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lichen inventory and species diversity at coastal ecosystems at No. 63 Benab, Berbice, Guyana

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Abstract

This study evaluates and compares lichen diversity at two coastal sites at No. 63 Benab, Berbice, Guyana. The study was completed in three phases. Phase one included the collection of the lichen specimens from the sites; Phase two consisted of the identification of the lichen specimens obtained from field visits and Phase three included the analysis of the data done on R version 4.2.2 (R-Studio) and Microsoft Excel to determine which of the sites had the highest lichen diversity. Sampling plots of 2000 meter square (2000 m²) for each of the two agroecosystems was demarcated and at both locations, 50 m x 40 m study plots were established, and samples of healthy mature trees were identified from each plot to determine species richness, evenness, and diversity of the lichen communities. The trunks of 40 healthy individual trees were intercepted with twine in the North, South, East, and West quadrants (each measuring 50 by 100 cm). A total of fifty-two thousand three hundred eleven (52,311) lichens were identified belonging to fifteen (15) families, twenty-three (23) genera and thirty (30) species. This research based on the available literature has reported seventeen (17) new species of lichens from eleven (11) families as first-time record of species that were not previously identified for Guyana and so adds to the biodiversity of lichen flora in the country. First time records for Guyana included four (4) species of lichens from the family Parmeliaceae; one (1) species from the family Chrysotrichaceae; one (1) species from the family Teloschistaceae; two (2) species from the family Lecanoraceae; one species from the family Arthoniaceae; two (2) species from the family; one (1) species from the family Stereocaulaceae; two (2) species from the family Phlyctidaceae; one species from the family Collemataceae; one species from the family Cladoniaceae and one species from the family Lichinaceae. The lichen communities were compared using the Simpson's Diversity Index, Shannon Diversity Index, Menhinick's Index and Pielou's Index. Statistical analyses were executed with the help of the R and Excel software and it was possible to distinguish between the two sites' species distribution, diversity, and abundance. The results showed that site # 1 lichen community had a higher species richness, species evenness, species diversity and abundance than site # 2. Four (4) species of lichens showed specificity towards twelve (12) species of host trees.

Keywords: Lichens; Species Diversity; Guyana; Species Abundance; Anthropogenic; Species Richness; Species Distribution; Species Evenness

1. Introduction

Lichens grow very slowly and can survive for many centuries while withstanding changes in harsh environments [4] [5] [28]. 'Lichen' is a Greek word and it refers to the surface growth of a tree's bark. Around 300 BC, an early botanist, Theophrastus presented the word 'lichen' as a plant group in the scientific world [4] [5] [36] [37]. Globally, there are approximately fifteen to twenty thousand (15,000-20,000) species of lichens. Some lichens are scarcely found in nature, and many are specialized to particular ecosystems and recur infrequently across the landscapes [4] [5] [19] [38].

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Over the years, lichens were once classified as a single organism and in other cases, they were gradually mistaken as bryophytes (mosses), while others were mistaken for seaweeds due to prior descriptions based on their physical appearance. However, in the early 1800s, when microscopes were developed and made available, scientists began to investigate and note the detailed physiology, anatomy and biology of lichens which established their true nature [4] [5] [19] [34] [51] [52].

A fungus's symbiotic interactions with an alga or a cyanobacteria result in the formation of lichens and are thought to be sophisticated living organisms [4] [5] [35] [45] [46] [57]. The mycobiont is a heterotrophic fungus that forms the primary lichen body and is considered to be the dominant partner and it determines the lichen's basic characteristics, including the thallus's form and the kind of fruiting body. The algae or cyanobacteria is autotrophic and this is called the photobiont which usually lies between the upper and lower fungal cortex [4] [5] [19] [26] [27] [34] [45] [51] [52] [54] [57].

The photobiont provides nutrition by photosynthesis and thus nourishes the fungus and allowing it to spread. The mycobiont provides the structural support and necessary protection for its algal partner [4] [5] [45] [46] [47] [54] [57]. A mutual water transportation and other essential liquids exist from the mycobiont to the photobiont [4] [5] [45] [46] [48] [57].

In many species of lichens, the upper cortex has specialized apertures that may range from brightly colored to muted or even black. These structures are referred to as the 'fruit bodies' or 'fruiting body' because they release tiny airborne spores. A new lichen thallus is formed whenever the spores come into contact with an appropriate photobiont (cyanobacteria or algae) [4] [5] [19] [46] [51] [52] [54]. However, numerous lichens, produce propagules that detaches and are capable of growing into a new lichen body. The most common form of propagule is soralia which is seen as a powdery structure and the upper cortex develop pustules. These are effective to release tiny clusters of soredia (cells from algae) which are held together by the hyphae of the fungus. Another structure which lichens utilize for vegetative reproduction are isidia and these contain both the algal and fungal cells [4] [5] [9] [13] [19] [51] [52]. In many ecological niches on differing physical or biological substrates, where fungi, algae, or cyanobacteria might not be able to exist alone, this lichen union enables a diverse floral group to thrive [4] [5] [47]. Some lichens have a high desiccation tolerance, thus can survive extremely high or low temperatures.

Lichens colonize different environments and are usually termed pioneer species. Nearly all terrestrial ecosystems have them present ranging from the tropical hot regions such as: hot deserts, rocky coasts, tropical rainforests to the cold polar regions e.g., arctic tundra and even high-altitude environments. Lichen species can also be found in other harsh environments like toxic slag heaps [4] [5] [23] [34]. Additionally, worldwide, an estimated 13,000-20,000 species of lichenized fungus exist and more than fifty percent (50%) that are documented are *Ascomycetes* [4] [5] [22].

Lichens come in many varieties of hues, from brightly coloured to dark and are grouped based on the many growth forms they exhibit. The majority of lichens, known as crustose lichens, grow in a crust-like fashion. Some are shrub-like and are known as fruticose lichens, while others are leafy and are known as foliose lichen. Lichens can grow in a variety of additional different growth forms [4] [5] [9] [19] [34] [51] [52] [54]. The wide variety of substrates that lichens use to grow is another way to classify them. For example, on rocks saxicolous lichens best flourish, terricolous lichens develop and thrive on the surface of the soil, lignicolous lichens are present on wood that has lost its bark, and foliicolous lichens grow on the leaves of vascular plants. Corticolous lichens thrive on the barks of vascular plants [4] [5] [46] [51] [52]. Corticolous microlichens form the biggest group in the lichen world and are also the least known to the scientific world [2] [4] [5] [19] [22].

Lichens are essential for the functionality of the ecosystems where they dwell by partnering with different trees and surrounding plants [4] [5] [47] [51] [52]. For a long time, lichens have been utilized as a key biological indicator in order to detect anthropogenic disturbance, such as air pollution, nitrogen (N) deposition, acid rain, and a number of other environmental variables [4] [5] [8] [9] [12] [14] [19] [26] [27]. Further, lichens were greatly used as a food source and to also extract chemicals like dyes or antimicrobial substances [4] [5] [28] [33]. However, lichens have little medicinal value in the world of science [4] [5] [7]. In addition, lichens and their habitats play an integral role in biodiversity and they are also important to organisms from a variety of species that are yet to be discovered and documented such as invertebrates from the phylum Arthropoda and warm-blooded vertebrates like mammals and birds [4] [5] [19] [38].

