

EFFECTS OF LAND-USE PRACTICE ON WETLAND SOIL HYDROLOGY, SALINITY, AND BIOGEOCHEMISTRY IN THE PRAIRIE POTHOLE REGION

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in Partial Fulfilment of the requirements
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in the Department of Soil Science
University of Saskatchewan
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By

Shayeb Shahariar

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ABSTRACT

Land-use practice shift to short rotation willow (SRW) can accelerate vegetation linked alterations in the shallow groundwater table (GWT), salinity, and nutrient availability in the hydrologically dynamic wetland riparian zones. Additionally, SRW has higher potentials to sequester soil organic carbon (SOC) and can further impact greenhouse gas (GHG) emissions and extracellular enzyme activities (EEAs). In a field experiment, the effects of SRW were evaluated by measuring the depth to GWT; groundwater and soil electrical conductivity (EC), nutrients (N, P, K, and S), and SOC content with different fractions and chemical compositions in soils (60-cm depth) collected along transects during the first rotation (3-year cycle) and compared with adjacent annual crop (AC), and pasture (PA) in two (site A and B) semi-arid prairie pothole region (PPR) wetland systems. In a microcosm experiment, GHG (CO_2 , CH_4 , and N_2O) emissions and EEAs [β -glucosidase (BG), N-acetyl glucosaminidase (NAG), and alkaline phosphatase (AP)] were measured in intact soil cores (30-cm depth) collected from the same field sites, and treated with declining water tables (2 to 26 cm depth), and salinity levels (S_0 = control, S_1 = 6 mS cm^{-1} , and S_2 = 12 mS cm^{-1}).

The GWT responded to precipitation patterns. Both groundwater and soil EC varied significantly ($p < 0.05$), but no consistent land-use patterns were observed between sites. Land-use practices only impacted the GWT depth significantly ($p < 0.001$) in site B. Soil EC did not vary significantly ($p > 0.05$) for depths, years, and months. Under SRW, soil $\text{NH}_4^+\text{-N}$ and K^+ contents were lower, whereas $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, and $\text{SO}_4^{2-}\text{-S}$ were higher. The groundwater $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, K^+ , and $\text{SO}_4^{2-}\text{-S}$ were significantly higher ($p < 0.05$) under SRW, whereas $\text{PO}_4^{3-}\text{-P}$ did not differ significantly ($p > 0.05$) across land-uses. Total SOC was higher in PA in both sites, but significant ($p < 0.05$) only in site B. The light fraction organic carbon (LFOC) and particulate organic carbon (POC) followed a similar land-use pattern (i.e., $\text{PA} > \text{SRW} = \text{AC}$). The SOC and water extractable organic carbon (WEOC) were significantly higher ($p < 0.05$) at 0-15 cm across

land-uses. The ratios of phenolic and amides to polysaccharides were significantly higher ($p < 0.05$) under SRW than AC and PA in site A. The higher alkyl-C to O-alkyl-C ratio suggested a higher degree of decomposition and better stability of SOC at 15-30 cm. The GHG emissions were significantly ($p < 0.001$) affected by the soils from different land-uses in the order of PA > AC = SRW. Compared to control, emissions of CO₂ and CH₄ were significantly lower ($p < 0.05$), while N₂O was significantly higher ($p < 0.05$) under higher salinity treatments (i.e., S1 and S2). Emissions under declining GWT were significantly ($p < 0.001$) variable and specific to each gas. The SRW soils had significantly lower ($p < 0.05$) global warming potential (GWP) than AC and PA. Soil EEAs significantly ($p < 0.05$) impacted by different land-uses (i.e., PA > AC = SRW), suggested that the effects resulted from background SOC. Soil EEAs were significantly ($p < 0.05$) reduced under higher depth to GWT except reverse for BG in site B. However, they did not differ significantly among salinity treatments. This research demonstrates that land-use linked variation in GWT and salinity can have a consequential effect on the riparian wetland soil nutrients, SOC, GHG emissions, and EEAs in the PPR.

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DEDICATION

This dissertation is dedicated to my parents, who always supported me to reach this stage.

To my Parents:

“Your encouragement and unconditional support made it possible to reach this phase.

Thank you for your unconditional love and supports.”

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LIST OF ACRONYMS

AAFC	Agriculture and Agri-Food Canada
AC	Annual crop
ADC	Agroforestry development center
ANOSIM	Analysis of similarities
ANOVA	Analysis of variance
AP	Alkaline phosphatase
BG	β -glucosidase
BMPs	Beneficial management practices
CAP	Constrained analysis of principal coordinates
CEC	Cation exchange capacity
DOC	Dissolved organic carbon
DON	Dissolved organic nitrogen
EEs	Extracellular enzymes
EEAs	Extracellular enzyme activities
EC	Enzyme commission
EC _{gw}	Groundwater electrical conductivity
EC _{soil}	Soil electrical conductivity
EC _a	Apparent electrical conductivity
EM	Electromagnetic
ESP	Exchangeable sodium percentage
ESRI	Environmental systems research institute
ET	Evapotranspiration
FTIR	Fourier transform infrared
GHG	Greenhouse gas
GPS	Global positioning system
GWT	Groundwater table
HF	Heavy fraction
HCA	Hierarchical cluster analysis
LC	Labile carbon
LF	Light fraction
LFOC	Light fraction organic carbon
LFON	Light fraction organic nitrogen
MP-AES	Microwave plasma-atomic emission spectrometry
MUB	4-methylumbelliferone
MUF	4-methylumbelliferyl

NAG	N-acetyl glucosaminidase
NMDS	Non-metric multidimensional scaling
OC	Organic carbon
PA	Pasture
PCA	Principal component analysis
PERMANOVA	Permutation multivariate analysis of variance
POC	Particulate organic carbon
PPR	Prairie pothole region
PVC	Poly vinyl chloride
RC	Recalcitrant carbon
RDA	Redundancy analysis
RTK	Real-time kinematic
SC	Slow carbon
SIMPER	Similarity percentage
SOC	Soil organic carbon
SOM	Soil organic matter
SRW	Short rotation willow
VSWC	Volumetric soil water content
TDS	Total dissolved salts
TSC	Total soil carbon
TIC	Total inorganic carbon
TN	Total nitrogen
VPA	Variation portioning analysis
WEOC	Water extractable organic carbon
WEON	Water extractable organic nitrogen

PREFACE

This dissertation is based on the five primary research chapters, followed by synthesis, excluding the global introduction (Chapter 1) and a literature review (Chapter 2). Out of five core research chapters, the first two chapters (Chapter 3 and 4) are case study types and written in the style of short communications. The rest three chapters (Chapter 5, 6, and 7) are hypothesis-driven in and written in manuscript format; these are in preparation for submission into *Geoderma*, *Global Change Biology*, and *Wetlands* journals, respectively. All these manuscript documents are written in the first-person plural to recognize the contribution of multiple authors.

Chapter 5: Shayeb Shahariar*, Derek Peak, Raju Soolanayakanahally, Angela Bedard-Haughn. ***Soil carbon beneath contrasting riparian land-uses in the prairie pothole region.***

Geoderma (anticipated submission date is September 10, 2020).

Chapter 6: Shayeb Shahariar*, Richard Farrell, Raju Soolanayakanahally, Angela Bedard-Haughn. ***Elevated salinity and water table drawdown significantly affect greenhouse gas emissions in soils from contrasting land-use practices in the prairie pothole region.***

Global Change Biology (submitted on September 8, 2020).

Chapter 7: Shayeb Shahariar*, Bobbi Helgason, Raju Soolanayakanahally, Angela Bedard-Haughn. ***Soil enzyme activity as affected by land-use, salinity, and groundwater***

fluctuations in wetland soils of the prairie pothole region. *Wetlands* (submitted on August 15, 2020).

Contributions of authors to the manuscripts were as follows:

- **Shayeb Shahariar** conceptualized, designed, and performed all the experiments; analyzed all the data, prepared all figures and tables, wrote and reviewed drafts of all the manuscripts, and made final editorial decisions regarding all text and graphs, approved all the final drafts, and acted as the corresponding author (*) for all the manuscripts.

