

QATAR UNIVERSITY

COLLEGE OF ARTS AND SCIENCES

NUTRIENT CONSUMPTION OVER TIME IN PLANT LITTER

DECOMPOSITION IN DRYLANDS: EFFECTS OF DIFFERENT SPECIES

MIXTURES AND PHOTODEGRADATION

BY

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ABSTRACT

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Title: Nutrient Consumption over Time in Plant Litter Decomposition in Drylands: Effects of Different Species Mixtures and Photodegradation

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Plant litter decomposition in soil is a dynamic process that is affected and regulated by several factors. Soil microbial activity, light intensity, litter composition and habitat all affect the rate of decomposition of litter. In this study we aimed to elucidate the effect of photodegradation, and different litter species mixtures on decomposition rate of nitrogen and carbon. Plant leaf litter of *Conocarpus lancifolius* and *Avicennia marina*, mixture of two species (*Conocarpus lancifolius* and *Ziziphus spina-christi*) and mixture of four species (*Conocarpus lancifolius*, *Avicennia marina*, *Ziziphus spina-christi*, and *Acacia ehrenbergiana*) were placed in litter bags to degrade below ground and on the surface. Samples were harvested after 50, 100, 205, and 345 days after which nitrogen and carbon concentration of the litter was analyzed. The study revealed that nitrogen and carbon on the surface decomposed faster for *A. marina* and the four species mixture compared to the one below ground which shows that sunlight exposure increases the rate of decomposition of nitrogen irrespective of its species. The species composition also influences the N % and C % decomposition in the plant litter.

The study revealed that N and C on the surface decomposed faster for four species mixture as compared to the one below ground which shows that sunlight exposure increases the decomposition of C compared to N of plant litter irrespective of

its species. The study also revealed that the mixture of different species can influence the decomposition of N % and C %.

Keywords: Nutrient Consumption, Microbial decomposition, Plant Litter, Arid Climate

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CHAPTER 1: INTRODUCTION

Work on the decomposition method has steadily grown since the litter bag technique was introduced in the 1960s, which allowed to quantify mass loss and also to measure decomposition levels and evaluate rates between locations, litter forms and many types of procedures (Prescott, 2010). The atmospheric CO₂ concentrations has been on the rise since the industrial revolution (Kochsiek, 2010). The litter decomposition plays a big role in the carbon cycling (Field et al., 1998). Therefore, it is very crucial to understand the factors controlling decomposition rates in the dry lands which comprises of more than 40% of the earth's surface area (Právělie, 2016; Reynolds, 2001), accounting net primary production of 30 percent and almost 20 percent of the world's soil organic C pool (Field et al., 1998; Lal, 2004).

Three main drivers influencing the decomposition of litter are: the climate, litter quality and the presence & availability of decomposing species (Bottner, 1995). Two simultaneous and basic sets of processes are involved in the litter decomposition. (1) Corresponding mineralization through succession of micro-organisms. (2) Downward leaching in the soil of soluble compounds, where compounds such as C and nitrogen (N) are converted to mineral or organic matter. Thus it is a complicated process that requires both biotic and abiotic factors (Aerts, 1997; Berg et al, 2005).

In terms of biotic factors, many studies have showed that plant litter is grazed by soil invertebrates such as mites and ants in majority of desert ecosystems (Elkins et al., 1982; Silva et al., 1985; Tracy et al., 1998; Walter et al., 1975). Arid environments comprise of adjacent patches of differing flora that vary in C content, nutrients availability, soil conditions, and access to UV, thus are therefore prone to choose numerous microbial species (Gallo et al., 2006).

For abiotic factors, decomposition of litter is generally presumed to rely on

precipitation and temperature which was shown to be largely correct for a significant number of ecosystems (Parton et al., 2007). However, abiotic causes are now recognized as major forces causing substantial functional deterioration, primarily by photodegradation via ultraviolet radiation (UV) exposure (Arriaga et al., 2007; Austin et al., 2006; Gallo et al., 2006; Vossbrinck et al., 1979). The process of photodegradation is where solar radiation degrades many compounds including plant litter, both by UV and visible ranges.

The possible significance of abiotic drivers under arid conditions is illustrated in recent studies on drylands (Austin et al., 2011; Austin et al., 2006; Hewins et al., 2013), whereas studies during wetter conditions indicates that abiotic drivers can be less significant than factors like rainfall induced biological activity (Araujo et al., 2015). Increasing numbers of studies in arid and semi-arid habitats have found that UV radiation (280–400 nm) photodegradation significantly contributes to surface litter decomposition (Austin et al., 2006; Brandt et al., 2010; Gallo et al., 2009). A study that manipulated rainfall and UV radiation in a semi-arid areas found that photodegradation increased the level of decay in dry conditions but not under wetter conditions (Brandt et al., 2007). Furthermore, during dry periods the microbial activity can be low thus the effect of photodegradation may increase the litter decay due to microbes not being able to take up the nitrogen which then gets lost from the system (Brandt et al., 2007; Parton et al., 2007). Several hypotheses have been suggested to clarify the decomposition of litter in arid environments more rapidly than expected, including precipitation spikes (Austin et al., 2004), consumption by arthropods and soil burial (Throop et al., 2007). Nonetheless, evaluations of these theories cannot explain why levels of decomposition in arid areas are faster than expected (MacKay et al., 1994; Schaefer et al., 1985). Even though studies show that photodegradation poses a great role for dry areas compared to

the wet areas, other conditions such as the timing of rainfall, microbial community, soil type and canopy cover in wet areas are also important (Brandt et al., 2010; Foereid et al., 2010; Gallo et al., 2006; Hewins et al., 2019).

