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Assessment of Corticolous Lichen Diversity in Romblon State University, Main Campus, Odiongan, Romblon

By Alwin F. Maulion
Romblon State University

Abstract- Corticolous lichens grow on barks of living host trees. Their ubiquitous distribution and sensitivity to pollutants makes them ideal model organisms to biomonitor air pollution and assess air quality. But before these lichens can be used as bioindicators, it is imperative that baseline information on their diversity in a given area must be established for comparison. In this study, the diversity of the corticolous lichens was evaluated in two different sites of Romblon State University Main Campus, Odiongan, Romblon. Sampling ladders with 10 x 10 cm contiguous quadrats were used to assess the diversity of corticolous lichens from different tree species within 10 x 10 m plots set-up in the study sites. Results showed that site 2(LDV= 170.2) showed the highest lichen diversity values than site 1 (LDV= 117.9). At least nine different species of corticolous lichens were reported in this research study. Our research is one of few studies in the Philippines looking at the application of corticolous lichens in biomonitoring environmental quality.

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I. Introduction

Lichens are mutualistic associations of a fungus and an alga or cyanobacterium and occur as crusty patches or bushy growths on trees, rocks and bare ground. The names given to lichens strictly refer to the fungal partner; the algae have separate names. Lichens are very sensitive to sulphur dioxide pollution in the air. Since industrialisation, many lichen species have become extinct in large areas of lowland Britain, one example being the beard moss *Usnea articulata*. This is mainly due to sulphur dioxide pollution, but the loss of habitat, particularly ancient woodland, has also led to reductions in some species. Lichens are sensitive to sulphur dioxide because their efficient absorption systems result in rapid accumulation of sulphur when exposed to high levels of sulphur dioxide pollution. The algal partner seems to be most affected by the sulphur dioxide; chlorophyll is destroyed and photosynthesis is inhibited. Lichens also absorb sulphur dioxide dissolved in water. Lichens are nature's pioneers especially in plants. They are not a single organism the way most other living things are, but rather it is a combination of two organisms which live together intimately. Most of the lichens are composed of fungal filaments, but living among the filaments are algal cells, usually from a green alga or a cyanobacterium and they are poikilohydric, meaning they are capable of surviving extremely low levels of water content.

According to Kershaw (1985) environmental conditions such as climate, substrate, light and moisture play important roles in the distribution of lichen. Lichen species with similar distribution models tend to have similar ecological requirements. Boundreaultet al., (2008) found that the dominance of bryophytes at trunk base and the dominance of lichens at breast height are related to different humidity levels along a tree. So, lichens bark structure influences epiphyte colonization and growth. Lichens in plants are not considered plant pathogens. Only a few cases of parasitic activity by lichens have been reported. The fungal partner of lichen was suspected of killing twigs and small branches of elm by infecting the cork cambium, which is found just below the bark. But this suspected pathogenic activity was never proven.

Instead, lichens are an important part of the ecosystem providing substrate or later succession species, microhabitats and food for herbivores. More importantly for recovering ecosystems, many lichen species have cyanobacteria photobionts or cyanobacteria that closely associated with them are therefore important in nitrogen cycling in which the natural circulation of nitrogen by living organism (Romagni and Gries, 2000). They have several important functional roles in forest ecosystems and they may constitute an important component of the total biodiversity (Dettki and Esseen, 2003). They increase structural complexity, modify canopy water regimes, influence nutrient cycling and provide habitat, food and nest material for many animals (Galloway, 1992; Rhoades, 1995) and are amongst the most significant indicators of air pollution (Richardson, 1992, Wolseley et al 1995, Upreti1995) because lichens are very sensitive to pollution in the air. One indication is when there are too many harmful things in the air, lichens die. If there are many lichens it probably means the air is clean. But, if there are only a few lichens in the neighborhood, the air is probably clogged with automobile fumes or industrial wastes. The bioindicator features of lichens are suitable for determining special ecological conditions such as substrate and air pollution. In recent

Author: Romblon State University. e-mail: alwinmaulion79@gmail.com

years, studies done on these issues have indicated that community structure and diversity of epiphytic lichens vary due to the differences in the environmental conditions and the preferences to substrate of lichens (Pirintsos et al., 1993; Burgaz et al., 1994; Pirintsos et al., 1995; Loppi et al., 1997).

They are also used as a food in many parts of the world. Some species of lichens such as *Cetrariaislandica* was an important human food in northern Europe, and was cooked as bread, porridge, pudding, soup, or salad and *Bryoriafremontii* was an important food in parts of North America, where it was usually pit cooked. Northern peoples in North America and Siberia traditionally eat the partially digested reindeer lichen (*Cladina spp.*) after they remove it from the rumen of caribou or reindeer that have been killed. Rock tripe(*Umbilicaria spp.* and *Lasalia spp.*) is lichen that has frequently been used as an emergency food in North America, and one species, *Umbilicariaesculenta*, is used in a variety of traditional Korean and Japanese foods.

In sense of biological activity, lichens have attracted much attention in investigations because of their antiviral, antibiotic, antioxidant, antitumor, allergenic and plant growth inhibitory activities (Boustie and Grube, 2005; Muller, 2001) and they produce secondary compounds, including pigments that reduce harmful amounts of sunlight and powerful toxins that reduce herbivory or kill bacteria. There are reports dating almost 2000 years old of lichens being used to extract purple and red colors. The pHindicatorlitmus is a dye extracted from the lichen genus *Rocellatinctoria* which was used in dyeing silken and woollen goods by boiling. Extracts from many Usnea species were used to treat wounds in Russia in the mid-twentieth century. The substance olivetol is found to be naturally present in certain species of lichens. This is a property it shares with the cannabis plant, which internally produces the related substance olivetolic acid (before using it to biosynthesis tetrahydrocannabinol (THC).

However, lichens have been essentially ignored by the modern pharmaceutical industry, despite the fact that lichen produce a large number of secondary metabolites with diverse structures and that studies have provided evidence of biological activity extracts from whole native lichens.

Some species of lichens are one of the most threatened organisms. The main threats that apply to biodiversity in general are also true for lichens, e.g. habitat degradation and loss (Groomet al., 2006), habitat fragmentation (Bergamini et al., 2005), overexploitation (Upreti et al., 2005), species invasions (LaGreca and Stutzman, 2006), and climate change. For instance, climate change is likely to have dramatic effects on distribution and abundance of lichen populations (Ellis and Coppins, 2007; Ellis et al., 2007).

Overexploitation of lichen populations for human uses is a serious problem, even if the demand is not increasing, but the size or quality of the habitat is declining. Habitat degradation and loss is the most serious threat to biodiversity in general (Groom et al., 2006) and in lichens in particular (Wirth, 1976, 1999). Loss of habitat leads to a reduction of local population sizes, and saxicolous, terricolous and epiphytic species are all similarly affected. Habitat loss has been identified as the most widespread threat to lichens, clear-cuts of old or natural forests accounting for 63 % of lost sites (Wolseley, 1995). Deforestation and degradation of lichen habitats by the replacement of natural forests with plantation forests have both a drastic effect on species richness and composition of lichen communities (Rose, 1992).

Monitoring programs and more specific concerns about environmental monitoring are required to ensure that lichens ecosystem are conserved and manage sustainably to maintain their environmental benefits in the ecosystem.

Unfortunately, in the case of Romblon State University there are no studies regarding lichens diversity and distribution that are made to catch the attention of the public agency in the government. Assessment of these organisms was the effective tool in giving the information about environment monitoring.

II. Statement of the Problem

This study was conducted to assess the diversity and of corticolous lichens found in the Romblon State University, Main Campus.

Specifically it aims to find answers to the following questions;

1. What is the diversity of corticolous lichens in Romblon State University (Main Campus), Odiongan, Romblon?
2. What are the different lichens species found in the study area?
3. Is there a presence of lichen indicator species in the study area?

III. Significance of the Study

The primary concern of this study was to assess the diversity and the different identification of lichens found in Romblon State University, Main Campus.

Findings of the study would help to determine the different kinds of lichens species present inside the Romblon State University, Main campus. With this, we could provide a basis for identification of lichens found in RSU, Main Campus.

The study would enable the researchers to be familiarized with the lichen species and to become aware of the environmental conditions that lichens contribute such as bioindicator of air pollution.