- **Derek Peak** contributed reagents, materials, and analytical support for FTIR of SOC measurement; reviewed drafts of the manuscript, made comments, and approved the final draft.
- **Richard Farrell** contributed reagents, materials, and analytical support for GHG measurement, reviewed drafts of the manuscript, made comments, and approved the final draft.
- **Bobbi Helgason** contributed reagents, materials, and analytical support for enzyme assays, reviewed drafts of the manuscript, made comments, and approved the final draft.
- **Raju Soolanayakanahally** provided technical supports for field and soil core samples collection, reviewed drafts of the manuscripts, made comments, and approved all the final drafts.
- **Angela Bedard-Haughn** provided all necessary financial support and supplies required for both the field and greenhouse experiments, guided the fundamental ideas for this work, reviewed drafts of the manuscripts, made comments, and approved all the final drafts.

1 INTRODUCTION

Wetlands are one of the most critical and complex ecosystems on earth in which numerous physical, chemical, and biological processes operate. In addition, wetlands are a vital landscape feature due to their unique role in regulating hydrological and biogeochemical cycles. They provide numerous ecosystem services such as water and nutrient storage and cycling, flood mitigation, groundwater recharge, carbon sequestration, habitat for wildlife and biodiversity (Mitsch & Gosselink, 2015). Wetland soils are often defined as hydric soils, which are formed under flooding and saturation that is long enough to develop anaerobic conditions for a significant portion of the year (Vepraskas & Craft, 2016). Based on the soil organic carbon (SOC) content, wetland soils can be either mineral soils (SOC < 12 to 20 % on a dry weight basis) or organic soils (Mitsch & Gosselink, 2015). Mineral wetland soils typically develop redoximorphic features, which is mediated by microbiological processes, and their rate depends on 1) prolonged anaerobic conditions, 2) necessary soil temperature, and 3) substrate for microbial activity, i.e., soil organic matter (SOM) content (Mitsch & Gosselink, 2015). Wetland soil is the medium where many biogeochemical transformations take place and are the primary storage of chemicals resulted from the decomposition of SOM (Reddy & DeLaune, 2008). Wetland soils can be sources, sinks, and transformers of carbon, nitrogen, phosphorus, sulfur, iron, and manganese; however, they are primarily valued for their ability to be a significant sink for nutrients. For instance, wetland carbon sequestration can improve the global carbon balance; however, denitrification and methanogenesis contribute to the emission of greenhouse gases from wetland soils (Mitsch & Gosselink, 2015).

The prairie pothole region (PPR) of the North American glaciated Great Plains is a landscape characterized by millions of small freshwater mineral soil wetlands that are popularly known as “potholes” or “sloughs” (Richardson et al., 1994). Precipitation is the primary source of water for the semi-arid PPR wetland systems, though most of the wetlands have some connection with

the groundwater (Winter, 1989). Because of the hydrologic connection to the groundwater (Thorslund et al., 2018), the PPR landscapes are interconnected wetland complexes; however, they can be either groundwater recharge, discharge, and through-flow in type (Richardson et al., 1994). The PPR comprises some of the most productive agricultural soil in North America. Agricultural practices are the dominant economic force that determines the land-use practice and have enormously impacted the wetland ecosystems within the PPR region (Bartzen et al., 2010, Guntenspergen et al., 2002). Intensive agricultural production through wetland drainage has remained the primary cause of wetland loss in this region (Dahl, 2010). However, other agricultural practices that have impacted wetlands in this regions include sedimentation (Gleason & Euliss Jr, 1998), movement of agrochemicals (Grue et al., 1989), nutrient inputs (Neely & Baker, 1989), variation in shallow groundwater table (Euliss & Mushet, 1996), and the alteration of riparian vegetation (Kantrud et al., 1989). The “sloughs” or “potholes” in the PPR are often surrounded by native willow (*Salix* Spp.) vegetation; a phenomenon traditionally known as “willow rings” (Kantrud et al., 1989). However, a “saline ring” around the slough can be developed, predominantly during dry summer months, due to the gradient triggered by higher evapotranspiration rates from the riparian vegetation (i.e., “willow rings”). The transport of naturally occurring salts to the near-surface soil layers can occur by capillary moisture flow and thus cause soil salinity (Stolte et al., 1992).

Short rotation willow (SRW), in the form of agroforestry, is a unique land-use practice of increasing interest in North America, including the Canadian PPR (Thevathasan et al., 2012). It has been projected that the fast-growing high biomass producing herbaceous-woody bioenergy crop harvested in a short rotation cycle (usually 3-year), i.e., SRW, will be a vital source of renewable energy in the coming decades (Volk et al., 2004). Additionally, the establishment of SRW land-use practice has been proposed to safeguard water quality (Dimitriou et al., 2009b), sequester carbon (Lockwell et al., 2012, Oelbermann et al., 2004), mitigate greenhouse gas

(GHG) emissions (Harris et al., 2017), enrich soil extracellular enzyme activities (EEAs) (Stauffer et al., 2014), and improve soil quality and health (Dollinger & Jose, 2018). However, SRW is also known to have both high water and nutrient demands due to its fast growth and high biomass-producing capability (Ledin, 1998). The establishment of phreatophytic plantations such as SRW, with higher evapotranspiration demand and deep root systems, therefore, can alter vegetation linked groundwater table depth fluctuations and elevate soil salinity (Jobbágy & Jackson, 2007, Noretto et al., 2013) in PPR riparian zones. There are two million hectares of degraded marginal riparian lands in Saskatchewan that are not suitable for cultivation or crop production due to excess moisture and/or salinity (Amichev et al., 2014b). Hence, these marginal riparian lands can be planted with SRW that has the potential to sequester carbon through biomass production, offset GHG emission, and improve soil quality in addition to producing renewable energy (Amichev et al., 2012, Stolarski et al., 2011). Thus, it is vital to assess the impact of the establishment of SRW land-use practice in the riparian zones of PPR wetland systems.

Land-use practice changes are of great interest in recent decades due to their potential impacts on carbon balance, soil quality, and environmental change. The productivity and sustainability of the agroecosystem critically depend on soil quality and health (Bünemann et al., 2018).

Sustainable land-use practices such as SRW agroforestry systems have shown substantial evidence of improved soil quality and health worldwide over the last four decades (Dollinger & Jose, 2018). Globally, there are many significant environmental and socio-economic challenges, such as food security, land degradation, and climate change. However, land-use practice in the form of agroforestry presents an integrated and practical tool to address many of the upcoming future challenges (Nair & Garrity, 2012). Furthermore, a comprehensive assessment is needed for the estimation of the environmental and agroecological benefits of establishing the SRW in the form of agroforestry in the riparian zones of PPR wetland systems. This, in turn, will support

beneficial land management decisions in the region. The following conceptual model (Figure 1.1) provides an overview of the key concepts and hypothesized relationships among the variables that were studied in this research project; this framework is used throughout the dissertation with additional detail for each chapter provided as part of that chapter’s preface.

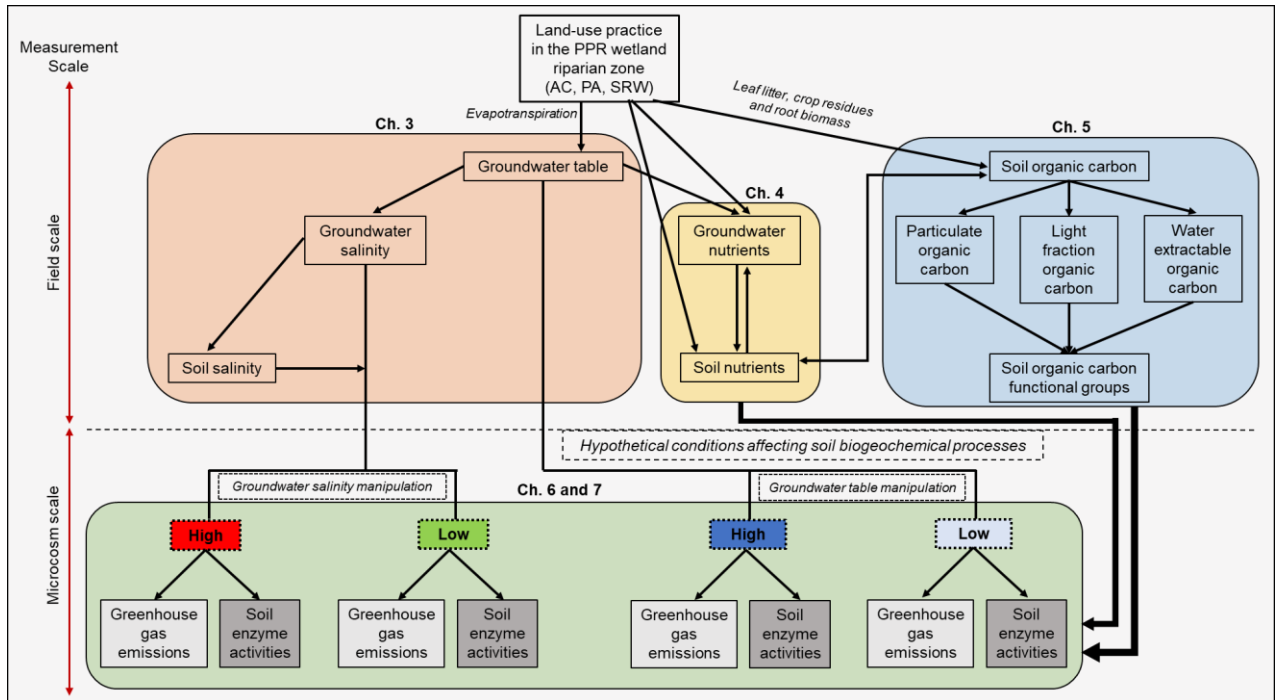


Figure 1.1 Conceptual model showing the effects of land-use practices on the measured variables in the riparian zones of the PPR wetland system from this study.