The importance of associations between decomposition levels or various biotic and abiotic variables based on a variety of other factors, like litter composition, microsite features (i.e. extent of rockiness or steepness of the slope) and plant composition. The importance of associations between decomposition levels or various biotic and abiotic variables are based on a variety of other factors, like litter composition and microsite features (i.e. extent of rockiness or steepness of the slope) (Aerts, 1997; Pérez-Harguindeguy et al., 2000; Schaefer et al., 1985). The factors mentioned above can be divided into two main categories, first being the litter quality and secondly the microsite features. Studies have shown that the quality of litter such as its physio-chemical characteristics has a strong correlation with the rates of its decomposition. For example plants with less structural tissue or bigger leaf area, high nutrient content and lower secondary compounds typically decompose more easily (Pérez-Harguindeguy et al., 2000; Wright et al., 2002). (Aerts, 1997; Pérez-Harguindeguy et al., 2000; Schaefer et al., 1985). The factors mentioned above can be divided into two main categories, first being the litter quality and secondly the quality of the habitat. The key factors affecting the decomposition in the plains was that it had the highest solar irradiance and termite activity. Studies have shown that the quality of litter such as its physio-chemical characteristics has a strong correlation with the rates of its decomposition. For example plants with less structural tissue or bigger leaf area, high nutrient content and lower secondary compounds typically decompose more easily (Pérez-Harguindeguy et al., 2000; Wright et al., 2002).

One of the theories suggest that lack of a negative correlation on lignin content

and mass loss often found in arid systems can be explained by a greater role of photodegradation in arid systems versus mesic systems (Moorhead et al., 1994; Whitford et al., 1981). Several reports have shown that UV radiation exposure enhances the degradation of litter lignin (Austin et al., 2010; Day et al., 2007; Henry et al., 2008). Therefore, litter of plants with higher lignin concentration can be more vulnerable to photodegradation than plant litter with lower lignin concentrations. Yet only a number of UV exposure studies have been known to use over one litter species (Brandt et al., 2007; M.E. Gallo et al., 2006). Whereas some studies have suggested the plant litter composition can be a significant factor affecting photodegradation levels, others say litter surface area is much more essential than its composition (Gallo et al., 2006).

A study conducted by Martínez-Yrizar et al in 2007, proved a clear correlation between the effect of habitat and litter quality. It was conducted in the Sonoran Desert in Mexico, which had 3 study sites within the area of 800 ha. The three study sites were distinctly selected for the difference in their environment. The first site was named “Plains”, partaking a large exposed bare ground with very low litter production. Only bearing dominant trees and widely spaced shrubs. The second site is known as “Arroyos” represented by xero-riparian vegetation which had the highest plant biomass and litter fall. Lastly, the third site was designated as the “Hillside” was represented by the Foothills Thornscrub. In comparison to the first and second site, the third site had intermediate litter fall and biomass. Furthermore, the study had three main plant litter conditions 1) Plant litter from *Encelia farinose*, 2) *Olneya tesota* and 3) mixture of the litter from five different species. The study showed the decomposition rates as k values, calculated by linear regression and $t_{1/2}$ representing the time required for decomposition 50% of the litter in months. The results indicated that the Plains site had an average decomposition rate of 3 times faster than the other two habitats, with the fastest

decomposition rate shown in *E. farinosa* followed by *O. tesota* and the mixture of species. Furthermore, Plains site was also the fastest in terms of the time required for decomposition of 50% of the plant litter. It had an average of 2.9 months followed by Hillside site with 7.3 months and lastly Arroyos with an average of 9.7 months. The key factors affecting the decomposition in the plains was that it had the highest solar irradiance and termite activity. In terms of the plant litter species, again *E. farinosa* was the fastest at decomposing 50% of its litter by 4.3 months in average but was even faster in the Plains site (1.8 months). Followed by was the mixture of species sample and *O. tesota* litter. (Martínez-Yrizar et al, 2007). The results from this study addresses many of the factors which will be studied in this project. One of which is the photodegradation of the plant litter in arid environments. Even though this study was conducted in the same desert, but the difference in the habitat type which were around 1 km apart from each other can manipulate the results widely. The Plains study site can somewhat depict the environment of Qatar since it was mostly exposed ground with few dominant trees and spaced shrubs as mentioned earlier. Due to that the light can directly reach the samples and photodegradation may take place and thus speed up decomposition rate as shown in the results. Similarly, the half-life of the 3 experimental litter groups placed in the Plains site were much lower compared to the other two sites (Martínez-Yrizar et al, 2007). Furthermore, the second factor that will be studied in our study is variances in decomposition rates of individual plant litter species, and mixture of plant litter from various species. As observed in Table 1, a similar approach has already been conducted in the Sonoran desert, Mexico (Pérez-Harguindeguy et al., 2000). The results from their experiment showed that the *E. farinosa* litter had the fastest decomposition rate and the shortest half-life compared to the *O. tesota* litter and the mixture of species. The findings revealed that the leaf litter of *O. tesota* could be identified by the end of the

experiment while *E. farinose* remaining litter was significantly deteriorated. This demonstrates that there is a direct effect of the litter chemistry and the leaves physical properties on litter decomposition rates (Pérez-Harguindeguy et al., 2000).

1.1 Objectives

1. Identify the difference in nutrient breakdown of plant litter placed on surface and below the ground.
2. Study the role of photodegradation on plant litter in arid climate.
3. Study the impact of different plant litter mixtures on nutrient breakdown.

1.2 Hypothesis

1. A mixture of plant litter species would decompose faster as compared to only single plant litter.
2. Photodegradation would lead to faster breakdown of N & C in litter placed above ground compared to plant litter placed under the ground.
3. Nitrogen would breakdown faster over time compared to carbon.

CHAPTER 2: METHODOLOGY

2.1 Study Site

The experimental site was established at the Biological Field of Qatar University in Doha, Qatar (GPS: 25°22'19.6"N 51°29'28.5"E). The Qatari climate is known to be arid, with precipitation abundant between October and May and typically mostly dry between June and September (Cheng et al., 2017; Mamoon et al., 2014). In Qatar, the rainfall is low, an average of roughly 80 mm/year. The temperature varies in January from 7°C (45°F) to approximately 45°C (113°F) in summer.

2.2 Experimental Species

The experiments is conducted using four different species, Sp. 1 - *Conocarpus lancifolius* also known as damas was introduced to Qatar in Mesaieed Industrial City but now it is much more widespread (Norton, 2009). It is also known to produce dense foliage and is a saline-tolerant tree that makes it very suitable for this study. Sp. 2 - *Avicennia marina* also known as dwarf mangrove, is a native species often found in sheltered bays and creeks along the tide. The areas remaining with *Avicennia marina* are in NE Qatar, Ras Laffan, Al Dhakhira, Al Khor, and Al Wakra (Norton, 2009). Sp. 3 - *Ziziphus spina-christi* also known as sidra tree, occasionally found in farms and roadside plantations but seldom find to be naturalized (Norton, 2009). Sp. 4 - *Acacia ehrenbergiana* also known as salam, is a native species found frequently in the rod at with other large shrubs and trees. It has many uses such as the stems being used for firewood and its pods & leaves being grazed by animals (Norton, 2009).