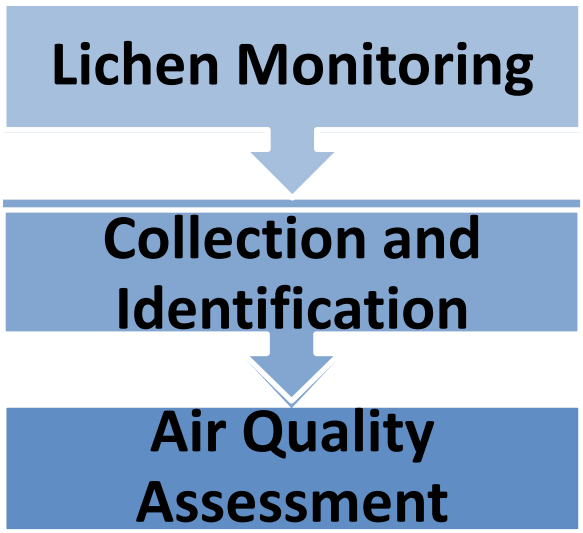
The result of this study would be a source of reference and guidance for replication of those who are interested to conduct other studies regarding lichens.

IV. Scope and Delimitation of the Study

The study was conducted to assess the diversity and distribution of lichen species found in RSU, Main Campus. The study focused entirely on lichens growing on trees. Making of quadrats in different aspect (N, E, W, S) of trees are determined in the study.

The study was limited to assessment of corticolous lichen species diversity found in RSU, Main Campus. The duration of the study lasted for three (3) months.

Conceptual Framework



a) Related Literature

Lichen was introduced into Greek literature about 300 B.C by Theophrastus primarily to describe outgrowths from the bark of olive trees, and this is the first written record on lichens (Hawksworth and Hill, 1984). Up to end of the 16thcentury, descriptions of lichens were entirely based on their physical appearance and were often incorrectly described as type of mosses or seaweeds.

Lichens were alluded to by only a few writers during the next 2000 years. This reflected not only the small amount of study in natural history, but also the relative lack of economic worth of lichens. However, the advent of microscope in the beginning of 18thcentury enabled detailed anatomical studies of lichens, which revealed their special dual character consisting of algal and fungal partners. This led to a series of more refined definitions.

Schneider (1897) wrote a history of lichenology, recognizing the following periods:

1. From the earliest times to the end of the seventeenth century.
2. From 1694, when Tournefort, the first to separate lichens taxonomically from the bryophytes, arranged plants into classes called genera, to 1729.
3. From 1729, when Micheli divided lichens into different orders, to 1779.
4. From 1779, when Weber established definite and reasoned lichen genera based on thestructure of thallus and fruits, to 1825.
5. From 1825, when Wallroth and Meyer each published works dealing with detailed morphological, ecological, and biochemical observations, to 1868.
6. From 1868, when Schwendener discovered the dual nature of lichens, to 1894.

The difficulty in finding a universal definition for 'lichen' results from the variability of fungal-algal associations and the range of symbiosis. A number of definitions of lichens are provided in contemporary literature (Hawksworth and Hill 1984, Orange 1994, Purvis 2000, Ulloa and Hanlin 2002, Wolseley et al. 2002, Allaby 2004, Gilbert 2004, Lawrence, 2005). However, the interpretation of lichen as an association of two organisms living in symbiotic relationship seems to be the most common dimension of these definitions. Indeed, in the Ainsworth and Bisby's Dictionary of the Fungi (Kirk etal. (eds.) 2001) lichen is defined as a stable self-supporting association of a fungus (mycobiont) and an alga or cyanobacterium (photobiont). More precisely, lichen is described as an ecologically obligate means able to exist under only one set of environment conditions, stable mutualism between an exhabitant fungal partner and an inhabitant population of extracellularly located unicellular or filamentous algal or cyanobacterial cells.

Perhaps a more widely accepted idea was given by Imshaug (1951)who defined a lichen as "an entity capable of reproducing itself, and consisting of two organisms, an alga and a fungus, living together in a state of symbiosis, as is manifested by some change in the anatomy, morphology, or physiology of at least one of its components."

Lichens do not have independent scientific names; the fungal and photosynthetic partners each have separate names, and names given to lichens are considered as referring to the fungal partner alone. The classification of lichens is therefore integrated into the system of Fungi. Current nomenclature is consistent with the recognition of lichens as a nutritional rather than a taxonomic group. The nomenclature of fungi including lichen–forming fungi is governed by the international code of botanical nomenclature (Kirk et al. (eds.) 2001).

b) Use as Bio-indicators

Lichens are widely used as environmental indicators or bio-indicators. If air is very badly polluted with sulphur dioxide there may be no lichens present, just green algae may be found. If the air is clean,

shrubby, hairy and leafy lichens become abundant. A few lichen species can tolerate quite high levels of pollution and are commonly found on pavements, walls and tree bark in urban areas. The most sensitive lichens are shrubby and leafy while the most tolerant lichens are all crusty in appearance. Since industrialisation many of the shrubby and leafy lichens such as *Ramalina, Usnea* and *Lobaria* species have very limited ranges, often being confined to the parts of Britain with the purest air such as northern and western Scotland and Devon and Cornwall.

c) Zonation of Lichens

A lichen zone pattern may be observed in large towns and cities or around industrial complexes which corresponds to the mean levels of sulphur dioxide experienced. The table shows the lichen zone scale of Hawks worth & Rose (1970). Particular species of lichen present on tree bark can indicate the typical sulphur dioxide levels experienced in that area. For example if there are no lichens present, the air quality is very poor (zone 1), whilst generally only crusty lichens such as *Lecanora conizaeoides* or *Lepraria incana* can tolerate poor air quality (zone 3). In moderate to good air, leafy lichens such as *Parmelia caperata* or *Evernia prunastri* can survive (zone 6) and in areas where the air is very clean, rare species such as 'the string of sausages' *Usnea articulata* or the golden wiry lichen *Teloschistes flavicans* may grow (zone 10).

It is important to note that the zone chart in Table 1 applies to areas where sulphur dioxide levels are increasing. If sulphur dioxide conditions are falling, lichens rarely colonise in exactly the same sequence; lichens are slow growing and may take a year or two to recolonise bark or other substrates following a reduction in air pollution levels, and tiny recolonising specimens can be difficult to spot and identify.

During the early and mid-twentieth century, air pollution levels were much greater than they are today in towns and cities of the UK. Sulphur dioxide levels were highest in the inner city areas becoming less polluted out towards the edges of the urban areas. At such times, the lichen zone scale would often highlight zone 1 as the inner city area, moving through the zones to the cleaner air at the edge of the city. From the 1970s onwards, sulphur dioxide levels have been falling markedly in the central and outer areas of cities, such that there may be no differentiation between levels in central and outer areas of many cities. The fall in sulphur dioxide levels between the 1970s and the 1990s has led to a number of lichens recolonising in areas from which they had previously been eliminated.

d) Ecology of Lichens

In general, three major life forms of lichen thallus are recognized, crustose (crust-like biofilm), foliose (leaf-like), and fruticose (branched tree-like, shrubby, pendulous; thalli (Hawksworth et al. 1995; Büdel and Scheidegger, 1996). The fourth type, gelatinous thallus, is restricted to some cyanobacterial lichens (Büdel and Scheidegger, 1996). Even without roots, lichens can efficiently extract nutrients (phosphorus, magnesium, calcium, potassium, sulfur, and iron) from recalcitrant surfaces (Richardson, 1975). Rhizinae on lichen thalli may have a function in the uptake of nutrients. Lichens often grow in habitats with extreme light, dryness, or temperature, which are less favorable or unsuitable for higher plants (Kershaw, 1985; Vrablikova et al. 2006).

Lichen thalli are poikilohydrous, which means that their water status passively follows the atmospheric humidity (Nash, 1996; Kappen, 2000). The presence of water rapidly activates lichen metabolism (Nash 1996, Schlensog et al. 2004). Recovery of the photosynthetic apparatus after the dark winter takes only minutes in Antarctic lichens, whereas in mosses it is a longer process (Schlensog et al.2004). Incredible adaptations enable some cold-adaptedgreen algal lichens to activate their photosynthesis at −20°Cwith water vapor obtained from snow. Photosyntheticactivity can be high by at 0°C (Kappen et al. 1996; Kappen, 2000; Richardson, 2002). Certain strategies increase the fitness of some lichen over others in dry habitats. The right choice of the photobiont, the water holding structures, and a tolerance to osmotic stressare some of the survival strategies. While green algae in lichens are able to activate their photosynthesis with water vapor, cyanobacteriain lichens need liquid water (Rundel 1988; Richardson 2002). This explains why algal lichenssurvive in dryer habitats than cyanobacterial lichens green; which in humid tropics represent nearly half of the known lichen species. Some cyanobacteriallichen species with gelatinous polysaccharides-containingthalli and green algal lichens with cushions' water-storing thalli are able to extend their daily metabolism compared to thin, easily drying lichen species (Richardson, 2002).

e) Lichen Symbiosis

Lichens are the symbiotic phenotype of nutritionally specialized fungi that acquire; in an ecologically obligate symbiosis, fixed carbon from a population of green algal or cyanobacterial cells (Dembitsky, 2003; Honegger, 1998; Yuan et al., 2006).

