf branch and Leaf yield

Treatment (variety)	Plant height (cm.)	Number of leaf branch (branch)	Leaf yield (kg./Acre)
Kalasin	102.74	22.89 c	4560 ab
Mukdahan	109.26	22.83 c	4320 b
Sakon Nakhon	107.96	35.19 a	3920 b
Nakhon Panom	107.39	30.85 b	5520 a
F-test	Ns	*	*
CV%	8.64	3.80	13.24

Compare the difference by Duncan's new multiple range test ns = no significance

The Nakhon Phanom variety whose pods are straight was chosen to be the indigo plant that yielded the most amount of fresh leaves. Though, its branches had fewer branches than the Sakon Nakhon variety which yielded the same type of straight pods, its leaves were found bigger than the Sakon Nakhon, Kalasin, and Mukdahan varieties. Moreover, the Nakhon Phanom variety had much more smaller l. That makes the produce of fresh leaves weight more than other varieties. As the varieties that produce curved pods like the Kalasin and Mukdahan varieties were compared to each other, it was found that the Kalasin variety yielded more indigo leaves. This is because the size of the Kalasin variety indigo leaf is bigger

than the Mukdahan variety. As both varieties eaves of the compound leaf than other varieties from 3-21 leaflets were compared with the same curved pod variety such as the Sakon Nakhon variety, it was found that the Sakon Nakhon variety yielded the highest produce. From the study of botany characteristic, an increase of leaves produce, color quantity and dye powder of the curved pod indigo plants, it was found that growing the curved pod indigo plant with a spacing 25×60 cm would result in fresh indigo leaves produce as much as 10,949.31 kg/Acre [4]. Thus, in case of choosing straight pod indigo plants for producing a dye, the Nakhon Phanom variety is recommended; while the curved pod indigo plant variety that yields the highest produce is the Sakon Nakhon variety.

3.2 A test of an indigo dye amount of each of the 4 varieties

From the comparison of an indigo dye amount per 100 g of leaves among the 4 varieties, it was found that there was no significant difference in statistics. However, the indigo dye amount of the Kalasin variety per 100 g of leaves was 0.109 g or highest among all the varieties. All other amounts gained from the Mukdahan, Sakon Nakhon, and Nakhon Phanom varieties were 0.104, 0.103, and 0.103 g, respectively (Fig. 1). As an amount of indigo dye of each of the 4 indigo varieties was considered, it was found that the Kalasin variety yielded the most amount of indigo dye as high as 0.109 g per 100 g. of its leaves. Yet, it did not yield the highest produce of fresh leaves, because it yielded 4560 kg/Acre. However, indigo leaves from the Kalasin variety, which is a curved pod indigo plant, are relatively bigger. It was found that leaves of the curved pod indigo plant are bigger and thicker than those of the straight pod varieties. Food, thus, is stocked more in such leaves. That results in a higher amount of indigo dye than that gained from the leaves of the straight pod indigo plant that has thinner leaves. As the indigo dye was compared with the dye from leaves of *Marsdenia tinctoria* R. Br., it showed that the latter yields indigo dye less than the indigo plant does.

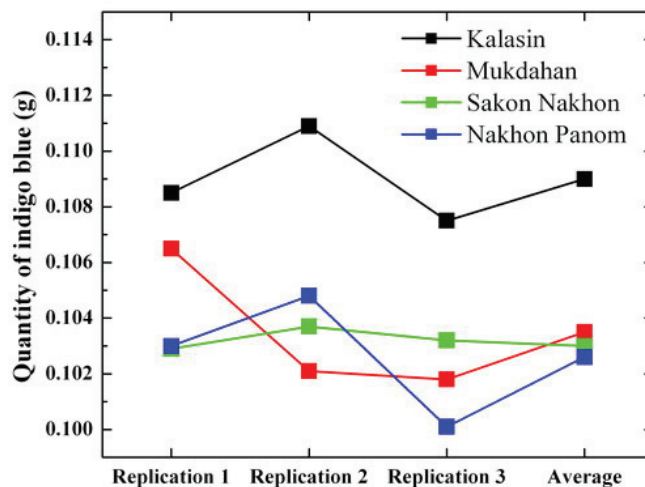


Fig. 1. Quantity of indigo blue of 4 indigo plant varieties, Kalasin variety, Mukdahan variety, Sakon Nakhon variety and Nakhon Panom variety

3.3 A study of other plants that yield the indigo color

3.3.1 *Marsdenia tinctoria* R. Br. is in the Asclepiadaceae family. It is a climber, having dark bark; and its trunk is green. It has a big-sized simple leaf with elliptic-shape. The apex of the leaf is acuminate and the base of the leaf is cordate. The leaf sizes are $5-9.5 \times 10.3-18.5$ cm², and its surface is a little bit wavy.

