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Review

Diversity of Plant Colorant Species in a Biodiversity Hotspot in Northern Thailand

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Abstract: Growing concerns regarding health and eco-safety have led to a resurgence in the recognition of natural products. Although plant colorants are often mentioned in ethnobotanical studies in northern Thailand, they have not been comprehensively examined. This study aims to (i) investigate the colorant plants used by ethnic minorities in northern Thailand and (ii) compare colorant plants used among the groups in northern Thailand with other regions worldwide. Data on colorant plants used by ten ethnic groups across 142 villages in northern Thailand were extracted in a systematic review of 42 published references. The importance of the colorant plant species was evaluated using the ethnobotanical index "use value" (UV), and the homogeneity of knowledge was assessed using the "information consensus factor" (ICF). The similarity of colorant plants used among the ethnic minority groups and beyond was evaluated using Jaccard's index. The 42 published references included information about 104 colorant plant species belonging to 85 genera and 43 families from seven provinces in northern Thailand. This represents nearly half of the 212 colorant plant species reported across all 72 provinces in Thailand. The most important species were *Strobilanthes cusia* and *Morinda angustifolia*, which are sources of blue and red colors. Fabaceae, Rubiaceae, and Lamiaceae were the plant families with most species used as colorants. The colorant plant species yielded ten different color shades, with blue being the most prevalent. The Jaccard's index varied from 0 to 0.27 among the northern Thai ethnic minorities and from 0 to 0.13 for regions outside of northern Thailand. The regions located closest to northern Thailand exhibited the highest degree of similarity. This study provides valuable insights into the traditional knowledge and usage of dye plants in northern Thailand, contributing to preserving the cultural heritage and providing basic knowledge for the sustainable use of natural colorants for modern applications.

Keywords: traditional knowledge; natural colorant; plant diversity; Thai ethnic minorities

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1. Introduction

Biodiversity hotspots are geographic areas with a significant concentration of biodiversity. These hotspots are typically regions with many endemic species. Thailand is one of the most renowned biodiversity hotspots in the Indo-Burmese region. The country is home to numerous ethnic communities and overlaps with two major biogeographical regions: the Indo-Chinese region and the Sundaic region [1–3]. With forests covering 32% of its area, Thailand hosts some 11,000 plant species [4]. Plants are essential to human life and communities, serving as vital resources for basic needs, such as food, housing, clothing, and medicine [5]. Over time, humans have learned about plant uses, accumulated knowledge and experience, and have passed this traditional knowledge on to the next generations through oral tradition and cultural practices [6–8].

Plant resources play a vital role in the survival and prosperity of human life and societies. They serve as crucial providers of food, housing, clothing, and medicine [9]. Textiles are among the requisites of human life. Throughout history, people across the globe have developed methods for coloring textiles using colorants derived from nature. Plant colorants have been integral to textile culture, employed in coloring silk, wool, and cotton [10,11]. In the 19th century, synthetic colorants were discovered by British chemist William Henry Perkin, with mauveine, made from aniline, being the first synthetic dye [12]. This discovery caused competition between natural and synthetic colorants. Synthetic colorants gained popularity over their natural counterparts due to lower costs and greater availability [13,14], leading to the abandonment of natural colorants and the traditional knowledge associated with them. Apart from coloring textiles, plants have been used to color a variety of other products, such as food, cosmetics, medicines, etc.

Increased awareness of health and eco-safety has recently produced increased attention to natural products because they are seen as benefiting mental and physical well-being. As a result, demand for natural colorants is rising, as they are seen as environmentally friendly and thought to exhibit minimal toxicity. In addition, plant colorants have received renewed interest due to their limited adverse effects on human health and the environment [13,15,16], with the food industry witnessing a growing consumer demand for natural food coloring. Subsequently, numerous studies have focused on plant pigments to identify potential alternatives to artificial colorants [17,18].