2.3 Experimental Design

The leaves from the plant species were dried at 48h at 70C. Thereafter, 2g of the dried leaf material was sealed in commercial empty tea bags made by nylon (hereafter litter bags). The weight was obtained using a balance with 0.0001 g accuracy. A total of 160 samples, 40 samples for each species mixture were allocated for two experimental conditions. Above ground (20 samples/species mixture) and buried at 8-10cm depth in the soil below ground (20 samples). In each of those two experimental conditions, there were four setups with five replicates each. The first and second setup consisted of plant litter from one species in each experimental group *C. lancifolius* or *A. marina*. The third setup included the mixture of plant litter from *C. lancifolius* & *Z. spina-christi* plant litter and finally the fourth setup was mixture of plant litter from *C. lancifolius*, *A. marina*, *Z. spina-christi* and *A. ehrenbergiana* as shown in in Table 1.

Furthermore, the species are assigned to the following abbreviations to represent the data obtained: *Conocarpus lancifolius* (Sp. 1), *Avicennia marina* (Sp. 2), *Ziziphus spina-christi* (Sp. 3), and *Acacia ehrenbergiana* (Sp. 4). The samples were placed in the field experiment on 28th February 2010 and collected on 18th April 2019 (50 days), 6th June 2019 (100 days), 21st September 2019 (205 days), and 9th February 2020 (345 days). After each collection, the samples were placed in the freezer for later chemical analyses.

Table 1. Overview of 8 experimental setup indicating the condition, species and number of litter types

Experimental Condition	Experiment #	Species	# of litter type
Above Ground	E1a	<i>C. lancifolius</i>	1
	E2a	<i>A. marina</i>	1
	E3a	<i>C. lancifolius</i> & <i>Z. spina-christi</i>	2
	E4a	<i>C. lancifolius</i> , <i>A. marina</i> , <i>Z. spina-christi</i> & <i>A. ehrenbergiana</i>	4
Below Ground	E1b	<i>C. lancifolius</i>	1
	E2b	<i>A. marina</i>	1
	E3b	<i>C. lancifolius</i> & <i>Z. spina-christi</i>	2
	E4b	<i>C. lancifolius</i> , <i>A. marina</i> , <i>Z. spina-christi</i> & <i>A. ehrenbergiana</i>	4

2.4 Litter Analysis

To obtain the initial concentrations of the experimental species, fresh litter (undecomposed) were collected separately and then combined as per the experimental setup (Table 2). Each experimental setup was conducted three times to obtain an average result. The samples were then placed in a oven for 48 hours at 70 °C. Once the samples completely dried, these were then pulverized into powder form. And then, using the FLASH 2000 Organic Elemental Analyzer, 3.5 mg of each sample was analyzed. N% w and C% w were the findings obtained.

All the data was obtained except for the above ground and below ground experiments from the last collection date for Sp. 1 and mixture of 4 species. In addition, only the below ground experiment data for Sp.2 and the mixture of Sp. 1 & 3 have been obtained. Owing to disruptions by ants and dogs, these samples were destroyed.

2.5 Statistical Analysis:

To determine the normal distribution of the study Shapiro-Wilk's test ($p > 0.05$) (Shapiro et al, 1965; Razali et al., 2011) and visual inspection of their histogram were analyzed. The Inverse Distribution Function of normal (idf.Normal) in SPSS was used to transform the data to be normally distributed To test for the effect of litter type/mixture, time and position (above or below ground), on N% and C% breakdown, three factor univariate ANOVA was used. All analyses was performed in IBM SPSS statistics (Version 26).

Table 2. Initial results of the undecomposed plant litter

Species	#	N% W	C% W
<i>C. Lancifolius</i>	1	1.37	31.20
	2	1.75	44.32
	3	1.50	31.70
	Average	1.54	35.74
<i>A. marina</i>	1	2.40	38.67
	2	2.25	38.08
	3	1.98	34.31
	Average	2.21	37.02
<i>C. lancifolius & Z. spina-christi</i>	1	1.45	33.15
	2	1.55	36.07
	3	1.61	39.20
	Average	1.54	36.14
<i>C. lancifolius, A. marina, Z. spina-christi & A. ehrenbergiana</i>	1	2.13	40.15
	2	1.70	36.90
	3	1.58	34.75
	Average	1.80	37.27

CHAPTER 3: RESULTS

The data of remaining N % and C % were not normally distributed for all the experimental conditions on ground and below ground. Thus the data was transformed and is represented in Tables 2, 3 & 4. The analyzed data is in the appendix section (Tables 6, 7 & 8) for reference.

3.1 *Conocarpus lancifolius*

Initial N % and C % of *C. Lancifolius* litter contained an average of 1.542 % (s.d. \pm 0.193) and 35.741 % (s.d. \pm 7.436), respectively (Table 2). Following the first collection in April 2019, mean remaining N % & C% of was 2.166 % (s.d. \pm 0.496) & 34.448 % (s.d. \pm 6.572) for above ground and 1.982 % (s.d. \pm 0.080) & 35.893 % (s.d. \pm 3.092) for below ground respectively (Table 3 & 4). The second collection in June 2019, average remaining N % & C % was 1.727 % (s.d. \pm 0.528) & 33.051 % (s.d. \pm 5.980) for above ground and 2.003 % (s.d. \pm 0.219) & 43.467 % (s.d. \pm 5.611) for below ground respectively (Table 3 & 4). The last collection in September 2019, average remaining N % & C % of was 2.187 % (s.d. \pm 0.203) & 37.680 % (s.d. \pm 2.319) for above ground and 1.814 % (s.d. \pm 0.603) & 37.450 % (s.d. \pm 6.642) for below ground, respectively (Tables 3 & 4).

Secondly, only one of the collection periods showed higher decomposed N % above ground compared to the below ground samples. Which was the second collection period, it had 0.27 % N more decomposed above ground. While the first and third collection periods had higher N decomposition below ground, 0.19 % and 0.38 % N, respectively. Overall the average results of all collection periods indicated that the decomposition was 0.10 % higher compared to above ground (Tables 3 & 4).