According to Hawksworth et al. 1995, lichen is an ecologically obligate, stable mutualism between an exhabitant fungal partner and an inhabitant population of extracellularly located unicellular or filamentous algal or cyanobacterial cells.

Scholler (1997) described how in the 18thcentury lichens on the bark of trees and rocks were recognized as physically joined algae and filaments of fungi. Indeed, this dual character of lichens was recorded as comprising algae and fungi living in a symbiotic relationship. This symbiotic description provided a more

specific explanation of the living arrangement between both partners.

Fink (1913) gave his own idea in the following statement:

> "The lichen is a fungus which lives all or a part of its life in parasitic relation with an algal host and also sustains a relation with an organic or an inorganic substratum."

The lichen symbiosis probably evolved around 400– 600 million years ago (Yuan et al. 2005). Lichens can be considered as ecosystems where the interaction of partners results in behavior and life forms that are not found in the isolated partners (Nash 1996).

Lichens are not regarded as a taxonomic group, butlichen taxonomy is based on the taxonomy of the fungal partner, the mycobiont (Tehler, 1996). In a course ofevolution, about 13,000 extant fungal species (Hawksworth, 2001) have specialized in gaining their carbon and about1, 500 species also in gaining their nitrogen from a photosynthesizingpartner (Hawksworth et al. 1995).Nearly 19% of all fungi are lichenized (Lutzoni et al. 2001; Hawks worth et al. 1995). The fungal diversity alone offers a great metabolic potential for new ecological and biotechnological discovery.

More than 98% of lichenized fungal species belong to phylum Ascomycota, a few to orders of phylum Basidiomycota and some to Mitosporic fungi (Hawks worth et al. 1995; Tehler 1996). Most of the lichenized fungi (mycobionts) form lichen symbiosis with green alga (Chlorophyta; Lewis and McCourt 2004), only about 10% with cyanobacteria, and 3% with both green alga and cyanobacteria (by Scheider et al. 1987 as cited by Woess 1988). Most of the tripartite lichen thalli consist of lichen fungi and green alga while the cyanobacteria are spatiallyseparated from alga in internally or externally occurringfungal compartments called cephalodia (Büdel and Scheidegger 1996). Some mycobionts can also changetheir photosynthesizing partner from green alga tocyanobacterium and vice versa and this leads to changesin thallus morphology. This behavior was suggested to be due to an environmental adaptation and related to ecological compatibility of the photobiont (Honegger1996; Stenroos et al. 2003).

Future studies with careful evaluation of cyanobacterial taxonomy (Oren, 2004) and carefully chosen DNA markers should result in a clearerpicture of the taxonomic diversity of lichen photobionts (Oksanen, 2006). There are challenges in finding appropriate DNA markers that have descended directly from a common ancestor that provide sufficiently but not too much nucleotide variation and have conserved sites for primer design (Oksanen et al.2004; Sánchez-Baracaldo et al. 2005).

f) Reproduction of Lichens

Lichens reveal various reproductive strategies where the mycobiont and its photobionts either disperse separately, in the case of sexual reproduction (horizontal transmission ofphotobionts) or where the lichen symbionts are co-dispersed with clonal, symbiotic propagules (vertical transmission of photobionts; Yahr et al., 2004). Lichens reproduce either with fungal spores (Büdel andScheidegger1996; Murtagh et al. 2000) that have to find a suitable photobiont or by vegetative propagules including both partners (Büdel and Scheidegger 1996). Crustoselichens grow slowly, ≤0.87 mm/year (Karlen and Black, 2002); other growth forms from0.06 to 36.5 mm/year Richardson, 1975). With a few exceptions, where photobionts grow betweesn the meiosporangia of the mycobiont and are co-dispersed with the ascospores (Ahmadjian, 1993), sexual reproduction is always associated with horizontal transfer of photobionts. A high number of species develop symbiotic propagule types such as is idia or soredia that facilitate clonal reproduction of the symbiosis (Budel and Scheidegger, 2008). In many species, these diaspores are multifunctional and can develop intoregeneration structures (Ott et al., 1993). Species with a predominantly clonal reproductive mode can exhibit extensiveclonal genetic structure. Some predominantly sexually reproducing lichen fungi may lack any structure at the local scale (Werth and Sork, 2008).

Zoller et al. (1999) were the first to recognize that lack of ascomata in strongly fragmented and geographically isolated populations of *Lobaria-pulmonaria* (''lungwort'') might be due to missing mating partners.

Sexual reproduction in lichens refers specifically to the sexuality of the lichen-forming fungus. During fungal sexual reproduction, ascospores are formed in ascomata (Ascomycetes) or, in basidiolichens, basidiospores are formed. Some lichen-forming fungi are capable of both selfing and outcrossing (homothallism), while others are obligatory outcrossers (heterothallism) (Zoller et al., 1999).

Some lichen-forming fungal species exhibit contrastingre productive strategies in different parts of their ranges (Poelt, 1970, 1972; Tehler, 1982; Mattsson and Lumbsch, 1989; Lohtander et al., 1998; Kroken and Taylor, 2001, Cornejo et al., 2009). Often, due to their different reproductive mode, these were described as separate species, when in fact they are conspecific. One example of these so-called ''species pairs'' is the sexual lichen *Porpidiaflavocoerulescens* and its clonal counterpart, *P. melinodes* (Buschbom and Mueller, 2006).

g) Ecological Diversity and Distribution

i. *World Status*

Lichens are widespread in many forests ecosystem (Dettki and Esseen, 2003).Lichens are the most successful symbiotic organisms in nature, dominating 8% or more of the earth's terrestrialarea (Ahmadjian,1995). According to Upreti (1998) India is a rich centre of lichendiversity, contributing nearly 15% of the 13,500 species of lichens so far recorded in the world and its share of just 2×4% of global land surface. Since then, they have colonized almost all habitats and extreme conditions, from epiphytic (growing on trees) to end olithic (growing under the surface of rocks), and from Antarctica to the highest mountains and sea shores (Nash III, 1996).

Lichens occur commonly as epiphytes on trees and other plants, and in some ecosystems epiphytic lichen biomass may exceed several hundred kg ha^{-1}(Coxson, 1995). In addition, they frequently colonize bare soil, where they are an important component of cryptogrammic soil crusts in arid and semi-arid landscapes(Belnap and Lange, 2003). Furthermore, lichensoccur almost ubiquitously on rocks with the most obvious ones occurring asepiliths, either growing over the surface or embedded within the upper fewmillimeters. A few lichens even occur to endolithically within the upper fewmillimeters of the rock, such as occurs in Antarctica (Friedmann, 1982).

Lichens occur in most terrestrial ecosystems of the world, but their biomasscontribution varies from insignificant to being a major component ofthe whole ecosystem (Kershaw, 1985).

In the study of Giao (2009) Eighty three (83) species of macrolichens are reported from Langbian Mountain and Ngoclinh Mountain, located in the Western Highlands of central Vietnam, including 61 new records for Vietnam (Aptroot and Sparrius,2006) estimated at least 1000 species lichen is in Vietnam.

New microlichen species for Thailand are described by Sparrius and Saipunkaew (2005).

Dodge (1973) reported 86 genera including 424 species of lichens from Antarctica and its adjacent islands.

According to Negi (2000) India is a rich center of lichen diversity, contributing nearly15% of the 13,500 species of lichens so far recorded in the world.

ii. *Philippine Diversity*

In Philippines, the total number of lichen species credited now is 790 (DENR1999; Tacio 2004).

Recently, Dulnuan (2006) reported 3 species of lichens with fruticose type of growth from a total of 52 lichen genera collected in Ifugao, Mountain Province.

Earlier study by Herre (1957) reported only 68lichen species from 26 provinces in Luzon (12), Visayas (6), and Mindanao (8). Majority of the species were foliose (45 species) and crustose (36 species) type of lichen growth. Eleven species were recorded as fruticose, while only one species was noted as squamulose. Among the fruticose type of lichens reported were species of Cladonia(2), Stereocaulon(2), and Usnea(7). Herre(1963) also reported from Bataan, Ilocos Sur, and Misamis, Negros Oriental, and Rizal five species ofUsnea: *U. hossei, U.longissima, U. marivelensis, U. misamisensis, and U. squarrosa.*

iii. *Biological Activities*

The challenge for today's pharmaceutical industry lies in the discovery and development of new pharmacological active molecules due to resistance to available antibiotics (Bahera et al., 2005). Similar to higher plants, lichens were used since antiquity as natural drugs, together with some marine organism and frog venom, are important sources of biologically active compounds (Barner, 2000). Their efficacy is due to the synthesis of unique secondary compounds, a number of which have important biological roles (Perry et al., 1999).