^a The arrows indicated the direction of the relationships between the variables.

^b The numbers within the same colored area relate to the chapter of the dissertation, where each variable is described.

^c AC =annual crop, PA =pasture, SRW = short rotation willow, PPR = prairie pothole region.

The dissertation is organized in a manuscript-based format consisting of five separate research studies with a general introduction, comprehensive literature review, and concluding synthesis of all significant findings.

Chapter one is the global introduction to the dissertation and lays out the objectives, research questions, and scope of the performed studies.

Chapter two presents a comprehensive review of the relevant literature on the topic. It provides an overview of prairie pothole region (PPR) agroecosystems, prevailing land-use practices, wetland soil biogeochemical cycling (with emphasis on the soil hydrology, salinity, nutrients, GHG emissions, soil EEAs), and short rotation willow (SRW) as a potential land-use practice.

Chapter three comprises the findings of a two-year field study examining spatial and temporal variability of soil hydrology and salinity under SRW plantation in the riparian zones of PPR wetland systems compared to adjacent land-use practices (i.e., pasture = PA, and annual crop = AC).

Chapter four presents the seasonal and annual variation of groundwater and soil nutrients with the two-year field study under SRW compared to AC and PA in the riparian zones of PPR wetland systems.

Chapter five compares the quantity and chemical composition of soil organic carbon (SOC) after the first rotation cycle of SRW plantation compared to contrasting riparian land-use practices (AC and PA) in the PPR wetland systems.

Chapter six, a microcosm study, quantifies the greenhouse gas emissions (CO_2 , N_2O , and CH_4) under a declining groundwater table and elevated salinity using intact soil cores collected at the end of the field experiment.

Chapter seven, also a microcosm study, assesses the EEAs [β -glucosidase (BG), N-acetyl glucosaminidase (NAG), and alkaline phosphatase (AP)] under varying groundwater table and salinity condition with the same intact soil cores discussed in chapter six.

Chapter eight synthesizes the key findings from each of the separate field and microcosm scale studies presented in this dissertation, including key conclusions and scope of future research.

2 LITERATURE REVIEW

2.1 Wetlands of the Prairie Pothole Region

Wetlands provide several essential ecosystem services, including water and nutrient retention and cycling, flood attenuation, water filtration, groundwater recharge, carbon sequestration, wildlife habitat, and biodiversity (Euliss Jr et al., 2011, Mitsch & Gosselink, 2015). Despite less than 9% of the earth's land area being covered by wetlands, they support incredibly high numbers of species (Zedler & Kercher, 2005). Even though wetlands have many ecological functions, the number and area of wetlands are decreasing globally (Verhoeven et al., 2006). Agricultural land-use is the primary cause of significant wetland losses in the world, while other causes include modification through hydrological alteration, salinization, eutrophication, sedimentation, filling, and/or invasion of exotic species (Zedler & Kercher, 2005).

The prairie pothole region (PPR) contains millions of small, isolated wetlands, generally known as potholes, left behind during the glacial retreat. They occupy topographic depressions of the hummocky and rolling semi-arid to a sub-humid climate region with potential evapotranspiration exceeds precipitation (van der Kamp et al., 2016, Winter, 1989). The PPR spans approximately 900,000 km² from North Central Iowa in the US to Central Alberta in Canada through most of Southern Saskatchewan (Figure 2.1) and comprises some of North America's most productive agricultural land (Gleason et al., 2011). Prairie potholes are important as they can facilitate the detention of runoff, reduce streamflow peaks and flooding, improve water quality, and provide significant wildlife habitat (Rosen et al., 1995). The PPR is an essential habitat for the production of ducks, which represents 10% of the total North American breeding range (Klett et al., 1988). Also, the PPR are essential features for agroecosystems that collect and store water from the surrounding landscape during rain or snowmelt and able to filter sediments and nutrients (Winter, 1989).



Source: Renton et al. (2015)

Figure 2.1 Map of the North American prairie pothole region.

Landscape features of wetlands in the PPR are changing due to natural and anthropogenic processes (Mitsch & Gosselink, 2015). The number of wetlands in the PPR has been reduced significantly since European settlement and development of agriculture on the Great Plains (Dahl, 2010). Large spans of native grasslands were typical before European settlement, but fertile soils and accessibility made the PPR ideal for agriculture. Chemical, physical, and biological alterations, primarily associated with agricultural practices, continue to threaten wetland function and integrity in this region (Guntenspergen et al., 2002). It has been reported that 71% of wetlands in the PPR have been cultivated to expand agricultural production within Canada (Mitsch & Hernandez, 2012). Furthermore, the threats to PPR wetlands are many and continue today, even when there are substantial environmental and scientific reasons for their protection and restoration.

2.2 Hydrology

“Hydrology is probably the single most important determinant of the establishment and management of specific types of wetlands and wetland processes” (Mitsch & Gosselink, 2015). The spatial variability of soil water in a wetland system is the reflection of the spatial variability of hydrological processes, which is controlled by various factors such as climate, topography, vegetation, and soil properties (Mohanty & Skaggs, 2001). Hydrology of prairie wetlands is highly sensitive to the effects of land-use practice changes (Conly & van der Kamp, 2001). Hydrology is governed by interchanges between precipitation, evapotranspiration, overland flow, and subsurface flow into and out of the wetlands; these interchanges are highly variable with space and time (van der Kamp & Hayashi, 2009, Woo & Rowsell, 1993). Hydrology of the prairie wetlands is mostly dependent on precipitation and snowmelt water (Winter, 1989). During the winter season, small depressions of the PPR collect water as snowdrifts or by surface snowmelt water runoff over frozen ground, and in the summer, water is depleted by evapotranspiration from the open water and wetland vegetation (Hayashi et al., 1998a). During

summer, precipitation is considerably less than evaporation, and thus surface runoff is rare except in extreme precipitation events and generally has no substantial influence on the wetland water balance; however, it depends on the size of collection basin, topography etc. (Conly & van der Kamp, 2001). In the PPR, the permeability of the clay-rich glacial deposits is very low, as a result, subsurface flow in and out is a small component of the water balance of most of the wetlands (van der Kamp & Hayashi, 1998, Woo & Rowsell, 1993)

Natural wetlands (usually temporary and seasonal ponds) in the PPR are generally surrounded by a ring of phreatophytic shrubs, particularly *Salix* Spp. (willow), which have an essential role in regulating the movement of shallow subsurface water through uptake and transpiration of water (Meyboom, 1966b). During summer months, vegetation in the riparian zones of the wetlands will laterally pull water and transpire it to the atmosphere (Millar, 1971), which can cause water levels to decline in pothole wetlands gradually. It has been estimated that local riparian vegetation can transpire 70% of the water that infiltrates beneath wetlands (Parsons et al., 2004). However, transpiration rates can be reduced in the absence of phreatophytic vegetation ring or if there are fallow agricultural lands (Hayashi et al., 1998a). The water losses from the wetland due to the combined effect of evaporation and transpiration are between 3-5 mm per day (Parkhurst et al., 1998, Rosenberry & Winter, 1997). However, the water loss from comparatively larger wetlands could be slower due to the lower area/shoreline ratio and larger heat storage capacity, causing less loss through transpiring vegetation (Millar, 1971).