The leaves are arranged opposite. The flower shape is a greenish-yellow umbel like raceme [5]. It blooms between April and July (Fig. 2).

3.3.2 The indigo plant called *Memecylin edule* Roxb. is in the Memecylaceae family. It is a shrub about 2-3 m tall, having blackish-brown bark and some length-wise cracks along its branches and trunk. Its leaf is simple and obovate-shaped and its apex is retuse, at base acute. The leaf sizes are $2.5-3.2 \times 3.5-6.4 \text{ cm}^2$, the lamina is thick and brittle. The shape of flowers which is developed looks like a cup shape. The petals of flower are purple or dark purplish blue, comprising 4 petals for one flower and it blooms between March and April. Its fruit is round-shaped, looks green when not mature and looks from dark-purple to blackish brown when mature [7] (Fig. 2).



Fig. 2. Leaf and stem of (a) *Marsdenia Tinctoria* R. Br. and (b) *Memecylin edule* Roxb

3.3.3 A study of an amount of dye color gained from leaves of the two varieties: *Marsdenia tinctoria* R. Br. and *Memecylin edule* Roxb. From the investigation of an amount of dye color gained from other varieties that yield indigo color, it was found that the leaves of *Marsdenia tinctoria* R.Br. when soaked in the water for 24 hr. in the room temperature yield 0.012 g per 100 g of its leaves. As they are soaked for 30 hr, they would yield more indigo color of 0.016 g per 100 g of its leaves. As they are soaked for 36 hr, the amount of indigo color would reach to 0.016 g per 100 g of its leaves. The *Marsdenia tinctoria* R.Br. s leaves when soaked in the water at the temperature of 40 °C as long as 24 hr were found giving the most amount of indigo blue equal to 0.025 g per 100 g of its leaves. When they were soaked as long as 30 hr, the indigo blue gained was as much as 0.016 g per 100 g of its leaves. That is the decreased amount. When they were soaked for 36 hr the increased amount of indigo blue became 0.019 g per 100 g of its leaves (Fig. 3). It is showed that the most appropriate temperature and time for soaking the leaves that yield the most amount of indigo blue are 40 °C and 24 hr, respectively. Yet, the amount of indigo blue would decrease when soaked longer. That is congruent with the practice of those local people whose prowess of bringing the *Marsdenia tinctoria* R. Br. leaves to be boiled first is clearly evident before pouring solution to mix with the indigo blue in the pot for making it stronger [5, 6]. It is showed that the high temperature will help dissolve more indican substance from the *Marsdenia tinctoria* R. Br. leaves [7, 8]. However, its leaves must be soaked in the warm water not longer than 24 hr. for its optimum result of gaining more dye color. The *Memecylin edule* Roxb. leaves on the contrary cannot yield the indigo color (Fig. 3).

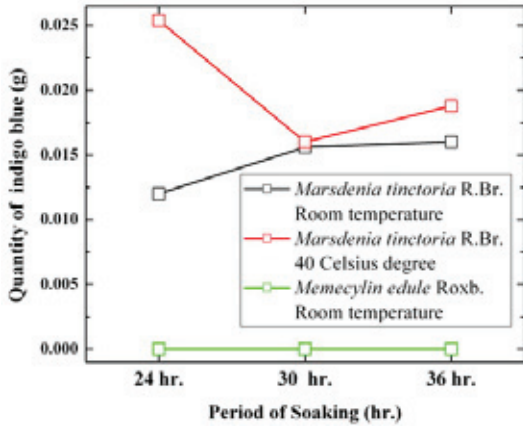


Fig. 3. Quantity of indigo blue of *Maesdenia tinctoria* R.Br. and *Memecylin edule* Roxb