According to Elix (1996) lichens produce a wide range of organic compounds that can be divided into two groups calledprimary metabolites and secondary metabolites. Primary metabolites are proteins, lipids, carbohydrates, and other organic compounds that are essential to the lichen's metabolism and structure. Some of these metabolites are produced by the lichen's fungal partner and others by the lichen's algal or cyanobacterial partners. Secondary metabolites are produced by the fungus alone and secreted onto the surface of lichen's hyphae either in amorphous forms or as crystals. If these substances are only found in lichens, then they are called lichen substances (Ozturket al., 1999). Burkholder et al. (1944) reported for the first time the presence of antibiotic substances in lichens.

The chemistry of about one third of all lichen species has been studied up to now and about 350 secondary metabolites are known from lichens. The chemical structures of approximately 200 of them have been established. They are extracellular products of relatively low molecular weight crystallized on the hyphal cell walls. Also they are usually insoluble in water and can be extracted into organic solvents. They amount to between 0.1 and 10% of the dry weight of the thallus, sometimes upto 30% (Galun, 1988).

After the discovery of penicillin from a fungus, numbers of lichens were screened for antibacterial activity in the 1940s and 1950s . For example, usnic acid has been used as atopical antibacterial agent and also it showed antimicrobialactivity against Gram-positive organisms 'in vitro (Lauterwein et al., 1995). Protoli-chesterinic acid exhibited in vitro activity against Helicobacter pylori (Ingolfsdottiret al., 1997). Lauterwein et al. (1995) investigated in vitroactivities of vulpinic acid and usnic acid against some aerobic and anaerobic microorganisms. Fournet et al. (1997) studied the activity of the lichen compounds usnic acid, pannarine

and 1_-chloropannarine against promastigotes forms of three strains of *Leishmaniaspp*. In addition lichens have been used for medicinal purposes throughout centuries. For example, *Lobariapulmonaria*, *Cetrariaislandica*, and *Cladonia*species were reputed to be effective in the treatment of pulmonary tuberculosis. Both enantiomeric forms of usnic acid inhibited the growth of *Mycobacterium tuberculosis* and *Mycobacterium tufu* in vitro at a relatively low concentration (Krishna and Venkataramana, 1992). In vitro activities of five common lichen compounds were screened for *Mycobacterium aurum* by Ingolfsdottir et al. (1998).

Two recent reviews summarize its antimicrobial, antiprotozoal, antiviral, antiproliferative, antiinflammatory, analgesic, antipyretic, and antitumoractivities as well as some other propertiessuch as UV protection, allergenic potential, toxicity (Cocchiettoet al., 2002; in Ingolfsdottir, 2002). Ingolfsdottir's review presents a comprehensive listfor the antimicrobial activity of (+)-usnic acid and(D)-usnic acid against gram positive and gramnegative, anaerobic bacteria, mycobacteria, andyeast/fungi with the relevant references.

Ghioneet al. (1988) reported the antibacterial activity ofusnic acid against *Streptococcus mutans*, *Streptococcuspyogenes*, and *Staphylococcus aureus*.

Lauterwein et al. (1995) determined in vitro activitiesof (+)-usnic acid, (D)-usnic acid, and vulpinic acid against aerobic and anaerobic microorganisms. They found that these lichen compounds did not inhibit gram negative rods or fungi at concentrations lower than32 μg/ml but were active against clinical isolates of *Enterococcus faecalis*, *Enterococcusfaecium*, *Staphylococcus aureus*, and someanaerobic bacteria.

The usnic acids (Ingolfsdottir, 2002) have also been found to exhibit antihistamine, spasmolytic, and antiviral properties as well as being active against gram-positive bacteria and streptomycetes. Indeed they are used in commercially available antiseptic creams including ''Usno'' and ''Evosin.'' Usnic acid is reported to be more effective than penicillin salves in the treatment of external wounds and burns and is also used to combat tuberculosis. The active centers of the usnic acid molecule seem to be the benzofuran or dihydrodibenzofurannucleus, the phenolic hydroxy groups and the 4,4a-double bond in the dihydroaromaticring. The antibiotic action of usnic acidis due to the inhibition of oxidative phosphorylation, an effect similar to that shown by dinitrophenol. More recently Shibuya et al. (1983) showed that4-O-methylcryptochlorophaeic acid was a powerful inhibitor of prostaglandinbiosynthesis and a potentially useful anti-inflammatory drug.

Lichen substances are also known to exhibit anti-tumor activity. Usnic acid has low level activity against lung carcinoma. Pannarin inhibited cell growth and induces cell death in human prostate carcinoma DU-145 cells (Maier et al. 1999).The orcinol derivatives tenuiorin and methyl orsellinate present in extract of *Peltigera leucophlebia* (Nyl.) Gyeln. (Peltigeraceae) exhibited in vitro inhibitory activity against 15-lipoxygenase from soybeans. A correlation has been observed between 5-lipoxygenase inhibition and anti-proliferative effects for related lichen metabolites. On this account, tenuiorin and methyl orsellinate were further tested for anti-proliferative activity on cultured human breast, pancreatic and colon cancer cell lines. Methyl orsellinate lacked anti-proliferative activity but tenuiorindepicted moderate activity (Ingolfsdottir et al. 2002). Bianthraquinone glycosides, colleflaccinosides isolated from *Collemaflaccidum* (Ach.) Ach. (Collemataceae) collected in Israel and Russia, were reported to have antitumor activity (Rezanka and Dembitsky, 2006).

However, the most active anti-tumor lichen substances are water soluble polysaccharides which appear to bepartially O-acetyled homo-D-glucans (Nash, 1996).

Various plant-derived and lichen-derived compounds that are known to have antimicrobial activity against "normal" microbes constitute one noteworthy group of candidates that might have activity also against MRSA or VRE or against resistant bacteria in general. One such compound is (+)-usnic acid, an old lichen-derived drug with antimicrobial and many other interesting biological activities (Ingolfsdottir et al. 1998; Huneck1999). Topical formulations of this drug, either as such or in salt form, have been subject to pilot clinical studies(Cocchietto et al. 2002), and the drug is usedin antifeedant products (Durazo et al. 2004), mouth rinses,and dentifrices (Grasso et al. 1989) as well as in cosmetics(Najdenova et al. 2001).

According to Crockett et al. (2003) and Rankovic et al. (2007) used lichens as medicine in treatingwounds, stomach diseases, and whooping cough in America andin Europe.

Quisumbing (1951) earlier reported the medicinal properties of fruticose lichen *Usneaphilippina*. Santos et al. (1964) tested the biological activities of these lichens and other fruticose lichens, e.g., *Usneasp*., *Ramalina sp*. and *Stereocaulon sp.*, and reported their inhibitory activitiesagainst Gram-positive bacteria such as *Micrococcus pyogenesvaraureus* 209 P (syn=Streptococcuspyogenes), penicillin-resistant *Micrococcus pyogenesvaraureus*, *Bacillus subtilis*, and the acid-fast bacilli, *Mycobacterium tuberculosis* 607.

Santos and Mondragon (1969) also conducted thin layer chromatographic analysis ofthese lichens and detected the following lichen acids: salazinicacid, stictic acid, usnic acid, barbatic acid, protocetraric acid, zeorin, atranorin, lecanorin, and homosekikaic acid. However, itwas not reported whether any of these metabolites is responsible for its antibacterial activities.

Antifungal activity of lichen extracts and lichen acids against plant pathogenic fungi was reported

(Gulluce et al., 2006; Halama and VanHaluwin, 2004; Oh et al., 2006).

Lichens have a large variety of uses and for some of them, ethnopharmacological properties are reported as for *Cetrariaislandica* still indicated as a cough remedy (Van Haluwyn and Lerond, 1993).Studies reporteda variety of very interesting properties e.g. antibiotic (Ogmundsdottir et al. 1998), antioxidant (Hidalgo et al.1994), anti-HIV (Neamati et al. 1997).

Caperatic acid and extracts of the lichens *Flavoparmelia baltimorensis* and *Xanthoparmelia cumberlandia have* antiherbivore activities against the snail *Palliferavaria* (Lawrey 1983, 1989). Methyl b-orcinolcarboxylate, ethyl hematommate and 5-chlorohematommate show nematocidic activity on larvae of Toxocaracanis (Ahad et al. 1991). Giez et al. (1994) and Emmerich et al. (1993) studied the effect of lichen substances on the growth and development of the polyphageous insect Spodopteralittoralis: atranorin, pulvinic acid dilactone, calycin, parietin, evernic, psoromic, physodic, 3-hydroxyphysodic, fumarprotocetraric, stictic, norstictic, salazinic, vulpinic, rhizocarpic, and usnic acids.