2.3 Hydrochemistry and Salinity

The water chemistry of the PPR wetlands is highly variable in space and time and closely related to the hydrologic regime of the wetlands (LaBaugh et al., 1987). The variation in the chemical characteristics of prairie wetlands indicates that their hydrologic functions are probably complex. In the PPR, wetland water levels have distinct seasonality (Winter & Rosenberry, 1998). Hence the chemical composition of water in wetlands also varies seasonally and inter-

annually (LaBaugh et al., 1998). Due to drought and deluge, the major ion chemistry of ephemeral, temporary, seasonal, and semi-permanent wetlands can be changed (LaBaugh et al., 1996). Long-term data from the Cottonwood Lake area in North Dakota, USA suggested that a wetland that contained sulfate-type water changed to a bicarbonate type water following the deluge; conversely, another wetland that usually contained bicarbonate-type water changed to a sulfate type of water by the second year of the drought (Winter & Rosenberry, 1998). Salt dynamics in a wetland are driven by its hydrology, which cycles seasonally but also changes as a result of climatic variability, changes in land-use, and management practices (Nachshon et al., 2014). Studies have further suggested that hydrological processes in the shallow subsurface area of the PPR wetlands play a crucial role in salt transportation (Hayashi et al., 1998b, Heagle et al., 2013, Nachshon et al., 2013). However, regional groundwater flow in deep aquifers also has major impacts on salt transport and accumulation in the PPR (Euliss et al., 2004). Nachshon et al. (2013) developed a synthesized conceptual model to describe salt dynamics occurring within the glacial till part of the prairies under various land-use and climatic conditions; this model was based on the studies of Berthold et al. (2004), Hayashi et al. (1998b), Heagle et al. (2013), Keller et al. (1991).

Generally, a recharge wetland that has higher water potential than the groundwater affects the supply of solutes from the wetland, which results in relatively low salinity and shallow groundwater (Hayashi et al., 1998b, Heagle et al., 2007, Parsons et al., 2004). Lower water potential than the groundwater in a discharge wetland acts as a hydraulic barrier to prevent leaching of the quantity of solutes from the wetland, and it results in a high salinity of water (van der Kamp & Hayashi, 2009). Studies on the chemical composition of groundwater in clayey-silty glacial till indicate that salt concentrations increase from depression-focused recharge areas to discharge areas due to differences caused by microtopographically controlled ratio of vertical water flux to oxidation rate (Hendry et al., 1986, Keller et al., 1991). The dissolution of carbonate

and sulfide minerals can also change the salinity of the prairie soils (Arndt & Richardson, 1989) and is variable throughout the prairie wetlands (LaBaugh et al., 1998).

There is often no regional streamflow in the prairie region; hence studying the single wetland is an excellent way to understand the system responses to a change (Nachshon et al., 2014).

Temporal dynamics of surface and subsurface hydrology and chemistry – including salinity – affect the composition and distribution of vegetation (Kantrud et al., 1989). In the riparian zones, evapotranspiration by willow ring is a dominant pathway by which water leaves the wetland (van der Kamp & Hayashi, 2009, Woo & Rowsell, 1993). Hence, the removal or absence of willow eliminates the transpiration component leaving more water in the wetland, which leads to increased shallow groundwater flow from the wetland and can give rise to potential salinization effects on soil (Stolte, 1997, Stolte et al., 1992). Transpiration from the wetland margin can allow the deposition of carbonate and sulfate salts at the surface of the soil (Arndt & Richardson, 1993). This accumulation of salts related to water movement can occur in a variety of soil environments (Richardson et al., 1992). For example, extreme wet periods may affect the water table underneath the uplands to rise above the level of the ponds, and the resulting flow of shallow groundwater can bring dissolved salts from the surrounding soils near to the ponds (Nachshon et al., 2014).

The Great Plain lakes of Western Canada exhibit a wide range of salinities and ionic compositions in which most ions display distinct trends, both spatially and with increasing salinity (Last, 1992). The most common soluble salts in Saskatchewan soils are magnesium and sodium sulfates (MgSO_4 and Na_2SO_4); calcium sulfate (CaSO_4) also occurs but is not as easily dissolved and is less harmful (Last & Ginn, 2005). All dissolved components in soils increase with increasing salinity, but at different rates. The relative proportions of Ca^{2+} and $\text{HCO}_3^- + \text{CO}_3^{2-}$ ions decrease with increasing soil salinity, whereas SO_4^{2-} ions increase with increasing salinity (Last, 1992, Pennock et al., 2014). A meta-analysis with the chemical composition of lake data

from northern Great Plains, with a significant portion from Saskatchewan lakes, revealed that the ionic proportions of Na^+ , Mg^{2+} , K^+ , and Cl^- has no significant relationship with salinity (Last, 1992). Trends and interrelationships among major ions showed that most solutes increase in concentration with increasing total salinity; however, SO_4^{2-} and Na^+ showed the most robust relationship with the salinity, whereas Ca^{2+} and CO_3^{2-} , the weakest (Last, 1992, Last & Ginn, 2005).

2.4 Land-use Practices

Intensive agricultural land-use practices have been found to disturb the natural vegetation, soils, and wildlife throughout the PPR (Bartzen et al., 2010, Guntenspergen et al., 2002). Wetland ecosystems are also vulnerable to land-use changes in the surrounding uplands (Covich et al., 1997). Several stressors related to intensified agricultural land-use and changes in land-use practices can diminish the capability of wetland ecosystems to play significant roles in sustaining environmental quality (Rosen et al., 1995). For example, the quality and functionality of wetlands in the PPR landscapes can potentially be degraded by agricultural stressors, such as altered hydrologic cycles caused by drainage ditches (Winter, 1989), runoff of agricultural chemicals (Grue et al., 1989), nutrient inputs (Neely & Baker, 1989), tillage (McConkey et al., 2003), and sedimentation (Gleason & Euliss Jr, 1998).

A study on soil moisture and hydrology in the prairie wetlands suggested that water balance can be affected by land-use changes on the surrounding uplands (Euliss & Mushet, 1996). For example, between 1980 and 1983 about one-third of the area of the St Denis National Wildlife Area (SDNWA), in Saskatchewan, Canada was converted to a permanent undisturbed cover of smooth bromegrass (*Bromus inermis*) and some alfalfa (*Medicago sativa*) to provide improved bird nesting habitat; the remainder of the area continued in cultivation. As a result of these land-use changes, wetlands within the grassed area dried out within a few years after conversion, whereas wetlands in the neighboring cultivated area retained water as before (van der Kamp et

al., 1999). Another study from Southern Alberta on the accumulation of soil and subsoil moisture under dryland agriculture demonstrated that the soil beneath permanent grass was much drier than beneath adjacent cultivated land (Christie et al., 1985). Change in salt content was relatively insignificant in this study, yet a total of 74.0 cm of additional moisture under the cultivated area of a Dark Brown Chernozem and 36.2 cm under a Brown Chernozem to a depth of 6 m in comparison to non-cultivated sites were found. All these studies revealed that the water balance in the PPR wetlands is susceptible to changes in land management.

Within the PPR landscapes, improved agricultural management strategies are often proposed ranging from tillage and fertilization modifications to complete land-use conversion for the reduction of greenhouse gas (GHG) emissions and improved water quality (Bedard-Haughn et al., 2006a). Therefore, the adoption of any land-use practice changes should be carefully selected. For example, to offset CO₂ emissions from agricultural land-use, no-tillage is very effective as a practice because of its ability to sequester carbon in soils (Luo et al., 2010, West & Marland, 2002), although increased N₂O emissions may offset this under decreased N use efficiency and management practices (Six et al., 2004).

2.5 Short Rotation Willow: A Potential Land-use Practice

The native willow (*Salix* Spp.) vegetation surrounding “slough” or “potholes” in the PPR is popularly known as “willow rings” (Kantrud et al., 1989, Stolte, 1997). The importance of native willow vegetation associated with wetland ecosystems is emphasized, yet a significant disappearance of wetlands caused by agricultural and urban land-use changes has reduced habitat throughout the PPR (Mirck & Schroeder, 2013). The ability to grow in nutrient-limited oligotrophic sites such as bogs, sand dunes, river sand, etc. by forming symbiotic associations with mycorrhizal fungi, which provide an additional supply of nutrients for plant growth, are essential characteristics of willows as a pioneer adaptive species (Heijden & Kuyper, 2003). Despite the critical ecological role of willow in PPR wetlands, historically, the willow rings have

been over-harvested for wood chips as part of the drainage of wetlands for agricultural crop production (Schroeder et al., 2009).