Traditional knowledge of plant colorant species has been maintained by various ethnic groups worldwide [19–24]. The conservation of the uniqueness and the revenue of natural colorant products are the reason that some peoples use colorants of plant origin [11]. Since 1986, there have been numerous ethnobotanical studies conducted in Thailand. Most of the research focuses on the utilization of plants in medicinal plants, food plants or various other use categories [2,25–27]. Despite the significance and variety of plant colorant species utilized by different ethnic minorities in northern Thailand, no research has collected and compared this traditional knowledge of Thailand's plant colorant species. Although our study is based on existing data, the compilation of the information and its extraction from publications, of which many were "grey" and hard to access, makes our study novel. This research addresses the following specific questions: (i) How many plant colorant species are used by the ethnic minorities in northern Thailand and to which plant families do they belong?; (ii) How similar are the colorant plant species used among the ethnic minority groups in northern Thailand, and how similar are these colorant species to those used in neighboring regions and countries and beyond?; (iii) How important are the different use categories and how is the colorant extracted from the plants?; (iv) Which plant parts are used to produce the color and which color do they produce?, and (v) Which were the most important colorant plant species and how are they used as plant colorant?

2. Materials and Methods

2.1. Data Collection

The data on traditional knowledge of plant colorant species were gathered from previous ethnobotanical research. Most of these studies were done as B.Sc., M.Sc., and PhD theses. The first thesis on plant colorants, which discussed the ethnobotany of the Karen ethnic group in Chiang Mai, was written in 1995 [28]. The primary sources were theses conducted at the Ethnobotany and Northern Thai Flora Laboratory within the Department of Biology at Chiang Mai University. Ethnobotanists from this laboratory have extensively studied plant uses in northern Thailand over the past three decades. The Sanga Sabhasri Library, located at the Queen Sirikit Botanic Garden, was an additional source of ethnobotanical theses and reports. The library houses an extensive collection of flora and ethnobotanical documents relevant to northern Thailand. In instances where the same data appeared in both a journal article and a thesis, we only used the data from the thesis to avoid redundancy.

Our comprehensive search extracted data from 42 references and encompassed ten ethnic groups across seven provinces in northern Thailand. Some of the references provided data from more than one province (Figure 1, Supplementary Table S1). The data concerning plant colorant species that were described in the 42 references had been collected from a diverse array of sources. We collected information on plant colorant species, including their local names, the parts used, methods of preparation, and the resulting color shades.

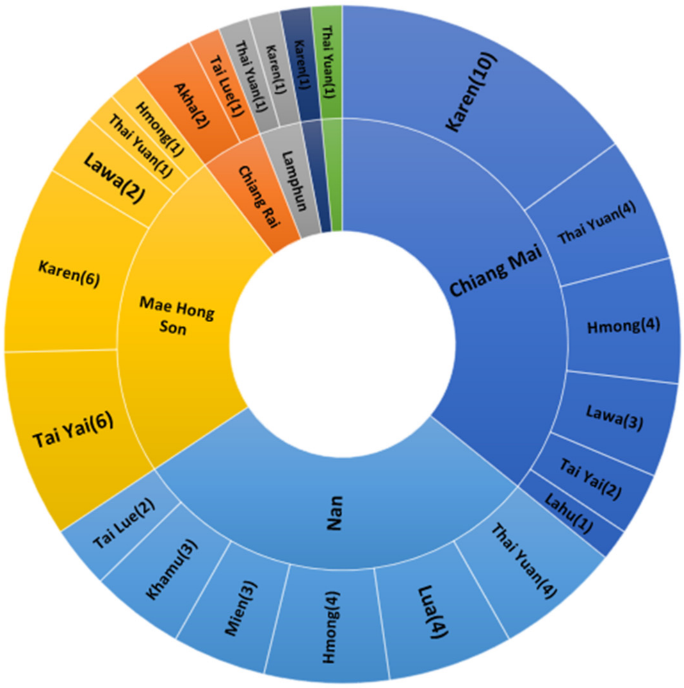

Figure 1. Data extracted from 42 references on colorant plants used by ten ethnic groups in northern Thailand. The inner circle represents the seven provinces from which the data originated, and the outer circle shows which of the ten ethnic groups were represented in the data for each province. The number in parentheses represents the number of villages where data collection was conducted.

2.2. Data Organization

All the scientific species and family names of the plant colorant species given from the primary references were verified and updated following The World Flora Online (WFO). In addition, the used plant parts as reported in the references were categorized and classified into stem, bark, leaves, inflorescence, infructescence, fruits, seeds, roots, gall, and exudates following the Economic Botany Data Collection Standard [29]. When data about the part of plants used or method of preparation were not provided in the studied references, we recorded them as unspecified.