Lichens have great importance in the world today. Lichens are utilized as a source of food for many animals such as caribou, reindeer, squirrel, musk ox, etc. which feed on reindeer moss (*Cladonia mangifea*) in the tundra. In aquatic habitats, fishes and other organisms utilize lichens as food. The Iceland moss (*Certaria islandica*) is widely used as food by people in Iceland, Sweden, Norway; *Lecanora esculenta* is considered as a holy food by the Jewish people and in Japan

the stone mushroom (*Endocarpon miniatum*) is used as a vegetable for preparing various dishes [4] [5] [6] [8] [32] [40] [43] [50] [55].

In order to create orcein, a biological stain, the lichen *Rocella tinctoria* is employed. Before litmus was made synthetically, the *Rocella tinctoria* could be used as a source of the pH indicator. Various lichen species are used to produce a variety of laboratory colors, including those for the litmus test, pH indicator, and other assays [4] [5] [8] [32]. Additionally, some lichen species—like *Ramalina* and *Evemia*—are utilized to produce fragrant incense and incense sticks. Some kinds of perfume or smells are made using other forms of lichen, such as *Lobularia pulmonaria* and *Evemia prunastri* [32]. As a natural treatment for a variety of rashes and skin disorders, lichens are also widely employed in the cosmetology and cosmetic industries [4] [5] [8].

The *Usnea* lichen is also capable to produce forest fires when the seasons are relatively hot. In areas where the humidity is high, some lichens thrive on concrete, stonework and window panes and overtime they can damage and deteriorate buildings by erosion [4] [5] [32]. Some lichens are utilized for their antibiotic properties like the *Usnea* and *Cladonia* where usnic acid is obtained and is pharmaceutically utilized to create ointments for the treatment of wounds and burns [4] [5] [32] [57]. In the pharmaceutical industry, numerous lichens are also frequently employed as anti-infectives to create antiviral, antibacterial, and other anti-inflammatory medications [4] [5] [8].

Lichens are the early colonizers in situations where primary succession is encouraged, such as dry, naked rocks, cliffs, mountains, etc. During the growth and developmental stages of lichens, they promote the erosion of cliffs and rocks by secreting unique acids that can penetrate the rocks with the aid of their hyphae. As a result, it produces miniscule crevices where the organic matter accumulates, hence, paving the way for growth of other organisms [4] [5] [17]. Consequently, by evaluating the size of lichens, petrologists and geologists are able to study and determine the lifetime and other exterior features of rocks and surfaces [4] [5] [8]. In 2018, Porada *et al.* reported, that non-vascular plants such as lichens and mosses, capture considerable amount of rainfall. They have the ability to alter the climate and the global water cycle by boosting evaporation of freely flowing water by about 61%. Additionally, because of the algal symbiont, they are crucial in the fixation of nitrogen. When it rains, some nitrates are washed away from the lichens and utilized by various plants that grow in the soil. Lichens help turn nitrogen from the atmosphere into nitrate [4] [5] [17].

A crucial characteristic of lichens is that they cannot tolerate pollution because they need fresh and clean environment, to adequately support their proliferation. Lichens can therefore take in carbon dioxide (CO₂) and heavy metals from the air [4] [5] [8] [17]. Subsequently, lichens play a major part of biodegradation by breaking down pollutants that are polluting the earth e.g., polyester (PS), lead (Pb), copper (Cu) and radionuclides. Lichens are also used in the degradation of various pathogens and other environmental reservoirs, which is capable of causing certain deadly infectious diseases in both plants, animals and in some cases human beings [4] [5] [8].

In some agroecosystems, sulphurous and nitrogenous oxides can cause damage to sensitive lichens [4] [15]. By observing and measuring the levels of pollutants in a specific lichen species, environmental scholars and researchers can use this property of lichen to estimate the degree of pollution in a given ecological community. Consequently, lichens are considered to be outstanding bioindicators of a healthy ecosystem [4] [5] [17]. The abundance of epiphytic lichens to evaluate the buildup of heavy metals present in the thalli of one species of crustose lichen, *Parmerlia caperata*, were used by Loppi & Corsini, 2003, in Pistoia, Central Italy as indicators to assess air quality and the effects of air pollution.

Flakus *et al.* (2011) conducted a study in South America where new species and records of *Lepraria* (*Stereocaulaceae*, lichenized Ascomycota) were documented. The study revealed two (2) new corticolous lichen species: *Lepraria nothofagi* from Argentina and *L. stephaniana* from pre-Andean Amazon forest of Bolivia. In addition, the study presents new records of sixteen (16) species of *Lepraria* from South America. *L. diffusa* and *Lepraria adhaerens* are new to the Southern Hemisphere; *L. borealis* is new to South America; *L. alpina* is new to Chile, Peru, Venezuela and Colombia; *L. caesioalba* (chemo type I) is new to Venezuela, *L. lobificans* new to Argentina, *L. pallida* new to Peru, and *L. sipmaniana* new to Bolivia and Chile. The Chilean records of *L. membranacea* appeared to belong to *L. sipmaniana*. Therefore, twenty-seven (27) species of *Lepraria*. are now known at present in South America.

A study was undertaken in 2019 by Bentez *et al.* to determine how the characteristics of the host trees affect the diversity of lichens growing on tree trunks in tropical dry forests. In this study, the diversity and make-up of epiphytic lichens on the trunks of five hundred thirteen (513) trees in disturbed and undisturbed dry forests of southern Ecuador are examined. Both lichen composition and richness were strongly linked with host tree characteristics such bark pattern and tree diameter. Furthermore, at various levels of disturbance, the diversity of epiphytic lichens was correlated with tree richness and canopy cover. According to the study's findings, the characteristics and species of the host tree are the key constraints on epiphytic lichen populations in Ecuador's seasonal dry tropical forests.

Etayo *et al.* (2020) described two (2) lichenicolous fungus from Brazil as being novel to science: *Stigmidium anguinellicola* on *Nyungwea anguinella* from Sergipe and *Cryptodiscus gassicurtiae* on *Gassicurtia coccinea* from Alagoas. In Brazil, nine (9) lichenicolous fungi have been documented on for the first time (or from South America). The annotated list includes the seventy-eight (78) lichenicolous fungus that have been previously reported from Brazil.

Lucking (1996) investigated the lichens that were gathered in Guyana as part of the Smithsonian International Cryptogamic Expedition. Eighteen (18) lichenicolous fungus and two hundred thirty-three (233) foliicolous lichen species were identified and reported from Guyana. In addition, six lichens— *Opegrapha matzeri* (lichenicolous on *Amazonomyces sprucei*), *Arthonia grubei*, *Calopadia pauciseptata*, *Badimia subelegans*, and *Pyrenidium santessonii* (lichenicolous on *Bacidia psychotriae*) and two lichenicolous fungi novel to science were identified. The study also revealed that of the new records for the Neotropics, one hundred fifteen (115) species are new to Guyana, bringing the total number of foliicolous species documented for the nation to two hundred eighty (280).

Detailed research of foliicolous lichens from Suriname and Guyana by Van den Boom & Sipman in 2014 led to the discovery of three (3) new species, new records, and new data. From the sampling of foliicolous lichens done by the first author in Suriname in 2014, a total of one hundred three (103) lichenized and lichenicolous fungi were discovered, one (1) undescribed specie was noted and eighty-nine (89) species were recorded for the first time for the country.

Subsequently, the second author then conducted field research in Guyana in 1992 and 1997, leading to the discovery of twenty-nine (29) first records, including a novel chemical strain of *Loflammea epiphylla* and two (2) undescribed species. Along with the newly identified species *Strigula transversoundulata*, *Enterographa paruiuae* and *Calenia surinamensis*, the additions for Guyana are also provided. There is also a comprehensive list for Suriname, which includes between one hundred thirty-three (133) and one hundred thirty (130) taxa.