In terms of the decomposed C %, the first two collections had higher decomposition above ground. The first collection had 1.44 % C more decomposed above ground where the second collection had 10.42 % C. On the contrary, the last

collection had higher remaining C % above ground compared to below ground but it was only 0.23 % higher. Overall the average results indicated that the above ground litter C % decomposed 3.88 % more compared the litter placed below ground (Tables 3 & 4).

3.2 *Avicennia marina*

For the second experimental group of *A. marina*, the initial litter had an average of N 2.221 % (s.d. \pm 0.212) and C 37.017 % (s.d. \pm 2.366) (Table 2). Following the first collection in April 2019, mean remaining N % & C% of was 2.449 % (s.d. \pm 0.208) & 44.700 % (s.d. \pm 5.107) for above ground and 2.712 % (s.d. \pm 0.066) & 45.007 % (s.d. \pm 2.560) for below ground, respectively (Table 3 & 4). The second collection in June 2019, average remaining N % & C % was 2.819 % (s.d. \pm 0.337) & 33.051 % (s.d. \pm 5.980) for above ground and 2.706 % (s.d. \pm 0.186) & 43.467 % (s.d. \pm 5.611) for below ground, respectively (Table 3 & 4). The last collection in September 2019, average remaining N % & C % was 2.187 % (s.d. \pm 0.203) & 37.680 % (s.d. \pm 2.319) for above ground and 1.814 % (s.d. \pm 0.603) & 37.450 % (s.d. \pm 6.642) for below ground, respectively (Tables 3 & 4).

Secondly, only two of the collection periods showed higher decomposed N % above ground compared to the below ground samples. The first collection and third collection had 0.26 % and 0.20 % N more decomposed above ground, respectively. Where the second collection had higher N decomposition below ground, 0.12 % N more than the above ground samples. Overall the average results of all collection periods indicated that the above ground N decomposed 0.11 % more compared the litter placed below ground (Table 3 & 4).

Lastly, all the three collection periods showed higher decomposed C % above ground compared to the below ground samples. The first, second and third collection

periods had 0.31 %, 2.78 % and 1.8 % C more decomposed above ground, respectively. Overall the average results of all collection periods indicated that the above ground C decomposed 1.63 % more compared the litter placed below ground (*Tables 3 & 4*).

3.3 Two species mixture

The mixture of *C. lancifolius* & *Z. spina-christi* litter's initial had an average of N 1.538 % (s.d. \pm 0.085) and C 36.139 % (s.d. \pm 3.025) (Table 2). Following the first collection in April 2019, mean remaining N % & C % of was 1.743 % (s.d. \pm 0.280) & 33.575 % (s.d. \pm 2.538) for above ground and 2.139 % (s.d. \pm 0.291) & 43.430 % (s.d. \pm 4.368) for below ground, respectively (Table 3 & 4). The second collection in June 2019, average remaining N % & C % was 2.077 % (s.d. \pm 0.199) & 41.672 % (s.d. \pm 1.718) for above ground and 2.118 % (s.d. \pm 0.168) & 44.575 % (s.d. \pm 4.947) for below ground, respectively (Table 3 & 4). The last collection was in September 2019, average remaining N % & C % of was 1.943 % (s.d. \pm 0.894) & 38.937 % (s.d. \pm 12.482) for above ground and 1.931 % (s.d. \pm 0.438) & 40.645 % (s.d. \pm 6.869) for below ground, respectively (Table 3 & 4).

Secondly, only two of the collection periods showed higher decomposed N % above ground compared to the below ground samples. The first collection and second collection periods had 0.40 % and 0.04 % N more decomposed above ground, respectively. Where the third collection had higher N decomposition below ground, 0.01 % N more than the above ground samples. Overall the average results of all collection periods indicated that the above ground N % decomposed 0.14 % more compared the litter placed below ground (*Table 3 & 4*).

Lastly, all the three collection periods showed higher decomposed C % above ground compared to the below ground samples. The first, second and third collection periods had 9.85 %, 2.90 % and 1.71 % C more decomposed above ground,

respectively. Overall the average results of all collection periods indicated that the above ground C % decomposed 4.82 % more compared the litter placed below ground (Table 3 & 4).

3.4 Four species mixture

The last experimental group which included the litter mixture of *C. lancifolius*, *A. marina*, *Z. spina-christi* & *A. ehrenbergiana*, had an initial average of N 1.804 % (s.d. \pm 0.287) and C 37.268 % (s.d. \pm 2.720) (Table 2). Following the first collection in April 2019, mean remaining N % & C% of was 1.958 % (s.d. \pm 0.259) & 36.081 % (s.d. \pm 5.165) for above ground and 2.194 % (s.d. \pm 0.200) & 44.182 % (s.d. \pm 5.403) for below ground, respectively (Table 3 & 4). For the second collection in June 2019, average remaining N % & C % was 1.963 % (s.d. \pm 0.265) & 39.615 % (s.d. \pm 1.242) for above ground and 2.186 % (s.d. \pm 0.206) & 44.051 % (s.d. \pm 4.936) for below ground, respectively (Table 3 & 4). The last collection in September 2019, average remaining N % & C % of was 1.803 % (s.d. \pm 0.113) & 34.832 % (s.d. \pm 3.648) for above ground and 2.077 % (s.d. \pm 0.230) & 42.877 % (s.d. \pm 4.327) for below ground, respectively (Tables 3 & 4).

Secondly, all the three collection periods showed higher decomposed N % above ground compared to the below ground samples. The first, second and third collection periods had 0.23 %, 0.23 % and 0.28 % N more decomposed above ground, respectively. Overall the average results of all collection periods indicated that the above ground N % decomposed 0.35 % more compared the litter placed below ground (Table 3 & 4).

Lastly, all the three collection periods showed higher decomposed C % above ground compared to the below ground samples. The first, second and third collection periods had 8.10 %, 4.43 % and 8.05 % C more decomposed above ground,

respectively. Overall the average results of all collection periods indicated that the above ground C % decomposed 6.86 % more compared the litter placed below ground (Tables 3 & 4).