2.3. Data Analysis

Because the data used in this study were from already published references, each of which had different numbers of informants, we used each village in the cited reference as a "pseudo-informant" in our analysis. Each "use report" refers to one traditional use of a plant colorant species mentioned in the cited reference. The ethnobotanical indices were calculated to determine the importance of each of the uses of plant colorant species.

2.3.1. Use Value

The importance of plant colorant species was evaluated by their use value (UV) [30], which is calculated as:

$$UV = \Sigma Uis/N$$

where U is the total number of use reports for a particular species and N is the total number of pseudo-informants.

2.3.2. Informant Consensus Factor

The homogeneity of knowledge among pseudo-informants regarding plant colorants used for each color shade was assessed using the information consensus factor (ICF). Originally developed for evaluating medicinal plants, the ICF measures the consensus among informants concerning the use of a particular plant species for a specific purpose [31]. The ICF is calculated using the following formula:

$$\mathrm{ICF} = (\mathrm{Nur} - \mathrm{Nt})/(\mathrm{Nur} - 1)$$

where Nur is the number of use reports in each color shade, and Nt is the number of taxa used in each color shade. A higher ICF value indicates greater agreement among informants about using a specific plant species for a particular color shade [32,33].

2.3.3. Jaccard's Index

We used Jaccard's index (JI) [34] to measure similarity between ethnicities, determining the proportion of shared plant colorant species between two ethnic groups. The Jaccard's index is calculated as follows:

$$\mathrm{JI} = c/(a + b - c)$$

where a is the number of colorant plant species used by ethnic group A, b is the number of colorant plant species used by ethnic group B, and c is the number of plant colorant species used by both ethnic groups, i.e., A and B.

3. Results

3.1. Diversity of Colorant Plants

A total of 324 use reports from the 42 references encompassed ten ethnic groups and 142 villages in Chiang Mai, Chiang Rai, Lamphun, Phrae, Nan, Mae Hong Son, and Tak provinces in northern Thailand. We found a high diversity of colorant plants (104 species) used by the ethnic groups (Table 1).

Table 1. Number of colorant plant species and colors produced, as reported for each of the ten ethnic minority groups in northern Thailand for which data were extracted from 42 references.

Ethnicity	No. of Species	Colors	References
Akha	2	Red, blue	[35,36]
Hmong	8	Blue, purple, green	[37–42]
Karen	66	Red, orange, yellow, green, blue, purple, pink, brown, grey, black	[28,42–56]
Khamu	6	Red, yellow, blue, brown, black	[38,57,58]
Lawa	19	Red, yellow, blue, brown, grey, black	[51,59,60]
Lua	7	Red, yellow, blue, brown, black	[38,48,61]
Mien	7	Red, green, blue, brown	[38,45]
Tai Lue	6	Red, blue, black	[62,63]
Tai Yai	4	Red, green, blue	[64–68]
Thai Yuan	19	Red, yellow, green, blue, purple, brown, black	[41,45,67,69–75]

There were 324 use reports related to 104 plant colorant species from 85 genera in 43 plant families in northern Thailand. Fabaceae included the highest number of plant colorant species (16 species, 50 use reports), followed by Rubiaceae (10 species, 33 use reports) and Lamiaceae (6 species, 17 use reports). There were 24 families (56%) that were represented by a single plant colorant species (Table 2).

Table 2. Numbers of plant colorant species in each plant family, used by 10 ethnic minority groups, inhabiting seven provinces in northern Thailand.

Family	No. of Species
Fabaceae	16
Rubiaceae	10
Lamiaceae	6
Acanthaceae, Combretaceae, Phyllanthaceae	5
Anacardiaceae, Fagaceae, Moraceae	4
Pentaphylacaceae	3
Apocynaceae, Arecaceae, Asteraceae, Dipterocarpaceae, Ebenaceae, Malvaceae, Musaceae, Primulaceae	2
Adoxaceae, Amaranthaceae, Bignoniaceae, Bixaceae, Celastraceae, Dilleniaceae, Euphorbiaceae, Hydrangeaceae, Iridaceae, Lauraceae, Lecythidaceae, Magnoliaceae, Melastomataceae, Menispermaceae, Myrtaceae, Oleaceae, Pandanaceae, Passifloraceae, Proteaceae, Salicaceae, Smilacaceae, Staphyleaceae, Styracaceae, Theaceae, Zingiberaceae	1

The plant colorant species were of various habits, such as trees (48 species) which was the most common habit, followed by shrubs (31 species), herbs (14 species), climbers (9 species), and palms (2 species).

