An Exploratory Study Using the Ground Layer Indicator Method in Montana Rangelands

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AN EXPLORATORY STUDY USING THE GROUND LAYER INDICATOR METHOD IN MONTANA RANGELANDS

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EXECUTIVE SUMMARY

Most terrestrial surfaces have a "ground layer" composed of lichens, mosses, liverworts, hornworts, free-living algae, free-living cyanobacteria, bacteria and/or micro-fungi (Elbert et al. 2012; Smith et al. 2015). Those that occupy the soil surface are called "biological soil crust". Others occupy the wood, rock, and dead organic material on the ground surface. Collectively this network of unrelated organisms, referred here as the Ground Layer, provides many biological and ecological functions. They represent a large percentage of biological diversity on the Earth. In arid regions they occupy the nutrient-poor zones between where individual vascular plants grow (Belnap et al. 2001). As a living mulch, biological soil crusts retain soil moisture, discourage annual weed growth, and reduce soil erosion caused by wind or water (Belnap et al. 2001). Cyanobacteria, either as free-living or in symbiosis with fungi (lichens) or mosses contribute fixed nitrogen to the soil and are major players in the cycling of nitrogen (Belnap et al. 2001; Elbert et al. 2012; Smith et al. 2015). All contribute to carbon uptake, sequestration (storage), and release (Elbert et al. 2012; Zhao et al. 2016). Rock dwelling species accelerate chemical weathering, may release phosphorus, and aid in soil formation (Elbert et al. 2012; Porada et. al 2013). Biological soil crusts occupy the space between sagebrush shrubs which may serve as travel corridors for the Greater Sage-grouse (*Centrocercus erophasianus*) (Connelly et al. 2016). A few soil dwelling lichens provide food for pronghorn (*Antilocapra americana*) (Sharnoff and Rosentreter 1998; Yellowstone Science 2007).

Biological soil crusts serve as indicators of rangeland health (Belnap et al. 2001). It is the structure and composition of the crust that provides information that may complement, explain, or indicate something about a site's characteristics and disturbance history that makes them useful for rangeland management and evaluation (Belnap et al. 2001). Their small size and perception of being difficult to identify has discouraged land managers from incorporating biological soil crusts into management methods (Belnap et al. 2001, Belnap and Lange 2001). In an effort to overcome these perceived problems and find a method to capture the functional significance for the entire ground layer in forests and rangelands, the Ground Layer Indicator was developed. The Ground Layer Indicator applies non-destructive sampling to assess functional groups (not species) and estimates their cover, biomass, carbon content, and nitrogen content at both plot and landscape scales (Smith et al. 2015). An adaptation of the original method was developed specifically for lands possessing less than 10% tree cover (rangelands) as a modification to the U.S. Forest Service Forest Inventory and Analysis (FIA) program procedures; this adapted method is the Ground Layer Indicator for Rangelands (GLIR).

In September 2016, an opportunity to conduct the first moss and lichen surveys in Mussellshell County, Montana also provided a chance to implement an exploratory study using the GLIR method. Led by the author of the Ground Layer Indicator method, five GLIR plots were subjectively placed in different grassland community types to assess the ecological functional of the ground layer. This report documents the exploratory study, its methods and results, and discusses the GLIR approach relative to rangeland management.

Functional groups are composed of species that share the same primary ecosystem function(s) and growth form(s). On the Milton Ranch, 12 functional groups were identified (**Table E-1**). All functional groups found consisted of lichens, mosses, and/or cyanobacteria. Additional functional groups are expected to occur in Montana, particularly in less arid rangelands.

Table E-1. ***The 12 functional groups and their ecological functions as found in the ground layer on the Milton Ranch.***

Functional Group Code	Primary Function(s) of Group
CBIND	Micro-lichens that bind moss and detritus and contribute to soil organic matter. examples: *Diploschistes muscorum*, *Bilimbia lobulata*, *Lepraria vouasuxii*
CCYANO	Cyanobacteria that are free-living, filamentous, fix atmospheric nitrogen, and bind soil particles. This group also includes free-living algae which can form a crust by binding soil particles. examples: *Microcoleus*, *Scytonema*, *Nostoc flagelliforme*
CN	Micro-lichens that fix atmospheric nitrogen because they contain cyanobacteria (also referred to as cyanolichens). examples: *Collema tenax*, *Enchylium coccophorum*, *Placynthium nigrum*
CO	Micro-lichens that are orange colored, whether growing on rock or soil. Some orange-colored micro-lichens may indicate nutrient (over-) enrichment of nitrogen dioxide or sulphur dioxide. examples: *Caloplaca* , *Xanthoria*
CROCK	Micro-lichens that colonize rock, aiding in soil formation and rock weathering, are not orange, and do not fix nitrogen. examples: *Acarospora*, *Candelariella*, *Lecanora*
CSOIL	Micro-lichens that grow into the soil and anchor soil particles, limiting soil erosion, and do not fix nitrogen. examples: *Aspicilia reptans*, *Placidium*, *Psora*
LLFOL	Macro-lichens that exhibit a foliose growth form. They provide invertebrate habitat, forage for pronghorn, and cover bare soil. These lichens grow horizontally on the ground. examples: *Physcia*, *Physconia*, *Phaeophyscia*, *Xanthoparmelia*
LLFRU	Macro-lichens that exhibit a fruticose growth form. They provide invertebrate habitat and forage for caribou. These lichens grow vertical from the ground surface. examples: *Circinaria hispida*, *Cladonia*
FM	Feather mosses occur on soil and intercept rainfall and may cool soil. examples: *Brachytheciastrum*, *Hypnum*, *Pylaisia*
MT	Tall, compact mosses occur on soil and accrue soil, and colonize bare soil. examples: *Ceratodon purpureus*, *Grimmia*, *Pterygoneurrum*, *Tortula*
MTL	Shorter, somewhat sprawling mosses occur on soil, intercept precipitation, and cool soil temperatures. example: *Syntrichia*
NOS	Cyanobacteria that are free-living, large lobed (foliose), fix atmospheric nitrogen, and colonize disturb soil. example: *Nostoc commune*

The five plots were named after the pasture/paddock in which they occurred and were located in different habitats as defined by Phillips (2010): *Grasslands of Introduced Grasses*, *Yucca Shrubland*, *Native Grasslands on Silty Soils*, *Sagebrush Shrubland*, and *Native Grassland.* Collectively the five plots averaged 250 ± 377 kg/ha of ground layer biomass on the Milton Ranch. The least ground layer biomass (10 kg/ha) was found in the West Lackey Paddock 5 pasture occupied by the *Grasslands of Introduced Grasses* plant community. In comparison, the other four plots occurred in native plant communities and exhibited 17 to 40 times (178 to 405 kg/ha) more ground layer biomass, though its statistical significance was not tested given the low sample size. This study found that two important components of Greater Sage-grouse habitat, healthy big sagebrush and biological soil crust, were found in the historic lek that occurs in the South Griffith Paddock 4 occupied by the *Sagebrush Shrubland* plant community.

The GLIR method directly estimates carbon sequestration, in that living and dead organisms in the ground layer store carbon in their tissues (biomass). As expected, carbon content increased in proportion to ground layer biomass. The West Lackey Paddock 5 plot within the *Grassland of Introduced Grasses* plant community had the least amount of carbon storage, less than 5 kg/ha, within the ground layer. The South Griffith 4 plot within a *Sagebrush Shrubland* plant community showed the highest carbon content of 180 kg/ha in the ground layer.

The GLIR method estimates nitrogen content which primarily came from three functional groups (CCYANO, CN, and NOS). The pattern in nitrogen content found at the five plots mirrored the pattern for biomass. The West Lackey Paddock 5 plot within the *Grassland of Introduced Grasses* plant community had very little nitrogen content, at less than 1 kg/ha, within the ground layer. The South Griffith 4 plot within a *Sagebrush Shrubland* plant community showed the highest nitrogen content of 4 kg/ha in the ground layer.

All 11 functional groups were present in the North Big Wall 4 plot found in *Native Grassland* and the South Big Wall 3 plot found in the *Yucca Shrubland*. The remaining South Griffith 2 and 4 plots found in *Native Grasslands on Silty Soils* and *Sagebrush Shrubland*, respectively, had 10 functional groups with only the 'micro-lichens on rock' (CROCK) functional group missing. Despite the relatively low biomass of ground layer organisms found in the *Grasslands of Introduced Grasses* (West Lackey 5) plot, nine functional groups were present. The report further discussions plausible reasons for the differences observed in functional groups among the five plots.

The biomass by functional group was averaged across the five plots. The 'micro-lichens that grow on rock' had the least biomass (0.1 kg/ha) while 'fruticose macro-lichens' had the most biomass (103 kg/ha). The report summarizes findings for each functional group.

To put the five GLIR plots into a grazing management context, livestock data from 2012 to 2015 was obtained for each pasture/paddock. Livestock grazing ranged from 1 to 20 days per paddock and the timing of grazing for any particular paddock varied within and between years. Ground layer biomass was abundant on each plot except for the West lackey 5 plot, and possible explanations are discussed in the report. The process for developing baseline conditions using grazing data and permanent GLIR plots to monitor changes is discussed.

Key recommendations include:

- The GLIR method was found to be appropriate for use on Montana rangelands to collect biomass, carbon content, and nitrogen content data on ground layer organisms. Collecting this data by functional group (not species) simplifies the information while maintaining its applicability to long-term monitoring of rangeland health and condition.

- As anticipated from the limited time and resources available, the number of GLIR plots was insufficient and their subjective placement prevented a statistical analysis and limited any interpretation of the data. The original method does require a study design that aligns data collection with management goals, a sufficient number of plots to characterize an area, and a stratified random sampling design to ensure sound statistical analysis and interpretation.

- It is recommended that the GLIR method be implemented in a pilot study that encompasses a large ranch or federal/state land management parcels. Examples include the Milton Ranch or as a supplemental indicator for the BLM's Assessment, Inventory, and Monitoring (AIM) project in Montana and other western states. The pilot study should establish permanent plots to develop baseline conditions and to assess changes with time or with management actions.

- The GLIR requires that crew performing the method be trained by a certified teacher. Annual training of BLM, USFS, and NRCS staff would develop a crew proficient in the method. Depending upon the level of experience, field crews can be trained in the methodology in 2-5 days. Trained field crews would become skilled in identifying the basic types of organisms that make up the ground layer (such as green algal lichens, cyanolichens, short and tall mosses, free-living cyanobacteria, mosses, and liverworts) and in varying habitats where they occur. However, field crews would not need to know species identification for these organisms. For previously trained crew members, a 1-day annual refresher training would likely be required to maintain the necessary skill set to ensure data quality.

Acknowledgements

The opportunity to organize the first moss and lichen surveys for Mussellshell County and to examine the use of the Ground Layer Indicator for Rangelands in Montana was a collaborative effort, and there are many people and their affiliations to recognize.

This project was made possible by a cooperative agreement between the Bureau of Land Management and The University of Montana – Montana Natural Heritage Program. I am very appreciative to Wendy Velman, Botany Program Lead for the Montana / Dakotas State BLM Office, for supporting this effort to bring biological soil crusts into the conversation of rangeland management. I am very grateful to Bill and Dana Milton for instigating collaboration, inspiring scientific study, and for their stewardship that seeks to develop a healthy relationship between land and cattle. I also thank the Milton Family for hosting the team of 10 lichenologists, bryologists, and botanists, in not only providing housing, but also the space for us to use our microscopes and books to identify the many specimens.

I am very grateful to Northwest Lichenologists for being able to support forays and provide expertise! Special appreciation goes to Dr. Robert Smith who developed the Ground Layer Indicator, led the task of implementing it on the Milton Ranch, conducted the data analysis, and engaged us in conversations. Thanks to Dr. Daphne Stone, Dr. Roger Rosentreter, Dr. Bruce McCune, Dr. Katherine Glew, Ann DeBolt, and Wendy Velman for conducting field work, and to Wildfire Wanderning for assisting with data and logistics. Many thanks goes to Dr. Joe Elliott for verifying the many moss specimens.

I am very appreciative to several folks who took the time to review and critique earlier versions of this report: Dr. Robert Smith (Postdoctoral Research Ecologist-Oregon State University), Dr. Roger Rosentreter (retired State Botanist for the Idaho BLM and Lichenologist), Dr. Jayne Belnap (USGS Scientist/Lichenologist), Wendy Velman, and Bill Milton.