Further, in 2021, a study was conducted by Bacchus and Da Silva. They investigated the corticolous lichen diversity in suburban and urban sites in New Amsterdam, Berbice, Guyana. In this study, a total of forty-one (41) trees were sampled and a total of fourteen thousand nine hundred seventy-eight (14978) individual lichens were identified from thirteen (13) genera, ten (10) families and eighteen (18) species were recognized and reported. The study's findings included a checklist of the corticolous lichen species found in New Amsterdam and the assertion that urban corticolous lichen communities exhibit higher species richness, species evenness, and alpha diversity than their suburban counterparts.

Bhagarathi *et al.* (2022) conducted a review of lichens and the elements that influenced their distribution in the neotropics. Fifty-eight lichen families were noted and a grand total of four hundred twenty-seven (427) lichen species were documented in the overall study, which examined twelve (12) neotropical countries. The review also concluded that a wide range of abiotic factors, including pH, temperature, latitude, humidity, moisture, light availability and topography, as well as the effects of anthropogenic activities like pollution and deforestation, play a significant role in influencing lichens and their distribution both in the neotropics and around the world.

Additionally, another review was conducted in 2023 by Bhagarathi *et al.* In this review, the biology and chemistry of lichens were examined along with their ethnopharmacological, pharmaceutical, therapeutic and ecological potential. The review concluded that lichens serve key roles in ecological processes that contribute to ecosystem health and sustainability and they perform important roles in primary productivity, nitrogen fixation, and seed germination. Furthermore, lichens provide a good habitat for numerous microfauna (such as arachnids, insects, and small reptiles like lizards) and macrofauna (such as birds and squirrels). Lichens are also both pioneer and keystone species, and they play a critical role in biomonitoring to detect air quality and pollutants. As a result, biomonitoring organizations, enterprises, and other stakeholders can use them as indicator species to develop strategic strategies for environmental evaluation.

The general objective of this study is to evaluate and compare the lichen abundance and distribution at two sites at No. 63 Benab, Berbice, Guyana. Recognizing that lichens play an integral role in nature since they contribute to many natural processes and act as major keystone species in ecosystems [34], this proposed research will be of great importance to Guyana since it will provide essential data to make accurate plans for environmental management of healthy ecosystems and be of use in pollution monitoring. The area selected for this research is close to many anthropogenic activities and the ecosystems are agroecosystems. It can also serve as a basis when comparing the lichen species diversity of similar forests sites/ areas. This study can be of great contribution as part of the scientific literature coming from Guyana for researchers who are pursuing similar studies in the future in the field of lichenology.

Given the current limited information on lichens in coastal areas in Guyana, the study will add to the body of information that already exists and at the same time, contribute to not only Guyana but the world at large. It will inform people about

lichens that are found on agricultural crop plants in tropical countries and provide a basic botanical inventory that can be used for community diversity study of lichens.

Lichens have not been extensively studied in the coastal areas in Guyana despite they have been dwelling with us for a number of years. There is not enough research on lichens being consistently done in Guyana over the years, hence, there is a lack of data regarding the diversity of lichen species in Region 6, Guyana. Over the years, some work was done in urban and suburban areas in Region 6. This lack of data concerning coastal lichen diversity makes it a difficult task for the Government of Guyana as well as other managing entities to make accurate plans with regards to environmental management and conservation and draw assessments of the species diversity as it concerns lichens.

2. Material and methods

2.1. Study Location

This particular investigation was conducted at No. 63 Benab, precisely along Berbice's East Coast. No. 63 Benab in Region 6 is located at East Berbice Corentyne, coordinates: -57.1487° or $57^{\circ}8'55.3$ W and 5.9838° or $5^{\circ}59'1.6$ N (Figure 1 and Figure 2).



Figure 1 Map of Guyana showing the location of No. 63 Benab (Google Maps)

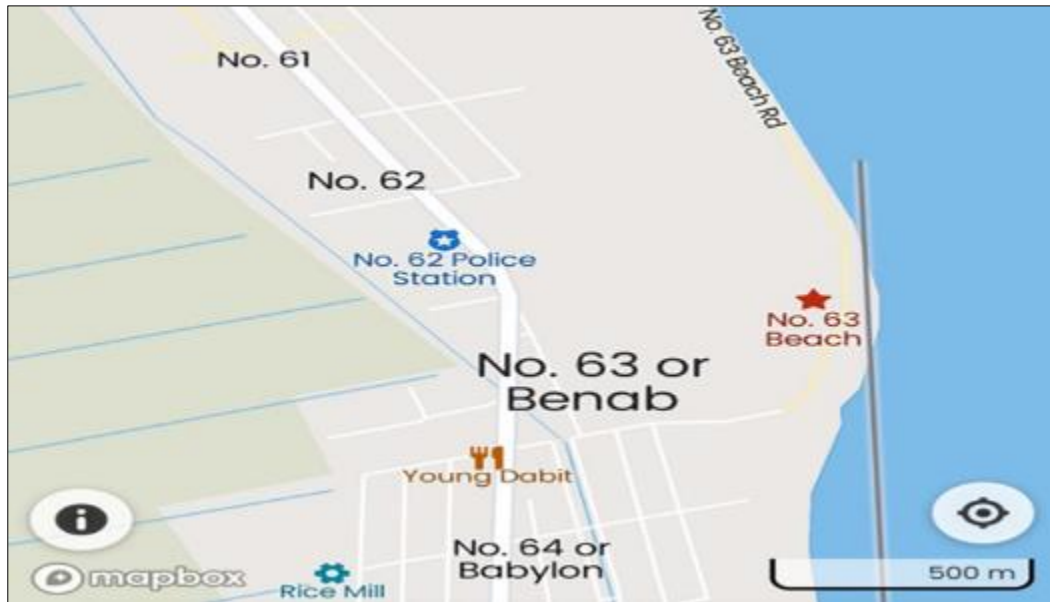


Figure 2 A closer view of the study location used for this research (Google Maps)

2.2. Experimental Design

The experimental design utilized in this research was Random Sampling. Trees were selected using two (2) criteria; the first was that trees selected for this research must be undamaged especially on the bark area and the second was that the trunk of the selected name must be 50 cm girth or more.

2.3. Sampling and Data Collection

The field work for this research was carried out during a five-month period from May-September, 2022. The study was carried out at the exact location (Figure 1 & Figure 2) where sampling plots of 2000 meter square (2000 m²) for each of the two agroecosystems was taken and study sites of 50 m × 40 m plots were established (Figure 3).

Within each of the designated plots, healthy, mature trees were selected for sampling. Undamaged free-standing trees with girths equal to or higher than 50 cm, measured at a height of 2 m above ground, were chosen for this experiment to ensure that completely mature trees were sampled [1] [2] [44].

Each host tree that was sampled was identified, at the very least, to its Genus and Species [2] [24]. By measuring the amount of sunlight that actually reaches the trunk, the sun's exposure was calculated while accounting for the shade that is cast by neighboring structures, bushes, and trees [25].

In order to survey the lichens, quadrats made with twine measuring 50 cm × 100 cm were used in the sampling procedure. Each quadrat was positioned North, South, East, and West of the tree and raised 3 feet (ft) above the ground (Figure 4) [2] [24]. Sampling was done within the designated quadrats as well as on the soil and any rocks present within the square of the quadrat. The very bottom of the tree's trunk was avoided because they variations between individual trees [2] [25].

All lichen species and their frequencies within each 50 cm x 100 cm quadrat were recorded on a lichen survey datasheet that was created for this field research. Every target lichen species had its cover calculated to the closest cm² and then expressed as a percentage of the investigated trunk area [2] [25].