Willow (*Salix* Spp.) has been increasingly used in ecological restoration work, soil erosion control, biomass production for feedstock, and energy production (Kuzovkina & Quigley, 2005). The vital physiological characteristics that affect the suitability of use of willow for environmental works include: extensive fibrous root system which reduced soil erosion (Rytter & Hansson, 1996); higher water use (Ledin, 1998); high evapotranspiration rates during the growing season (Ledin, 1998, Persson & Lindroth, 1994); efficient uptake of nutrients, high filtering capacity for nitrogen and ability to facilitate denitrification in the root zone (Aronsson & Perttu, 2001, Aronsson et al., 2000); tolerance of saturated soils and oxygen shortage in the root zone (Jackson & Attwood, 1996); drought and salinity tolerance (Vandersande et al., 2001); and tolerance of increased concentration of carbon dioxide and methane in some species (Maurice et al., 1999).

Short rotation willow (SRW) is a specific land management system of increasing interest in North America and European countries in which high-density fast-growing biomass producing crops are harvested under short rotation cycles for energy purposes (Lockwell et al., 2012). The standing stock of aboveground carbon from willow agroforestry is usually higher than the equivalent land-use, and it may also increase soil carbon sequestration potential (Guo & Gifford, 2002, Oelbermann et al., 2004, Paul et al., 2003) and increase humus content in the soil. Short rotation willow is currently being explored as a source of biomass energy in Canada; however, it is unclear whether afforestation with willow plantations will enhance or diminish soil C storage and nutrient availability (Ens et al., 2013a). The low soil nutritional status of non-amended SRW production could modify soil biological activity, microbial communities, and biogeochemical cycles (Stauffer et al., 2014). For example, the effects of SRW on soil physiochemical properties, including nutrient availability and soil carbon storage, were compared to forestry,

grassland, and agricultural land-use in a field experiment in the north of France. The results suggested that SRW could promote soil biological and physiochemical properties such as increased microbial abundance, enzyme activities, nutrient availability, and SOC compared to annual arable crops; however, in the short term, soil properties measured were lower compared to the forest and grassland soils (Stauffer et al., 2014). The SRW was recognized as both water and nutrient demanding (Ledin, 1998) and can represent a carbon sink over a long time compared to other land-use practices; therefore, it could also induce N and K depletion over the short term period (Ens et al., 2013a, Lockwell et al., 2012). Studies with SRW plantation suggested that the rate of evapotranspiration was higher than under arable crops; however, the values vary markedly and are related to site-specific factors such as local conditions in terms of soil type, temperature, precipitation, groundwater level, plant species, age of the plant, and their interactions (Dimitriou et al., 2009a, Dimitriou et al., 2009b).

The core ways of improving SOC pool for a given land area are: 1) land-use practice changes on degraded marginal agricultural land back to forest or grasslands, and 2) land management known as “best practice” such as reduction of tillage intensity on croplands, prevention of drainage, and introduction of sustainable forestry (Dawson & Smith, 2007, Janzen, 2004).

Therefore, the establishment of SRW within the marginal riparian land of the PPR agroecosystem could potentially sequester SOC, promote biological activity, and offset GHG emissions (Amichev et al., 2012, Stauffer et al., 2014). There are millions of hectares of abandoned and degraded marginal lands, including the wetland riparian zones across the PPR, which are not suitable for crop production either due to high salinity or excess moisture (Amichev et al., 2014b). More than two million hectares of degraded marginal land are available in Saskatchewan, Canada, which has the potential of C sequestration through SRW biomass production in addition to supplying fuelwood (Amichev et al., 2012, Stolarski et al., 2011).

2.6 Soil Organic Carbon: Pools, Dynamics, and Sequestration

Wetlands cover only a small share of the terrestrial land surface (less than 9%), even though it is estimated that they store 20-30% of the Earth's soil pool of carbon (Bridgham et al., 2006, Lal, 2011). Based on the location and climatic conditions, undisturbed wetlands can be both sources and sinks of soil C (Kayranli et al., 2009). The freshwater mineral soil wetlands of the PPR can lead to the accumulation of 18 Tg C yr⁻¹ because of slow decomposition under the anoxic soil environments (Bridgham et al., 2006). Nevertheless, pressure related to intensified agricultural land-use practices has been found to disturb the native vegetation with the loss of soil organic carbon (SOC) within the PPR (Janzen et al., 1998, VandenBygaart et al., 2003).

Land-use change redistributes the equilibrium between carbon inflows and outflows in an ecosystem until a new equilibrium is reached. Consequently, a soil may act either as a carbon source or as a carbon sink according to the ratio between inflows and outflows during this process (Guo & Gifford, 2002). Carbon sequestration generally occurs due to the accumulation of non-mineralizable organic materials as soil humus (Ramesh et al., 2019). Re-establishment of permanent grass in cultivated ephemeral wetlands could provide maximum C sequestration, but it is uncertain how this type of land-use change might affect GHG emissions within the Canadian Prairie landscape (Bedard-Haughn et al., 2006a). While plantation of SRW on abandoned farmland in southern Quebec, Canada acted as a soil carbon sink over the long-term when replacing no-till crop rotation but induced N depletion over the short-term; however, the conversion from abandoned alfalfa fields into SRW did not enhance soil carbon sequestration (Lockwell et al., 2012).

Soil organic carbon is a crucial component of the soil-plant ecosystem and is closely associated with soil properties and biogeochemical processes (Wang & Wang, 2011). Soil organic carbon pools can be divided according to turnover rate into a) labile, b) slow, and c) recalcitrant (Six et al., 2002b). They can also be divided by density fractionation into a) light fraction (LF) and b)

heavy fraction (HF), which can be further subdivided according to their location within the soil matrix into a) free light fraction (located between aggregates) and b) occluded light fraction (located within soil aggregates) (Golchin et al., 1994). Quantification of SOC is complicated by the fact that it is made up of numerous carbonaceous compounds that react differently to changes in agricultural management practices (Ghani et al., 2003). Also, it is challenging to distinguish changes in the short-term SOC if there are high background levels of C already present in each soil (Gregorich et al., 1994). However, compared to total SOC, labile carbon (LC) pools are relatively small and have a fast turnover rate; these pools are of interest since they tend to react disproportionately to changes affecting the balance of C mineralization and accumulation in soil (Lockwell et al., 2012). Labile carbon can be used as preliminary indicators to assess whether a new type of management will behave like a C sink or a C source when compared to the previous system (Gregorich et al., 1994). The light fraction of SOC is a useful indicator of labile C. For example, a strong correlation between the LF content and the respiration rate, N mineralization and microbial N content of soils was found in a study with soils from three long-term crop rotations in Saskatchewan, Canada (Janzen et al., 1992).

Labile carbon pools dominated by soil microbial biomass are sensitive to changes in land-use and management practices and are therefore used as a tool to assess C dynamics and soil quality under willow plantations (Lockwell et al., 2012). Changes in land-use and soil management practices lead to changes in SOC characteristics. Results from a study (Bolinder et al., 1999) revealed that light fraction-N and macro-organic matter-N were the most sensitive SOC characteristics when conservation management practices were compared to conventional management; however, these changes often occur gradually and are challenging to detect in the short term.

Changes in plant species, primary productivity, litter quantity, and quality and soil structure change under different land-use practices can have significant direct and indirect effects on

SOC pools and dynamics (Ramesh et al., 2019). However, the impact of land-use changes on organic carbon pools in the mineral soil also depends on long-term site-specific factors (e.g., climate, topography, and parent material) and is often predominated by the high spatial heterogeneity of soil organic carbon (Schwendenmann et al., 2007). A study was conducted to assess the impact of changes in land-use practices on soil carbon stocks and dynamics shortly after SRW establishment at one site and after multiple rotations at another; no significant difference in SOC was found between an abandoned alfalfa field and other cropping systems (i.e., buckwheat, 1- and 2-year old willow stands) at the establishment site (Lockwell et al., 2012). In this study, the proportion of amino sugar in the total SOC of the 1- and 2-year old SRW was higher than for both controls at depths of 0-20 cm.