Heteroglycans and a beta-glucan isolated from *Thamnolia vermicularis* var. *subuliformis* were tested for in vitro immune modulation activity and reported to have various influences on the immune system (Omarsdottir,, et al, 2007).

Previous phytochemical studies on *Usnea longissima Linn*. also known as Old Man's Beard, resulted in the isolation of several lichen acids, with anti-inflammatory, analgesic, antipyretic, anti-tumor, anti-cholesterol and nematocidal properties (Yamamoto et al., 1995; Nishitobaet al., 1987).

According to Nash (1996), lichen substances have a harmful property. In northern Europe the lichen *Letharia vulpine* was used traditionally as a poison for foxes and wolves. The toxic principle is the pulvinic acid derivative vulpinic acid, which is not only poisonous to all meat eaters but also to insects and molluscs. Surprisingly this compound is ineffective against rabbits and mice. The secalonic acid derivatives are also highly poisonous. These substances are mycotoxins and, like vulpinic acid, may have evolved to serve a twofold ecological role. Thus, in addition to screening incoming light, they are highly poisonous to grazing herbivores (Nash, 1996).

Contact dermatitis, a severe skin rash, is well known among forestry and horticultural workers in North America, forming part of a syndrome known as' woodcutter's eczema'' or ''cedar poisoning.'' These complaints are an allergic response resulting from exposure to various lichen substances. Among the lichen substances responsible are usnic acid, evernic acid, fumarprotocetraric acid, stictic acid, and atranorin. Usnic acid, for instance, is a common lichen substance in the corticolous species of Alectoria, Evernia, and Usnea, which are widespread in the forests of North America. A dusting of soredia on clothing causes allergic reactions in the wives of lumbermen not directly exposed in the forests. Atranorin and stictic acid are also capable of photosensitizing human skin as well as being contact allergens. This can lead to photo contact dermatitis, where the allergic reactions become much more acute when the persons are exposed to the lichen substances in combination with light (Hale, 1983; Richardson, 1988).

Periodically hundreds of elks die in western North America when these largeruminants are forced out of their normal winter habitats by excessive snows and at lower elevations primarily find *Xanthoparmelia chlorochroa* to eat. Although the toxin is fully resolved, the abundance of salazinic acid is suspected. In contrast, these animals eat other epiphytic lichens without apparent ill effects (Nash, 1996).

iv. *Lichens in Perfume*

Large amounts of two lichen species are being processed in the perfume industry (mainly at Grasse, France): 1900 tons/year of *Pseudevernia furfuracea* ("tree moss"; 1997 level) and about 700 tons/year of *Evernia prunastri*. The lichen extracts have a certain "green" aspect caused by esters of substituted aromatic acids and act as fixatives. The combined lichen material and tree bark is subsequently extracted with an organic solvent and treated with ethanol. The concentrate of this solution contains a mixture of essential oils and depside derivatives (degradation products). The final extract with its sweet ''mossy'' smell is used in some perfumes to ensure persistence on the skin, as the major ingredients do not evaporate readily. The lichen extract may amount to 1–12%of the finished perfume. The precise identity of the scented component remains a trade secret but comprises a very small proportion (c. 0.04%) of the total extract, the majority of which comprises borneol, cineole, geraniol, citronellol, camphor, naphthalene, orcinol, orsellinate esters and their homologues (Moxham 1980; Richardson 1988; Hiserodt et al. 2000).Usnic acid is used as a preservative in cosmetic creams (Seifert and Bertram 1995), and atranorin, pannarin, gyrophoric, acid and usnic acid are applied in suntan preparations (Fernandez et al. 1996).

v. *Lichens in Dyeing*

Lichens were used as a source of dyestuff from the time of the ancient Greeks and probably earlier (Henderson 1999), but are of little economic importance today. Historically *Roccellamontagnei*, common fruticose lichen on rocks,provided valuable red or purple dyes in the Mediterranean region. These dyes were produced by ''fermenting'' the Roccella or chemically equivalent species (*Ochrolechiatartarea*, *O. androgyna*, or *Parmotrematinctorum*) with dilute ammonia solution. The macerated lichen and dilute ammonia were sealed in a container containing twice the volume of air. The

purple color developed after a week and was used as a direct dye (orchil) for protein fibres (wool and silk). The simple para-depsideserythrin (*Roccella*) and lecanoric acid (*Ochrolechia* and *Parmotrematinctorum*) present in these lichens are responsible for these colors. Rapid basehydrolysis of the lecanoric acid or erythrin by ammonia gives ammoniumorsellinate and then orcinol (by decarboxylation). Subsequent oxidative coupling in the presence of ammonia gives rise to the dyestuff, orcein, which comprises a mixture of three major chromophores, 7-hydroxyphenoxazone, 7-amino-phenoxazone and 7-aminophenoxazine (Hale, 1983). The common acid-base indicator litmus, formerly widely used in chemistry laboratories, is closely related to orcein but represents a more complex mixture of polymeric compounds with the 7-hydroxyphenoxazone chromophore and its anion being responsible for the sensitivity of the color to pH (Nash, 1996).

vi. *Lichens as Environmental Indicators*

Lichens have been recognized as being very sensitive to air pollution for many years (Nimis et al. 2002).

According to Garty, 2001, lichens adsorb and are sensitive to heavy metals. Coccomyxa photobiont species were more sensitive to metals than Trebouxia species and this may affect the habitat preference of lichens containing these green algae.

Lichens are used in environmental monitoring of industrial pollution (Garty, 2001). Monitoring methods include quantification of lichen populations, examination of lichen morphology, and heavy metal analyses of natural or transplanted thalli (Garty, 2001). The emission of ethylene is one of the measures of air pollution stress even though ethylene biosynthesis and its control in lichen are not fully understood (Ott et al. 2000; Oksanen, 2004). A new method for environmental monitoring involves the reduction of triphenyltetrazolium chloride to colored triphenylformazan in lichen (Backor and Fahselt, 2004).This measure of lichen dehydrogenase activityindicates environmental stress in lichens and their isolated bionts.

The strongest case for using lichens as bioindicators of air pollution involves sulfur dioxide (Grace et al. 1985a; Seaward 1993; Hawksworth, 2002; Nash and Gries, 2002). Some forms of coal (and other fuel products) have particularly high levels of sulfur, and its oxidation leads to the formation of sulfur dioxide, one of the major gases associated with acid rain. In fact sulfur dioxide has only an average atmospheric residency time of about 12 hours, because its high solubility in water leads to its trapping in water vapor aerosols and rapid conversion to sulfuric acid, one of the stronger acids (Nash, 1996).

h) Related Studies

Macrolichens cover and their distribution pattern on two common *Quercus semecarpifolia* and *Rhododendron arboreum* trees from the moist temperate forest (Chopta) of Garhwal Himalaya. Out of three d. b. h. classes trees (diameter at breast height), d. b. h. between 0.1-0.30 m, has found maximum cover of macro-lichens at southeast aspect (Nature and Science, 2009).

Recently, on 26 April 2012, scientists reported that lichen survived and showed remarkable results on the adaptation capacity of photosynthetic activity within the simulation time of 34 days under Martian conditions in the Mars Simulation Laboratory (MSL) maintained by the German Aerospace Centre (DLR).

In a pioneering study, Culberson et al. (1988) attempted to elucidate the sexual cycle in *Cladonia chlorophaea* using chemical markers. Earlier investigations on North American populations of *C. chlorophaea* had distinguished 14 distinct chemotypes, which were interpreted as sibling species. Culberson et al. (1988) analyzed secondary products in progeny of individuals of *C. chlorophaea* taken from populations of mixed chemotypes. According to Vartia (1973) lichens prevent to decay wood by fungi. He reported that characteristic secondary metabolites of lichens, such as usnic, divaricatic and lichesterinic acids, inhibit the growth of some filamentous fungi. It was therefore expected that lichen mycobiont cultures would yield growth inhibitors of wood decaying fungi (Yamamoto et al. 2002a).

In 2008, scientists from the European Space Agency (ESA) sent a suitcase-sized Expose-Experiment package to the International Space Station (ISS) filled with organic compounds and living organisms to test their reaction to outer space. The samples returned to Earth in 2009. Lichen has proven to be tough cookies – back on Earth, some species continue to grow normally. ESAs Rene Demets explains: “These organisms go into a dormant state waiting for better conditions to arrive.”