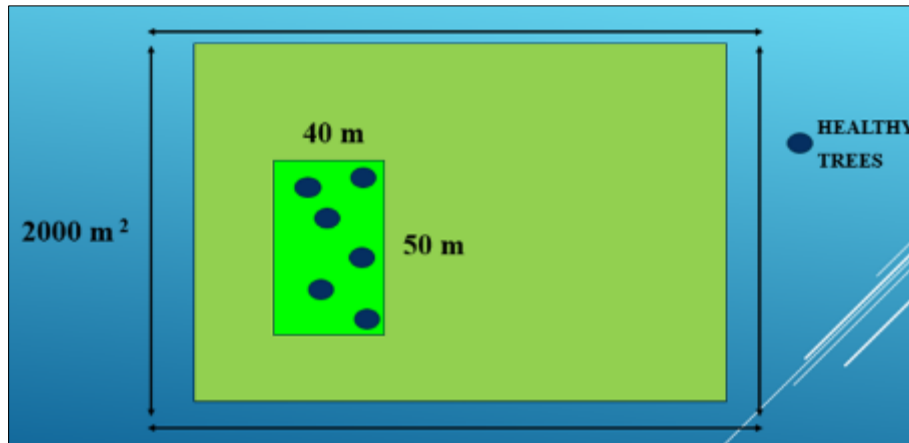


Figure 3 Layout of Sampling Sites and Study Plots

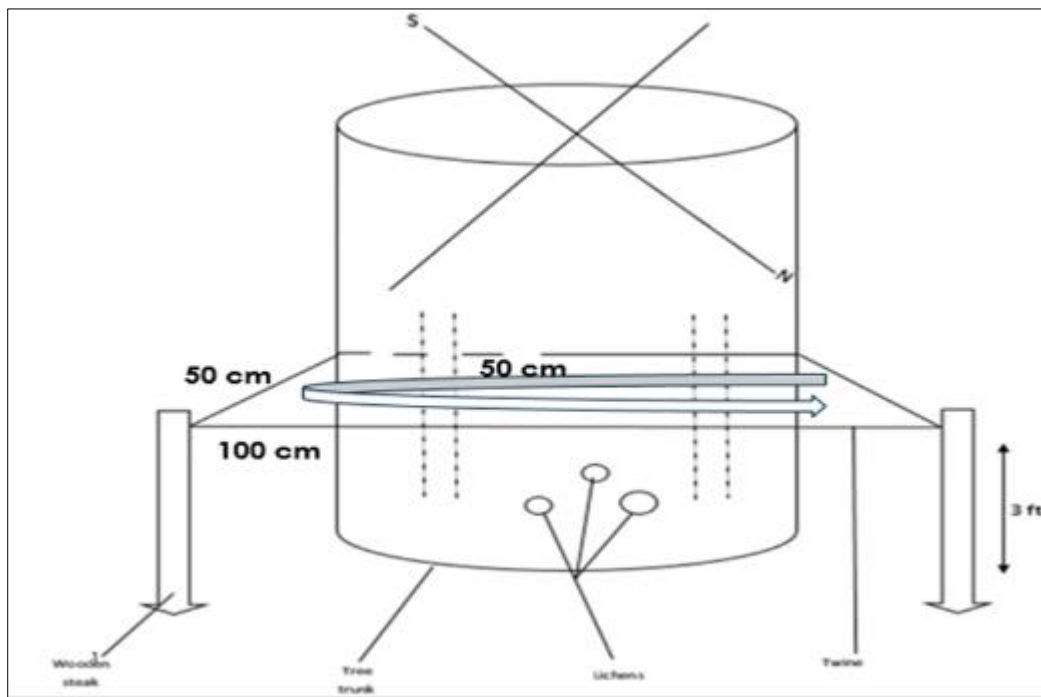


Figure 4 Outline of how each sampling quadrat was set-up to sample lichens on tree trunk and surrounding area [23].
Adopted from [2] [42]

2.4. Lichen Identification

On-site morphological observations of the thalli and apothecia of the lichen specimens were made using a magnifying lens in order to identify the lichen specimens. Identification was done at least to genus and to the species where-ever applicable. Identification was further done using the *Mosses and Lichens a popular guide to the identification and study of our commoner mosses and lichens, their uses, and methods of preserving* by Nina L. Marshall (1919), *Forestry Commission Handbook 4: Lichen in Southern Woodlands* by Broad (1989), *A Reference Notebook: Identifying Mixed Hardwood Forest Lichens* prepared by Irwin M. Brodo and Brian Craig (2001), *Collector's Handy Book: Algae, Fungi, Diatoms, Lichens, Desmids and Mosses* by Johann Nave (n. d.). Additionally, the following dichotomous keys and identification pamphlets were utilized in the identification process: *Lichens Two Lives* by Todd Wesley (2005), *Field oriented keys to the Florida lichens* by Rosentreter *et al.* (2015), *Heathland Lichens* by Brian Eversham (2015) and *Lichen Identification Guide* (2015).

Lichens possess over seven hundred (700) secondary metabolites and some of these are species-specific, hence, spot tests were used to assist in the lichen identification. Spot tests usually utilize reagents that are capable of detecting various lichen substances by generating characteristic color changes without specifying the type of lichen substance, however, acting as an indicator of certain substance groups. These include the reagents iodine solution (which goes dark blue when certain polysaccharides are present), potassium hydroxide (which exhibits unique colors when depside and depsidones are present), and calcium hypochlorite solution which turns pink or red when certain depsides are present [24].

Spot tests were conducted by first scraping the cortex of the lichen sample with a scalpel in order to expose the medulla and then adding the reagent using a pipette in order to observe the color change under a microscope [24]. If any lichens were unable to be identified during the field expedition, then a sample was collected and taken to the laboratory for further identification. After identification, all data obtained was tabulated on the lichen data sheet that was utilized in the field survey.

2.5. Data Analysis

The diversity index is a quantitative metric that reflects the number of distinct species present in a dataset while also taking into account the distribution of fundamental units, such as individuals, among those species. When both the number of species and evenness increase, the index value rises. When all species are equally plentiful for a given number of species, the diversity index value is maximized [42].

The number of species in a given area or the proportion of species among all the individuals of all the species present was also used to quantify species diversity. A diversity index can be used to express this relationship [42]. Menhinick's index, which measures species richness, Simpson's diversity index, Shannon diversity index, and Pielou's index of species evenness were used to compare species diversity [24].

The Menhinick's index is as follows:

$$D = \frac{S}{\sqrt{N}}$$

Where, D = Menhinick's index; S = number of different species in sample; N = the total number of individuals in sample.

The Simpson's Diversity Index is as follows:

$$D_{sim} = 1 - \left(\frac{\sum n(n-1)}{N(N-1)} \right)$$

Where, D_{sim} = Simpson's Diversity Index; n = number of individuals of a particular species; N = total number of individual organisms (all species combined).

The Shannon Index follows the formula:

$$H' = \sum_{i=1}^S p_i \ln p_i$$

Where, p_i = proportion of the total sample belonging to the i^{th} species; $p_i = n_i/N$; n_i = frequency of species i ; N = total number of species. The Shannon Diversity Index value spans from 1.5 to 5.0, where higher values signify greater diversity and vice versa [24].

The formula for Pielou's evenness is:

$$J' = \frac{H'}{\log_e S}$$

Where, J' = Pielou's evenness, S = Total Species.