TABLE OF CONTENTS

APPENDICES

FIGURES

FIGURES

TABLES

An Exploratory Study Using the Ground Layer Indicator Method in Montana Rangelands

1.0 INTRODUCTION

Biological soil crusts are a natural component of many landscapes across North America (Belnap and Lange 2001, Smith et al. 2015, Weber et al. 2016). They can be composed of lichens, mosses, liverworts, hornworts, free-living algae, free-living cyanobacteria, bacteria, and/or micro-fungi. This network of vastly different organisms forms a surface layer that lives on or within soil particles.

In rangelands, this layer can be viewed from functional, structural, and compositional perspectives (Belnap et al. 2001). The biological soil crust layer functions as living mulch, retains soil moisture, discourages annual weed growth, reduces soil erosion caused by wind or water, fixes atmospheric nitrogen, and contributes to soil organic matter. Structurally, moss rhizoids, lichen rhizines, fungal hyphae, and cyanobacteria filaments weave together and bind soil particles. In arid regions they occupy the nutrient-poor zones between individual plants. Compositionally they are composed of many species and contribute significantly to biological diversity in any landscape.

In the western U.S., rangeland managers monitor the ecological trend and health of vegetation using indicator plants (USDA 1937; Stoddart et al. 1943). Biological soil crusts can also serve as indicators of rangeland health. In comparison to vascular plants, biological soil crusts are less influenced by short-term climatic conditions, making them good indicators of long-term environmental factors. It is the structure and composition of the crust that provides information that may complement, explain, or indicate something about a site's characteristics and disturbance history that makes them useful for rangeland management and evaluation (Belnap et al. 2001).

Their small size and perception of being difficult to identify has discouraged land managers from incorporating biological soil crusts into management methods (Belnap et al. 2001, Belnap and Lange 2001). In an effort to overcome these perceived problems and find a method to capture the functional significance for the entire ground layer in forests and rangelands, the Ground Layer Indicator was developed. The Ground Layer Indicator applies non-destructive sampling to biological soil crusts and assesses functional groups (not species) to estimate cover, biomass, carbon content, and nitrogen content at both plot and landscape scales (Smith et al. 2015). This method also broadens the scope to include the entire non-vascular layer that covers the ground, including organisms that dwell on soil (biological soil crusts), wood, rock, and dead organic material. An adaptation of the original method was developed specifically for lands possessing less than 10% tree cover (rangelands) as a modification to the U.S. Forest Service Forest Inventory and Analysis (FIA) program procedures; this adapted method is the Ground Layer Indicator for Rangelands (GLIR).

In September 2016, an opportunity to conduct the first moss and lichen surveys in Mussellshell County, Montana also provided a chance to implement an exploratory study using the GLIR method. Since the late 1800s numerous moss and lichen surveyors have navigated around Montana, yet for Musselshell County no floristic surveys and only a few collections have ever been documented (Elliott 1993, McCune et al. 2014, MTNHP 2015). Members from Northwest Lichenologists convened at a private livestock ranch to survey rolling, dissected grassland, open ponderosa pine forest, and rock outcrops for non-vascular species and to collect ecological data from GLIR plots (**Figure 1**). Led by the author of the Ground Layer Indicator method, five GLIR plots were placed in different grassland community types to assess the ecological functional of the ground layer. This report documents the exploratory study, its methods and results, and discusses the GLIR approach relative to rangeland management.

2.0 METHODS

From September 13-16, 2016, the Ground Layer Indicator for Rangelands pilot study was implemented along with surveys to document the mosses, lichens, liverworts, and cyanobacteria on the Milton Ranch. The Milton Ranch is about 10 miles northeast of Roundup, Montana in Mussellshell County (**Figure 1**). Species occurrence data was collected to document locations, abundance, habitat, and other details and is housed in the botany database at the Montana Natural Heritage Program (MTNHP) in Helena, Montana and is available on MTNHP website (http://mtnhp.org). Voucher specimens are deposited at the University of Montana Herbarium in Missoula, Montana.

The exploratory study subjectively located GLIR plots at five sites that were near to areas surveyed for non-vascular species, existing Milton Ranch vegetation transects, and/or 2010 plant community plots. The five GLIR plots were named after the pasture and paddock in which they occurred: North Big Wall Pasture, Paddock 4 (N Big Wall #4), South Big Wall Pasture, Paddock 3 (S Big Wall #3), South Griffith Pasture, Paddock 2 (S Griffith #2), South Griffith Pasture, Paddock 4 (S Griffith #4), and West Lackey Pasture, Paddock 5 (W Lackey #5) (**Figure 1**).

2.1 Plot Layout

A modified plot layout of the original Ground Layer Indicator method was set-up at the five sites (**Figure 2**). From a randomly located central point, three transect tapes were stretched for 120 feet (36.58 meters (m)) in the north (360 degrees), southeast (120 degrees), and southwest (240 degrees) directions and anchored on both sides with a chaining pin. Each tape was kept as straight, taut, and low to the ground as possible. The cover and depth of each biological soil crust functional group was measured within a 20x50-centimeter (cm) microquad frame. On each transect the first microquad was placed at 9.8 feet from center and thereafter at 9.8-foot (3 m) intervals. A total of 32 microquads were observed, such that the north, southeast, and southwest transects contained 10, 11, and 11 microquads respectively. To avoid damage to the biological soil crust layer, walking occurred along the west side and microquads were placed on the east side of the transect tape.

Figure 1. ***Approximate locations of the Ground Layer Indicator for Rangelands plots (★) on the Milton Ranch (bordered by thick black line).***

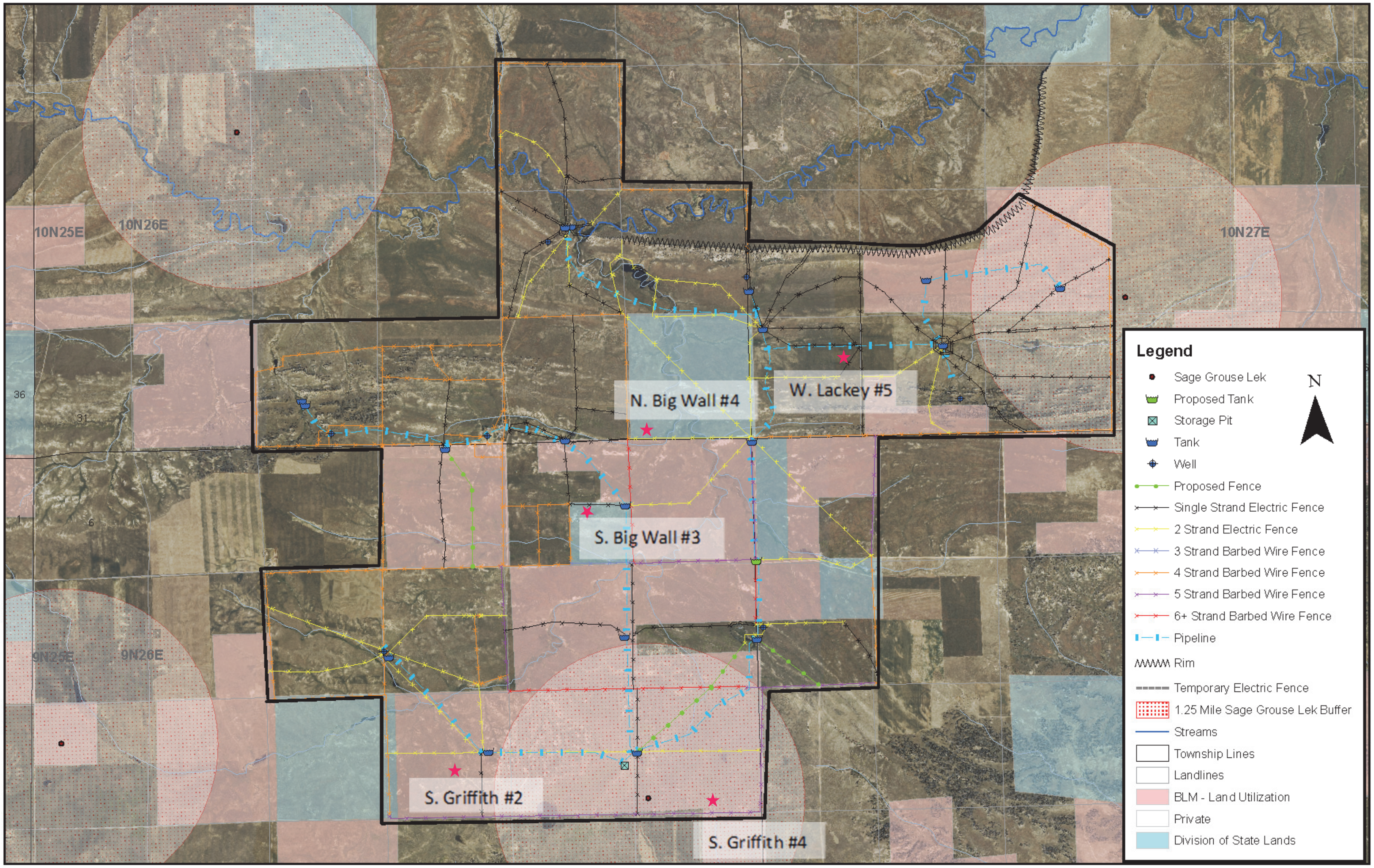
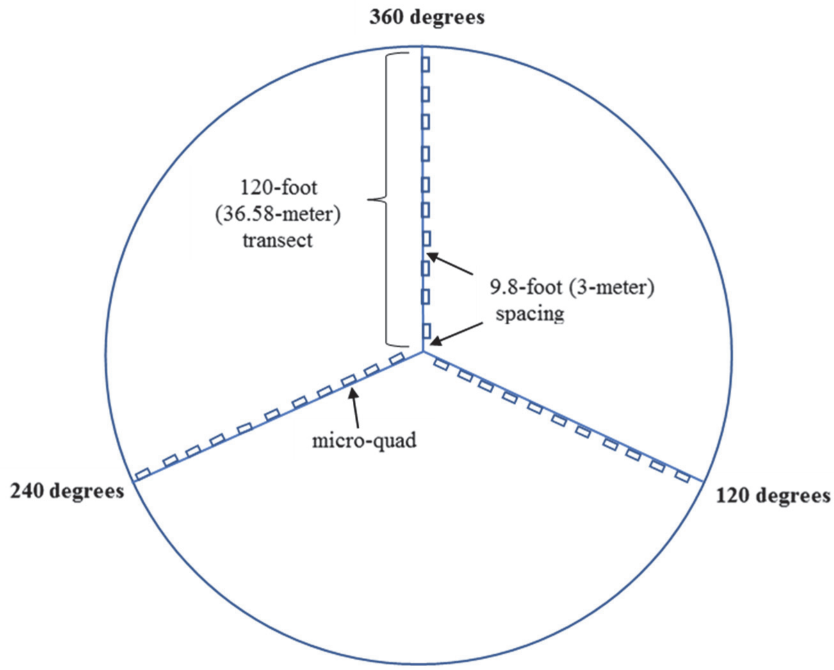


Figure 2. ***Plot layout for the Ground Layer Indicator for Rangelands method.***



2.2 Data Collection

Plot and microquad data was recorded onto a data form (**Figure 1** in **Appendix A**). The microquad frame was placed with the long side parallel to the transect. It lays flat to the ground surface, but may encompass internal terrain (bunchgrass tufts, hummock-hollow formations, or other small features).

In each microquad the depth and cover of each functional group and a trampling code was recorded (**Table 1**; **Figure 1** in **Appendix A**). Depth was measured using a metal chaining pin that has been etched at each Depth Class increment (with doubling increments defined at 0.125, 0.25, 0.50, 1, 2, 4, 8 and 16 inches). The *Depth Class* measurement includes all living and dead material for which identifiable moss, liverwort, cyanobacteria, or lichen structures are visually distinguishable. Unrecognizable, decomposed plant matter, peat, organic soil, or mineral soil that may form in deeper layers was not measured. The *Cover Class* was measured by looking directly over (vertical) the microquad and recording it as a percentage within categories. Functional groups may vertically overlap, making it possible for total cover to exceed 100%. An exhaustive search for every tiny sprig was not required. When mats of a functional group make up more than 50% cover in the microquad, the middle (median) value from five test measurements was recorded. For very thin biological soil crust or other mosses/lichens that are present only as thin crusts or films, the depth class was recorded as a trace. Ultimately, the goal is to accurately estimate the volume and density of ground layers, for later calculation of biomass and nutrient content.