3.2. The Similarity of Plant Colorant Species among Ethnic Groups

Jaccard's index values for pairs of the ethnic groups varied from 0.00 to 0.27 (Table 3). The highest Jaccard's index for plant colorant species was observed between Hmong–Khamu and Hmong–Tai Lue pairs (0.27), followed by the Hmong–Akha pair (0.25). *Strobilanthes cusia* was identified as the most commonly used plant colorant species across all ethnicities.

Table 3. Jaccard's similarity index was calculated for pairs of ethnic groups in a sample of plant colorant species used in ten ethnic groups in northern Thailand.

	Akha	Hmong	Karen	Kha-mu	Lawa	Lua	Mien	Tai Lue	Tai Yai	Thai Yuan
Akha		0.25	0.01	0.14	0.00	0.13	0.13	0.14	0.00	0.05
Hmong			0.04	0.27	0.04	0.07	0.15	0.27	0.09	0.13
Karen				0.06	0.09	0.04	0.04	0.04	0.01	0.09
Khamu					0.04	0.18	0.18	0.20	0.00	0.09
Lawa						0.08	0.08	0.04	0.00	0.06
Lua							0.08	0.08	0.00	0.08
Mien								0.08	0.10	0.08
Tai Lue									0.11	0.19
Tai Yai										0.05

Compared to other regions and countries beyond northern Thailand, we found that many used the same species as colorants. Jaccard's index values were 0.00–0.13 (Table 4). The highest similarity was observed within the geographically closest areas. The high similarity indices indicate a significant resemblance in plant colorant species' use and application [76]. The differences in climatic conditions, local knowledge of plant colorant species, topography, and the flora in the specific region obviously influence the degree of similarity observed [77].

Table 4. Comparison of colorant plant species encountered in seven provinces in northern Thailand with similar data from other regions of Thailand, and other countries in Asia, Africa and the Americas.

Study Area	No. of Species in Study	No. of Species in the Present Study	Shared Species	Jacard's Index	Reference
Northeast Thailand	56	104	18	0.13	[78]
Western Myanmar	25	104	8	0.07	[23]
India	165	104	15	0.06	[21]
Northern Vietnam	43	104	7	0.05	[20]
Madagascar	128	104	13	0.05	[79]
Eastern Uttar Pradesh, India	20	104	5	0.04	[80]
Northwest Yunnan	23	104	4	0.03	[81]
China	19	104	3	0.03	[82]
Reunion Island	194	104	10	0.03	[24]
NW and W Himalaya	26	104	3	0.02	[83]
Central Mexico	28	104	2	0.02	[22]
Guizhou China	23	104	2	0.02	[11]
Peru	30	104	0	0.00	[84]
China	13	104	0	0.00	[18]
West Kalimantan	13	104	0	0.00	[85]

3.3. Use Categories, Preparation, and Extraction of Plant Colorant Species

The highest number of use reports were for textile coloring (87%; 271 use reports), food coloring (6%; 18 use reports), material coloring (5%; 14 use reports), and other uses for coloring (2%; 7 use reports) (Figure 2 and Supplementary Table S2).