Beta diversity was calculated using Whittaker's index of diversity:

$$\beta_w = \frac{S}{\alpha}$$

Where, β_w = Whittaker's index of diversity, S = total number of species in an area; α = mean sample species number. The variation in species composition between two or more sampling locations is known as beta diversity. As a measure of heterogeneity, it is utilized [24].

The data that was obtained from the study was subjected to statistical analysis using R software version 4.2.2 (R-Studio) and Microsoft Excel. To compare the necessary data of the research appropriate charts, tables and graphs were generated using R version 4.2.2 (R-Studio) and Microsoft Excel.

3. Results and discussion

3.1. Species/ distribution of lichens and new records

A total of fifty-two thousand three hundred eleven (52,311) lichens were recorded from the two (2) study sites. As shown on Table 1 and Table 2, fifteen (15) families, twenty-three (23) genera and thirty (30) species were recorded from forty (40) sampled host trees. Five (5) families had more than one genus: Parmeliaceae (*Parmelia sulcata*, *Flavoparmelia soredians*, *Flavoparmelia caperta*, *Melanohalea exasperatula*, *Hypotrachyna laevigata*, *Usena barbata* & *Parmelia tiliacea*), Arthoniaceae (*Cryptothecia striata*, *Arthonia cinnabarina*, *Arthonia purinata* & *Arthonia radiata*), Candelariaceae (*Candelaria concolor* & *Candelariella reflexa*), Graphidaceae (*Graphina anguina* & *Graphis elegans*) and Collemataceae (*Collema furfuraceum* & *Lathagrium cristatum*). Twelve (12) different species of trees were sampled.

This research in coastal ecosystems at No. 63 Benab, Berbice brought about first-time record of species that were not recorded for Guyana and adds to the biodiversity of lichen flora in the country. When comparing the literature from the few past researches that were conducted in Guyana regarding lichens, seventeen (17) new species of lichens from eleven (11) families were documented for the first time. Four (4) species of lichens from the family Parmeliaceae (*Parmelia sulcata*, *Melanohalea exasperatula*, *Usena barbata* and *Parmelia tiliacea*); one (1) species from the family Chrysotrichaceae (*Chrysothrix candelaris*); one (1) species from the family Teloschistaceae (*Xanthoria parietina*); two (2) species from the family Lecanoraceae (*Lecanora chlarotera* and *Lecanora muralis*); one species from the family Arthoniaceae (*Cryptothecia striata*); two (2) species from the family Candelariaceae (*Candelaria concolor* and *Candelariella reflexa*); one (1) species from the family Stereocaulaceae (*Lepraria lobificans*); two (2) species from the family Phlyctidaceae (*Pertusaria albescens* and *Pertusaria amara*); one species from the family Collemataceae (*Lathagrium cristatum*); one species from the family Cladoniaceae (*Gymnoderma lineare*) and one species from the family Lichinaceae (*Lichina pygmaea*) were all recorded for the first time in Guyana.

Table 1 Number of species and genera at each study site

#	Family	Total # Of Genera	Total # Of Species	Site 1		Site 2	
				# Of Genera	# Of Species	# Of Genera	# Of Species
1	Parmeliaceae	5	7	5	7	2	3
2	Caliciaceae	1	1	1	1	1	1
3	Chrysotrichaceae	1	1	1	1	1	1
4	Teloschistaceae	1	1	1	1	1	1
5	Lecanoraceae	1	3	1	3	1	3
6	Arthoniaceae	2	4	2	4	1	1
7	Candelariaceae	2	2	2	2	2	2
8	Stereocaulaceae	1	1	1	1	1	1
9	Graphidaceae	2	2	2	2	2	2
10	Monoblastiaceae	1	1	1	1	1	1

11	Phlyctidaceae	1	2	1	2	1	1
12	Collemaataceae	2	2	2	2	1	1
13	Cladoniaceae	1	1	1	1	0	0
14	Lichinaceae	1	1	1	1	0	0
15	Ramalinaceae	1	1	1	1	1	1

Table 2 Species frequency distributed over each site sampled & overall total of each site

Family	Species	Site # 1	Site # 2	Total at the 2 Sites
Parmeliaceae	<i>Parmelia sulcata</i>	65	0	65
	<i>Flavoparmelia soledians</i>	681	125	806
	<i>Flavoparmelia caperta</i>	1697	528	2225
	<i>Melanohalea exasperatula</i>	440	0	440
	<i>Hypotrachyna laevigata</i>	181	119	300
	<i>Usena barbata</i>	52	0	52
	<i>Parmelia tiliacea</i>	107	0	107
Caliciaceae	<i>Dirinaria applanata</i>	3247	4209	7456
Chrysotrichaceae	<i>Chrysothrix candelaris</i>	1575	76	1651
Teloschistaceae	<i>Xanthoria parietina</i>	261	45	306
Lecanoraceae	<i>Lecanora chlarotera</i>	583	848	1431
	<i>Lecanora muralis</i>	5095	3761	8856
	<i>Lecanora conizaeoide</i>	5366	5705	11071
Arthoniaceae	<i>Cryptothecia striata</i>	26	0	26
	<i>Arthonia cinnabarina</i>	557	0	557
	<i>Arthonia purinata</i>	92	0	92
	<i>Arthonia radiata</i>	983	963	1946
Candelariaceae*e	<i>Candelaria concolor</i>	1489	1085	2574
	<i>Candelariella reflexa</i>	1287	282	1569
Stereocaulaceae	<i>Lepraria lobificans</i>	2018	1067	3085
Graphidaceae	<i>Graphina anguina</i>	492	185	677
	<i>Graphis elegans</i>	798	72	870
Monoblastiaceae	<i>Anisomeridium bifforme</i>	489	92	581
Phlyctidaceae	<i>Pertusaria albescens</i>	1201	380	1581
	<i>Pertusaria amara</i>	254	0	254
Collemaataceae	<i>Collema furfuraceum</i>	1013	27	1040
	<i>Lathagrium cristatum</i>	180	0	180
Cladoniaceae	<i>Gymnoderma lineare</i>	113	0	113
Lichinaceae	<i>Lichina pygmaea</i>	1599	0	1599
Ramalinaceae	<i>Bacidia laurocerasi</i>	379	422	801
TOTAL		32320	19991	52311

Crustose lichens species were the most recorded throughout the two (2) sites (46 %) while crustose-leprose lichens (7 %) and fruticose lichens (7 %) were the least to be recorded. Thirty-nine percent (40 %) of the recorded lichens recorded were foliose lichens (Figure 5).