According to the Bergquist of Journal Sentinel, 2011, a laboratory study has found that lichens on Wisconsin's landscape break down the infectious proteins that are responsible for causing chronic wasting disease, or CWD - the devastating neurological disorder that was discovered in Wisconsin's wild deer population in 2002. The study by researchers at a federal government animal health laboratory in Wisconsin showed that certain lichen organisms contain an enzyme that is capable of degrading prions.

V. Research Methodology

This study utilizes an experimental research design. Corticolous lichens were collected from different species of trees from two study sites. The collected specimen will be identified by observing its characteristics using “Consortium of North American

Lichen Herbaria and "A Guide to the Study of Lichens by Schneider".

a) Research Locale and Time

This study was conducted at Romblon State University, Main Campus from the month of August to October, 2013. The location map of the study is presented in Figure 3.

Figure 1: MAP OF ROMBLON STATE UNIVERSITY Main Campus

b) Selecting tree species

Tree species are selected according to circumference of trunks must not be less than 110 cm, and injured trees are not suitable for the survey.

c) Surveying lichen diversity on tree trunks

Lichen diversity (LD) was surveyed on the selected trees, using a surveying quadrat. This quadrat consisted of four independent quadrat segments; each 50cm in height and 10cm in width. Quadrat segments were placed on the North, East, South and West side of thetrunk 100cm above the ground. Each quadrat segment was subdivided into five quadrat squares 10 x 10cm (Figure 2) and the presence of lichen species was recorded in each quadrat square.

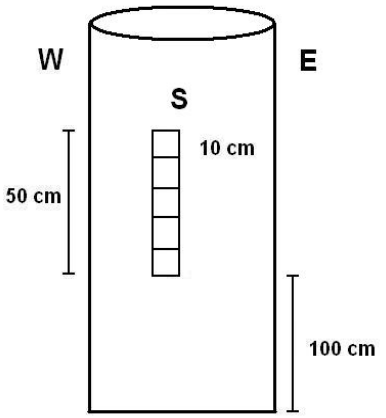


Figure 2: Surveying quadrat segment with five quadrat squares (adapted from Asta et al. 2002).

d) Collection and Identification

All samples of lichens collected in the RSU, main campus were processed for identification in the laboratory.

Thalline Spot tests was applied for identifying lichen species that have a color reaction using 10% solution of Potassium hydroxide (K), 4%solution of Sodium hypochlorite or chlorine (C) and the Lugol' s iodine.

e) Thalline Spot test procedures

1. Remove a piece of lichen from the specimen.
2. Place the sample on a white filter paper.
3. Add a minute amount of spot test reagent with a dropper.
4. Observe colour changes quickly.

f) Species Identification

The lichen species were identified by comparing them with the characteristics of lichens published in the book of Schneider entitled "A Guide to the Study of Lichens" and the documented samples of "Consortium of North American Lichen Herbaria".

g) Data Analysis

i. *Calculation of lichen diversity values*

Following the procedures of Asta et al. (2002 a, b) LD values for each sample plot were calculated.

Within each sample plot a sum of frequencies of all lichen species for each aspect on each tree was calculated. For each tree there were four Sums of Frequencies (SFi) on the North (SFiN), East (SFiE) South (SFiS) and West (SFiW) side of the trunk. Then the arithmetic Mean of the Sums of Frequencies (MSF) for each aspect (North, East, South, and West) in sample plot j was calculated following the formula:

$$MSF_{Nj} = (SF_{1Nj} + SF_{2Nj} + SF_{3Nj} + SF_{4Nj})$$

where;

MSFN: is the mean of the sums of frequencies of all trees of plot j for each aspect(e.g. North)

SF: is sum of frequencies of all species recorded for each aspect (e.g. North) of tree i
N: is the number of surveyed trees with a given aspect in unitj

The LD value of sample plot j (LDVj) was then calculated as the sum of the MSFs ofall aspects:

$$LDV_j = (MSF_{Nj} + MSF_{Ej} + MSF_{Sj} + MSF_{Wj})$$

The comparison of lichen species in trees were taken and analyzed by getting the overall population of lichens per site. Results from the study were presented in graphical form by showing the standard form.

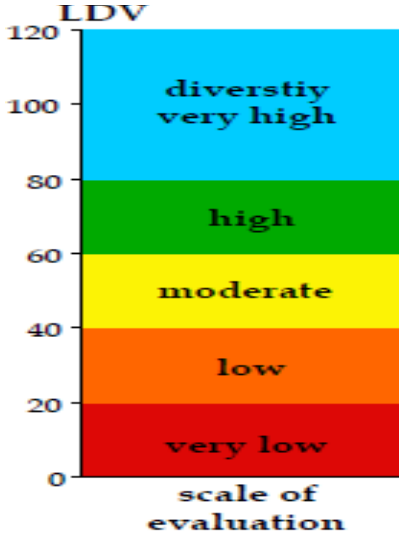
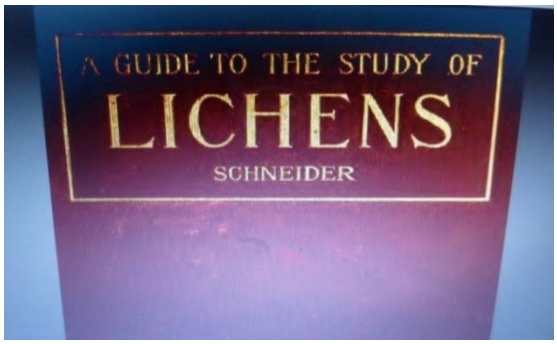


Figure 3: Assessment of Lichen Diversity (Asta et. al.)

VI. Results & Discussions

a) The Identification of Lichen Species

The lichen species were identified by comparing it with the characteristics of lichens published in the book of Schneider entitled "A Guide to the Study of Lichens" and the documented samples of "Consortium of North American Lichen Herbaria".



Collected from Consurtium of North American
Study site Lichen Herbaria
Common name: Blemish lichens
Scientific name: Phlyctis argena (white)

Description:

Thallus: crustose, typically continuous, even to tuberculate, but rarely coarsely tuberculate or tuberculate-plicate, sometimes scurfy, usually distinctly rimose in thick specimens, thin to thick, forming extensive patches up to several dm in diam.; prothallus: often conspicuous, white, felty, composed of radiating hyphae forming a marginal border to about 1 cm wide. *Surface:* white, sometimes with a brownish tinge sorediate.

Collected from Study site

Consurtium of North American Lichen Herbaria

Common name: Common green shield lichen
Scientific name: Parmelia caperata

Description:

Thallus: adnate to loosely adnate, foliose, 5-20 cm in diam., sometimes forming extensive patches, irregularly lobate. *Lobes:* subirregular, elongate, plane to subconvex, separate, 5-13 mm wide, contiguous to somewhat imbricate; apices rotund, crenate, eciliate. *Upper surface:* yellow green to pale yellow, occasionally green-gray (in shade), smooth but becoming rugose and folded with age, dull to somewhat shiny; epruinose and emaculate. *Soredia:* laminal, granular to wart-like, initially in circular soralia but becoming diffuse and confluent; *Isidia:* absent. *Medulla:* white with continuous algal layer. *Lower surface:* black centrally, brown and naked peripherally; ***Rhizines:*** dense to sparse centrally to edge of brown zone, black, simple, sometime brown or white tipped.

Collected from Study site

Consurtium of N. American Lichen Herbaria

Common name: Drinaria lichen
Scientific name: Drinaria applanata

Description:

Thallus: foliose, appressed to agglutinated, loosely appressed at the lobe tips, up to 6 cm in diam., pinnately or subpinnately lobate. *Lobes:* radiating, confluent, flat or convex, but sometimes concave towards the lobe tips, 0.5-2 mm wide, distinctly flabellate towards the lobe tips. *Upper surface:* gray, bluish gray or almost white, with a punctiform, rarely patchy white pruina or epruinose, sorediate. *Soredia:* farinose, in laminal, globose or elongated soralia. *Pseudocyphellae:* distinct, marginal, rarely also laminal, usually restricted to the peripheral parts of the lobes, sometimes reticulately confluent. *Medulla:* white, the lowest part sometimes orange, especially towards the lobe tips. *Lower surface:* black in center, paler towards lobe tips, erhizinate. *Apothecia:* very rarely present, laminal on thallus, 0.5-1.5 mm wide.