Table 3. Remaining N% & C% of 3 collection dates for below ground experimental groups

Collection Dates	<i>C. lancifolius</i>		<i>A. marina</i>		<i>C. lancifolius</i> & <i>Z. spina-christi</i>		<i>C. lancifolius</i> , <i>A. marina</i> , <i>Z. spina-christi</i> & <i>A. ehrenbergiana</i>	
	N% W	C% W	N% W	C% W	N% W	C% W	N% W	C% W
4/18/2019	1.94	37.64	2.70	43.17	1.79	48.15	2.30	51.83
	1.87	38.40	2.80	49.14	1.93	45.17	2.29	45.34
	2.01	35.23	2.75	43.56	2.54	37.08	2.15	43.97
	2.00	37.42	2.63	43.30	2.27	41.07	2.36	43.04
	2.09	30.78	2.68	45.87	2.16	45.69	1.87	36.73
Average	1.98	35.89	2.71	45.01	2.14	43.43	2.19	44.18
6/8/2019	2.18	43.83	2.44	42.20	2.17	49.98	2.28	41.96
	2.23	36.25	2.83	49.54	2.37	42.67	2.22	42.43
	2.04	46.88	2.61	41.62	2.11	43.69	2.01	40.96
	1.84	50.49	2.91	45.51	1.97	48.78	2.47	52.83
	1.73	39.89	2.72	54.40	1.96	37.75	1.96	42.08
Average	2.00	43.47	2.70	46.65	2.12	44.57	2.19	44.05
9/21/2019	1.32	35.88	2.35	32.05	1.24	28.95	1.86	36.49
	1.29	29.39	2.65	41.18	2.43	41.73	2.28	45.01
	1.92	47.34	2.57	42.55	2.09	44.70	2.08	44.54
	1.77	34.96	2.42	35.76	1.85	41.40	2.33	47.60
	2.77	39.67	2.87	48.45	2.03	46.45	1.84	40.74
Average	1.81	37.45	2.57	40.00	1.93	40.65	2.08	42.88

Table 4. Remaining N% & C% of 3 collection dates for above ground experimental groups

Collection Dates	<i>C. lancifolius</i>		<i>A. marina</i>		<i>C. lancifolius</i> & <i>Z. spina-christi</i>		<i>C. lancifolius</i> , <i>A. marina</i> , <i>Z. spina-christi</i> & <i>A. ehrenbergiana</i>	
	N% W	C% W	N% W	C% W	N% W	C% W	N% W	C% W
	1.500	24.550	2.084	38.409	1.552	32.848	1.728	35.726
	2.549	41.267	2.536	41.994	2.166	35.829	1.988	38.195
4/18/2019	2.495	39.018	2.653	42.003	1.597	36.513	1.634	32.484
	2.444	38.837	2.467	42.087	1.656	36.140	2.185	41.290
	1.909	26.529	2.449	41.958	1.900	39.135	2.192	39.779
Average	2.179	34.040	2.438	41.290	1.774	36.093	1.945	37.495
	2.204	40.078	2.557	41.742	2.182	41.128	2.267	39.384
	2.040	37.331	2.330	36.988	2.259	40.900	1.878	39.610
6/8/2019	1.196	24.640	2.923	41.432	1.815	39.917	1.632	39.648
	1.162	23.528	2.845	41.916	2.038	40.051	1.928	40.170
	2.006	38.650	2.833	42.510	2.026	41.619	2.093	40.599
Average	1.722	32.845	2.698	40.918	2.064	40.723	1.960	39.882
	2.289	38.342	2.139	39.394	0.339	0.000	1.938	39.699
	2.033	40.433	2.564	40.208	4.081	83.591	1.698	38.943
9/21/2019	2.347	39.565	2.545	39.288	1.873	40.017	1.664	31.245
	2.218	38.354	2.362	40.603	1.865	39.723	1.824	36.986
	1.906	39.217	2.186	35.570	1.656	35.690	1.900	37.197
Average	2.159	39.182	2.359	39.013	1.963	39.804	1.805	36.814

3.5 Three Factors ANOVA

The three factor ANOVA analysis on the decomposition of N found that the time and the type of the litter were statistically significant at $P = 0.000$ ($F(3, 11.755) = 9.938$) and $P = 0.000$ ($F(3, 11.755) = 30.047$), respectively. While the position of the litter did not have a statistically significant effect at $P = 0.169$ ($F(1, 11.755) = 1.920$) on the decomposition of N % (Table 5). Furthermore, the effect from the interaction of time \times position ($F(3, 11.755) = 0.559$, $P = 0.643$), time \times litter type ($F(9, 11.755) = 0.888$, $P = 0.538$), position \times litter type ($F(3, 11.755) = 0.923$, $P = 0.432$) and time \times position \times litter type ($F(9, 11.755) = 1.018$, $P = 0.430$) did not have a statistically significant effect on the decomposition of N (Table 5).

In terms of the decomposition of the C %, the time, position and the type of the litter were all statistically significant at $P = 0.001$ ($F(3, 3081.968) = 6.087$), $P = 0.000$ ($F(1, 3081.968) = 12.943$) and $P = 0.003$ ($F(3, 3081.968) = 4.982$) on the decomposition of C %, respectively (Table 5). Furthermore, the effect from the interaction of time \times position ($F(3, 3081.968) = 1.512$, $P = 0.215$), time \times litter type ($F(9, 3081.968) = 1.115$, $P = 0.358$), position \times litter type ($F(3, 3081.968) = 0.826$, $P = 0.482$) and time \times position \times litter type ($F(9, 3081.968) = 1.113$, $P = 0.360$) did not have a statistically significant effect on the decomposition of C (Table 5).