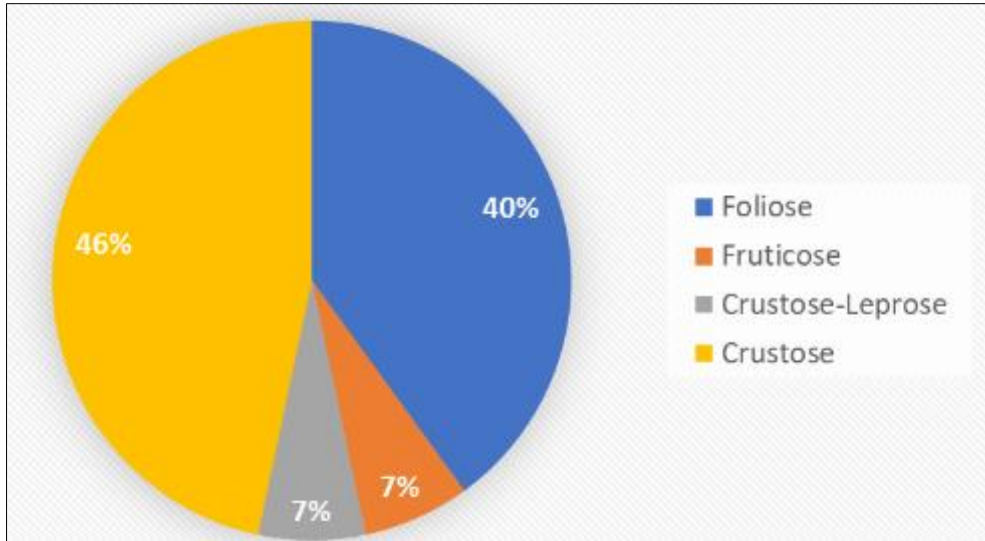


Figure 5 Species distribution of lichens according to thallus type

Site #1- This site is located close to the public road and showed heavy anthropogenic activities. During consecutive visits to the site, humans were continuously traversing through the area. Many activities such as planting of trees and land clearing was regularly done. All of the trees within this site were healthy and none of them showed signs that they were directly affected by anthropogenic influence. Twenty (20) trees at this site were randomly selected. There were eleven tree species: *Mangifera indica*, *Artocarpus camansi*, *Citrus aurantiifolia*, *Citrus reticulata*, *Citrus sinensis*, *Citrus limon*, *Psidium guajava*, *Annona muricata*, *Azadirachta indica*, *Melicoccus bijugales* and *Cocos nucifera*. Thirty (30) different species of lichens (Table 4) were identified and recorded.

Site #2- This site showed traces of heavy anthropogenic activities because it was located close to the beach. During field visits, there was constant human activity and the area was dominated by coconut trees (*Cocos nucifera*). There was much coconut harvesting activities taking place at this site. There was evident land pollution in the form of plastic, bottles and cans at this site. Some of the trees showed signs of being affected by anthropogenic activity since areas of the bark were fragmenting and appeared charred and some tree branches appeared to have been chopped off and the area healed over. Twenty (20) trees were randomly selected and there were three (3) species: *Cocos nucifera*, *Mangifera indica* and *Tamarindus indica*. Nineteen (19) different species of lichens were identified and recorded.

3.2. Estimated Species Diversity

3.2.1. Shannon Diversity Index (Site # 1)

Table 3 Result of Shannon Diversity Index at Site #1

SITE # 1						
Species	Number	pi	lnpi	pi * lnpi	H	H
<i>Parmelia sulcata</i>	65	0.00	-6.20	0.00	-2.80	2.80
<i>Flavoparmelia soledians</i>	681	0.00	-3.90	-0.10		
<i>Flavoparmelia caperta</i>	1697	0.10	-2.90	-0.20		
<i>Melanohalea exasperatula</i>	440	0.00	-4.30	-0.10		
<i>Hypotrachyna laevigata</i>	181	0.00	-5.20	0.00		
<i>Usena barbata</i>	52	0.00	-6.40	0.00		

<i>Parmelia tiliacea</i>	107	0.00	-5.70	0.00		
<i>Dirinaria applanata</i>	3247	0.10	-2.30	-0.20		
<i>Chrysothrix candelaris</i>	1575	0.00	-3.00	-0.10		
<i>Xanthoria parietina</i>	261	0.00	-4.80	0.00		
<i>Lecanora chlarotera</i>	583	0.00	-4.00	-0.10		
<i>Lecanora muralis</i>	5095	0.20	-1.80	-0.30		
<i>Lecanora conizaeoide</i>	5366	0.20	-1.80	-0.30		
<i>Cryptothecia striata</i>	26	0.00	-7.10	0.00		
<i>Arthonia cinnabarina</i>	557	0.00	-4.10	-0.10		
<i>Arthonia purinata</i>	92	0.00	-5.90	0.00		
<i>Arthonia radiata</i>	983	0.00	-3.50	-0.10		
<i>Candelaria concolor</i>	1489	0.00	-3.10	-0.10		
<i>Candelariella reflexa</i>	1287	0.00	-3.20	-0.10		
<i>Lepraria lobificans</i>	2018	0.10	-2.80	-0.20		
<i>Graphina anguina</i>	492	0.00	-4.20	-0.10		
<i>Graphis elegans</i>	798	0.00	-3.70	-0.10		
<i>Anisomeridium biforme</i>	489	0.00	-4.20	-0.10		
<i>Pertusaria albescens</i>	1201	0.00	-3.30	-0.10		
<i>Pertusaria amara</i>	254	0.00	-4.80	0.00		
<i>Collema furfuraceum</i>	1013	0.00	-3.50	-0.10		
<i>Lathagrium cristatum</i>	180	0.00	-5.20	0.00		
<i>Gymnoderma lineare</i>	113	0.00	-5.70	0.00		
<i>Lichina pygmaea</i>	1599	0.00	-3.00	-0.10		
<i>Bacidia laurocerasi</i>	379	0.00	-4.40	-0.10		

Site # 1 yielded a total of thirty-two thousand three hundred-twenty (32,320) lichens. Thirty (30) species of lichens were collected and identified at the end of the field work from this site. *Lecanora conizaeoide* (5366 individual lichens) was most recorded at this site, while *Cryptothecia striata* (26 individual lichens) was the species to be least recorded at this site. Using Microsoft Excel, the Shannon Diversity Index (H) was utilized to calculate the species diversity and the H value obtained was 2.80 (Table 3).

3.2.2. Shannon Diversity Index (Site # 2)

At site # 2, nineteen (19) species of lichens were identified and nineteen-thousand nine hundred ninety-one (19,991) species of lichens were recorded. *Lecanora conizaeoide* (5705 individual lichens) was the species of lichen to be most recorded at this site while *Collema furfuraceum* (27 individual lichens) was the species to be least recorded. When Microsoft Excel was used, the Shannon Diversity Index (H) was used to calculate the species diversity at this site and the H value obtained was 2.10 (Table 4).

Table 4 Result of Shannon Diversity Index at Site #2

SITE # 2						
Species	Number	pi	lnpi	pi * lnpi	H	H
<i>Flavoparmelia soledians</i>	125	0.00	-5.10	0.00	-2.10	2.10
<i>Flavoparmelia caperta</i>	528	0.00	-3.60	-0.10		
<i>Hypotrachyna laevigata</i>	119	0.00	-5.10	0.00		
<i>Dirinaria applanata</i>	4209	0.20	-1.60	-0.30		
<i>Chrysothrix candelaris</i>	76	0.00	-5.60	0.00		
<i>Xanthoria parietina</i>	45	0.00	-6.10	0.00		
<i>Lecanora chlarotera</i>	848	0.00	-3.20	-0.10		
<i>Lecanora muralis</i>	3761	0.20	-1.70	-0.30		
<i>Lecanora conizaeoide</i>	5705	0.30	-1.30	-0.40		
<i>Arthonia radiata</i>	963	0.00	-3.00	-0.10		
<i>Candelaria concolor</i>	1085	0.10	-2.90	-0.20		
<i>Candelariella reflexa</i>	282	0.00	-4.30	-0.10		
<i>Lepraria lobificans</i>	1067	0.10	-2.90	-0.20		
<i>Graphina anguina</i>	185	0.00	-4.70	0.00		
<i>Graphis elegans</i>	72	0.00	-5.60	0.00		
<i>Anisomeridium biforme</i>	92	0.00	-5.40	0.00		
<i>Pertusaria albescens</i>	380	0.00	-4.00	-0.10		
<i>Collema furfuraceum</i>	27	0.00	-6.60	0.00		
<i>Bacidia laurocerasi</i>	422	0.00	-3.90	-0.10		

Table 5 Comparison of Species Diversity at the two (2) sites

SITE # 1	SITE # 2
SDI-H	SDI-H
2.80	2.10
TOTAL SDI-4.9	
SIMPDI-H	SIMPDI-H
0.92	0.83
TOTAL SIMPDI-1.75	

When the H values obtained from the two (2) sites were compared (Table 5), Site #1 had an H value of 2.80 and Site #2 has a value of 2.10. Therefore, according to the results obtained when the Shannon Diversity Index was calculated, Site #1 was noted to have the most lichen diversity and Site #2 was noted to be the site with the least lichen diversity. A chart was generated by Microsoft Excel (Figure 6) to illustrate the comparison of the H values calculated from Sites #1 and #2 and it clearly showed that Site #1 has the highest diversity of lichens.