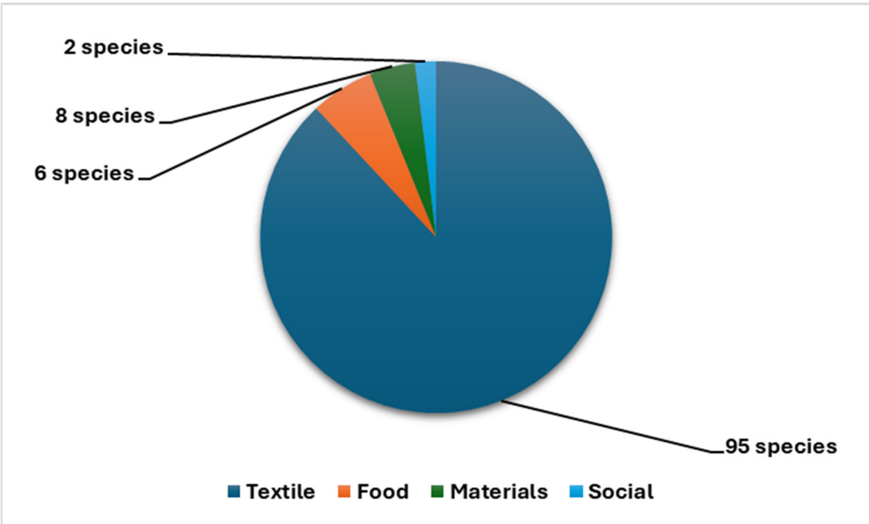

Figure 2. Number of colorant plant species used in each of four categories by ten ethnic minority groups in northern Thailand.

Some of the data extracted from the 42 primary sources lacked detailed descriptions of preparation methods and applications, with only 108 use reports (35%) providing complete information. Preparation methods typically depended on the available part of the plant materials. Raw materials were cut into small pieces, and then the plant pigments were extracted either by boiling with water or employing a cold method, in which the plant materials were soaked in water to achieve the desired color (Figure 3).

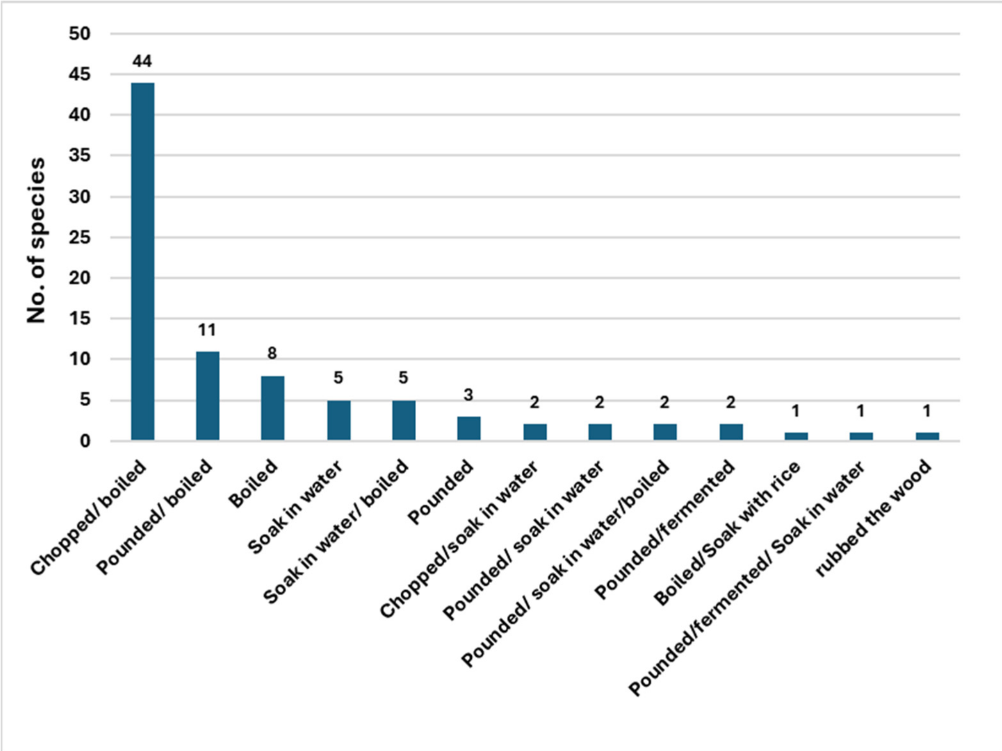

Figure 3. The number of colorant plant species used in 13 preparation methods by 10 ethnic groups in northern Thailand.

3.4. Plant Parts Used and Colors Obtained

The most common plant part used as colorant was bark, mentioned in 43 use reports, followed by leaves (19 use reports), fruits (13 use reports), and stems (11 use reports) (Figure 4).