The accumulation of SOC primarily depends on the input of soil organic residues and its rate of degradation (Janzen, 2006). However, the molecular structure, and biological and environmental factors such as litter quality and microbial community, are closely related to C dynamics and control the rate of SOC decomposition processes (Schmidt et al., 2011). The quality, function, and biochemical stability of SOC depend on the relative abundance of recalcitrant versus labile C chemical functional groups (Beer et al., 2008), and the chemical composition of SOC changes when the soil organic residues under different vegetation types from contrasting land-use practices decompose (Lafleur et al., 2015). Hence, the characterization of SOC may provide an insight into its nature, dynamics, and its ability to contribute to biogeochemical cycling in the agroecosystems (Ondrasek et al., 2019).

2.7 Wetland Soil Biogeochemical Cycling

Wetland soils can serve as a reservoir of both water and nutrients. Soil microorganisms drive biogeochemical cycles, and enzymes are the primary mediators of these processes, including organic matter decomposition, C and N mineralization, and elemental cycling (Ondrasek et al., 2019). Biogeochemical processes related to C and N cycling are vital as they can result in GHG

emissions as well as accumulation of C and nutrients (Mitsch & Gosselink, 2015). However, C, N, and P mineralization can vary considerably with soil moisture conditions (aerobic vs. anaerobic) and among different types of wetlands (Bridgham et al., 1998).

2.7.1 Carbon Cycling

Carbon cycling within the wetland has potential interest due to globally significant feedbacks between various aspects of wetland carbon dynamics and global change (Janzen, 2004).

Interest in the global carbon cycle has increased in recent years due to climate change, including land-use change and GHG emissions (Bridgham et al., 2006). Wetland ecosystems provide an ideal environment for the sequestration of atmospheric carbon, yet they also are natural sources of greenhouse gas emissions, mainly methane (Badiou et al., 2011, Mitsch et al., 2012). Evidence suggests that prairie wetlands can be both sources and sinks of carbon, depending on their location, climate, and hydrology (Badiou et al., 2011, Bedard-Haughn et al., 2006a, Euliss et al., 2006, Kayranli et al., 2009). Among the different pools of organic carbon (OC) in wetland ecosystems, dissolved organic carbon (DOC) is the largest, usually about ten times greater than particulate organic carbon (POC) (Dean & Gorham, 1998). However, POC is the dominant source of OC in the soil sediments.

Carbon mineralization is the process of converting organic residues into CO₂ and CH₄ as a result of microbial respiration, with subsequent evolution of the gas from the soil (Schimel & Schaeffer, 2012). In wetland systems, typically low organic carbon decomposition and mineralization occur due to high carbon accumulation rates under anaerobic conditions coupled with high vegetation productivity (Bridgham et al., 2006, Reddy & DeLaune, 2008). The rate of CO₂ production from organic residues is regulated through aerobic and fermentative respiration (Dawson & Smith, 2007). Availability of OC, plant residue composition, nutrient availability, and temperature determine the rate of OC decomposition and mineralization to DOC, CO₂, and CH₄ (Blanco-Canqui & Lal, 2004). These parameters interact with soil physiochemical factors,

including moisture, pH, redox potential, activators (e.g., cations) that influence the activity of the soil microbial community, and extracellular enzymes for organic residues decomposition (Bonnett et al., 2006). Changes in the wetland soil hydrologic conditions that regulate organic residues decomposition have the potential to change GHG effluxes, which may contribute to global climate change. In a study with sedge meadow soil in Northern wetlands, the highest carbon mineralization rate and methane production were found under anaerobic conditions (Updegraff et al., 1995). Another study found a higher organic carbon mineralization rate ($C-CO_2/C_{org}$) under SRW land-use after three years of growth as compared to the forest, grassland or arable land-uses, which suggested higher organic matter biodegradability and availability in the SRW system (Stauffer et al., 2014).

Methanogenesis is an important wetland soil biogeochemical process because CH_4 is a potent GHG (Bridgham et al., 2006) with 25 times the global warming potential of CO_2 . Generally, under strict anaerobic conditions, CH_4 (by methanogenesis) rather than CO_2 is the dominant end-product of organic residues decomposition (Dawson & Smith, 2007, Freeman et al., 2004). Wetlands are the largest source of CH_4 emission, contributing about 20% of the annual global emission to the atmosphere (Wang et al., 1996). Two major metabolic pathways can produce CH_4 ; however, in freshwater wetlands, the primary mode of production is the splitting of acetate that comes from the fermentation of organic residues (Reddy & DeLaune, 2008). Net CH_4 emission involves not only methanogenesis but also methanotrophy. The activity of both methanotrophs and methanogens are dependent on temperature, pH, water level, and nutrient availability (Rask et al., 2002). In the PPR, most wetlands are situated within agricultural fields where they are farmed except during extremely wet periods and generally receive runoff from surrounding agricultural areas (Euliss et al., 2006). Evidence suggested that historical land-use change affects CH_4 emission; for example, the conversion of cropland to perennial grassland can reduce CH_4 emission from upland soils (Dobbie et al., 1996). Hence, converting cultivated

cropland to perennial vegetation within wetland catchments may reduce nutrient enrichment buffer effects and possibly lower emissions CH_4 from wetland systems.

2.7.2 Nitrogen Cycling

Nitrogen (N) cycling processes, such as mineralization, nitrification, and denitrification, determine the retention or loss of N from the wetland (Reddy & DeLaune, 2008). The N cycle in wetlands is sensitive to many environmental factors, including hydrology and the resulting pattern of soil moisture and other soil properties (Ehrenfeld & Yu, 2012). Evidence has suggested that change in land-use practice can have both positive and negative effects on the characteristics of soil organic carbon, soil N content, and cycling (Compton et al., 1998). In particular, the microbial communities driving soil processes and N dynamics are different for agricultural versus other land-use practices (Uri et al., 2008).

Nitrogen mineralization is a microbially mediated process in which NH_4^+ is liberated from soil organic matter (Zak & Grigal, 1991). Gross N mineralization, nitrification, and consumption processes co-occur, and the amount of N released into the soil depends on the relative magnitudes of these processes (Recous et al., 1998). The quantity of inorganic N available for plant growth is regulated by N mineralization, which is mostly controlled by C/N ratio, plant litter chemistry, soil moisture, and soil temperature in wetland soils (Chapin et al., 2003, Zak & Grigal, 1991, Zak et al., 1990). Land-use management or disturbance also has a significant effect on soil N mineralization as it regulates these controlling factors (Zak & Grigal, 1991). Change in oxygen concentration associated with soil hydrologic change due to land-use management (Conly & van der Kamp, 2001) could contribute to the differences in N mineralization in wetland soils. In an incubation study at 30°C for nine weeks under waterlogged conditions, readily-mineralized N production was significantly higher in samples from plots with groundwater tables at 20 or 50 cm than in those where the water level was at the surface; the

authors suggested that the increases in N availability as a result of lowering the water table could be attributed mainly due to deeper plant rooting (Williams & Wheatley, 1988).

Nitrification is the two-step oxidative (aerobic) soil microbial process of transformation of ammonium (NH_4^+) to nitrate (NO_3^-) (Smith, 1997). The spatial arrangement and physical connection between uplands and wetlands allow for the linkage of N cycling processes. Hence, nitrification can occur within upland ecosystems, and NO_3^- which is highly mobile within the soil, can be leached or transport to wetlands systems (Zak & Grigal, 1991). Nitrification is inhibited under waterlogged conditions; however, it can occur in soil with intermediate water content (Smith, 1997). Nitrification is generally favored by more available NH_4^+ (the initial substrate for nitrification), moderate pH, and well-aerated soils, but declines as soils become very dry (Barnard et al., 2005). Nitrification rates also depend on the land-use and management and various other soil properties such as the C/N ratio, plant litter chemistry, soil moisture, and temperature variation (Zak & Grigal, 1991, Zak et al., 1990). The actual rate of conversion of NH_4^+ to NO_3^- is the gross nitrification rate and can be very high even in soils with a negligible net nitrification rate (Verchot et al., 2001). Therefore, gross N nitrification measurement may give a more precise overview of the nitrification process (Han et al., 2012). In an incubation study with soils from different land-use practices out of 15 soil parameters, soil moisture, soil temperature, NH_4^+ content, NO_3^- content, microbial biomass N, microbial respiration rate, and dissolved organic N content were found to be significantly correlated with the gross nitrification rate (Cookson et al., 2007).