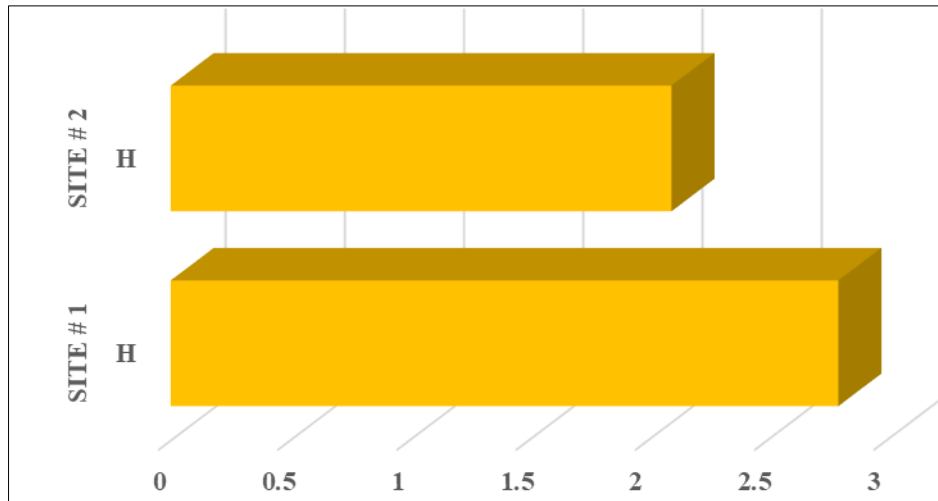


Figure 6 Comparison of the Species Diversity at the two (2) sites

The raw data of the results was subjected to R software version 4.2.2 and R-Studio. The Shannon Diversity index (H) obtained when the code [lichenSD <- diversity (lichen1, index="Simpson") #Shannon-Wiener Diversity – species and evenness lichenSD] was executed is 2.823105 for Site #1 and 2.102173 for Site #2. The results from the software are identical to the previous results (Table 3 & Table 4) obtained from Microsoft Excel.

Additionally, the Simpson Diversity Index (D) was calculated using the code [-lichen_simpson<- diversity lichen1, "simpson") lichen_simpson]. The values generated by the software for Site #1 is 0.9163291 and 0.8270018 for Site #2. The higher the value for this index, the greater the diversity of the species. Therefore, Site #1 has a greater lichen diversity than Site #2.

The Site # 1 community was more diverse than the Site # 2 community, according to a comparison of the lichen alpha diversity using Simpson's Diversity Index (1-D) (Table 5). Between the two (2) sites, Site #2 demonstrated the least diversity. As D increases, diversity diminishes due to the factors of evenness and dominance in Simpson's index; hence, the index is typically considered as its complement 1-D and has a value between 1 and 0. The sample gets closer to the monoculture's limit as 1-D gets closer to 0 [31]. No. 63 Benab has a 1-D = 1.75 total diversity score, which indicates an extremely varied neighborhood.

The alpha diversity among each unique site was also compared using the Shannon-Wiener Diversity Index. In most ecological research, this index's values range from 1.5 to 3.5, rarely going above 4 [31]. The measure rises as community diversity and fairness both rises. The Shannon-Wiener Diversity index values for each location all fell within the acceptable range. The Site # 1 community displayed greater diversity than the Site # 2 community, according to this index, which followed the same trend as the Simpson's index (Table 5). For the two (2) locations, Site #1 displayed the highest level of diversity, followed by Site #2, which had the lowest level of diversity.

3.3. Estimated Species Richness

Table 6 Comparison of Species Richness at the two (2) sites

SITE # 1	SITE # 2
# Of Species	# Of Species
30	19
Menhinick's Index-0.00057349	Menhinick's Index-0.00036321
TOTAL Menhinick's Index-0.0009367	

Site #1 had a total of 30 lichen species present and Site #2 had a total of nineteen (19) species of lichen present (Table 6). The results showed that site #1 has a higher specie richness than site #2. A chart was generated by Microsoft Excel

(Figure 7) to illustrate the comparison of the number of species present at both Sites #1 and #2 and it clearly showed that Site #1 has the most species of lichens.

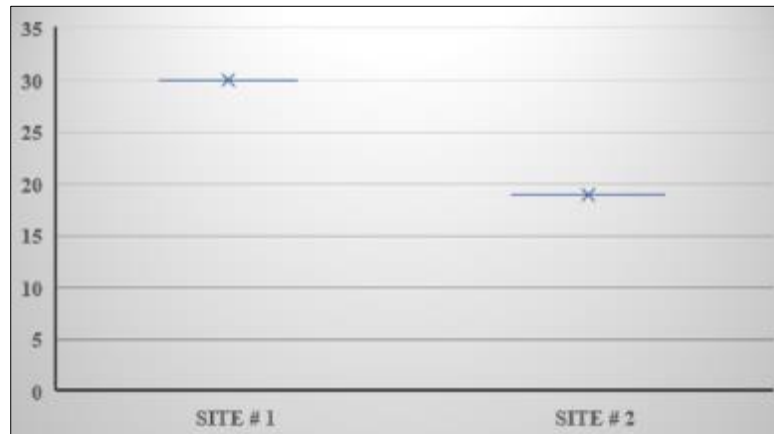


Figure 7 Comparison of the Species Richness at the two (2) sites

The data set was subjected to analysis by R software version 4.2.2 and R-Studio and the Pielou index was used to calculate the species richness at Site #1 and Site #2. The code [## Species richness (S) and Pielou's evenness (J): lichensp.rich <- specnumber(lichen1) lichensp.rich] was run and the values 30 and 19 was obtained for Site #1 and Site #2 simultaneously.

The study has added to the cumulative knowledge of lichen flora in Guyana with a total of fifty-two thousand three hundred eleven (52,311) individuals representing thirty (30) species (Table 2). This is one of the first studies on lichen diversity in a coastal ecosystem which is impacted by anthropogenic activities within Guyana. The species richness accounted for in this study may be unable to be directly compared to other results from other similar studies due to differences in sampling procedures. Part of the host tree trunks were sampled in this study, however in other studies the entire tree may have been sampled.

Lecanoraceae, *Graphidaceae*, and *Arthoniaceae* had the highest percentages of crustose lichen individuals and were the most prevalent lichens (Figure 5). However, the *Parmeliaceae*, *Caliciaceae*, *Collemaaceae*, and *Lichinaceae* families have the highest abundances of foliose lichens (39%) (Figure 5). It is impossible to speculate on the potential reasons of the observed species richness because many limiting factors, including site conditions, pollution, light intensity, life expectancy of the species, and canopy cover, were not addressed in this study.

