

Diversity of Epiphytic Lichen in University of Phayao, Thailand

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Abstract: The diversity of lichen in University of Phayao, Thailand has been surveyed and documented by collecting throughout the rainy seasons from May to October 2018. The lichen samples were randomly taken from the bark of trees. The species of lichen were identified and grouped on the lichen growth. 210 samples of total lichen were taxonomically classified based on anatomical, morphological and chemical constituent substances by spot test, ultraviolet and thin layer chromatography, divided to two groups and identified of 8 families 16 genera and 36 species. 6 families 11 genus and 29 species of crustose were found. Most of the frequency families of crustose were Graphidaceae (14 species), Lecanoraceae (6 species), Trypetheliaceae (3 species), Physciaceae (3 species), Pertusariaceae (2 species) and Chrysothricaceae (1 species), respectively. Two families 5 genera and 7 species of foliose were found. The families of foliose were Physciaceae (5 species), and Parmeliaceae (2 species). This is a key for identification of lichens in plant genetic conservation area at University of Phayao.

Key words: Lichen, diversity, University of Phayao, Thailand

1. Introduction

Lichens are the co-occurring organisms (mutualism) including of fungi and algae [1] that become a new creature with different shapes according to species and location. They are found in common areas, cold weather to the polar in the Arctic circle including hot weather and arid zone up to desert or warm and humid [2]. They are estimated that there are approximately 25,000 species of lichens around the world [3]. Thailand is a country with a lot of lichens because Thailand is in a tropical zone. There is a diversity of forest

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resources in each of regions that is a suitable condition for lichen growth. From the report of the number of lichens in Thailand is found 1,292 species [4]. Epiphytic lichens are importantly biotic components in many forest ecosystems which are high species diversity, obvious on biomass structures and ecological biodiversity in northern ecosystems [5]. Lichens also have many benefits. Some lichens are used as medicines from their secondary metabolite properties that have important biological activities such as antimicrobial, antipyretic, cytotoxic, antitumor, analgesic, antiviral, allergenic compounds [6], for the perfume industry and some as acid-alkaline indicators [7]. Moreover, lichens are used as an indicator of atmospheric pollution or could use the indication of environmental condition [8], [9] including to still have rapidly response to concentration level of atmospheric pollution [10] and as a bioindicator of air quality [11]. The atmospheric pollution of monitoring in that area was not able to use only lichen diversity it must apply the monitoring data of atmospheric pollution in along with the area [12].

Lichens based on both morphological and chemical investigations by identifying of lichen species is an interesting taxonomical study that uses morphological and chemical analysis [13]. Chromatographic study on the chemical composition of lichens emphasized the presence of many compounds belonging to various chemical categories [14]. Culberson and Kristinsson (1970) suggested thin-layer chromatography (TLC) of lichen substances also treat, specific groups of compounds that were found to be convenient for distinguishing a limited number of specific compounds for confirmatory identifications [15]. Moreover, thin layer chromatography (TLC) have greatly improved the speed and certainty of recognition of lichen substances that are simple to use, relatively inexpensive and remains the most readily accessible and widely used method for identification of lichen metabolites as chemical investigations on an integral part of all serious taxonomic studies [16]. However, Lichen species usually identifies using different methods which together can reveal the species identity from identification keys [13].

The purpose of this research is the study on the diversity of epiphytic lichen in University of Phayao, Thailand that has been surveyed and documented by collecting throughout the rainy seasons. The study was started from May to November 2018. The species of lichen were identified and grouped on the lichen growth. The lichens were taxonomically classified based on anatomical, morphological and chemical constituent substances by spot test, ultraviolet and thin layer chromatography.

2. Materials and Methods

2.1 General topography

University of Phayao is in the northern region at Muang district, Phayao province, Thailand, located at 19.0285° N and 99.8963° E. The temperature of this region has varied a condition during 20°C to 38°C whereas the annual rainfall in approximately 35 to 2017 mm. The forest characteristic study of plant genetic conservation area at University of Phayao is a dry deciduous forest with a height of 527 meters above sea level (MAMSL), the humidity was approximately 76%, The temperature was approximately 35 °C and Geographic coordinates are approximate of Latitude: 19.0374216 and Longitude: 99.925759.