Table 5. Results of three factor univariate ANOVAs evaluating litter type/mixture, time and position (above or below ground), on N % and C % breakdown

Source	Type III sum of squares	<i>df</i>	Mean square	<i>F</i>	<i>P</i> -value
Breakdown of N %					
Corrected Model	15.655a	31	0.505	4.812	0.000
Intercept	580.412	1	580.412	5530.313	0.000
Time	3.129	3	1.043	9.938	0.000
Position	0.201	1	0.201	1.920	0.169
Litter Type	9.460	3	3.153	30.047	0.000
Time × Position	0.176	3	0.059	0.559	0.643
Time × Litter Type	0.839	9	0.093	0.888	0.538
Position × Litter Type	0.291	3	0.097	0.923	0.432
Time × Position × Litter Type	0.961	9	0.107	1.018	0.430
Error	11.755	112	0.105		
Total	655.654	144			
Corrected Total	27.410	143			
$R^2 = 0.571$ (Adjusted $R^2 = 0.452$)					
Breakdown of C %					
Corrected Model	2233.748 ^a	31	72.056	2.619	0.000
Intercept	211685.369	1	211685.369	7692.734	0.000
Time	502.459	3	167.486	6.087	0.001
Position	356.156	1	356.156	12.943	0.000
Litter Type	411.262	3	137.087	4.982	0.003
Time × Position	124.843	3	41.614	1.512	0.215
Time × Litter Type	276.243	9	30.694	1.115	0.358
Position × Litter Type	68.161	3	22.720	0.826	0.482
Time × Position × Litter Type	275.606	9	30.623	1.113	0.360
Error	3081.968	112	27.518		
Total	231051.995	144			
Corrected Total	5315.716	143			
$R^2 = 0.420$ (Adjusted $R^2 = 0.260$)					

d.f., degrees of freedom; *F*, F-statistics; *P*-value, significance level; litter type, different experimental condition

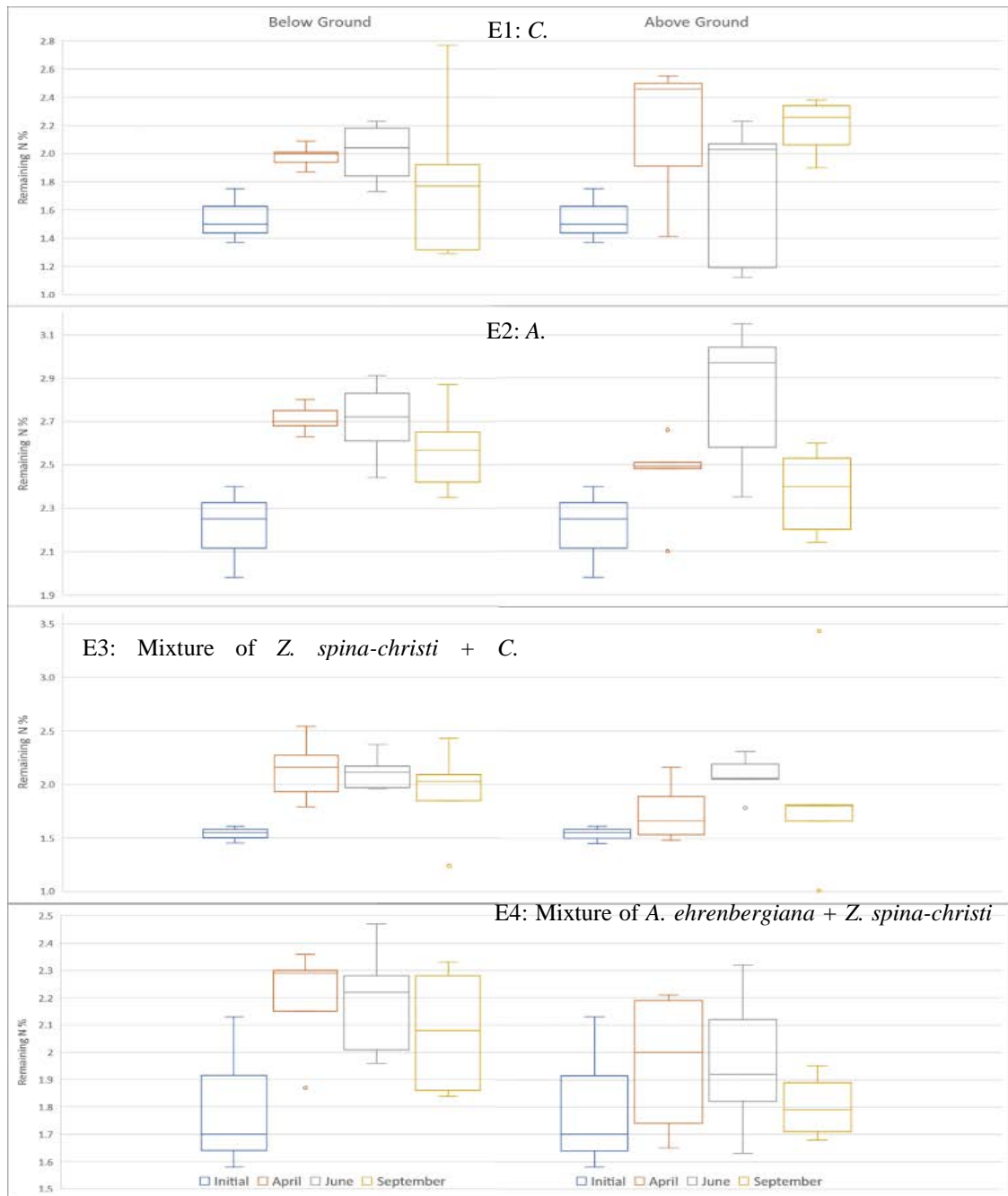


Figure 1. Boxplots of remaining N % from the start of the experiment till September 2019. The Boxplots also show the difference in the remaining N% of all the different experimental conditions such as below & above ground for *C. lancifolius*, *A. marina*, mixture of *Z. spina-christi* + *C. lancifolius* and mixture of *A. ehrenbergiana* + *Z. spina-christi* + *C. lancifolius* + *A. marina*.