Site # 1 has eleven species of host trees: *Mangifera indica*, *Artocarpus camansi*, *Citrus aurantiifolia*, *Citrus reticulata*, *Citrus sinensis*, *Citrus limon*, *Psidium guajava*, *Annona muricata*, *Azadirachta indica*, *Melicoccus bijugales* and *Cocos nucifera* – however Site # 2 was predominantly populated with *Cocos nucifera* as the major host tree and *Mangifera indica* and *Tamarindus indica* were also present.

Species diversity indices are unitless, which makes it difficult to understand what a given value means. The indices are, therefore, used to compare among sites. The square root of the number of species to the total number of individuals forms the basis of Menhinick's Index for Richness (Priyadarshi, 2019). Overall, the Site # 1 community of lichens has the highest Menhinick's Index than the Site # 2 community (Table 6). Site # 1 has the highest individual count as well as the highest species count while Site # 2 has the lowest individual as well as species count. The overall species richness index for No. 63 Benab was 0.0009367 (Table 6).

3.4. Estimated Species Evenness

Table 7 shows the results obtained when the species evenness was calculated using Microsoft Excel. The results clearly showed that site #1 has a higher species evenness than site #2. A chart was generated by Microsoft Excel (Figure 8) to compare the species evenness data of Sites #1 and #2.

The data was subjected to the R software version 4.2.2 and R-Studio and the code [J.lichen <- lichenSD/log(lichensp.rich) #Pielous Evenness J.lichen] was used to calculate the species evenness at the two sites. The results obtained from the

analysis yielded a value of 0.8300326 for Site #1 and 0.7139468 for Site #2. These values obtained were the same as the values obtained when calculations were done via Microsoft Excel (Table 7).

Table 7 Comparison of Species Evenness at the two (2) sites

SITE	Shannon Diversity Index (SDI)	Species Richness (SR)	Species Evenness (SDI/LN[SR])
#1	2.823105	30	0.830032686
#2	2.102173	19	0.713946872

Pielou's Index for Species Evenness was utilized to compare species evenness. This just compares the Shannon-Wiener's Index's actual diversity value to the widest range of diversity values, which is 0 to 1. Pielou's index value decreases when species abundance within the sample varies more between different taxa [21]. The Site # 1 community has a higher species evenness than the Site # 2 community (Table 7). Pielou's index is highly dependent on sample size and is very sensitive to rare taxa [21].

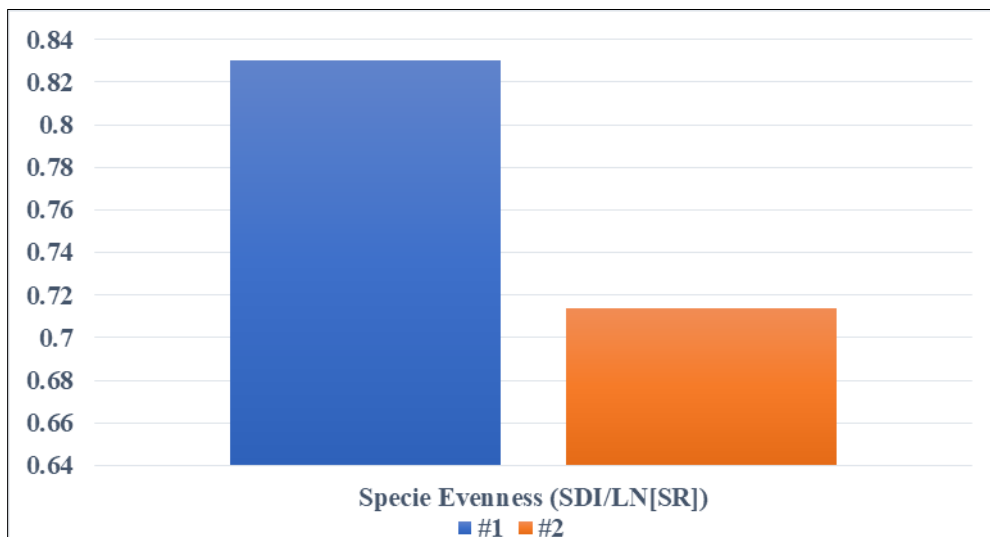


Figure 8 Comparison of the Species Evenness at the two (2) sites

3.5. Influence of Rainfall on Lichen Abundance

Lichens continue to grow and are more visible during periods when dew, mist, and rain water are present [49]. Most of the fieldwork pertaining to this research was completed during the rainy season, between the months of May-August. Most lichen specimens were recorded during this period because they were readily visible on their host trees since rain water was available in adequate amount.

The primary factor underlying lichens' sensitivity to climate is their poikilohydric nature, which directly regulates pertinent eco-physiological processes affecting growth rates and species distributions [20]. In particular, thallus water saturation and desiccation are influenced by ambient temperature and moisture levels, which are intimately correlated with lichen physiology [18]. Even while alternative hydration sources, including dew and air humidity, may be significant [12], lichens are photosynthetically active when wet, hence the amount of precipitation directly affects their growth rate, biomass accumulation, and variety. Therefore, moisture (rainfall) has a significant impact on and dependence on the distribution, quantity, diversity, richness, and evenness of lichen species.

According to the literature obtained from past studies that were conducted, this research pales in comparison since it was conducted during the period of intense rainfall on the coastal areas of Guyana. Additionally, the rich lichen diversity and abundance that recorded a grand total of fifty-two thousand three hundred eleven (52,311) individual lichens from fifteen (15) families, twenty-three (23) genera and thirty (30) species at the two (2) sites at No. 63 Benab, Berbice, Guyana are highly influenced by the high amount of precipitation in the area.

4. Conclusion

The knowledge about the variety of lichens in Guyana has been expanded by this study. More lichens were present at Site #1 than at Site #2. It was observed that the lichen community at Site #1 had a higher species richness, species evenness, and species diversity than the lichen community at Site #2 at No. 63 Benab. However, significant taxa might not have been documented and taken into account because of the sampling strategy and effort used for this study.

There was a higher number of crustose lichens as compared to foliose, fruticose and squamulose. Site # 1 with a higher diversity of tree species showed a higher diversity of lichens. Site # 2 was dominated by *C. nucifera* and was also noted to have the lowest lichen distribution, lichen abundance, species richness, species evenness and species diversity. This may be indicative that lichen diversity in an area is correlated to the diversity of the host specificity.

Moisture (rainfall) appear to play an integral and important role on the lichen species distribution, diversity, richness and evenness because lichens are photosynthetically active when wet and this directly controls relevant eco-physiological processes influencing growth rates and species distributions.

Recommendations

Based on the study and the results obtained, the following recommendations are made:

- Future studies should be conducted to research other parts of the host tree and their lichen diversity as well as other substrates on which lichens grow.
- Sampling should be done during the dry season instead of only during wet season and results should be compared to observe if lichen abundance is affected by such seasonality.
- Future research should test environmental parameters such as temperature, moisture, air quality, water quality and salinity to determine if any of these factors contribute to the lichen diversity, richness or evenness in a specific area.
- It is also necessary to sample and study lichens from other administrative regions to examine the diversity of lichens in Guyana,
- Future research should examine bark traits that are known to affect lichen appearances to ascertain whether there is in fact a relationship between lichens and tree hosts in terms of host tree specificity.
- It will be important to evaluate the lichen variety in other parts of Guyana as conservation efforts develop as this will help in making decisions to conserve these communities by informing conservationists and the government about the current state of these communities.
- Lichens are useful bio-indicators of ecological health because they can measure things like air quality. As a result, lichens can be used in studies that aim to evaluate and monitor ecosystems, particularly those in anthropogenic settings.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors hereby declare that this manuscript does not have any conflict of interest.

Statement of informed consent

All authors declare that informed consent was obtained from all individual participants included in the study.

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