2.2 Sampling method

The data were collected from March to November 2018 in the field because it is the rainy season that performs the most complete collection of samples. A random sampling method was used to document the lichen diversity by laying 10 x 50 m. belt transects in the field. In each belt transect, all the substrates were thoroughly searched for epiphytic lichens. The diversity of epiphytic lichens on the trunk and host plants were recorded with their diameter at breast height (DBH). The collected epiphytic lichens specimens were packed in paper bags, dried and identified using standard collected samples. After the collection, the epiphytic lichens specimens were brought to a laboratory and identified them by morphological and chemical studies.

2.3 Spot test according to Orange et al. (2010)

Coloring test on tallus with chemicals (Spot test) is testing of lichen composition that may react with certain chemicals to cause color reactions. This is the identification of lichen species by dripping the prepared chemicals on the samples. The chemicals were K (Potassium hydroxide), C (Calcium hypochlorite), KC (Potassium hydroxide and Calcium hypochlorite) and I (Iodine). Then, the color was observed that appeared after dripping the chemical for 30 seconds. For no reaction, it is used instead of the plus sign (+). For reaction response, it is used instead of the minus sign (-), For a positive reaction, K (Potassium hydroxide) will occur as red or rust color. C (Calcium hypochlorite) will occur as orange or green color, KC (Potassium hydroxide and Calcium hypochlorite) will occur as red or rust color, and I (Iodine) will occur as blue or purple color and record the results [17].

2.4 The fluorescence test under ultraviolet (UV) according to Orange et al. (2010)

The fluorescence test under ultraviolet (UV) is a test using ultraviolet light (350 nm) or light tester for the fluorescence test of lichens [17].

2.5 Thin Layer Chromatography (TLC) according to White and James (1985)

Chemical analyses of extracts were carried out by thin-layer chromatography (TLC) [18].

2.6 Identification keys

The lichen genera identification keys by Harry Sipman, *Malesian Lichen Genera* (2003) and Brodo et al.'s 2001 *Lichens of North America* were used as an identification guide [19], [20].

3. Results and Discussion

From the survey and sampling of the lichens in plant genetic conservation area at University of Phayao by surveying and random sampling was collected the lichens of 210 samples. The lichen group can be divided into 2 large groups. Crustose was the most discoverable lichen group in the area, next below was foliose by classifying the diversity of each of the lichen groups were as shown in table 1 and 2, and pie-chart on a percentage of lichen families that found in University of Phayao area as shown in figure 1.

The studies of lichens diversity by the random sampling method in plant genetic conservation area at University of Phayao were analyzed in that the forest characteristics was a deciduous forest by studying of both external and internal morphology, chemical examination with Spot test technique, Test method in lichens with thin layer chromatography (TLC), The fluorescence test under ultraviolet (UV) and quantification with the lichen genera identification keys.

The results found that Crustose was the most discoverable lichen group in the area. Graphidaceae was the most discoverable family in lichen families, next below was Lecanoraceae, Trypetheliaceae, Physciaceae, Pertusariaceae, and Chrysothricace, respectively. Foliose could be classified into 2 families, 5 genera and 8 species. Physciaceae was the most discoverable family in lichen families and next below was Parmeliaceae.

The lichen samples were randomly taken from the bark of trees. The species of lichen were identified and grouped on the lichen growth. 210 samples of total lichen were taxonomically classified based on anatomical, morphological and chemical constituent substances by spot test, ultraviolet and thin layer chromatography, divided to two groups and identified of 8 families 16 genera and 36 species.

Table 1 the summary of data on lichens diversity (Crustose) in the plant genetic conservation area at University of Phayao.

Thallus	Family	Genus	Species	No.	
Crustose	Chrysothricaceae	<i>Chrysothrix</i>	sp.1	up 179	
		Graphidaceae	<i>Acanthothecis</i>	sp.1	up 184
	<i>Diorygma</i>		sp.1	up 180	
			sp.2	up 185	
			<i>Graphis</i>	sp.1	up 021, 049, 187
	Lecanoraceae	<i>Graphis</i>	sp.2	up 032, 102	
			sp.3	up 044, 104, 206	
			sp.4	up 095, 107	
			sp.5	up 134, 141, 188	
			sp.6	up 163, 115, 191	
			sp.7	up 164, 207	
			sp.8	up 168	
			sp.9	up 190, 186, 204	
			sp.10	up 209	
			Lecanoraceae	<i>Thelotrema</i>	sp.1
	<i>Lecanora</i>	sp.1		up 012, 016, 075, 078	
		sp.2		up 035, 093, 127	
		sp.3		up 055	
		sp.4		up 077, 188	
		sp.5		up 178, 196	
		sp.6	up 181		
	Pertusariaceae	<i>Pertusaria</i>	sp.1	up 167, 014, 039, 143	
			sp.2	up 177, 087, 094, 167	
	Physciaceae	<i>Amandinea</i>	sp.1	up 171	
			sp.2	up 202	
	Trypetheliaceae	<i>Buellia</i>	sp.1	up 043, 099, 202, 208	
			<i>Laurera</i>	sp.1	up 123, 159, 170
				sp.2	up 176, 100
			<i>Trypethelium</i>	sp.1	up 169
total	6 families	11 genera	29 species		