Table 1. ***Cover and density class values and definitions using the Ground Layer Indicator Method.***

Cover Code	Percent Cover Class	Cover Description
0	absent	
T	>0 – 0.1%	trace (T) amount
1	>0.1 – 1%	size of two postage stamps
2	>1 – 2%	half-size of a standard business card
5	>2 – 5%	size of a business card
10	>5 – 10%	size of a US dollar bill
25	>10 – 25%	
50	>25 – 50%	
75	>50 – 75%	
95	>75 – 95%	
99	> 95%	Virtually complete cover
Depth Code	**Depth Class**	**Depth Description**
0	absent	
T	0 – 1/8 inch	trace (T): often used for a thin biological soil crust.
Q	>1/8 – 1/4 inch	quarter (Q) of an inch
H	>1/4 – 1/2 inch	half (H) of an inch
1	>1/2 – 1 inch	
2	>1 – 2 inches	
4	>2 – 4 inches	
8	>4 – 8 inches	
16	>8 – 16 inches	

2.3 Functional Groups

In biological soil crusts, a functional group is defined by organisms that share the same primary ecosystem function(s) and growth form(s); it avoids the need to identify species. In reality the ecological roles of each functional group are not mutually exclusive. For example, all functional groups intercept precipitation and lessen the erosive forces of rainfall. The value to defining functional groups is that species are lumped into a group that emphasize a primary, dominant function. A functional group usually is made up of multiple species. At the same time, each species of moss, lichen, liverwort, or cyanobacteria will belong to a single functional group.

On the Milton Ranch, 12 functional groups[1] could easily be differentiated within the entire ground layer (**Table 2**). Mosses, lichens, and cyanobacteria/algae belong to different functional groups based on their unique biology. For **mosses**, functional groups were further divided based on their growth form. For **lichens**, their morphology divided them into: **macro-lichens** that are relatively large species that grow on the top of bark, rock, or soil and can be removed and examined without damaging them, and **micro-lichens** that are relatively small species that grow by etching themselves into wood, rock, or soil and cannot be removed without crumbling their bodies or removing their substrate. Their functional groups were sub-divided based on the primary ecological function they provided. Macro-lichens are also called **foliose** or **fruticose lichens** based on either a “leafy” or “shrubby” growth form, respectively. Micro-lichens are called **crustose** or **squamulose lichens** based on their “crusty” growth form. For **cyanobacteria**, their morphology sub-divided them into two functional groups based on their growth form of “filamentous” or “leafy”.

[1] For other ground layer functional groups that may occur in Montana consult Smith et al. 2015.

Table 2. ***The 12 functional groups and their ecological functions as found in the ground layer on the Milton Ranch.***

Functional Group Code	Primary Function(s) of Group and Examples[1]	Present in GLIR Plots?
CBIND	Micro-lichens that bind moss and detritus and contribute to soil organic matter. examples: *Diploschistes muscorum*, *Bilimbia lobulata*, *Lepraria vouasuxii*	yes
CCYANO[2]	Cyanobacteria that are free-living, filamentous, fix atmospheric nitrogen, and bind soil particles. This group also includes free-living algae which can form a crust by binding soil particles. examples: *Microcoleus*, *Scytonema*, *Nostoc flagelliforme*	yes[2]
CN	Micro-lichens that fix atmospheric nitrogen because they contain cyanobacteria (also referred to as cyanolichens). examples: *Collema tenax*, *Enchylium coccophorum*, *Placynthium nigrum*	yes
CO	Micro-lichens that are orange colored, whether growing on rock or soil. Some orange-colored micro-lichens may indicate nutrient (over-) enrichment of nitrogen dioxide or sulphur dioxide. examples: *Caloplaca* , *Xanthoria*	yes
CROCK	Micro-lichens that colonize rock, aiding in soil formation and rock weathering, are not orange, and do not fix nitrogen. examples: *Acarospora*, *Candelariella*, *Lecanora*	yes
CSOIL	Micro-lichens that grow into the soil and anchor soil particles, limiting soil erosion, and do not fix nitrogen. examples: *Aspicilia reptans*, *Placidium*, *Psora*	yes
LLFOL	Macro-lichens that exhibit a foliose growth form. They provide invertebrate habitat, forage for pronghorn, and cover bare soil. These lichens grow horizontally on the ground. examples: *Physcia*, *Physconia*, *Phaeophyscia*, *Xanthoparmelia*	yes
LLFRU	Macro-lichens that exhibit a fruticose growth form. They provide invertebrate habitat and forage for caribou. These lichens grow vertical from the ground surface. examples: *Circinaria hispida*, *Cladonia*	yes
FM	Feather mosses occur on soil and intercept rainfall and may cool soil. examples: *Brachytheciastrum*, *Hypnum*, *Pylaisia*	no
MT	Tall, compact mosses occur on soil and accrue soil, and colonize bare soil. examples: *Ceratodon purpureus*, *Grimmia*, *Pterygoneurrum*, *Tortula*	yes
MTL	Shorter, somewhat sprawling mosses occur on soil, intercept precipitation, and cool soil temperatures. example: *Syntrichia*	yes
NOS[2]	Cyanobacteria that are free-living, large lobed (foliose), fix atmospheric nitrogen, and colonize disturb soil. example: *Nostoc commune*	yes[2]

[1] **Appendix B** provides a preliminary list of the species found on the Milton Ranch and their assigned functional group.
[2] Microquad data was collected separately for CCYANO and NOS functional groups but combined in the analyses.

2.4 Calculation of Biomass and Nutrient Content

Based on allometric equations, we nondestructively calculated biomass and nutrient content at the level of each functional group per microquad. First, bulk density was estimated as a nonlinear function of field-measured depth, based on a calibration curve from previous destructive sampling (Smith et al. 2015); this takes into account the fact that shallow biotic soil crusts tend to be quite compact and dense, while deeper mats tend to be looser and fluffier. Second, volume, a three-dimensional measure, was calculated as the product of depth and area as non-destructively measured in each microquad. Third, biomass was calculated as the product of bulk density and volume. Nutrient contents (carbon and nitrogen) were then determined for each functional group as a proportion of biomass following the nutrient analyses conducted by Smith et al. (2015). Microquad values were aggregated to determine plot-level totals and functional-group means.

3.0 RESULTS AND DISCUSSION

3.1 Site Assessment

On the Milton Ranch in September 2016, the ground layer consistently contained many lichens, mosses, and cyanobacteria that served a multitude of ecosystem functions. Macro-fungi were occasionally observed, but in too low of an abundance to record, and liverworts were not found. At each GLIR plot, the biomass, aerial cover, volume, depth, carbon and nitrogen content, and number of functional groups was calculated (**Table 3**).

3.2 Ground Layer Biomass

Collectively (all 5 plots together), an average of 250 ± 377 kg/ha of ground layer biomass was found on the Milton Ranch (**Table 3**). The standard deviation of 377 being much larger than the average of 250 kg/ha indicates that five plots were likely insufficient to adequately sample the range of variation on the Milton Ranch, given that biomass varies substantially from plot to plot. Our field observations found that ground layer organisms were prevalent on the landscape, but were actually patchy in coverage. Over short distances coverage changed from being nearly absent to present, and when present biomass fluctuated from a little to a lot. Each plot represented a different grassland or shrubland community type and soil types which also affect total biomass. Given the plot to plot variation in biomass, plant communities, and soil types, we believe that more plots are required to accurately characterize the ground layer on the Milton Ranch for the purpose of guiding management.

In comparison, another study using the Ground Layer Indicator method found that shrub-steppe and dry ponderosa pine forests of Oregon can yield a biomass of 318 ± 116 kg/ha (6 plots) and 603 ± 305 kg/ha (10 plots) respectively (Smith et al. 2015). Collectively, the five Montana plots averaged more lichen (162 ± 265 kg/ha) than moss (87 ± 213 kg/ha) biomass (**Table 3**), while in the shrub-steppe and dry ponderosa pine forests of Oregon, mosses (278 ± 115 kg/ha and 537 ± 275 kg/ha, respectively) was more abundant than lichens (66 ± 30 kg/ha and 39 ± 6 kg/ha, respectively). In this exploratory study, broad inferences cannot be made because the sampling design was insufficient to characterize the ground layer. However, future studies using a statistically designed sampling scheme of GLIR plots will be able to quantify the ground layer at

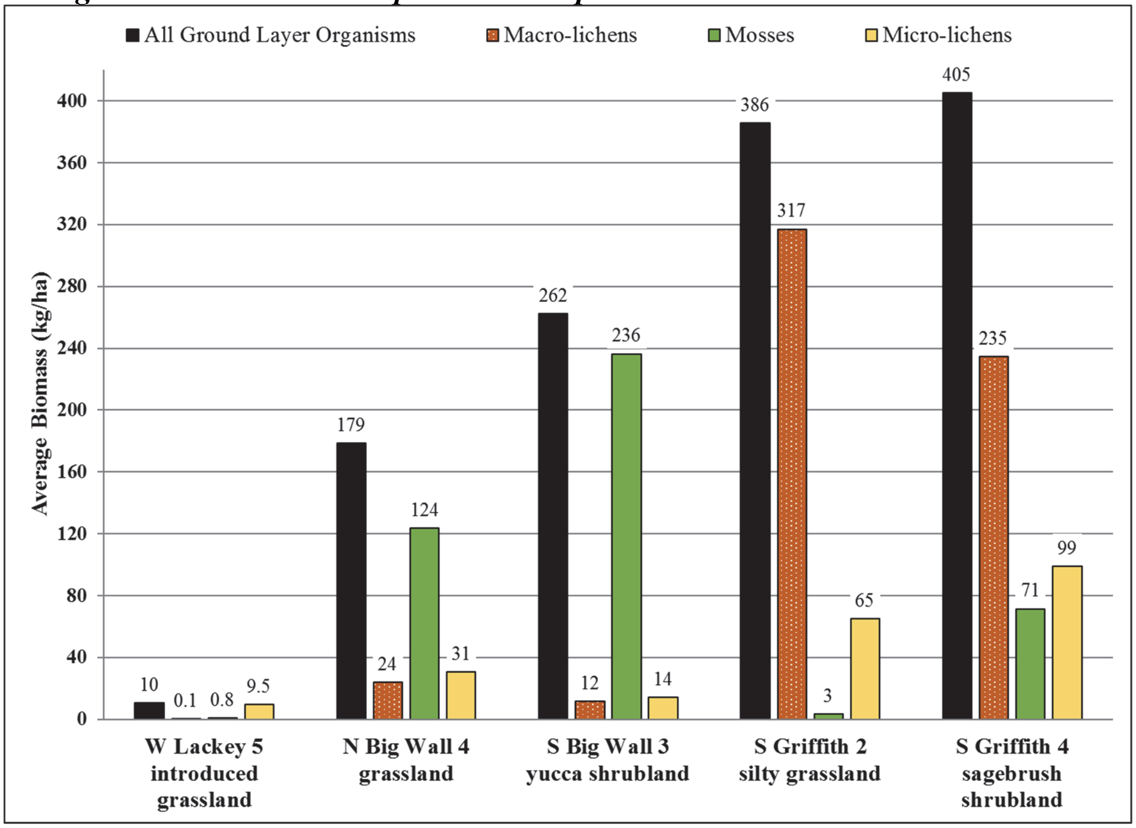
Table 3. ***Statistics from the 32 microquads at each of five plots on the Milton Ranch, Roundup, Montana. Values are averages plus or minus 1 standard deviation, unless indicated differently.***

Metric	**North Big Wall, Pad 4** *grassland*	**South Big Wall, Pad 3** *yucca shrubland*	**South Griffith, Pad 2** *silty grassland*	**South Griffith, Pad 4** *sagebrush shrubland*	**West Lackey, Pad 5** *introduced grassland*	**Average from 5 Plots**
Ground Layer Dry Biomass (kilogram per hectare (kg/ha))	179 ± 367	262 ± 503	386 ± 400	405 ± 596	10 ± 18	249 ± 377
Macro-lichen Dry Biomass (kg/ha)	24 ± 89	12 ± 25	317 ± 348	235 ± 542	0.1 ± 0.7	118 ± 201
Micro-lichen Dry Biomass (kg/ha)	31 ± 45	15 ± 30	65 ± 111	99 ± 117	10 ± 18	44 ± 64
Moss Dry Biomass (kg/ha)	124 ± 332	237 ± 507	3 ± 5	72 ± 216	0.9 ± 2	87 ± 213
Carbon (C) content (kg/ha)	79 ± 163	116 ± 223	171 ± 177	180 ± 264	5 ± 8	110 ± 167
Nitrogen (N) Content (kg/ha)	2 ± 4	3 ± 5	4 ± 4	4 ± 5	0.2 ± 0.3	3 ± 4
Ground layer Volume (cubic meter per hectare)	3 ± 6	5 ± 9	6 ± 7	7 ± 10	0.2 ± 0.3	4 ± 7
Ground Layer Percent Cover	5 ± 9%	7 ± 10%	11 ± 12%	12 ± 16%	1 ± 1%	7 ± 9%
Average Depth of Functional Groups (cm)	0.16	0.14	0.20	0.16	0.07	0.14
Number of Functional Groups (richness). In this study 12 was the maximum number possible.	11	11	10	10	9	10

sites and compare results between sites and across regions to help detect distributional differences. Such a sampling design might resemble the U.S. Forest Service's FIA program, which assigns one random plot location per cell of a pre-defined grid, or it could also be stratified by land ownership or forest/non-forest status.