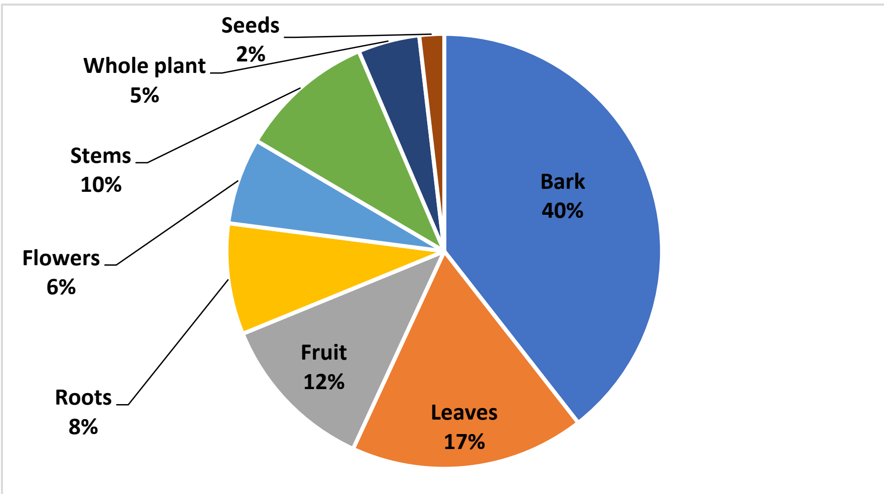

Figure 4. Different parts of plants used as colorants by ten ethnic minority groups in northern Thailand.

The data included information for 65 species about which color shade was produced by the colorant plant (Figure 5). Brown, yellow, and red were the most prevalent colors derived from 14 to 18 different colorant plant species. A middle group of 7–10 species produced green, blue, and purple colors. The last group, of 2–5 plant species per color, included black, grey, orange, and pink.

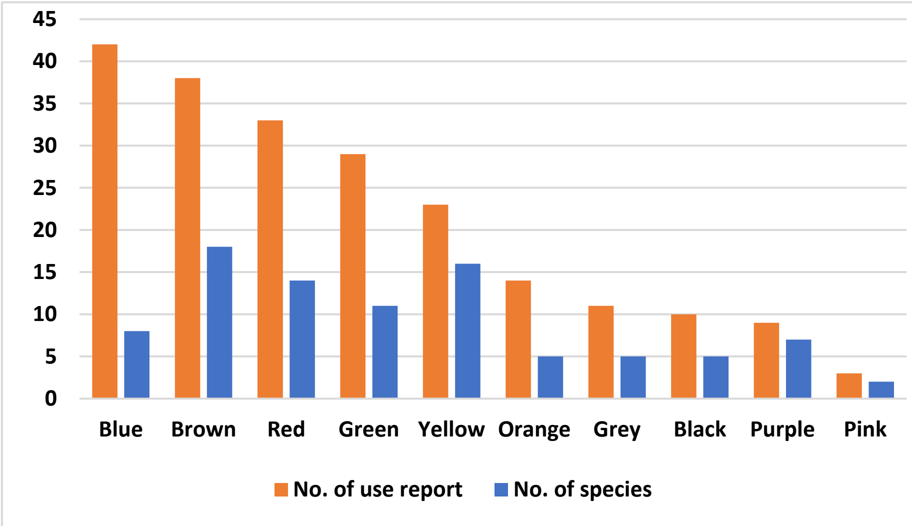

Figure 5. The number of species and the number of use reports used to produce ten color shades by ethnic groups in northern Thailand.

3.5. Important Coloring Plant Species and Homogeneity of Species Used

The use values (UV) for individual species ranged from 0.007 to 0.283 (Supplementary Table S2). Based on the UV, we categorized the coloring plants into three use levels: high (UV $\geq$ 0.093), middle (0.007 < UV < 0.093, and low (UV $\leq$ 0.007). Of the 104 colorant plant species assessed, three had high use values, 40 had medium use values, and 60 had low use values. Colorant plant species with high use value were Strobilanthes cusia (UV = 0.275), Morinda angustifolia (UV = 0.183), and Oroxylum indicum (UV = 0.092). Of all the plant colorant species that we found in this study, more than half (around 60) were cited in just one reference, e.g., Careya arborea, Dipterocarpus obtusifolius, Lablab purpureus, and Livistona speciosa were singletons.