Collected from Study site

Consurtium of American Lichen Herbaria

Common name: Gold dust lichen

Scientific name: Chrysothrix xanthina

Description:

Thallus: crustose-leprose, bright yellow, unstratified, adnate, diffuse, irregularly spreading, sometimes forming scattered granules, but usually ±continuous. Soredia fine, with individual granules minutely convex to spherical, 20–80 µm wide, not agglomerated. *Medulla:* not apparent. *Apothecia:* reported to be rare, to 0.5 mm wide, sessile, rounded; disc orange, plane to convex, heavily yellow-pruinose; margin very thin, ecorticate, soon becoming excluded; hymenium colourless, to 50 µm thick; epihymenium colourless, composed of a reticulate layer of richly branched paraphyses; hypothecium colourless, poorly developed; *Ascospores;* (2–) 3-septate, obovoid to ellipsoidal, straight or curved, often constricted in the middle, 9–14 × 3 µm.

Collected from American Study site

Consurtium of N. Lichen Herbaria

Common name: Blemished lichen
Scientific name: Phlyctis argena (gray)

Description:

Thallus: crustose, typically continuous, even to tuberculate, but rarely coarsely tuberculate or tuberculate-plicate, sometimes scurfy, usually distinctly rimose in thick specimens, thin to thick, forming extensive patches up to several dm in diam.; prothallus: often conspicuous, white, felty, composed of radiating hyphae forming a marginal border to about 1 cm wide. *Surface:* gray sometimes with a brownish tinge (in herbarium specimens only?), sorediate. *Soredia:* forming coarse consoredia, up to 90-125 μm in diam., often mixed with eroding cortex fragments, in pale yellow to greenish white (rarely pure white and sometimes becoming pink in the herbarium) irregular soralia often somewhat elongate and angular and usually delimited by a ±raised rim formed by the cortex and often to a cm or more wide or sometimes becoming confluent and accounting for most of the thallus.

Collected from Study site

Consurtium of North American Lichen Herbaria

Common name: pencil mark lichen

Scientific name: Graphis scripta

Description:

Thallus: crustose, continuous to slightly rugose. *Surface:* cream-colored, white or pale gray or grayish green, dull. *Apothecia:* raised from the thallus, lirellate. *Lirellae:* oblong, ±flexuous and branched, 1-3 x 0.2-0.4 mm. *Disc:* narrow to wide and open, dark gray to brown with whitish pruina. *Margin:* well developed, covering the lateral part of the ascocarps; excipular lips: black, entire, sometimes narrow. *Exciple:* poorly developed and not carbonized at the base, carbonized laterally, with entire excipular lips whose basal part is sometimes less developed and less carbonized. *Epihymenium:* brown, 5-10 μm thick. *Hymenium:* not inspersed, 90-100 μm tall; paraphyses: 1.5-2 μm thick, dense, tips distinctly brown or yellowish brown; subhymenium: hyaline, 10-20 μm thick.

Collected from Study site

Consurtium of North American Lichen Herbaria

Common name: none

Scientific name: Miriquidica atrofulva

Description:

Life habit: lichenized; Thallus: crustose or squamulose, usually composed of contiguous to scattered areoles, sometimes rimose; prothallus: sometimes present. Areoles: angular to roundish in outline or irregularly shaped. Surface: white, gray, brownish yellow or brown, lacking secondary reproductive structures. Cortex: eucortex or phenocortex, often with a distinct epinecral layer. Medulla: white to spotted brown, I-. Photobiont: primary one a chlorococcoid green alga, secondary one absent. Ascomata: apothecial, black or dark brown, immersed to sessile, lacking a thalline margin;

Collected from American Study site

Consurtium of North Lichen Herbaria

Common name: none

Scientific name: Parmelia soredians

Description:

Thallus: adnate to tightly adnate, to 4–8 cm wide. Lobes imbricate, 1–3 mm wide; margins shallowly incised; apices rotund. Upper surface: yellow-green, smooth, dull to slightly shiny, without dactyls and isidia; marginal lobes developing laminal rugae, the surfaces of which disintegrate to form orbicular soralia with granular soredia; rugae coalesce to form sorediate ridges which eventually develop into large, pulvinate soralia in thallus centre. Medulla: white. Lower surface: with moderately dense, simple rhizines. Apothecia and pycnidia: not seen.

Collected from Study site

Consurtium of North American Lichen Herbaria

Common name: Jelly lichen

Scientific name: Collema furfuraceum

Description:

Thallus: foliose, medium-sized to large, (1-)3-6 (-10) cm across, membrane-like, closely adnate, conspicuously, deeply and broadly lobate. Lobes: (0.2-) 0.5(-1) cm broad, thin, (50-)60-105 μm thick, apically rotund or extended, +overlapping. Upper surface: dark olive-green to brownish black, paler and +transparent when moist, strongly but broadly ridged (wrinkled); ridges: radiate, sometimes postulate in young parts of thallus, becoming long, narrow and flexuous, 0.1-0.3 mm wide, up to 1.5 mm tall, simple or branched.

SITE 1

Elevation: 36 masl

Location: N 12^0 23.792' E 121^0 59.037'

10 Trees/species were assessed

Table 2: Tree No.1

Polyalthia longifolia (Indian tree)	N	E	S	W
Phlyctis argena (white)	106	118	90	86
Parmelia caperata	110	136	107	103
Drinaria applanata	20	35	15	21
Chrysotrix xanthina	36	22	26	28
Phlyctis argena (gray)	12	9	10	12
Graphis scripta	31	26	22	20
Miriquidica atrofulva	4	4	2	4

SF: Sum of frequencies of all lichen species found at one aspect of tree
N=319, E= 350, S=272, W= 274
MSF: Mean of the sums of frequencies of all the sampled trees of unit
N= 31.9, E= 35, S= 27.2, W= 27.4
LICHEN DIVERSITY VALUE(LDV)
MSF(N)+MSF(E)+MSF(S)+MSF(W)
(31.9+35+27.2+27.4)
LDV = 121.5

Table 3: Tree No. 2

Swietenia mahogani (Mahogany)	N	E	S	W
Phlyctis argena (white)	100	92	88	86
Parmelia caperata	104	98	96	94
Drinaria applanata	12	26	11	18
Chrysotrix xanthina	32	21	22	26
Phlyctis argena (gray)	10	12	12	11
Graphis scripta	36	22	24	24
Miriquidica atrofulva	2	2	2	4

SF: Sum of frequencies of all lichen species found at one aspect of tree
N=296, E= 273, S=255, W= 263
MSF: Mean of the sums of frequencies of all the sampled trees of unit
N= 29.6, E= 27.3, S= 25.5, W= 26.3
LICHEN DIVERSITY VALUE(LDV)
MSF(N)+MSF(E)+MSF(S)+MSF(W)
(29.6+27.3+25.5+26.3)
LDV = 108.7

Table 4: Tree No.3

Areca catechu (Betel nut)	N	E	S	W
Phlyctis argena (white)	112	106	100	111
Parmelia caperata	120	112	116	115
Drinaria applanata	10	21	26	20
Chrysotrix xanthina	36	28	26	20
Phlyctis argena (gray)	11	11	14	10
Graphis scripta	32	30	26	22
Miriquidica atrofulva	2	0	2	1

SF: Sum of frequencies of all lichen species found at one aspect of tree N= 323, E= 308, S=310, W= 299
MSF: Mean of the sums of frequencies of all the sampled trees of unit
N= 32.3, E= 30.8, S= 31, W= 29.9
LICHEN DIVERSITY VALUE(LDV)
MSF(N)+MSF(E)+MSF(S)+MSF(W)
(32.3+30.8+31+29.9)
LDV = 124
Total LDV of SITE 1
LDV (T1) + LDV (T2) + LDV (T3)/3
(121.5+108.7+124)/3
SITE 1 Total LDV = 117.9
SITE 2
Elevation: 59 meters above sea level
Location: N 12^0 23.736' E 121^0 59.187'
10 trees/species were assessed

Table 5: Tree No.1

Polycias nodosa (Malapapaya Tree)	N	E	S	W
Phlyctis argena (white)	101	90	96	108
Parmelia caperata	106	98	102	90
Drinaria applanata	10	9	11	10
Chrysotrix xanthina	12	8	10	12
Phlyctis argena (gray)	90	88	96	100
Graphis scripta	102	97	99	90
Miriquidica atrofulva	2	6	6	8
Parmelia soredians	22	26	26	20
Collema furfuraceum	4	3	6	4

SF: Sum of frequencies of all lichen species found at one aspect of tree
N= 449, E= 425, S= 452, W= 442
MSF: Mean of the sums of frequencies of all the sampled trees of unit
N= 44.9, E= 42.5, S= 45.2, W= 44.2
LICHEN DIVERSITY VALUE(LDV)
MSF(N)+MSF(E)+MSF(S)+MSF(W)
(44.9+42.5+45.2+44.2)
LDV = 181.8