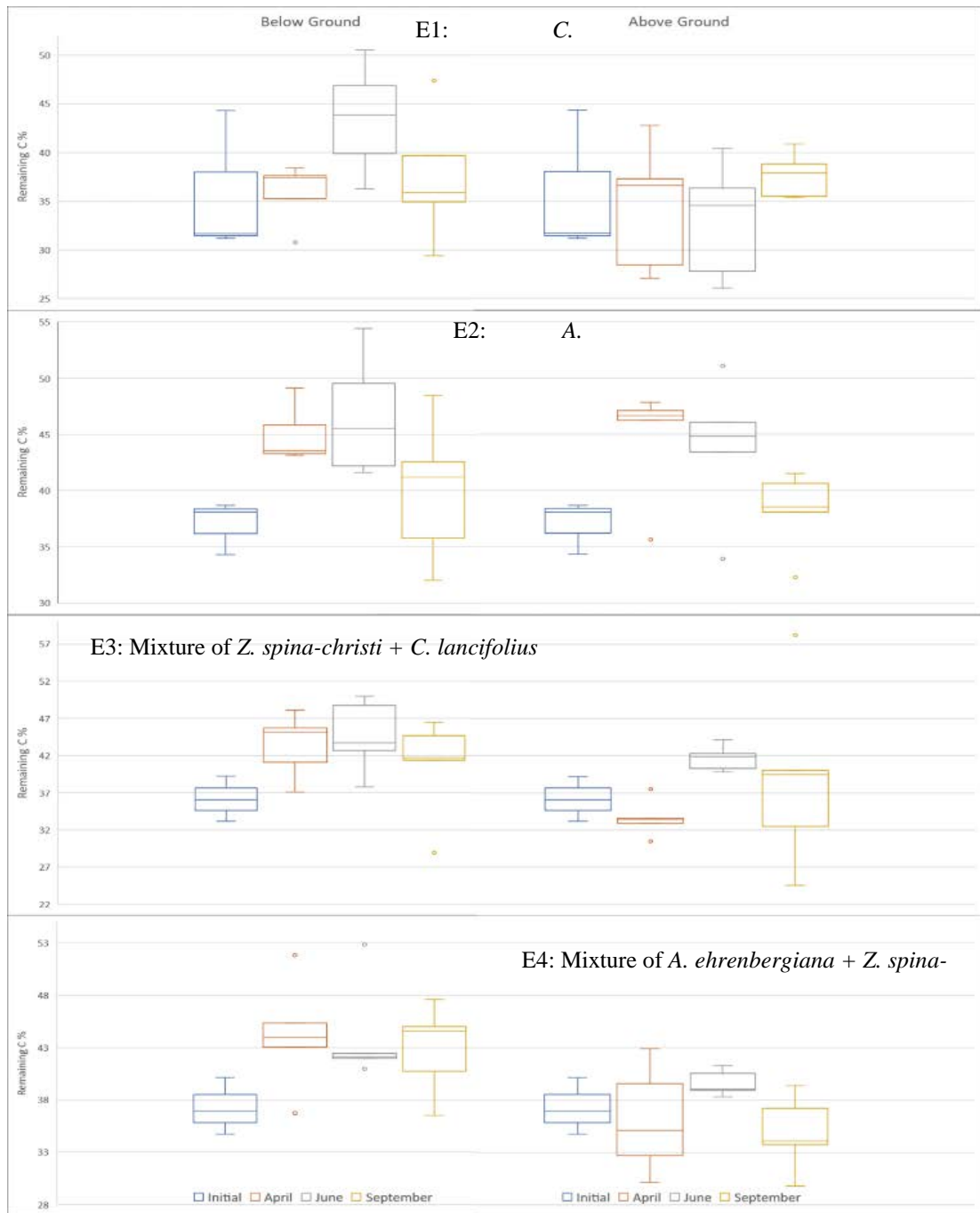


Figure 2. Boxplots of remaining C% from the start of the experiment till September 2019. The Boxplots show the difference in the remaining C% of all the different experimental conditions such as below & above ground for *C. lancifolius*, *A. marina*, mixture of *Z. spina-christi* + *C. lancifolius* and mixture *A. ehrenbergiana* + *Z. spina-christi* + *C. lancifolius* + *A. marina*.

CHAPTER 4: DISCUSSION

These results provide conclusive evidence that the higher number of litter species litter increases the decomposition rates of both the N and C, our hypothesis suggested that a mixture from various species of litter would increase the decomposition rates of the litter. Which was clear since mixture of two and four species litter decomposed much faster compared to the litter which only had single species litter. Furthermore, our second hypothesis suggested that litter placed above ground would decompose faster compare to litter placed below ground. This was also proven by seven of our experiments while only one experiment (*C. lancifolius*) showed higher decomposition rates for N below ground which could be attributed to human error since the values were withing the standard error range.

In terms of the breakdown of N % the highest decomposition was observed in the mixture of *A. ehrenbergiana* + *Z. spina-christi* + *C. lancifolius* + *A. marina* (E4) followed by mixture of *Z. spina-christi* + *C. lancifolius* (E3) and lastly *A. marina* (E2). These results indicate that the breakdown of N% is influenced by the number of species and its composition in the litter. Furthermore, all three of the experimental groups had higher decomposition of N above ground compared to below ground except for *C. lancifolius* (E1) litter, which had higher decomposition of N % below ground.

The results from the decomposition of C also indicated a much higher breakdown for all the experimental groups above ground. The highest decomposition was observed in the following order, mixture of four species, mixture of two species and the two single species (E4, E3, E1 and lastly E2). Both mixture of four and two species litter also decomposed the most above ground for N % while the litter of *A. marina* was least decomposed for C % and litter of *C. lancifolius* for N %. Both experimental groups consisted of single species, thus indicating that having one species

of litter reduces the decomposition of C and N overall.

The higher rate of decomposition above ground can be attributed to the presence of daylight, reduced vegetative cover and decreased cloud cover in arid and semi-arid climates (Arriaga et al., 2007; Austin et al., 2006; Gallo et al., 2006; Vossbrinck et al., 1979). Previously conducted studies have showed that prolonged exposure to intense sunlight (photoirradiation) promotes the decomposition of litter in arid and semi-arid lands (Gallo et al., 2009). Litter placed below ground does not get any sunlight therefore it would get decomposed slowly as compared to the litter placed above ground. *C. lancifolius* results are very fluctuating, with values of N % above ground being very contrasting (increasing in April, decreasing trend in June and a spike in September). This can be due to either experimental, sampling, instrumental error, or environmental effect (Lancerf et al., 2009). Another cause for such high rates of decomposition underground could be due to the microbial colonies of nitrogen fixing bacteria that decompose N and C underground (Hewins et al., 2019).

A higher decomposition in mixture of species litter does not necessarily mean that it is directly attributed to the number of species; some studies have stated that soils with lower N content can have different effects compared to soils with already rich N content. Soils with high N content is less prone to degrade while soil with low N content can potentially increase the decomposition of N rich plant litter (Bonanomi et al., 2014, 2017; Knorr et al., 2005; Lummer et al., 2012). Furthermore, some studies have also suggested the plant litter composition can be a significant factor affecting photodegradation levels, while others say litter surface area is much more essential than its composition (Gallo et al., 2006). These factors were clearly indicating that our results of high N and C decomposition in the mixture of litter species could be due to the difference in litter composition such as lower C and N content in the soil, higher C

and N content in plant litter and the difference in the surface area of the plant litter utilized.