Denitrification is a microbial respiratory reduction of NO_3^- and NO_2^- mainly to gaseous N_2O and N_2 in soil, which can be affected by N, temperature, and CO_2 and contribute to global change (Barnard et al., 2005). It is the primary pathway of producing N_2O , a potent greenhouse gas with 296 times the global warming potential of CO_2 that contributes to global warming and stratospheric ozone destruction (Smith, 1997). Denitrification rates are regulated by the

availability of NO_3^- , O_2 supply, and pH of the soil (Korom, 1992, Wallenstein et al., 2006).

Denitrification is generally favored by the high availability of labile C as a source of energy and NO_3^- as an electron acceptor. Wetland vegetation is an essential supplier of organic matter, hence fueling denitrification (Bastviken et al., 2005). Usually, denitrification requires anaerobic conditions; however, a study found that it has the greatest N_2O emission rate when 75% of soil pore space is filled with water (Khalil & Baggs, 2005). Fluctuations in soil hydrological conditions are reported to generate the highest N_2O emissions; 60-80% WFPS - where oxygen concentrations are not too low - is the optimum hydrological condition for N_2O emissions (Davidson et al., 2000, Pennock et al., 2010). A recent study suggested that micro-topography within the riparian zone has a significant influence on soil oxygen dynamics and, therefore, redox-sensitive biogeochemical processes such as denitrification (Duncan et al., 2013). The riparian zone could contribute more than 99% of total denitrification, yet it represents a small portion of a catchment area (Duncan et al., 2013).

In Canada, a significant proportion of total N_2O emissions are derived from the transformations of mineral N from agricultural land-use (Rochette et al., 2008). Changes in land-use practices, therefore, may have a significant influence on GHG emissions. One study found that the conversion from cultivated land to permanent grassland resulted in the most considerable reduction in N_2O emissions among other management practices due to reduced soil water availability for mineralization/nitrification and denitrification in permanent grassland (Grant et al., 2004). In another study with ephemeral prairie wetlands in central Saskatchewan, the mean N_2O emissions from cultivated wetlands during May to July decreased from 112.8 to 17.0 ng N_2O m^2 s^{-1} ; in contrast, in uncultivated wetlands, emission ranged from 31.8 to 51.1 ng N_2O m^2 s^{-1} , which suggested land-use differences in emission sources and rates (Bedard-Haughn et al., 2006b). Also, the trend of N_2O emission was correlated to water-filled pore space in cultivated

wetlands, which demonstrated the influence of soil moisture on emissions (Bedard-Haughn et al., 2006b).

2.7.3 Phosphorus Cycling

Phosphorus is removed from wetland waters or sediments through assimilation by living organisms and later released from those organisms (mineralization of organic phosphorus) through excretion, leaching, and decomposition. Phosphorus occurs in wetlands in two main forms: inorganic (as PO_4^{3-} and its polymers) and organic, either in dissolved or a portion of particulate matter (Reddy et al., 1999). In organic wetland soils, most soil P (> 95%) is in the organic form with cycling among P-forms controlled by biological forces (i.e., microbes and plants), whereas in mineral soils, most of the soil P may be bound to minerals containing Al, Fe, Ca, or Mg (Vepraskas & Craft, 2016). Precipitation as insoluble Ca or Mg-phosphates or adsorption to carbonates are the dominant transformations at $\text{pH} > 7$, and the solubility of Ca or Mg-P complexes is determined by soil or water pH rather than Eh (Moore & Reddy, 1994). Neither PO_4^{3-} -P nor organic P is volatile; thus, P loss to the atmosphere as phosphine (PH_3) gas is insignificant and unreported in prairie wetlands (Neely & Baker, 1989). Thus, most of the added P accumulates in the wetland systems. The P enrichment of P-limited systems leads to eutrophication and can cause N limitations and ecosystem stress (Reddy & DeLaune, 2008). A significant amount of P in the above-ground and below-ground biomass from the wetland vegetation is a significant source of organic P in wetlands (Reddy & DeLaune, 2008). The biotic process is the dominant pathway by which organic P is mineralized in wetlands that require enzymatic cleavage of phosphate esters through hydrolysis by extracellular enzymes such as phosphatases (Eivazi & Tabatabai, 1977). Land-use in the PPR is dominated by intensive agriculture from which Prairie wetlands frequently get high nutrient loadings, including P that is adsorbed and moves with the sediments from nutrient-rich agricultural runoff (Neely & Baker,

1989). Such nutrient rich sites can be managed by planting high P uptake and resorption willows (Da Ros et al., 2018).

2.8 Soil Enzyme Activities

Soil extracellular enzymes (EEs), such as hydrolases, play a vital role in the decomposition of SOM, transformation, mineralization, and nutrient cycling (Luo et al., 2017). A variety of soil EEs are produced by soil microorganisms to catalyze the SOC degradation, and acquire energy and nutrients (Sinsabaugh et al., 2008, Wallenstein & Burns, 2011). Soil enzymes are the primary mediators of soil processes and are considered as the keys of biogeochemical cycles, such as organic matter decomposition, C, N, P mineralization, and elemental cycling (Burns et al., 2013, Burns & Dick, 2002). Indication of soil enzyme activities can be used to describe changes in soil quality due to land-use management (Trasar-Cepeda et al., 2008) and climate-linked environmental changes (Henry, 2012) as they can perceive changes before they are detected by other soil analyses (Acosta-Martínez et al., 2007).

Studies have shown that enzyme activities are sensitive to changes in soil due to land-use practices (Acosta-Martínez et al., 2003a, Acosta-Martínez et al., 2003b, Bandick & Dick, 1999, Štursová & Baldrian, 2011, Wallenius et al., 2011); therefore it has been suggested as a potential indicator of soil health and quality (Bandick & Dick, 1999, Burns et al., 2013). Soil enzymes can provide information on the nutrient status and potential activities of soils, such as substrate availability, mineralization, and respiration activities (Burns et al., 2013, Marx et al., 2005). In an experiment, eleven soil enzymes were studied relative to soil management and soil quality at two sites, except α - and β -glucosidase and α - and β -galactosidase, activities were found higher in continuous grass fields than in cultivated fields (Bandick & Dick, 1999). Land-use practices that supply elevated levels of crop residues can significantly increase the soil extracellular enzyme activities (EEAs) (Bandick & Dick, 1999, Jordan et al., 1995). In cultivated systems, activity was higher where cover crops or organic residues were added as compared to

treatments without organic amendments, which suggested that the soil BG (β -glucosidase) activities best reflects the management effects on soil quality (Bandick & Dick, 1999). In a study with SRW compared to forestry, grassland, and agroecosystem, high phosphatase activity levels were observed in the forest plot compared to the other land-uses (Stauffer et al., 2014). In a study with soils (0-15 cm) collected from Canadian native Prairie grassland and a cultivated field 200-m away found that phosphatase (49%) and arylsulphatase (65%) were depressed due to the cultivation activities (Gupta & Germida, 1988).

Altered vegetation cover due to land-use practice change in the semi-arid PPR can alter shallow groundwater water balances through evapotranspiration and can lead to the accumulation of higher salts in the surface (LaBaugh, 1989, Stolte et al., 1992). Elevated soil salinity can reduce soil EEAs due to the direct effects of osmotic potential and specific ions on enzymes and indirectly by lowering microbial biomass (Rath & Rousk, 2015). For example, among 36 soils in an arid region of Spain, negative correlations were found between soil EC and dehydrogenase, phosphatase, urease, and β -glucosidase activities (García et al., 1994). Furthermore, alternate wet and dry conditions resulting from shallow groundwater fluctuation in a wetland soil can also affect soil EEAs. During the drought condition, the matric potential decreases in soil. Consequently, the low availability of water is often combined with high salt concentrations and causes limited resource supply and physiological stress to microorganisms as well as the biogeochemical processes (Schimel, 2018).

2.9 Significance and Objectives of this Ph.D. Research Project

The inclusion of SRW in the riparian zones of the PPR wetland systems may increase SOC sequestration; however, this does not always occur. Improved SOC storage via higher biomass production and leaf litter input, a well-managed SRW plantation could reduce GHG emissions in the marginal degraded land. However, any land-use practice or management decisions that affect wetland soil hydrology, salinity, and nutrient balance might also adversely interfere with

shallow groundwater hydrology, hydrochemistry, and biogeochemistry. Hence, the entire system may not act as a C sink and consequently affect the wetland soil biogeochemical processes.

Additionally, there are risks of alteration in soil hydrology linked with potentially higher evapotranspiration rate of SRW, which could also unfavorably affect wetland soil salinity.