On the Milton Ranch, the average ground layer biomass in the GLIR plots ranged from 10 to 405 kilograms per hectare (kg/ha) (**Figure 3**). The least ground layer biomass (10 kg/ha) was found in the West Lackey Paddock 5 pasture occupied by the *Grasslands of Introduced Grasses* plant community (Phillips 2010). In comparison, the other four plots occurred in native plant communities and exhibited 17 to 40 times (178 to 405 kg/ha) more ground layer biomass, though its statistical significance was not tested given the low sample size (**Figure 3**).

Figure 3. ***Biomass of all ground layer organisms, moss, macro-lichen, and micro-lichen averaged across the 32 microquads at each plot.***



- The West Lackey Paddock 5 pasture occurs in the *Grasslands of Introduced Grasses* plant community (Phillips 2010). This plot was unique because most of the Milton Ranch is occupied by native grass, shrub, or tree plant communities. The *Grasslands of Introduced Grasses* plant community represents about 8% of the Milton Ranch and occurs where lands were broken by the plow more than 20 years ago, planted with crops, and later re-planted with introduced perennials, mostly crested wheatgrass and alfalfa (Phillips 2010). This plant community is dominated by crested wheatgrass (*Agropyron cristatum*; 35% average canopy cover) and occurs on sandy, silty, and clayey soils (Phillips 2010). While crested wheatgrass remains the dominant plant cover, Wayne Phillips' assessment in 2010 showed there were strong indications that plant succession is advancing toward a native grassland type (Phillips 2010). Observations not only found a diversity of native grasses, forbs, and shrubs growing in with the crested wheatgrass, but also an increase in species diversity and abundance (Phillips pers. comm.). In addition to the introduced perennials, this paddock contains a livestock water tank, which is about 222 meters from the GLIR plot center. At about 112 meters from the GLIR plot center in the northeast direction, livestock trails are evident on the ground surface. Despite the paddock's history of habitat conversion and the plot's closeness to the water tank, the plot contained the basic ground layer organisms of macro-lichens, micro-lichens, cyanobacteria, and mosses (**Figure 3**).

- The North Big Wall Paddock 4 occurs in a *Native Grassland* plant community either on silty, sandy, or shallow soil. Unfortunately, the exact plant community type was not determined at the time of sampling. This plot had 179 kg/ha of ground layer biomass.

- The South Big Wall Paddock 3 occurs in the *Yucca Shrubland* plant community, which is found on particular sandy and shallow soils (Phillips 2010). This type is also unique as it represents only 5% of the Milton Ranch. This community type is dominated by yucca (*Yucca glauca*; 20% average canopy cover) and a diversity of grasses (about 60% canopy cover) and forbs (wildflowers) (Phillips 2010).

- The South Griffith Paddock 2 occurs in the *Native Grasslands on Silty Soils* plant community, which represents about 35% of the Milton Ranch (Phillips 2010). This community is dominated by needle-and-thread grass (*Stipa comata*), western wheatgrass (*Elymus smithii*), and thickspike wheatgrass (*Elymus lanceolatus*) (40% average canopy cover), and depending upon the location can be co-dominated by thread-leaved sedge (*Carex filifolia*), blue grama (*Bouteloua gracilis*) grass, and/or Japanese brome (*Bromus japonicus*) (Phillips 2010). Forbs are present, but with lower canopy covers (Phillips 2010).

- The South Griffith Paddock 4 occurs in the *Sagebrush Shrubland* plant community. The *Sagebrush Shrubland* plant community is found on silty, clayey, or clay pan soils and represents about 15% of the Milton Ranch (Phillips 2010). This community is dominated by big sagebrush (*Artemisia tridentata*; 35% average canopy cover), western and/or thickspike wheatgrasses (10-20% average canopy cover), and lesser amounts of a large variety of forbs and other grasses. In addition, this plot occurs within an historic lek that has not been used by Greater Sage-grouse (*Centrocercus erophasianus*) birds since the 1970s (Crowe personal communication; **Figure 1**). Leks are areas where male birds display competitive courtship behavior to attract female birds. The reasons for their apparent inactivity are not known (Crowe personal communication). In Montana preferred habitat for leks often are clearings surrounded by sagebrush of 20-50% canopy cover (MTNHP 2018). Nesting habitat also requires at least 20% sagebrush cover while brood habitat is in more open stands of sagebrush mixed with native bunchgrasses and perennial forbs (MTNHP 2018, Connelly et al. 2016). Biological soil crusts are an important component of Greater Sage-grouse habitat (Connelly et al. 2016). For reasons already mentioned biological soil crusts reduce soil erosion and discourage annual grass germination allowing areas between sagebrush to remain open for birds to travel (Connelly et al. 2016; Rosentreter pers. comm.). Of the five GLIR plots, this plot showed the highest biomass (and standard deviation) of ground layer organisms and exhibited 10 of the 11 functional groups (**Table 3**). Observations found big sagebrush to be prevalent and healthy. While this study shows important components of Greater Sage-grouse habitat are present, additional studies of the vascular and non-vascular components are needed to assess the quality of these areas for lek, nesting, or brooding use.

The North Big Wall Paddock 4 and South Big Wall Paddock 3 plots were dominated by moss functional groups whereas the South Griffith Paddocks 2 and 4 plots were dominated by macro-lichen functional groups (**Figure 3**). The micro-lichen/cyanobacteria functional groups were relatively low in cover, though highest at the South Griffith Paddocks. The differences in biomass and/or functional groups could reflect the plot's geography, presence or absence of disturbance, and/or interactions between living organisms, soil properties, and precipitation. To tease apart the influence that geography, disturbance, soil, and precipitation have on biological soils crusts would require a larger number of plots systematically randomized across regions that exhibit a range of biophysical variation.

3.3 Ground Layer Carbon Content

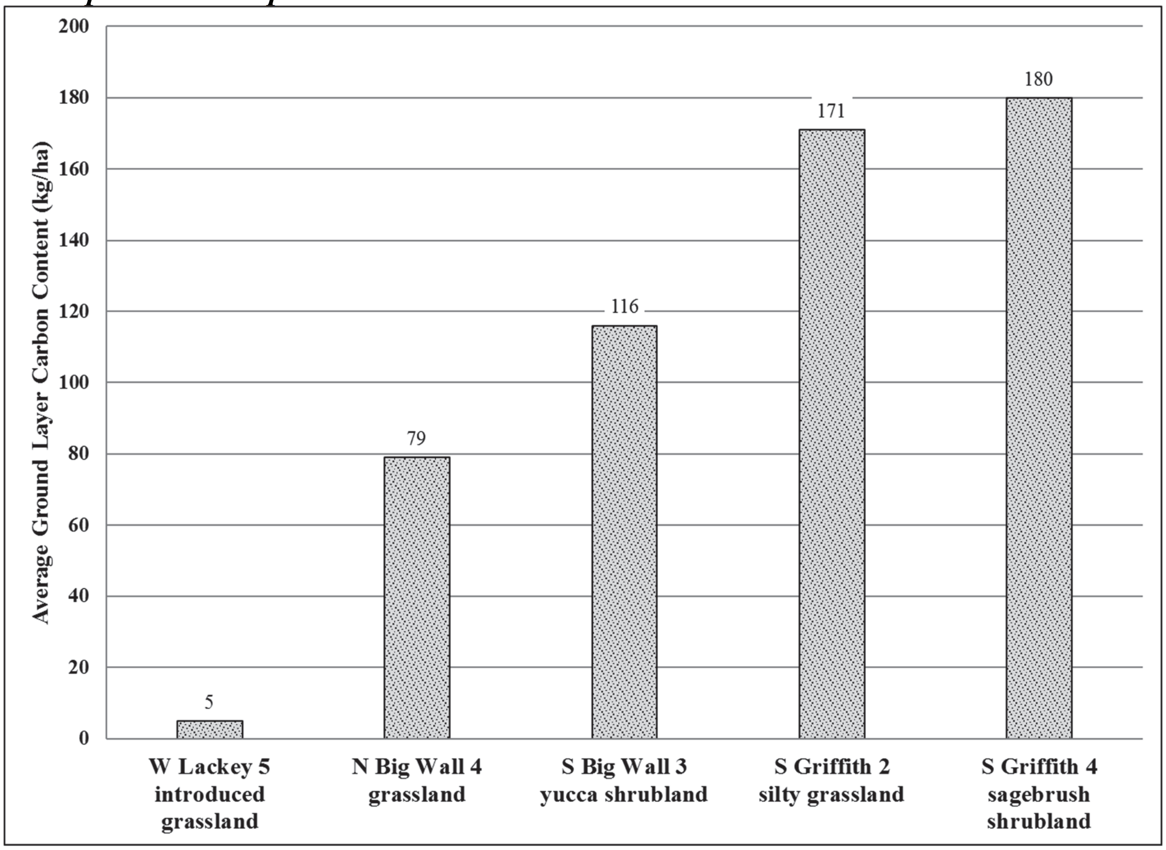
Vascular plants and biological soil crusts contribute to rangeland carbon uptake, storage (sequestration), and release. Decaying thalli, leaves, stems, and flowers/capsules release carbon that improves soil fertility and provides energy sources for soil microbial populations (Belnap et al. 2001). Vascular plants provide organic material directly beneath them, but seldom much in the larger interspaces between plants. In these interspaces, ground layer organisms often provide the primary source of carbon and biologically-available nitrogen where they are present (although nitrogen deposition from agricultural and industrial sources may also contribute). Carbon inputs depend upon the abundance and species composition of the biological crust, and on precipitation, humidity, time of year, temperature, and other factors. For example, carbon inputs are higher when mosses and lichens are present than when crusts are dominated by cyanobacteria (Belnap et al. 2001).

The GLIR method directly measures carbon sequestration, in that living and dead organisms in the ground layer store carbon in their tissues (biomass). The pattern in carbon content provided by the ground layer organisms at the five plots mirrored the pattern for biomass, which is expected since carbon is proportional to dry mass (**Figures 3** and **4**). The West Lackey Paddock 5 plot within the *Grassland of Introduced Grasses* plant community had the least amount of carbon content (storage), less than 5 kg/ha, within the ground layer (**Figure 4**). In comparison, the other two plots found in *Native Grassland* plant communities averaged higher carbon contents in their ground layers, 79 kg/ha and 171 kg/ha respectively (**Figure 4**). The South Big Wall 3 plot within the *Yucca Shrubland* plant community had an intermediate carbon content of 116 kg/ha while the South Griffith 4 plot within a *Sagebrush Shrubland* plant community showed the highest carbon content of 180 kg/ha in the ground layer (**Figure 4**). As expected, carbon content increased in proportion to ground layer biomass.

Ground layer organisms, along with their epiphytic[2] counterparts, are integral players in the global biogeochemical cycles of carbon and nitrogen (Elbert et al. 2012). Through photosynthesis and respiration these organisms contribute to the movement (flux) of carbon that varies among habitats or ecosystems. As mentioned earlier, the rates at which they sequester or release carbon depends upon rainfall, temperature, soil moisture, and soil radiation (Zhao et al. 2016). Relative to soils covered by ground layer organisms, barren soils have a reduced potential for carbon sequestration and soil fertility. Where the surface soil is barren, the carbon cycle is greatly impaired, reducing both options of sequestering carbon and improving soil fertility. In addition, the ability for terrestrial (vascular) plants to sequester carbon dioxide may be constrained by the amount of available fixed-nitrogen (Belnap 2001; Elbert et al. 2012). Therefore, biological soil crusts with nitrogen-fixing organisms may indirectly assist vascular plants in storing carbon (Elbert et al. 2012).

[2] Non-vascular species that grow on trees, shrubs, and other plants.