The information consensus factor (ICF) ranged from 0.25 to 0.83 (Table 5). The blue shade had the highest ICF value, at 0.83, based on 42 use reports and eight colorant plant species. Example of colorant plant species that give this color were Strobilanthes cusia and Indigofera tinctoria. To obtain the color, both were pounded and their leaves were fermented to obtain the colorant. The orange shade (ICF = 0.69, 14 use reports, 5 species) was obtained from Bixa orellana and a green shade (ICF = 0.68, 29 use reports, 10 species) was based on the use of Oroxylum indicum and Pandanus amaryllifolius.

Table 5. Informant consensus factor (ICF) value for coloring plants recorded among ten ethnic minority groups in northern Thailand according to data extracted from 42 published references.

Colors/Shades	No. of Use Reports	No. of Species	ICF
Blue	43	8	0.83
Orange	14	5	0.69
Green	29	10	0.68
Grey	11	5	0.60
Red	33	14	0.59
Black	10	5	0.56
Brown	38	18	0.54
Pink	3	2	0.50
Yellow	23	16	0.32

4. Discussion

Our data, derived from 42 references based on ethnobotanical studies in seven provinces in Thailand's northern region, included information on 104 colorant species. This number represents nearly half of the 212 plant colorant species reported from all

72 provinces of Thailand [86]. Fabaceae, Rubiaceae, and Lamiaceae made up one third of the colorant species in our dataset. Fabaceae, not surprisingly, was the best represented family with 16 colorant species, which agrees with its leading position among plant colorant species worldwide [20,23,87]. Fabaceae globally encompasses about 20,000 species and includes a variety of life forms, such as trees, shrubs, herbs, lianas, vines, etc. The usefulness of Fabaceae goes beyond colorant plants and it is leading in species numbers of medicinal plants, both globally and particularly in Thailand [2,27,88]. The other two families with many colorant species are also large globally distributed plant families. Rubiaceae, with over 13,000 species, is mostly known for its it many uses based on its content of alkaloids. Its common name (madder family) is based on the common madder, Rubia tinctorum, which produces a red pigment in its roots, which has been used as a colorant [89–91]. Lamiaceae, the mint family, is also large, with nearly 7000 species distributed globally. Species of that family also have many uses, mostly based on their content of ethereal oils and their pigments, such as chlorophyll and carotene, which are used as colorants [92–95].

Hmong, Khamu, Tai Lue, and Akha are ethnic groups who traditionally dress in black or indigo [96,97]. Colors like indigo blue were obtained from Strobilanthes cusia, Indigofera tinctoria, Clitoria ternatea and a few others. These plants were commonly used by the Hmong, Khamu, Tai Lue, and Akha. The number of plant colorant species used by the Hmong (8), Khamu (6), Tai Lue (6), and Akha (2) was low when compared to the other ethnic groups. However, only 1–2 species were used in each ethnic group, contributing to a considerable degree of similarity. Boiling in water was the most common method for color extraction, and it is a common extraction method elsewhere where plants are used for coloring [78,98]. Hot water extraction was the most frequently used method, offering a simple means to extract plant pigment from raw materials. When used for coloring textiles, most processes involved soaking fabrics in the solution in which the pigments were dissolved, making hot water extraction a suitable preparation method [78,99].

Ethnic groups in northern Thailand used a wide variety of plant parts for color extraction, indicating a diverse range of plant sources for obtaining various colors. Bark was the most common plant part used for colorant extraction, and coloring textiles with bark was often associated with the brown color shade. Brown was derived from many plant species, as also shown in a report on plant colorant species used by the Tai Lao ethnic group in northeastern Thailand outside the region studied in this research [78,100]. This shows that many plant sources are available for obtaining a brown color. The shades of yellow, brown, grey, and black are often based on the plants' content of tannins, which are predominantly found in plant bark [83]. These tannins generate different shades of color when used for coloring. The presence of tannins in a variety of plant species makes them a significant source of natural colorant, offering a spectrum of colors, used in traditional textile production for centuries.