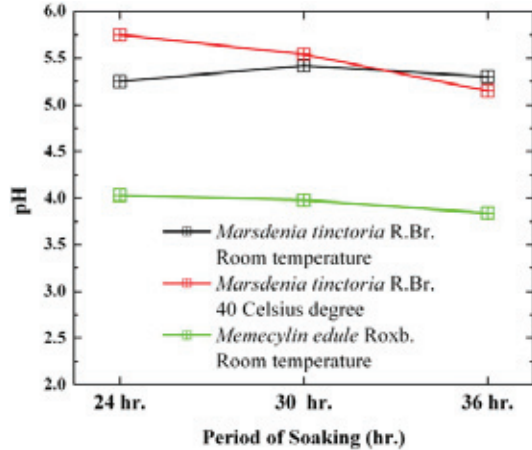


Fig. 4. pH of *Marsdenia tinctoria* R.Br. and *Memecylin edule* Roxb



(a)



(b)

Fig. 5. Indigo blue of (a) *Memecylin edule* Roxb and (b) *Marsdenia tinctoria* R. Br

From measuring the pH level of the *Marsdenia tinctoria* R.Br. solution, it was found that the solution gained from the soaked leaves at the room temperature for 24 hr had a pH value of 5.25. When leaves soaked for 30 hr, the pH value reached 5.42; while leaves soaked for 36 hr, the pH value will be lessened as equal to 5.30. As for the solution gained using the temperature of 40 °C for 24 hr, its pH value was 5.75. If the leaves were soaked for 30 hr, the solution gained will be decreased to the value of 5.54. If soaking was done longer up to 36 hr, the pH value would decrease to be 5.15. The solution gained from soaking the *Memecylin edule* Roxb. leaves in the room temperature, its pH value was 4.03. When the time of soaking extended to be 30 hr long, the solution gained would decrease to be 3.98. As the soaking took longer time up to 36 hr, the pH value would more decrease to be 3.84 (Fig. 4). The pH level of solution gained from soaking the *Memecylin edule* Roxb. leaves was considered, it would show that its leaves soaked at the room temperature as long as 36 hr would give a higher acidity level than that of the solution gained from soaking the *Marsdenia tinctoria* R.Br. leaves both in the room temperature and in the temperature of 40 °C. From the investigation of acid and base states of solutions gained from soaking the *Marsdenia tinctoria* R.Br. and *Memecylin edule* Roxb. leaves, it was found that the *Memecylin edule* Roxb. solution had the high acidity level of 3.84. This finding is congruent with the practice of that local wisdom whose prowess of using the *Memecylin edule* Roxb. as shown in Fig. 5 (a), leaves solution to mix with the indigo leaves solution in the indigo dye pot for making a dye stronger is obviously evident [7]. In

Fig. 5 (b), it is concluded that the indigo blue from the *Marsdenia tinctoria* R.Br. leaves can be used as a dye the same way as the indigo leaves [8] by soaking them into the warm water. The *Memecylin edule* Roxb. leaves on the contrary cannot yield the indigo color. The pH level of solution from the *Memecylin edule* Roxb. leaves is considered, the high acidity level was found. The solution from *Memecylin edule* Roxb. is natural mordant [9]. Thus, the solution is used for adjusting the acid-base states of dye for adjusting the pH value of the indigo dye solution in the pot.

4. Conclusions

The Nakhon Phanom variety yielded the highest amount of fresh indigo leaves produce with an average of 5520 kg/Acre, which makes a significant different amount as compared with those gained from the Kalasin, Sakon Nakhon, and Mukdahan varieties. The Kalasin variety yielded leaves produce with an average of 4560 kg/Acre, which is different from the Sakon Nakhon and Mukdahan varieties that yielded 4320 and 3920 kg/Acre, respectively. The Kalasin variety was found to give the most amount of indigo dye as equal to 0.109 g per 100 g. of the indigo plant leaves. The other varieties saying Mukdahan, Sakon Nakhon, and Nakhon Phanom give their own average of all the indigo dye amounts of 0.104, 0.103, and 0.103 g, respectively. However, they showed no significant difference in comparison. The plant that yields indigo blue, which can be used in place of a domestic indigo blue is a climber named *Marsdenia tinctoria* R.Br. whose leaves produce yields an indigo color of as much as 0.025 g per 100 g of its leaves soaked in the water at the temperature of 40 °C whereas, the *Memecylin edule* Roxb leaves that do not yield indigo color but high acidity are used to adjust the acidity state of the dye solution in the dying pot or natural mordant to suit the need of those who dye fabrics.

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