Figure 4. ***Carbon content provided by ground layer organisms, averaged across 32 microquads at each plot.***



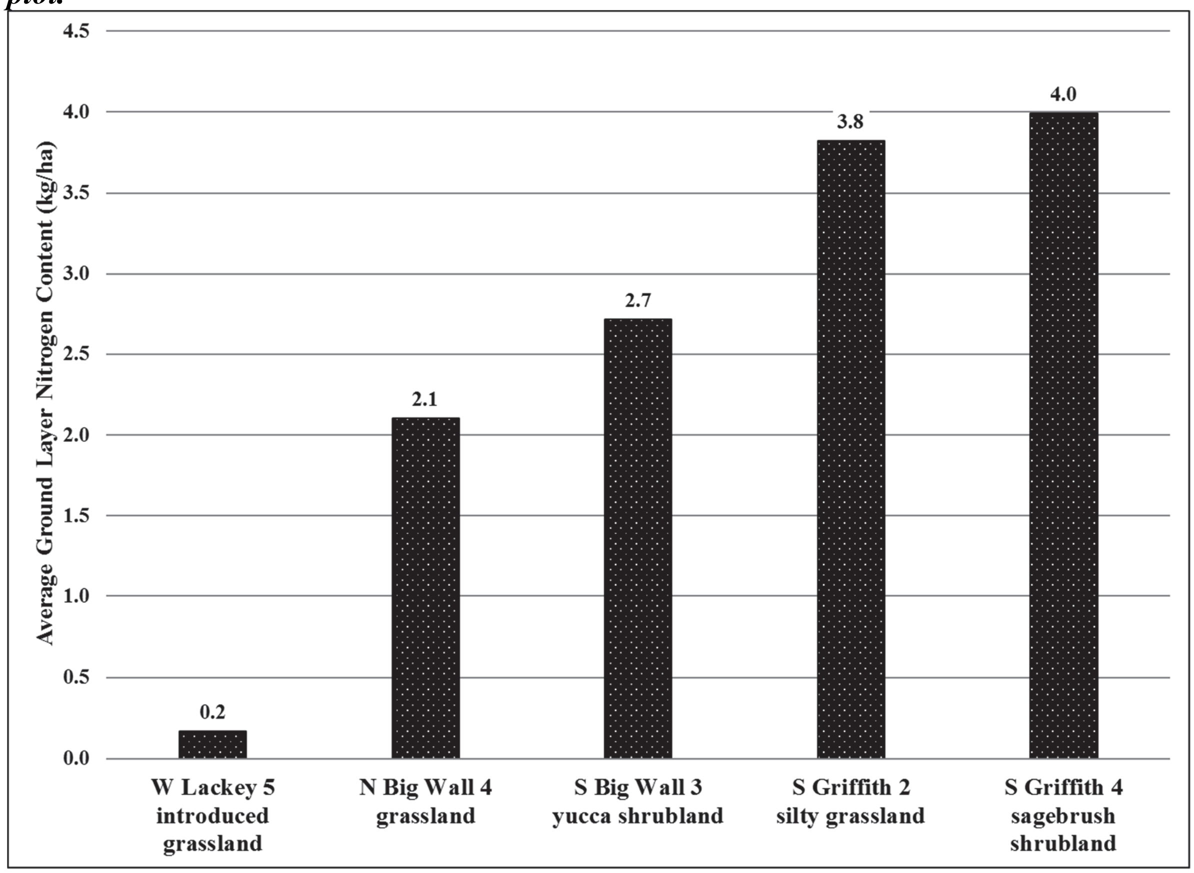
3.4 Ground Layer Nitrogen Content

Our atmosphere is the major global reservoir for nitrogen, making up 78% of our air. However, most living organisms cannot directly use atmospheric dinitrogen (N_2) and instead rely on processes that convert it into biologically useful ammonium or nitrate, which can be viewed as rangeland "fertilizer". Soils are often low in biologically useful nitrogen, and it is often the major limiting factor to plant growth (Freeman and Worth 1999). In arid environments, soil nitrogen concentrations are particularly low.

Cyanobacteria and cyanolichens fix atmospheric nitrogen and can release (leak) excess amounts of it into the soil during rain events. The fixed-nitrogen released to the soil can then be taken up by surrounding vascular plants, fungi, and bacteria (Mayland and MacIntosh 1966; Mayland et al. 1966; Stewart 1967; Jones and Stewart 1969). In general cyanobacteria and cyanolichens become more abundant in arid landscapes. Nitrogen-fixation rates vary with species composition, biomass, time of year, precipitation, and temperature. Biological soil crusts contribute nitrogen to soils directly under vascular plants and to the spaces between plants helping to maintain soil fertility (Harper and Pendleton 1993, Belnap 1994, Belnap 1995, Belnap and Harper 1995).

The pattern in nitrogen content found at the five plots also mirrored the pattern for biomass. Functional groups of free-living cyanobacteria and cyanolichens (lichens that contain cyanobacteria) substantially contribute to nitrogen accumulations (**Figures 3** and **5**). These functional groups are CCYANO, CN, and NOS (**Table 2**). Other functional groups (LLFOL, LLFRU, MTL) accumulate nitrogen and will release it when they decay. At each plot, nitrogen-fixing cyanobacteria and cyanolichens were present. The West Lackey Paddock 5 plot within the *Grassland of Introduced Grasses* plant community had very little nitrogen content, at less than 1 kg/ha, within the ground layer (**Figure 5**). In comparison, the other two plots found in native grassland plant communities averaged higher nitrogen contents in their ground layers, 2.1 kg/ha and 3.8 kg/ha respectively (**Figure 5**). The South Big Wall 3 plot within the Yucca Shrubland plant community had an intermediate nitrogen content of 2.7 kg/ha while the South Griffith 4 plot within a sagebrush shrubland plant community showed the highest carbon content of 4 kg/ha in the ground layer (**Figure 5**). While the nitrogen contents in the ground layer are very small, they are contributing useable nitrogen to the open spaces where either nitrogen-fixing vascular plants, such as species of *Astragalus*, *Lupinus*, *Trifolium*, and *Medicago,* or decaying vascular plant matter are absent.

Figure 5. ***Nitrogen content of ground layer organisms averaged across 32 microquads at each plot.***



3.5 Ground Layer Functional Groups

Although 12 functional groups of ground layer organisms were recognized on the Milton ranch, 11 were found on the five GLIR plots (**Table 4**). Only functional groups were recorded in the microquad plots, but later these functional groups were assigned to the species found during the surveys (**Tables B-1** and **B-2** in **Appendix B**). On the five plots, the number of functional groups varied from 9 to 11, indicating the array of ecological functions present (**Table 3**). In general, healthy habitats should support a large array of the possible ecological functions, though some sites won't be capable of supporting all functions. All 11 functional groups were present in the North Big Wall 4 plot found in *Native Grassland* and the South Big Wall 3 plot found in the *Yucca Shrubland*. The remaining South Griffith 2 and 4 plots found in *Native Grasslands on Silty Soils* and *Sagebrush Shrubland*, respectively, had 10 functional groups with only the 'micro-lichens on rock' (CROCK) functional group missing. This functional group is especially good at weathering rock to form soil, albeit very slowly. Despite the relatively low biomass of ground layer organisms found in the *Grasslands of Introduced Grasses* (West Lackey 5) plot, nine functional groups were present. The missing groups were 'micro-lichens on rock' and 'fruticose macro-lichens' (LLFRU), which seems noteworthy for land that was historically tilled and is currently near to a livestock water source. It is easy to conjecture that land chosen for tilling and planting could lack rock substrates whether caused by particular site characteristics, the act of tilling, current livestock disturbance, happenstance, and/or a complex set of reasons. Research has shown that the 'fruticose macro-lichens' are the most susceptible to mechanical disturbances, and may take longer to re-colonize (Belnap et al. 2001, Eldridge and Rosentreter 1999). In the absence of disturbance biological soil crusts undergo successional processes, becoming more complex in species diversity and physical structure (Belnap and Lange 2001). What drives the relative abundance and distribution of these functional groups on any plot is a complex answer that cannot be addressed with such few plots, and goes beyond the purpose of the GLIR method. Given a proper sampling design and other protocols, the interactions among climate, disturbance, and site characteristics can be identified.

Averaging across the five plots, functional group biomass ranged from 0.1 kg/ha for the 'micro-lichens that grow on rock' to 103 kg/ha for the 'fruticose macro-lichens' (**Table 4**; **Figure 6**). For each functional group, biomass is a result of how many times the group is found (frequency) and the amount of soil surface covered by the group. Although some groups naturally have members that grow large, even many of the smaller micro-lichens can grow and coalesce to cover large areas on the ground.

Figure 6. ***Average dry biomass by functional group for all five plots (160 microquads) combined.***

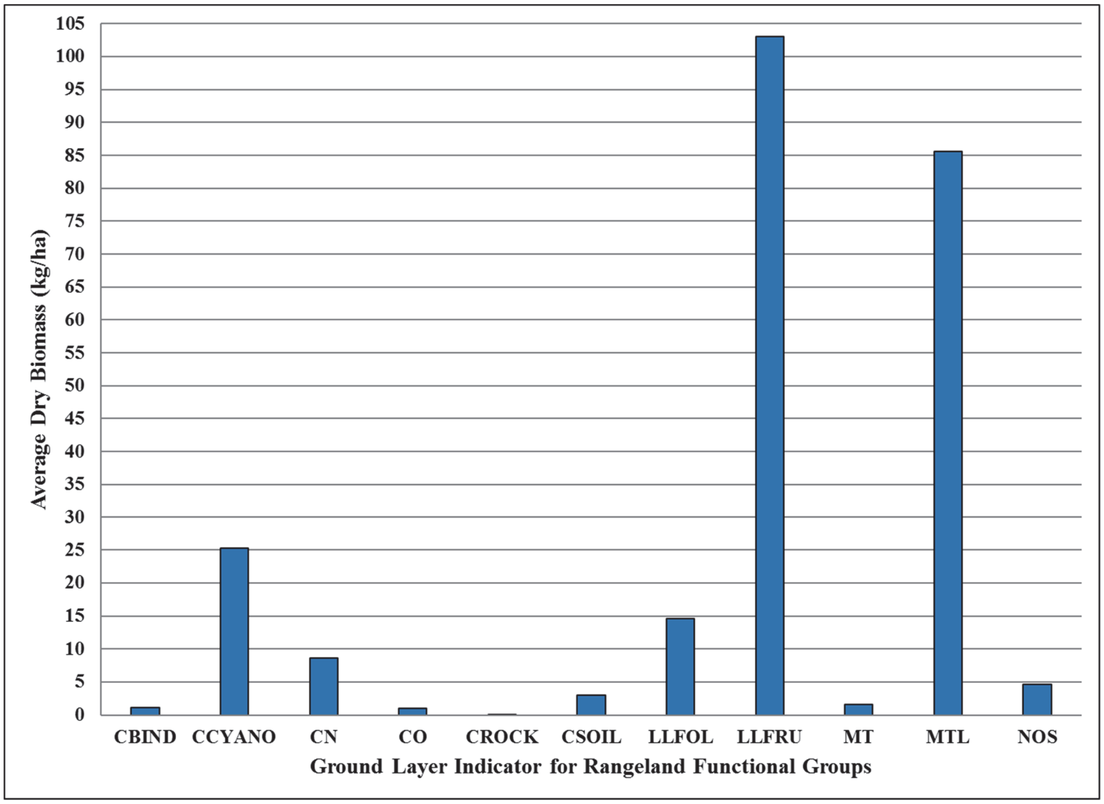


Table 4. ***Statistical data on functional groups from five plots (160 microquads) on the Milton Ranch, Roundup, Montana.*** *Calculations are averages plus or minus 1 standard deviation, unless indicated differently.*

Functional Group Code[1]	**Frequency** Number of occurrences in 160 microquads	**Dry Biomass** (kg/ha)	**Carbon (C) Content** (kg/ha)	**Nitrogen (N) Content** (kg/ha)	**Volume** (cubic meter per hectare)
CBIND	36	1.1 ± 0.5	0.5 ± 0.2	0	< 0.1
CCYANO	87	25.3 ± 8.4	11.2 ± 3.7	0.4 ± 0.1	0.4 ± 0.1
CN	68	8.6 ± 3.8	3.8 ± 1.7	0.2 ± 0.1	0.1 ± 0.1
CO	55	1 ± 0.3	0.4 ± 0.1	0	< 0.1
CROCK	2	0.1 ± 0.1	0	0	< 0.1
CSOIL	83	3 ± 0.9	1.3 ± 0.4	0	< 0.1
LLFOL	84	14.6 ± 6.8	6.5 ± 3	0.1 ± 0.1	0.3 ± 0.1
LLFRU	67	103 ± 31.7	45.7 ± 14.1	0.8 ± 0.2	1.7 ± 0.5
MT	44	1.6 ± 0.7	0.7 ± 0.3	0	< 0.1
MTL	62	85.6 ± 38.6	38 ± 17.1	0.9 ± 0.4	1.5 ± 0.7
NOS	50	4.6 ± 3	2.1 ± 1.3	0.1 ± 0.1	0.1

[1] Refer to **Table 2** for functional group definitions.