Table 6: Tree No.2

Swietenia mahogani (Mahogany Tree)	N	E	S	W
Phlyctis argena (white)	96	100	90	96
Parmelia caperata	102	98	88	90
Drinaria applanata	8	12	8	6
Chrysotrix xanthina	10	10	8	4
Phlyctis argena (gray)	80	86	86	90
Graphis scripta	96	99	97	97
Miriquidica atrofulva	2	0	2	4
Parmelia soredians	30	32	22	28
Collema furfuraceum	2	2	4	2

SF: Sum of frequencies of all lichen species found at one aspect of tree
N= 426, E= 439, S= 405, W= 417

MSF: Mean of the sums of frequencies of all the sampled trees of unit
N= 42.6, E= 43.9, S= 40.5, W= 41.7
LICHEN DIVERSITY VALUE(LDV)
MSF(N)+MSF(E)+MSF(S)+MSF(W)
(42.6+43.9+40.5+41.7
LDV = 168.7

Table 7: Tree No.3

Polyalthia longifolia (Indian tree)	N	E	S	W
Phlyctis argena (white)	96	90	104	100
Parmelia caperata	90	82	90	92
Drinaria applanata	8	8	6	8
Chrysotrix xanthina	10	6	6	4
Phlyctis argena (gray)	92	86	86	82
Graphis scripta	86	90	90	94
Miriquidica atrofulva	6	6	2	4
Parmelia soredians	20	22	18	24
Collema furfuraceum	2	0	1	1

SF: Sum of frequencies of all lichen species found at one aspect of tree
N= 408, E= 390, S= 403, W= 409
MSF: Mean of the sums of frequencies of all the sampled trees of unit
N= 40.8, E= 39, S= 40.3, W= 40.9
LICHEN DIVERSITY VALUE(LDV)
MSF(N)+MSF(E)+MSF(S)+MSF(W)
(40.8+39+40.3+40.9)
LDV = 161
Total LDV of SITE 1
LDV (T1) + LDV (T2) + LDV (T3)/3
(181.8+168.7+161)/3
SITE 2Total LDV = 170.2

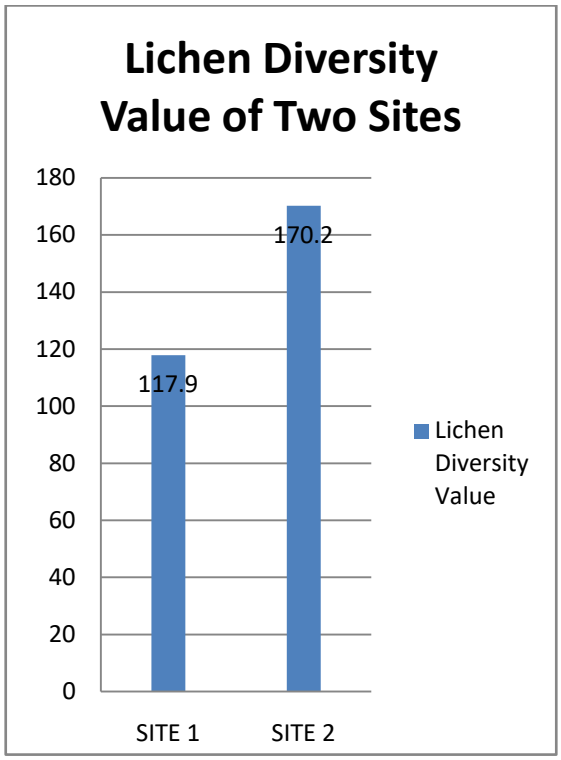


Figure 4: Shows that Site 2 has the higher Lichen Diversity Value than Site 1. Both sites have very high lichen diversity based on the scale of evaluation by Asta et. al.

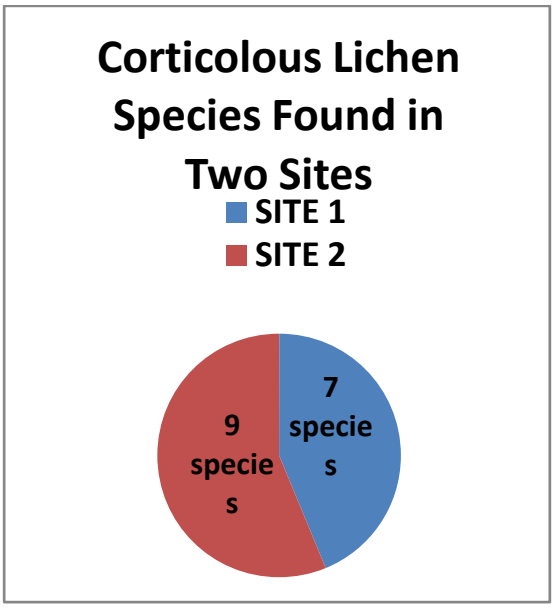


Figure 5: Show that there were 7 species of corticolous lichens in found Site 1 and 9 species in Site 2

Table 8: Thalline Spot Test

SPECIES	TYPE OF THALLUS	SPOT TEST REACTION		
		KOH Sol.	Chlorine Sol.	Lugol' s Iodine
Phylctis argena	Crustose	K+ (positive) black	C- (negative)	I+ (positive) gray
Parmelia caperata	Foliose	K+ (positive) dirty yellow	C+ (positive) copper green	I- (negative)
Drinaria applanata	Foliose	K+ (positive) yellow	C- (negative)	I+ (positive) reddish
Chrysothrix xanthina	Crustose	K+ (positive) yellow	C+ deep yellow	I- (negative)
Phlyctis argena	Crustose	K+ (positive) yellow	C- (negative)	I+ (positive) reddish
Graphis scripta	Crustose	K+(positive) black	C- (negative)	I- (negative)
Miriquidica atrofulva	Crustose	K+ (positive) pink	C+ (positive) orange	I- (negative)
Flavoparmelia soredians	Foliose	K+ (positive) yellow	C+ (positive) white	I- (negative)
Collema furfuraceum	Foliose	K-(negative)	C- (negative)	I- (negative)

As shown in this table, thallus of each species were identified and spot test reactions are noted that a +denotes color reaction and a – indicates that there is no color change.

Table 9: Common Lichen Indicator of Air Pollution (Hawks worth & Rose)

Polluted Areas	Moderately Polluted	Slightly Polluted	Clean Air
Buellia punctata	Evernia prunastri	Anaptychia ciliaris	Degelia plumbea
Cladonia coniocraea	Foraminella ambigua	Graphis elegans	Lobaria pulmonaria
Cladonia macilenta	Hypogymnia	Graphis scripta	Lobaria scrobiculata
Diploicia Canescens	physodes	Opegrapha varia	Pannaria rubiginosa
Lecanora conizaeoides	Lecanora chlarotera	Parmelia acetabulum	Permelia perlata
Lecanora dispersa	Lecidella elaeochroma	Parmelia caperata	Ramalina calicaris
Lecanora expallens	Parmelia glabratula	Phaeophyscia orbicularis	Ramalina fastigiata
Lepraria incana	Parmelia saxatilis	Physcia aipolia	Ramalina fraxinea
Xantoria parietina	Parmelia sulcata	Physconia distorta	Teloschistes flavicans
	Physcia adscendens	Physconia enteroxantha	Usnea species
	Physcia tenella	Pseudevernia furfuracea	
	Plastismatia glauca		
	Ramalina farinacea		

This table show that there were two common lichen indicator of air pollution present in the two study sites, Graphis Scripta and Parmelia caperata.

VII. Findings

1. Site 2 (LDV = 170.2) has the higher Lichen Diversity value than Site 1 (LDV = 117.9).
2. Both sites have Very High Lichen Diversity based on the scale of evaluation by Asta et. al.
3. Seven (7) corticolous lichen species found in Site 1 and nine (9) corticolous lichen species in Site 2.
4. Two (2) types of lichens were found in the study sites, crustose and foliose.
5. Two (2) species of lichen indicator found in the study sites, *Graphis scripta and Parmelia caperata.*

VIII. Conclusions

1. The diversity of corticolous lichens in the Romblon State University, Main Campus is Very High.
2. There were nine (9) species of corticolous lichens present in the Romblon State University, Main Campus.
3. Crustose and Foliose types of lichens were present in the study area.
4. There were two (2) lichen air pollution indicators present in the study area.
5. The air status of the study area is slightly polluted.

Recommendations

1. Other studies should be conducted by assessing all the factors in the growth of lichens including humidity, temperature, ph bark, light intensity and climate conditions.
2. Additional references must be provided to other researchers for their guidance.

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