From table 1, Crustose was found in the area of 6 families and 11 genera were as follow; family Chrysothricaceae found 1 genus (*Chrysothrix*), family Graphidaceae found 4 genera (*Acanthothecis*, *Diorygma*, *Graphis*, and *Thelotrema*), family Lecanoraceae found 1 genus (*Lecanora*), family Pertusariaceae

found 1 genus (*Pertusaria*), family Physciaceae found 2 genera (*Amandinea*, and *Buellia*), and family Trypetheliaceae found 2 genera (*Laurera*, and *Trypethelium*).

Table 2 the summary of data on lichens diversity (Foliose) in the plant genetic conservation area at University of Phayao.

Thallus	Family	Genus	Species	No.
Foliose	Parmeliaceae	<i>Bulbothrix</i>	sp.1	up 121, 083, 152, 172
		<i>Parmotrema</i>	sp.1	up 131, 028, 034, 036, 050, 062, 066, 068, 069, 108, 116, 125, 131, 156, 197, 199
	Physciaceae	<i>Dirinaria</i>	sp.1	up 013, 020, 025, 026, 124, 153, 166
			sp.2	up 024, 111, 114, 119, 173, 197
		<i>Hyperphyscia</i>	sp.1	up 007, 017, 022, 064, 135, 140, 144, 147, 151, 154, 166, 041, 071, 083, 109
		<i>Pyxine</i>	sp.1	up 008, 009, 011, 019, 063, 067, 074, 129
			sp.2	up 019, 030, 040, 145, 192
total	2 families	5 genera	7 species	

From table 2, Foliose was found 2 families and 5 genera in the area were as follows; family Parmeliaceae found 2 genera (*Bulbothrix*, and *Parmotrema*) and family Physciaceae found 3 genera (*Dirinaria*, *Hyperphyscia*, and *Pyxine*).

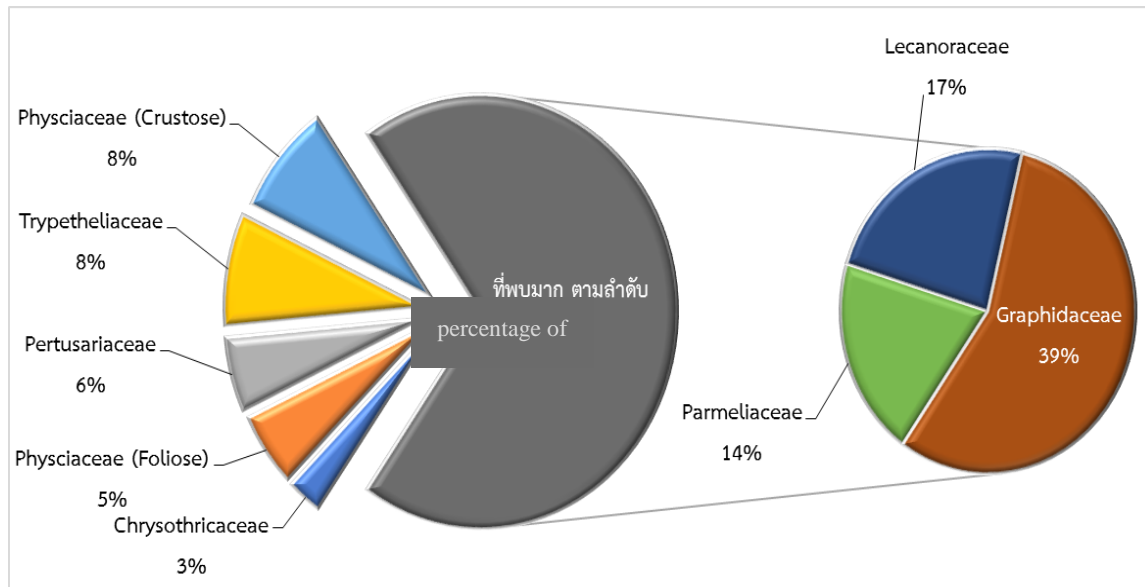


Fig. 1 Percentage of lichen families found in University of Phayao area.