In decreasing order of their biomass general comments on members and frequency (**Table 4**) are made for each functional group:

- The 'fruticose macro-lichen' (LLFRU) functional group had the highest average biomass of 103 kg/ha. The dominant member was likely *Cladonia pocillum*, which was only observed on the ranch in its sterile form. This group also includes the fruticose species of *Circinaria* which may provide food for pronghorn (*Antilocapra americana*) (Sharnoff and Rosentreter 1998). The 'fruticose macro-lichen' group was found in 67 of the 160 microquads, making it the 5th most frequently encountered functional group.
- The 'loose-turf moss' (MTL) functional group averaged 86 kg/ha. Only mosses belonging to the genus *Syntrichia* belong to this group. *Syntrichia* mosses often have a loosely upright growth habit and distinctive leaf tips that are translucent and reflect light. This group was found in 62 of the 160 microquads, making it the 6th most frequently encountered functional group.
- The 'cyanobacteria/algae' (CCYANO) functional group averaged 25 kg/ha. Its members all have a filamentous growth form, and likely consist of *Nostoc flagelliforme*, *Scytonema* species, and *Microcoleus* species. Although the 'cyanobacteria/algae' functional group ranked third in average biomass, it ranked the highest in frequency, being found in 87 of 160 microquads.
- The 'foliose macro-lichen'(LLFOL) functional group averaged 15 kg/ha. Its dominant members are five *Xanthoparmelia* species which can provide food to pronghorn (Bernt 1976; Thomas and Rosentreter 1989; Thomas and Rosentreter 1992). Its relatively low biomass is a bit surprising given that it was commonly collected in the floristic surveys and was second highest in frequency, being found in 84 of 160 microquads. Many of the *Xanthoparmelia* species found on the Milton Ranch will curl up when dry, and upon moisture will flattened.
- The 'micro-lichens that fix nitrogen' (CN) functional group averaged 9 kg/ha. These nitrogen-fixing lichens belong to the genera of *Collema*, *Enchylium*, and *Placynthium*. This group was the fourth mostly common encountered group, being found in 68 of the 160 microquads.
- The 'foliose *Nostoc* cyanobacteria' (NOS) functional group averaged 5 kg/ha. Only *Nostoc commune* belongs to this group, which is distinct in being a large-lobed (foliose growth form) cyanobacteria (nitrogen-fixer). It is an early successional species that colonizes disturbed areas and can also be more tolerant of disturbances (fire for example) than other ground layer organisms. It ranked eighth in frequency being found in 50 of the 160 microquads. For some data analyses, NOS was lumped with CCYANO.
- The 'micro-lichens that colonize soil' (CSOIL) functional group averaged 3 kg/ha. Its very diverse membership all share the characteristic of growing in soil, binding particles and reducing the initial erosive impacts from rain and wind. Prominent members might include the crustose species of *Buellia*, *Candelariella*, *Endocarpon*, *Fulgensia*, *Placidium*, *Psora*, *Toninia*, and *Rinodina*. Despite its low biomass, it was the third most frequently encountered group, being found in 83 of the 160 microquads.
- The 'turf moss' (MT) functional group averaged about 2 kg/ha. This group includes mosses that grow upright, colonizing and building up soil, especially in sparse post-

disturbance habitats. Its members include *Bryum argenteum* and *Pterygoneurum ovatum*. The ‘turf moss’ functional group ranked ninth in frequency, being found in 44 of the 160 microquads.

- The ‘micro-lichens that bind together moss and detritus’ (CBIND) functional group averaged about 1 kg/ha. This is a diverse group that also includes micro-lichens that parasitize moss. Its members include the crustose species of *Bacidia*, *Bilimbia*, *Diploschistes*, *Lepraria*, and *Thrombium*. The ‘micro-lichens that bind together moss and detritus’ functional group was ranked tenth in frequency, being found in 36 of the 160 microquads.
- The ‘orange micro-lichen’ (CO) functional group averaged 1 kg/ha. These are orange colored micro-lichens that grow by etching themselves into rock. Some members like higher levels of nitrogen and sulphur dioxide, whether from natural (such as bird droppings) or man-made sources (such as agricultural fertilizers or concentrated animal use). This ‘orange micro-lichen’ functional group was seventh in frequency, being found in 55 of the 160 microquads.
- The ‘rock colonizing micro-lichen’ (CROCK) functional group averaged only one-tenth of a kg per ha. This group was also the least encountered, being found in 2 of the 160 microquads. Despite the group’s low biomass and frequency, their membership is very diverse, and includes crustose lichens that grow on rock, but are not orange and are not nitrogen-fixers. Members may include species of *Acarospora*, *Buellia*, *Lecanora*, or *Verrucaria*.

As previously stated the average amount of carbon and nitrogen found within biological soil crusts in the five plots was 110 kg/ha and 2.6 kg/ha, respectively (**Table 3**). In viewing this information from a functional group perspective, all groups contribute to carbon storage or sequestration but in different proportions (**Figure 7**). For example, the ‘micro-lichens on rock’ group was rarely found on the GLIR plots, though in the landscape they were frequently encountered. For nitrogen, 6 of the 11 functional groups contribute in substantive amounts (**Figure 8**). Cyanobacteria and cyanolichens (CCYANO, CN, and NOS groups) fix atmospheric nitrogen and make it available to plants and other vegetation. Because all species require nitrogen, other functional groups (LLFOL, LLFRU, and MTL groups) accumulate and slowly release it through decomposition.

Figure 7. ***Average content of carbon by functional group for all five plots combined.***

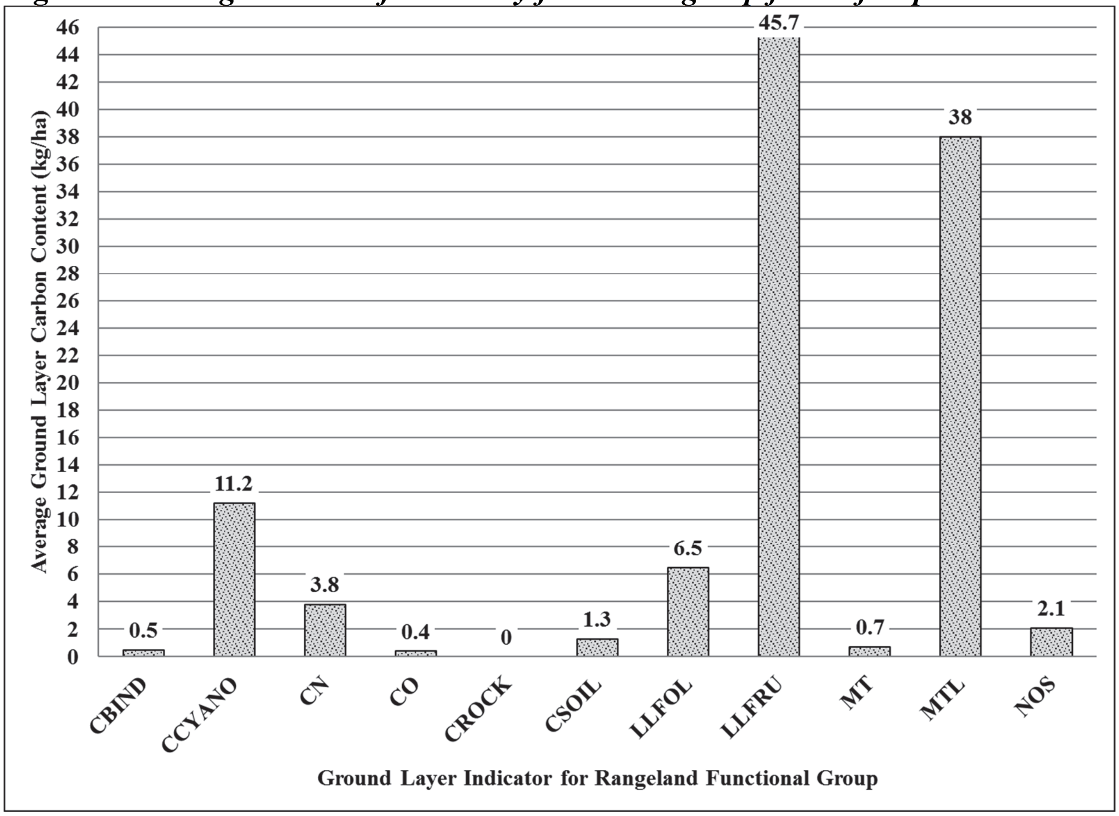
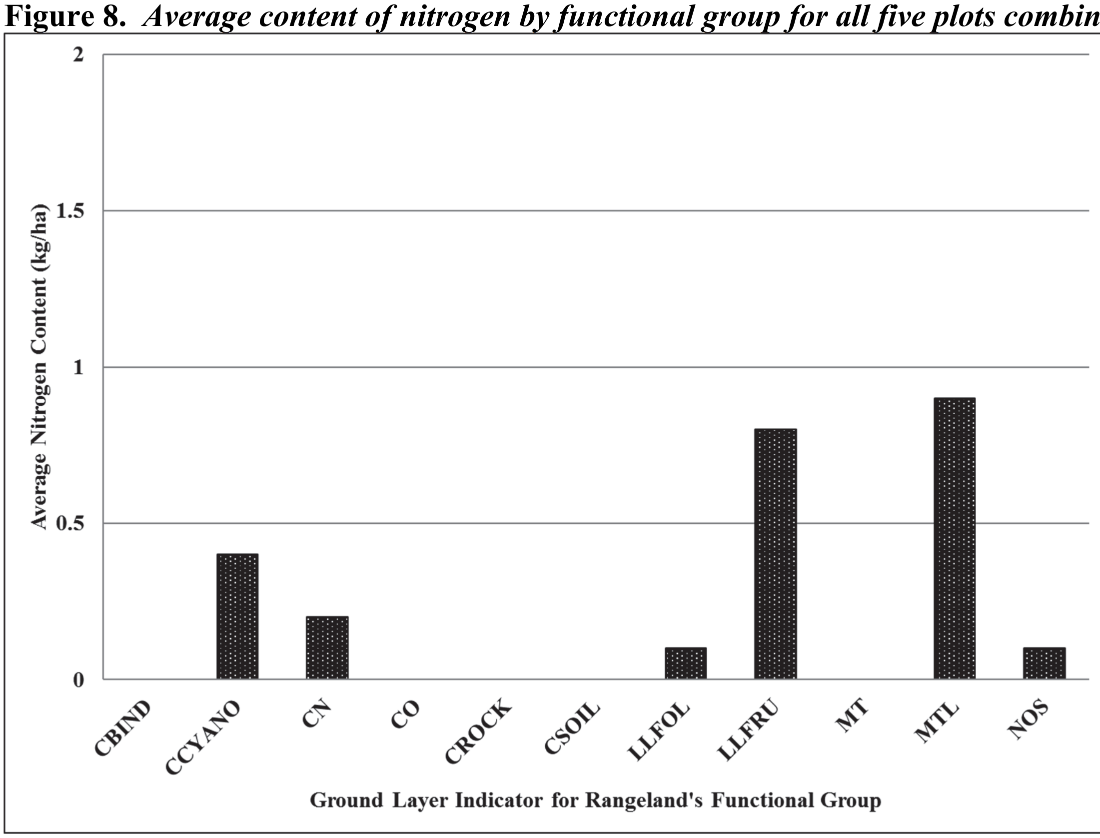


Figure 8. ***Average content of nitrogen by functional group for all five plots combined.***