In terms of overall interactions, there was no statistically significant interactions between time \times position, time \times litter type, position \times litter type and time \times position \times litter type on the decomposition of N and C. Thus, it means that the different factors were not influenced by the other factors.

The relationship between carbon and nitrogen decomposition between different species could not be established due to fluctuating results and the dubious initial % of C and N. The values for the initial and decomposed litter were less than the values obtained afterwards in subsequent months. This might be due to sampling error, handling error or even instrumental problems. Moreover, data collection was stopped due to the COVID pandemic therefore the results for February and other subsequent months could not be collected. Some of the experiments could not be continued due to destruction of experimental setup by ants and insects.

CHAPTER 5: CONCLUSION

Litter decomposition is affected by various abiotic and biotic factors. Establishing a relationship between these factors and decomposition rates has been a challenge especially in arid and semi-arid climates. The study proved that the plant litter placed above ground decomposes faster compared to the litter placed below ground. This study also revealed that a mixture of various species litter also increases the rates of decomposition compared to litter of just 1 species. Both factors have different effect on the decomposition rate of specific elements such as C and N in the plant litter.

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APPENDIX

Table 6. Original Initial results of the undecomposed plant litter

Species	#	N% W	C% W
<i>C. Lancifolius</i>	1	1.496	34.213
	2	1.736	41.703
	3	1.575	34.740
	Average	1.602	36.885
<i>A. marina</i>	1	2.362	39.535
	2	2.215	39.288
	3	1.954	37.291
	Average	2.177	38.705
<i>C. lancifolius & Z. spina-christi</i>	1	1.540	35.857
	2	1.603	38.551
	3	1.624	39.657
	Average	1.589	38.022
<i>C. lancifolius, A. marina, Z. spina-christi & A. ehrenbergiana</i>	1	2.102	40.045
	2	1.693	38.902
	3	1.615	37.680
	Average	1.803	38.876

APPENDIX

Table 7. Original data for remaining N% & C% of 3 collection dates for below ground experimental groups

Species	<i>C. lancifolius</i>		<i>A. marina</i>		<i>C. lancifolius</i> & <i>Z. spina-christi</i>		<i>C. lancifolius</i> , <i>A. marina</i> , <i>Z. spina-christi</i> & <i>A. ehrenbergiana</i>	
	N %	W	N %	W	N %	W	N %	W
Collection Dates								
	1.936	39.164	2.670	41.402	1.853	42.106	2.253	42.521
	1.899	39.388	2.708	42.330	1.935	41.762	2.249	41.784
4/18/2019	2.001	38.305	2.693	41.438	2.546	38.904	2.141	41.526
	1.960	39.086	2.628	41.420	2.237	40.539	2.336	41.292
	2.061	33.944	2.655	41.839	2.175	41.808	1.894	38.841
Average	1.971	37.977	2.671	41.686	2.149	41.024	2.175	41.193
	2.178	41.520	2.441	41.124	2.177	42.380	2.241	41.014
	2.210	38.582	2.722	42.336	2.337	41.233	2.194	41.158
6/8/2019	2.025	41.999	2.603	40.794	2.085	41.512	2.001	40.493
	1.879	42.456	2.767	41.789	1.953	42.325	2.448	42.579
	1.715	39.953	2.681	42.751	1.950	39.165	1.949	41.060
Average	2.001	40.902	2.643	41.759	2.100	41.323	2.167	41.261
	1.434	38.516	2.308	35.316	1.281	29.390	1.882	38.782
	1.389	31.065	2.644	40.590	2.382	40.845	2.248	41.753
9/21/2019	1.916	42.026	2.553	41.196	2.069	41.732	2.048	41.723
	1.779	38.180	2.370	38.459	1.881	40.601	2.282	42.029
	2.698	39.895	2.734	42.235	2.010	41.968	1.879	40.226
Average	1.843	37.936	2.522	39.559	1.925	38.907	2.068	40.903

APPENDIX

Table 8. Original data for remaining N% & C% of 3 collection dates for above ground experimental groups

Species	<i>C. lancifolius</i>		<i>A. marina</i>		<i>C. lancifolius</i> & <i>Z. spina-christi</i>		<i>C. lancifolius</i> , <i>A. marina</i> , <i>Z.</i> <i>spina-christi</i> & <i>A.</i> <i>ehrenbergiana</i>	
	N% W	C% W	N% W	C% W	N% W	C% W	N% W	C% W
	1.500	24.550	2.084	38.409	1.552	32.848	1.728	35.726
	2.549	41.267	2.536	41.994	2.166	35.829	1.988	38.195
4/18/2019	2.495	39.018	2.653	42.003	1.597	36.513	1.634	32.484
	2.444	38.837	2.467	42.087	1.656	36.140	2.185	41.290
	1.909	26.529	2.449	41.958	1.900	39.135	2.192	39.779
Average	2.179	34.040	2.438	41.290	1.774	36.093	1.945	37.495
	2.204	40.078	2.557	41.742	2.182	41.128	2.267	39.384
	2.040	37.331	2.330	36.988	2.259	40.900	1.878	39.610
6/8/2019	1.196	24.640	2.923	41.432	1.815	39.917	1.632	39.648
	1.162	23.528	2.845	41.916	2.038	40.051	1.928	40.170
	2.006	38.650	2.833	42.510	2.026	41.619	2.093	40.599
Average	1.722	32.845	2.698	40.918	2.064	40.723	1.960	39.882
	2.289	38.342	2.139	39.394	0.339	0.000	1.938	39.699
	2.033	40.433	2.564	40.208	4.081	83.591	1.698	38.943
9/21/2019	2.347	39.565	2.545	39.288	1.873	40.017	1.664	31.245
	2.218	38.354	2.362	40.603	1.865	39.723	1.824	36.986
	1.906	39.217	2.186	35.570	1.656	35.690	1.900	37.197
Average	2.159	39.182	2.359	39.013	1.963	39.804	1.805	36.814