From figure 1, the percentage of lichen families were found in University of Phayao area by according to the frequency of lichens (%) that can be found as follows: Graphidaceae (39%), Lecanoraceae (17%), Parmeliaceae (14%), Physciaceae (crustose) (8%), Trypetheliaceae (8%), Pertusariaceae (6%), Physciaceae (foliose) (5%), and Chrysothricaceae (3%), respectively.

Most of the frequency families of crustose were Graphidaceae (14 species), Lecanoraceae (6 species), Trypetheliaceae (3 species), Physciaceae (3 species), Pertusariaceae (2 species) and Chrysothricaceae (1 species), respectively. Two families 5 genera and 7 species of foliose were found. Most of the frequency families of foliose were Physciaceae (5 species), and Parmeliaceae (2 species).

From studying on the diversity of lichens in the plant genetic conservation area at University of Phayao by the random sampling method. The forest characteristic was a deciduous forest. The research was studied the morphological characteristics of external and internal morphology, performed the chemical inspection with Spot test technique, the test method for substances in lichens with Thin layer chromatography (TLC), the fluorescence test under ultraviolet (UV), and the analysis of lichens by using the classification guide (Key) to identify the most lichens. The results of this research were had in some ways consistent with the results of the other studies.

Grote et al. (2013) were studied the biodiversity of lichens, fungi, and mushrooms for created a diversity database. The research site was the Plant Genetic Protection Area of RSPG, Nampung Dam, Sakon Nakhon Province. Forest in the area was deciduous forest and mixed deciduous forest. They have correspondingly studied the morphological characteristics of external and internal morphology, the fluorescence test under ultraviolet (UV) and the test method for substances in lichens with Thin layer chromatography (TLC) which found the most of lichens in family Raphidaceae, followed by lichens in families Physciaceae, Parmeliaceae, Lecanoraceae, and Trypetheliaceae [21].

Mongkolsuk et al. (2013) were studied the biodiversity of lichens from Phu Luang Wildlife Sanctuary, Loei province at 400-1,500 meters altitude with differential forest types as hill evergreen forest, tropical rainforest, dry evergreen forest, mixed deciduous forest, dry dipterocarp forest, lower montane scrub and coniferous forest that were correspondingly studied the method using taxonomically classified of anatomical, morphological and chemical substances by spot test and thin layer chromatography. They have identified the lichens of 34 families 106 genera and 461 species [22].

Pitakpong and Maungsan (2018) were studied lichen species in a total study of 22 areas at the village in the surrounding area of Mab Ta Phut industrial estate, Rayong province, Thailand that explored 110 of mango trees were found the total lichens of 11 families, 20 genera and 26 species. From observations by suitable weather condition, air quality, habitat and environment for Lichen's life, thus was identically found lichens in the same group from both areas that were Crustose of 3 families and 4 genera were as follow; family Chrysothricaceae found 1 genus (*Chrysothrix*), family Lecanoraceae found 1 genus (*Lecanora*), and family Trypetheliaceae found 2 genera (*Laurera* and *Trypethelium*), and was Foliose of 1 family (Physciaceae) found 3 genera (*Dirinaria*, *Hyperphyscia*, and *Pyxine*) [1].

4. Conclusion

Based on the study of lichens diversity in the plant genetic preservation area, University of Phayao, Mueang District, Phayao province since May to November by surveying and random sampling found that forest characteristics were a deciduous forest. The samples were collected from 210 samples. Crustose was the most discoverable lichens group in the area that has 6 families, 11 genera and 29 species. Most of the frequency families were Graphidaceae (14 species), Lecanoraceae (6 species), Trypetheliaceae (3 species), Physciaceae (3 species), Pertusariaceae (2 species) and Chrysothricaceae (1 species), respectively. Two families 5 genera and 7 species of foliose were found. The families of foliose were Physciaceae (5 species), and Parmeliaceae (2 species). This is a key for identification of lichens in plant genetic conservation area at University of Phayao.

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