3.6 Grazing Assessment

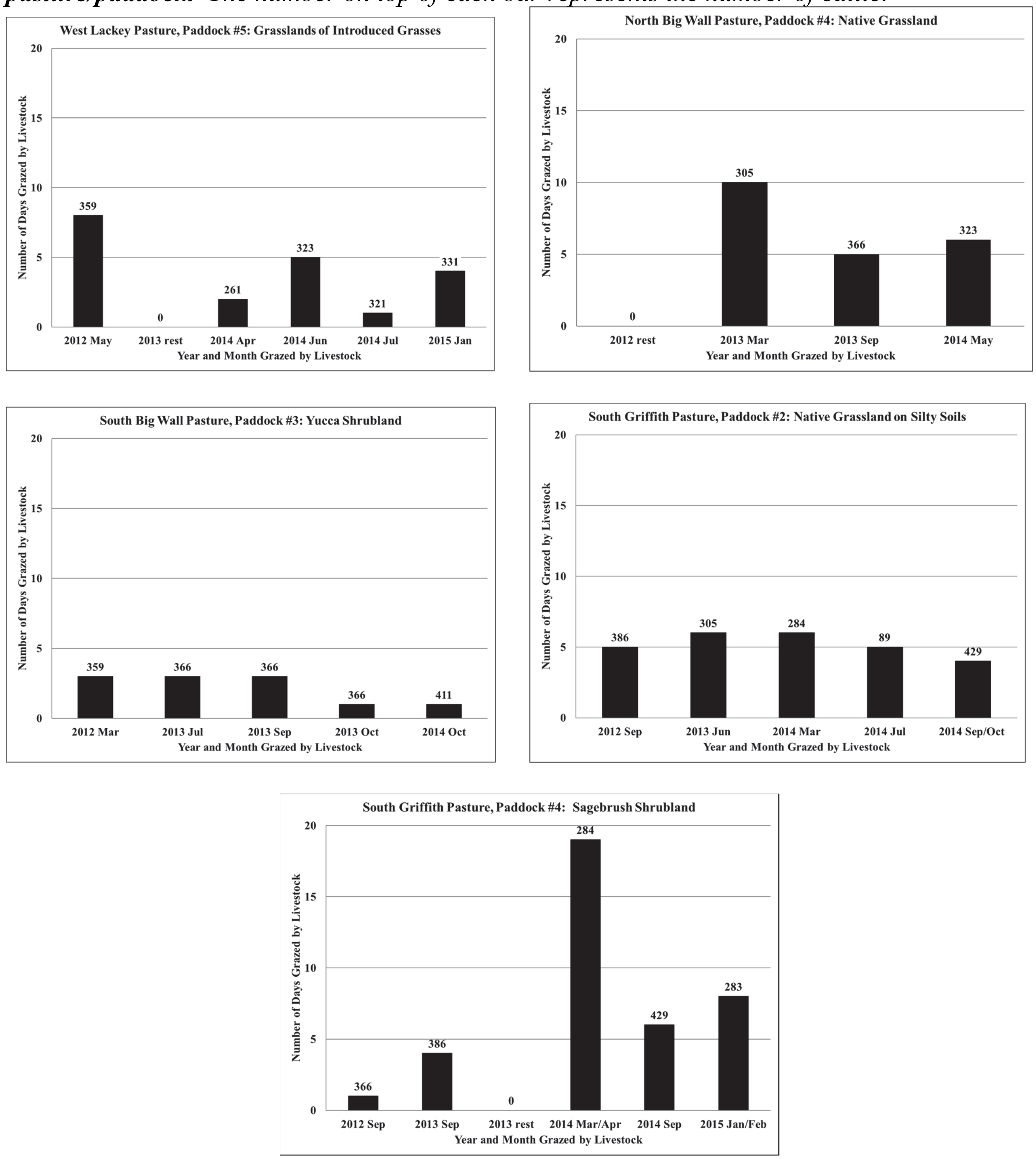
As with vascular plants, both the entire ground layer and its subset of biological soil crust communities can be used to track long-term environmental changes (such as temperature or precipitation) or physical disturbances (such as, fire, trampling, or soil compaction) (Smith et al. 2015). The type and structure of the biological soil crust is dependent upon geography, climate, time since disturbance, and species' distributions (Belnap et al. 2001). Species succession happens in biological soil crust communities as it does for plant communities (Belnap et al. 2001). The presence, absence, and abundance of early- or late-successional species can provide information regarding a site's disturbance history. When combined with data on vascular plant community composition, this information can assist a land manager in understanding a site's history, potential productivity, and ecological integrity.

To make effective assessments that inform how ranchers and public lands managers can efficiently operate, a baseline of data from which future comparisons can be made is required. The five plots were subjectively located on the Milton Ranch, and were not established to serve as a baseline for future rangeland monitoring tools. In practice, permanent GLIR plots will need to be established in concert with a vegetation management plan to order to obtain data that meets management objectives. To put the five GLIR plots into a grazing management context, livestock data was obtained for each pasture/paddock (**Figure 9**). The Milton Ranch consists of approximately 15,000 acres of land privately held and leased from the Bureau of Land Management and State of Montana for livestock grazing. For approximately the past 10 years, the land has been divided into about 9 pastures (allotments) that are sub-divided into approximately 69 paddocks of unequal size and dimension; paddocks range from 16 to 464 acres and on occasion will be temporarily split. On average the ranch grazes 700-800 cattle (350-400 cow-calf pairs) during the growing the season. In general cattle are moved between paddocks at a minimum of every three days. Every four years each paddock will get a full year's rest from grazing unless unusual conditions trigger an adjustment. The number of cattle that grazed in each paddock and the time of year was summarized for each pasture from about 2012 to 2015, which predates the 2016 pilot study. During the September 2016 study, no cows were observed in sampled paddocks.

Monitoring is the collection and analysis of repeated observations that assesses trends, either increasing, decreasing, or stable (Elzinga et al. 1998). When tied to specific management goals, monitoring of biological soil crusts can evaluate landscape changes over the defined time period. They usually are not designed to infer the cause of the observed changes. Although this pilot study was not designed to complement the existing vegetation management plan that the Milton ranch implements, such a network of GLIR plots could be readily added. A network of GLIR plots could serve to determine if there is a negative (declining), positive (increasing), or stable trend in the biomass, carbon content, or nitrogen content of the biological soils crusts, which themselves are indicators of rangeland health. The GLIR plots can also be established before an intervention to measure a response to changes in management or restoration.

On the Milton Ranch, livestock grazing within the paddocks is short-lived, ranging from 1 to 20 days, and the timing for any given paddock is variable within and between years (**Figure 9**). The functional group composition of the biological soil crust is expected to be influenced by the intensity and type of soil surface disturbance and time since the disturbance (Belnap et al. 2001).

Figure 9. ***The number of days and the month/year that livestock have grazed in each pasture/paddock.*** *The number on top of each bar represents the number of cattle.*



Biological soil crusts are more resilient to disturbance when conditions are moist and when the disturbance intensity is dispersed or short-term (Memmott 1998). Ground layer biomass was abundant at all GLIR plots except for the West Lackey 5 plot (**Figure 3**). Among these five plots, cattle numbers and their duration of use in the paddocks appear similar. Given this, the low biomass found in the West Lackey 5 plot could likely be caused from the past habitat conversion of soil tilling and planting. However, the plot was located somewhat near to the cattle's water source which would maintain a higher level of soil disturbance and negatively impact biological soil crusts. It remains unclear if the presence of nine functional groups represented by micro-and macro-lichens, mosses, and cyanobacteria supports Phillips' theory that this plant community is advancing toward a native grassland vegetation (Phillips 2010). Monitoring the biological soil crust community within each vegetation type would determine the trend and provide feedback for management actions.

4.0 SUMMARY

The management of North American rangelands has often focused on the distribution, species composition, and abundance of vascular plants, and has avoided the 'barren' areas between plants. Between plants often lives a ground layer composed of lichens, mosses, liverworts, hornworts, free-living algae, free-living cyanobacteria, bacteria, and/or micro-fungi that grow on soil, wood, rock, or other organic matter. Nutrient cycles and vascular plant performance are strongly influenced by the ground layer, particularly by organisms growing on soil that are termed biological soil crust (Bowker et al. 2006). It is the entire ground layer that binds surface soil particles to reduce erosion (Mazor et al. 1996), regulates the water runoff-infiltration balance (Warren 2001), and increases soil moisture retention (Alexander and Calvo 1990). Although soil erosion is widespread, directly impacts plant productivity, and has been a long-standing concern to range managers, range assessments that are more ecologically based are not commonly implemented (Bowker et al. 2006). In arid rangeland management, monitoring of the ground layer would quantify the ecological condition and its degree and direction of change.

The Ground Layer Indicator for Rangelands (GLIR) examines the entire non-vascular ground layer and is used to understand ecosystem function, specifically for potential carbon storage, nitrogen fixation, forage availability, soil stability, and site disturbance (Smith et al. 2015). It was designed to monitor the ground layer across rangelands at ranch scales as well as larger, state-sized scales. A exploratory study to implement GLIR in Montana was conducted at the Milton Ranch, northeast of Roundup, Montana in Mussellshell County. While systematic ground layer sampling is ongoing as part of the Forest Inventory and Analysis program in Alaska, the US Pacific Northwest, and the US Midwest (Smith et al. 2015; Calabria et al. 2016; Smith et al. 2017), this was the first use of this modified method in Montana.

Some of the findings from this exploratory study are summarized as follows:

- The Ground Layer Indicator for Rangelands examines the ecological roles and functions that the community of mosses, lichens, liverworts, hornworts, free-living cyanobacteria, free-living algae, bacteria, and micro-fungi who live in soil (biological soil crust), wood, rock, and other organic matter provide to rangelands.

- The GLIR method was found to be appropriate for use on Montana rangelands to collect biomass, carbon content, and nitrogen content data on ground layer organisms. Collecting this data by functional group (not species) simplifies the information while maintaining its applicability to long-term monitoring of rangeland health and condition.

- The method found 12 functional groups of which 11 were present on the GLIR plots. Liverworts, though expected, were not observed during the project. These functional groups emphasize the ecological role(s) that the group performs well relative to rangelands. They are separated by organism, growth form, substrate, and a few other ocular characteristics, thus eliminating the need to identify species. However, the method does require training by a certified teacher. Other functional groups are expected to occur in Montana, particularly in less arid rangelands (see Smith et al. 2015).

- The ground layer was examined at GLIR plots that represent *Grasslands of Introduced Grasses*, *Native Grassland*, *Native Grassland on Silty Soil*, *Yucca Shrubland*, and *Sagebrush Shrubland* plant community types.

- At each plot, the ground layer biomass, carbon content, nitrogen content, number of functional groups, and other attributes were summarized and graphed. Plot comparison across the five sites and between the native and non-native habitat sites were made and discussed. Applications of the method are also discussed. As anticipated the number of GLIR plots were insufficient to accurately characterize the ground layer at the scale of the Milton Ranch or to test for statistical significance.

- The non-vascular survey and GLIR plot located in the *Sagebrush Shrubland* plant community also occurred within an historic lek area. This study found that two important components of Greater Sage-grouse habitat, healthy big sagebrush and biological soil crust, are present.

- For each paddock that contained a GLIR plot the number of livestock, length of time grazed, and time of grazing was charted from 2012 to 2015. The process of developing baseline conditions from grazing data and permanent GLIR plots from which changes can be assessed is discussed.

Based on this exploratory project using the GLIR method, several recommendations are made to help guide future work on ground layer organisms in Montana:

- The Ground Layer Indicator method and its modified GLIR version was designed to make landscape estimates, such as at ranch- or state-size scales. As with all research or monitoring techniques, a well-designed approach that uses stratified random sampling, determines an acceptable risk of Type I and Type II errors, and uses a statistically acceptable plot density is necessary to develop.

- The exploratory study subjectively placed five GLIR plots near to areas being surveyed for non-vascular species, to vegetation transects currently used for managing rangeland on the Milton Ranch, and/or to the 2010 plant community plots. The intent was to provide some additional ecological and species information to the existing projects. As anticipated from the limited time and resources available, the number of GLIR plots was insufficient and their subjective placement prevented a statistical analysis and limited any interpretation of the data. The original method does require a study design that aligns data collection with management goals, a sufficient number of plots to characterize an area, and a stratified random sampling design to ensure sound statistical analysis and interpretation.

- Future monitoring using the GLIR methodology should permanently establish plots in order to develop baseline conditions from which changes can be assessed. Permanent plots can be monumented in a variety of ways, of which some methods make the plot unnoticeable to ranchers and animals.

- It is recommended that the GLIR method be implemented in a pilot study that encompasses a large ranch or federal/state land management parcels. Examples include the Milton Ranch or as a supplemental indicator for the BLM's Assessment, Inventory, and Monitoring (AIM) project in Montana and several other western states. The pilot study should establish permanent plots to develop baseline conditions and long-term ecological monitoring.

- The GLIR requires that crew performing the method be trained by a certified teacher. Annual training of BLM, USFS, and NRCS staff would develop a crew proficient in the method. Depending upon the level of experience, field crews can be trained in the methodology in 2-5 days. Trained field crews would become skilled in identifying the basic types of organisms that make up the ground layer (such as green algal lichens, cyanolichens, short and tall mosses, free-living cyanobacteria, mosses, and liverworts) and in varying habitats where they occur. However, field crews would not need to know species identification for these organisms. For previously trained crew members, a 1-day annual refresher training would likely be required to maintain the necessary skill set to ensure data quality.