When measured by the use value (UV), Strobilanthes cusia was the most important plant colorant species among ethnic groups in northern Thailand, used by many groups such as the Karen, Hmong, Akha, Khamu, Lawa, Lua, Mein, Tai Lue, and Thai Yuan. Furthermore, it was reported in 14 (33%) out of the 42 primary references. This could be because they were easy for people to find [89]. Strobilanthes cusia is an herbaceous, perennial plant distributed throughout South Asia and it is extensively cultivated and widely used in the Indo-Chinese peninsula. It is easy to propagate by cuttings and its leaves can be harvested 2–3 times a year [101–103]. The results of this study align with ethnobotanical studies in The Republic of Georgia in the Caucasus, indicating that cultivated plants have more known uses than wild plants. This is evident from the higher UV values of cultivated plants compared to those of wild plants [104]. Strobilanthes cusia holds cultural significance for various indigenous communities in East and Southeast Asia. Cultivated for centuries, it is likely to be the only natural dye still in use today in these regions, where it has been extensively cultivated for centuries. In addition to its plant colorant value, Strobilanthes cusia is used in traditional Chinese medicine, within a roughly 1000-year-old medicinal tradition [102,105,106]. This species was reported as a colorant plant in several ethnobotanical studies from across Asia, such as in India [107], China [18,81,82,103,108] and Myanmar [23].

We found that 61 of the 104 plant colorant species were singletons known from only one of the 42 original references. This heterogeneity was also observed in previous studies of medicinal plants and food plants [25,109]. The singletons could be viewed as genuinely rarely used, as secret, as part of specialist traditional knowledge, or having previously unshared uses, all of which could be due to their rarity or difficulty of use. The large number of singletons emphasizes the significance and urgent need for more ethnobotanical research, especially considering the current state of knowledge erosion and its rapid increase [25,110–112].

The blue shade had the highest ICF value because it had many use reports based on relatively few species; this color is derived from Strobilanthes cusia and Indigofera tinctoria, both with high use values. In contrast, the yellow color had many use reports but was derived from many different colorant species, resulting in a lower ICF value than the blue shade. A higher ICF value indicates significant importance, suggesting that Strobilanthes cusia was widely utilized by many informants. This high level of agreement among informants reflects a well-defined plant colorant heritage [113]. Nowadays, the food industry aims to diversify its color options using natural colorants. However, natural blue color is relatively rare and highly sought after in the food and beverage industry [114,115]. The findings of this study could be further used to explore and develop natural blue colorants for this industry.

5. Conclusions

Northern Thailand is rich in plant colorant species, harboring nearly half of the country's colorant species in only 7 of the over 72 Thai provinces. The region is also more diverse than most other regions in and outside of Thailand but surpassed by the much larger count for Reunion Island (194 sp), India (165 sp) and Madagascar (128 sp). The most common use of colorant plants in northern Thailand was for textile coloring. This is commonly done by hot water extraction, which is a method used throughout the world, probably because of its simplicity and effectiveness. Northern Thailand was also similar to other places in extracting plant colorants mostly from the bark of the plants, which was often associated with the production of brown hues. The most commonly used colorant plant species in northern Thailand were plants that are well known for colorant purposes all over Southeast Asia. *Strobilanthes cusia* is a traditional source of indigo colorant in the region. *Morinda angustifolia*, which was the second most important colorant species in northern Thailand, occurs naturally over a large part of Southeast Asia, where it is widely used to produce yellow, red, or brown coloring for textiles. The most frequently used plant families were Fabaceae, Rubiaceae, and Lamiaceae, all of which are large and globally distributed families with many uses. In conclusion, the outstanding feature of northern Thailand is not so much the individual colorant species found there as the high diversity of the colorant plant flora concentrated in a single small region.

The extent to which our compilation captures the full range of plants used for coloring remains uncertain. Future research should explore the traditional knowledge of natural plant colorants in additional communities and regions to augment and broaden our understanding of this subject. The diversity of plant colorant species used among ethnic groups in northern Thailand is remarkable; however, many plants used for colorant extraction and documented in the cited ethnobotanical studies have not been thoroughly investigated. Further research is essential to assess these plant-derived colorants' stability and their potential for development in the food, cosmetic, and pharmaceutical industries. Ultimately, this study may provide valuable background for rural development initiatives and the preservation and revitalization of traditional knowledge regarding plant colorants within local communities.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/d16040194/s1. Supplementary Table S1: An overview of the 42 sources that were consulted to analyze the data on plant colorants in northern Thailand; Supplementary Table S2: List of plant colorant species used by the ethnic groups in northern Thailand.

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