- Determine the feasibility of modeling the potential cover and composition of biological soil crusts in Montana, in support of on-the-ground assessment and monitoring efforts.

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Appendix A

Ground Layer Indicator for Rangelands Method Data Sheet

Datasheet for FIA Ground Layer: NONFOREST lands

Plot name: ________________________ Coords: ________________, ________________ Crew name: ____________________________ Date: ______________20_____

Functional groups

Type	FXNL GRP	Name	Examples
Moss	**MS**	Sphagnum peat-moss	*Sphagnum* only
	MN	N-fixing feather mosses	*Pleurozium, Hylocomium, Rhytidiadelphus* only
	MF	Feather (branched) mosses	*Drepanocladus, Thuidium, Brachythecium*
	MTL	Moss "loose" turf	*Syntrichia* only
	MT	Turf (upright) mosses	*Bryum, Polytrichum, Grimmia, Encalypta, Ceratodon*
Liverwort	**VF**	Flat (thalloid) liverworts	*Marchantia, Conocephalum*
	VS	Stem-and-leaf liverworts	*Anthelia, Cephaloziella*
Macro-lichen	**LF**	Forage lichens	reindeer-lichen *Cladonia, Alectoria, Bryocaulon*
	LN-fol	N-fixing foliose lichens	*Peltigera, Nephroma, Solorina*
	LN-fru	N-fixing fruticose lichens	*Stereocaulon* only
	LL-fol	Other foliose lichens	*Parmelia, Physcia, Xanthoparmelia*
	LL-fru	Other fruticose lichens	unbranched-*Cladonia*, most 'vagrant' lichens
Micro-lichen	**CO**	Orange crustose lichens	*Xanthoria, Candelaria*
	CN	Crust lichens, N-fixing	*Collema, Leptogium, Polychidium, Massalongia*
	CBIND	Crust lichens binding moss and detritus	*Trapeliopsis, Megaspora, Diploschistes*
	CSOIL	Crust lichens on 'bare' soil	*Psora, Placidium, Phaeorrhiza, Placynthiella*
	CROCK	Crust lichens on rock	*Acaraspora, Aspicilia, Lecidea, Rhizoplaca, Candelariella*
	CCYANO	Cyanobacterial/algal crust	*Microcoleus, Nostoc,* Chlorophyta

Cover classes

COVER CLASS	Percent cover
0	Absent
T	>0 – 0.1
1	>0.1 – 1
2	>1 – 2
5	>2 – 5
10	>5 – 10
25	>10 – 25
50	>25 – 50
75	>50 – 75
95	>75 – 95
99	>95

Depth classes

DEPTH CLASS	Depth (inches)
0	0
T	<1/8, trace
Q	>1/8 – 1/4
H	>1/4 – 1/2
1	>1/2 – 1
2	>1 – 2
4	>2 – 4
8	>4 – 8
16	>8

Site description:

Subplot	Transect	Microquad	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS
1	90°	1																		
		2																		
		3																		
		4																		
	270°	5																		
		6																		
		7																		
		8																		
2	360°	9																		
		10																		
		11																		
		12																		
	180°	13																		
		14																		
		15																		
		16																		

(continued on next page)

Datasheet for FIA Ground Layer: NONFOREST lands

Plot name: ______________________ Coords: ______________, ______________ Crew name: __________________________ Date: _____________20_____

(continued from previous page)

Functional groups

Type	FXNL GRP	Name	Examples
Moss	**MS**	Sphagnum peat-moss	*Sphagnum* only
	MN	N-fixing feather mosses	*Pleurozium, Hylocomium, Rhytidiadelphus* only
	MF	Feather (branched) mosses	*Drepanocladus, Thuidium, Brachythecium*
	MTL	Moss "loose" turf	*Syntrichia* only
	MT	Turf (upright) mosses	*Bryum, Polytrichum, Grimmia, Encalypta, Ceratodon*
Liverwort	**VF**	Flat (thalloid) liverworts	*Marchantia, Conocephalum*
	VS	Stem-and-leaf liverworts	*Anthelia, Cephaloziella*
Macro-lichen	**LF**	Forage lichens	reindeer-lichen *Cladonia, Alectoria, Bryocaulon*
	LN-fol	N-fixing foliose lichens	*Peltigera, Nephroma, Solorina*
	LN-fru	N-fixing fruticose lichens	*Stereocaulon* only
	LL-fol	Other foliose lichens	*Parmelia, Physcia, Xanthoparmelia*
	LL-fru	Other fruticose lichens	unbranched-*Cladonia*, most 'vagrant' lichens
Micro-lichen	**CO**	Orange crustose lichens	*Xanthoria, Candelaria*
	CN	Crust lichens, N-fixing	*Collema, Leptogium, Polychidium, Massalongia*
	CBIND	Crust lichens binding moss and detritus	*Trapeliopsis, Megaspora, Diploschistes*
	CSOIL	Crust lichens on 'bare' soil	*Psora, Placidium, Phaeorrhiza, Placynthiella*
	CROCK	Crust lichens on rock	*Acaraspora, Aspicilia, Lecidea, Rhizoplaca, Candelariella*
	CCYANO	Cyanobacterial/algal crust	*Microcoleus, Nostoc,* Chlorophyta

Cover classes

COVER CLASS	Percent cover
0	Absent
T	>0 – 0.1
1	>0.1 – 1
2	>1 – 2
5	>2 – 5
10	>5 – 10
25	>10 – 25
50	>25 – 50
75	>50 – 75
95	>75 – 95
99	>95

Depth classes

DEPTH CLASS	Depth (inches)
0	0
T	<1/8, trace
Q	>1/8 – 1/4
H	>1/4 – 1/2
1	>1/2 – 1
2	>1 – 2
4	>2 – 4
8	>4 – 8
16	>8

Subp	Tran.	Mqd.	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS	FXNL GRP	COVER CLASS	DEPTH CLASS
3	135°	17																		
		18																		
		19																		
		20																		
	315°	21																		
		22																		
		23																		
		24																		
4	45°	25																		
		26																		
		27																		
		28																		
	225°	29																		
		30																		
		31																		
		32																		

Appendix B

A Preliminary Checklist of the Moss, Lichen, and Cyanobacteria Taxa and their Functional Groups found on the Milton Ranch

Table B-1. ***Draft list of the mosses found on the Milton Ranch during the surveys from September 13-15, 2016 and their functional groups.***

MOSS SPECIES	FUNCTIONAL GROUP
Barbula convoluta	MT
Brachytheciastrum collinum	MF
Brachytheciastrum velutinum	MF
Bryum argenteum	MT
Ceratodon purpureus	MT
Didymodon fallax	MT
Didymodon tectorum	MT
Encalypta vulgaris	MT
Gemmabryum caespiticium	MT
Grimmia anodon	MT
Grimmia plagiopodia	MT
Hypnum cupressiforme	MF
Hypnum revolutum	MF
Hypnum vaucheri	MF
Jaffueliobryum wrightii	MT
Myurella julacea	MF
Pseudocrossidium obtusulum	MT
Pseudoleskea incurvata	MT
Pseudoleskeella tectorum	MT
Pterygoneurum ovatum	MT
Pterygoneurum subsessile	MT
Pylaisia polyantha	MF
Syntrichia caninervis	MTL
Syntrichia papillosissima	MTL
Syntrichia ruralis	MTL
Tortella alpicola	MT
Tortula hoppeana	MT
Tortula mucronifolia	MT

* MF = Feather Moss species. This group was not observed on the 2016 GLIR plots.

Table B-2. ***Draft list of the lichens and cyanobacteria found on the Milton Ranch during the surveys from September 13-15, 2016 and their functional groups.***

LICHEN or CYANOBACTERIA	FUNCTIONAL GROUP	LICHEN or CYANOBACTERIA	FUNCTIONAL GROUP
Acarospora fuscescens	CROCK	*Dermatocarpon miniatum*	LLFOL
Acarospora glaucocarpa	CROCK	*Diploschistes gypsaceus*	CBIND
Acarospora stapfiana	CROCK	*Diploschistes muscorum*	CBIND
Acarospora strigata	CROCK	*Diploschistes scruposus*	CBIND
Anaptychia elbursiana	LLFOL	*Enchylium coccophorum*	CN
Aspicilia reptans	CSOIL	*Endocarpon loscosii*	CSOIL
Bacidia bagliettoana	CBIND	*Endocarpon pusillum*	CSOIL
Bagliettoa calciseda	CROCK	*Fulgensia bracteata*	CSOIL
Bilimbia lobulata	CBIND	*Fulgensia desertorum*	CSOIL
Buellia dispersa	CROCK	*Fulgensia subbracteata*	CSOIL
Buellia elegans	CSOIL	*Heppia lutosa*	CSOIL
Buellia epigaea	CSOIL	*Heteroplacidium zamenhofianum*	CSOIL
Buellia punctata	CROCK	*Lecanora crenulata*	CROCK
Buellia venusta	CROCK	*Lecanora flowersiana*	CROCK
Caloplaca atroalba	CO	*Lecanora hagenii*	CROCK
Caloplaca citrina	CO	*Lecanora muralis*	CROCK
Caloplaca decipiens	CO	*Lecanora saligna*	CROCK
Caloplaca jungermanniae	CO	*Lecanora subintricata*	CROCK
Caloplaca lactea	CO	*Lecanora zosterae*	CBIND
Caloplaca microphyllina	CO	*Lecidella carpathica*	CROCK
Caloplaca pyracea	CO	*Lecidella cf euphorea*	CROCK
Caloplaca stillicidiorum	CO	*Lecidella patavina*	CROCK
Caloplaca tiroliensis	CO	*Lecidella stigmatea*	CROCK
Caloplaca tominii	CO	*Lepraria vouauxii*	CBIND
Caloplaca trachyphylla	CO	*Lobothallia alphoplaca*	LLFOL
Caloplaca xanthostigmoidea	CO	*Microcoleus*	CCYANO
Candelariella aggregata	CBIND	*Nostoc commune*	NOS
Candelariella antennaria	CROCK	*Nostoc flagelliforme*	CCYANO
Candelariella aurella	CROCK	*Parmelia saxatilis*	LLFOL
Candelariella rosulans	CROCK	*Phaeophyscia constipata*	LLFOL
Candelariella vitellina	CBIND	*Phaeophyscia nigricans*	LLFOL
Circinaria contorta	CROCK	*Phaeophyscia orbicularis*	LLFOL
Circinaria hispida	LLFRU	*Physcia biziana*	LLFOL
Cladonia cariosa	LLFRU	*Physcia dimidiata*	LLFOL
Cladonia coniocraea	LLFRU	*Physcia muscigena*	LLFOL
Cladonia fimbriata	LLFRU	*Physconia muscigena*	LLFOL
Cladonia imbricarica	LLFRU	*Physconia perisidiosa*	LLFOL
Cladonia pocillum	LLFRU	*Placidium lacinulatum*	CSOIL
Cladonia pyxidata	LLFRU	*Placidium rufescens*	CSOIL
Collema crispum	CN	*Placidium squamulosum*	CSOIL
Collema tenax	CN	*Placynthiella oligotropha*	CSOIL
Collema tenax group	CN	*Placynthium nigrum*	CN

Table B-2 (continued). ***Draft list of the lichens and cyanobacteria found on the Milton Ranch during the surveys from September 13-15, 2016 and their functional groups.***

LICHEN or CYANOBACTERIA	FUNCTIONAL GROUP
Psora decipiens	CSOIL
Psora tuckermanii	CSOIL
Rhizoplaca chrysoleuca	CROCK
Rinodina albertana	CSOIL
Rinodina bischoffii	CROCK
Rinodina pyrina	CROCK
Rinodina straussii	CROCK
Rinodina terrestris	CSOIL
Scytonema	CCYANO
Staurothele areolata	CROCK
Staurothele elenkinii	CROCK
Thelidium minutulum	CROCK
Thrombium epigaeum	CBIND
Toninia ruginosa	CSOIL
Toninia sedifolia	CSOIL
Verrucaria calkinsiana	CROCK
Verrucaria inficiens	CROCK
Xanthoparmelia camtschadalis	LLFOL
Xanthoparmelia chlorochroa	LLFOL
Xanthoparmelia neochlorochroa	LLFOL
Xanthoparmelia neowyomingica	LLFOL
Xanthoparmelia wyomingica	LLFOL
Xanthoria elegans	CO
Xanthoria fallax	CO