Necessarily, comprehensive research is required to evaluate the introduction of a new land-use practice within the PPR agroecosystems. Thus, this research project ultimately can advance the knowledge base and provide relevant information needed to develop long term wetland monitoring systems, prediction models, and management strategies under future land-use and climate change situations.

Therefore, this Ph.D. research project aimed to evaluate the impacts of contrasting land-use practices on wetland soil hydrology and salinity, nutrients content, organic carbon sequestration, GHG emissions, and enzyme activities involved in biogeochemical cycling in the riparian zones of the PPR wetland systems. The specific objectives examined the land-use practice effects at two scales:

Field-scale

To evaluate the effects of SRW establishment in the riparian zones of the PPR wetland systems in contrast to prevalent AC and PA on:

- 1) the variability of shallow groundwater table depth and salinity, and spatial and temporal distribution of soil salinity
- 2) the quantity, and seasonal and annual variation of groundwater and soil nutrients (N, P, K, and S)
- 3) the quantity, different size and density fractions, and chemical composition of SOC

Microcosm-scale

To evaluate the response of the PPR wetland soils collected from contrasting riparian land-use practices (i.e., SRW, AC, PA) and manipulated with:

- 1) the declining groundwater table in combination with different groundwater salinity levels on GHG (CO₂, CH₄, N₂O) emissions
- 2) the declining groundwater table in combination with different groundwater salinity levels on β -glucosidase, N-acetyl glucosaminidase, and alkaline phosphatase enzyme activities

Each of the objectives stated above were evaluated through specific experiments and are presented in the subsequent five chapters in this dissertation.

3 LAND-USE EFFECTS ON SPATIAL AND TEMPORAL VARIABILITY OF RIPARIAN WETLAND SOIL HYDROLOGY AND SALINITY

3.1 Preface

Short rotation willow (SRW) is a phreatophytic plant and known to have high water demand, and its rate of evapotranspiration (ET) is higher than annual crops. However, ET can vary markedly based on the SRW variety or species, age of the plantation, and site-specific confounding factors such as precipitation, temperature, soil types, and groundwater level. Seasonal variation in groundwater table depth can occur naturally. Furthermore, anthropogenic changes due to the introduction of a new land-use practice may intensify these fluctuations and affect the hydrochemistry (particularly the salinity) of a wetland system. Hence, projection of the impact of SRW plantation in the riparian zones of Prairie wetlands is vital to promote the prospect of bioenergy cultivation in the marginal degraded land in this region. An integrated impact assessment can also help the decision-making and management of both productive agricultural land and the valuable wetland habitat in the prairie pothole region (PPR). The conceptual model (Figure 3.1) summarizes the variables that were assessed in Chapter 3 and can potentially be impacted by land-use practices.

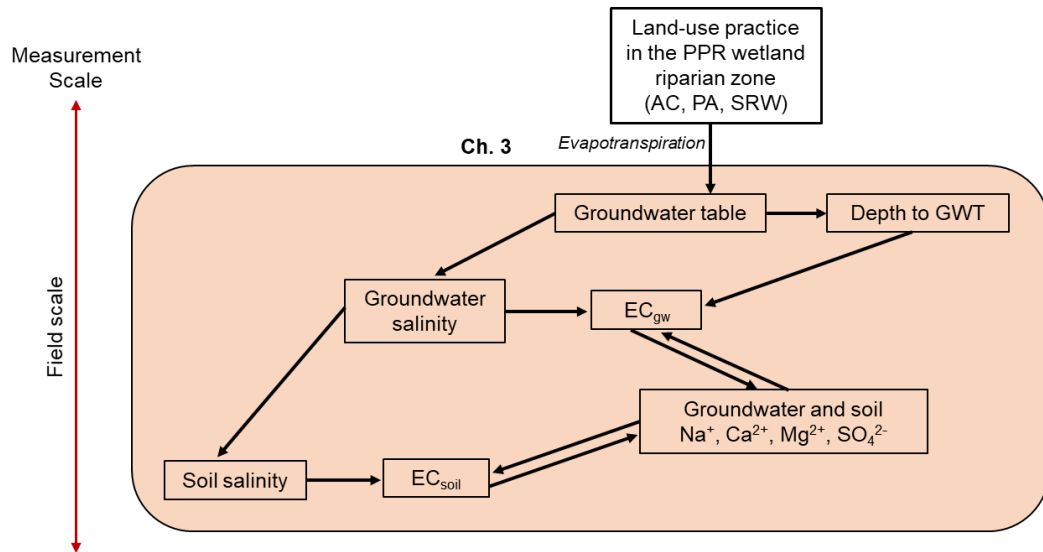


Figure 3.1 Conceptual model showing the potential effects of land-use practices on GWT, EC_{gw} , and EC_{soil} in the riparian zones of the PPR wetland system.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, GWT = depth to groundwater table, EC_{gw} = groundwater electrical conductivity, EC_{soil} = soil electrical conductivity, PPR = prairie pothole region.

3.2 Abstract

Shallow groundwater fluctuations in wetland systems occur naturally. However, anthropogenic changes such as land-use alteration in the riparian zone can further stimulate groundwater table depth variations and salts dynamics, potentially leading to soil salinization. In a multi-year field experiment, the impacts of three riparian land-use practices on shallow groundwater table fluctuations and salt accumulation (i.e., salinity) were compared in two semi-arid PPR wetland sites (A and B). The land-use practices were 1) short rotation willow (SRW), 2) annual crop (AC), and 3) pasture (PA); soil (0-60 cm depth) and groundwater salinity and depth to groundwater table (GWT) were measured via groundwater monitoring wells established along transects within each land-use. Land-use practices impacted the GWT depth significantly in site B ($p < 0.001$), but not significantly in site A ($p = 0.325$). No consistent land-use patterns were observed between sites, suggesting other factors controlled the observed GWT depth variability. Groundwater responded to precipitation patterns. Fluctuations in GWT depth were greatest between June and August due to higher precipitation and smaller during May and September

under reduced precipitation; however, it was not significant ($p > 0.05$). Higher precipitation events throughout the wet year (2014) resulted in a significant ($p < 0.05$) rise in the GWT (i.e., decreased depth to GWT) under all land-uses. The variations in groundwater electrical conductivity (EC_{gw}) were significant ($p < 0.05$) across land-uses; however, inconsistent between sites. The positive correlation between depth to GWT with groundwater and soil EC, as well as TDS, indicated higher salinity and accumulation of salts with increased depth to GWT. Soil EC_{soil} significantly ($p < 0.05$) varied among land-use practices; however, no consistent patterns were observed between sites. The variations between depths, years, and months were not significant ($p < 0.05$). Throughout the experimental field period, site B consistently exhibited higher soil EC_{soil} (two-fold) compared to site A. Nonetheless, at both depths (i.e., 0-30 and 30-60 cm), decreasing tendencies were observed in soil EC_{soil} with increasing SRW biomass and vice versa. The results from this study provide an understanding of land-use induced hydrological alteration and its implication on salinity, which provides essential context for wetland management in the PPR.

3.3 Introduction

Land-use alterations are one of the most critical anthropogenic causes of secondary salinization in the dry arid and semi-arid region (Sakadevan & Nguyen, 2010). Salinization of groundwater and soil are ancient and severe environmental problems that pose a threat to crop production and soil degradation within agroecosystems. Land-use practices, climate, and site-specific factors such as precipitation, temperature, soil types, and groundwater level are the main controls shaping the accumulation of salts (i.e., salinity) in soils at different temporal and spatial scales. A high water table with the contribution of additional dissolved salts to the shallow groundwater can further amplify soil salinity via the excess soil wetness caused by summer fallow and irrigation (Eilers et al., 1997). Additionally, certain vegetation types, such as short rotation willow (SRW), have significant potential/risk to disrupt soil water balance (i.e.,

hydrology) and can trigger soil salinization (Jobbágy & Jackson, 2007). As the groundwater table becomes shallower, capillary rise and evapotranspiration bring groundwater and solutes upwards to the root zone and increase the risk of salinization (Nosetto et al., 2013). On the other hand, land-use practices such as continuous cropping and permanent cover (e.g., growing perennial forage) in the upland area can prevent soil salinization in adjacent low-lying areas by limiting the amount of water leaching through the soil (Eilers et al., 1997).

Prairie pothole region (PPR) of the glaciated North American Great Plains consists of millions of small wetlands commonly-known as "sloughs" or "potholes" and provide crucial habitat of migratory waterfowl and productive agricultural land (Dahl, 2010). Wetlands in the PPR are situated in a semi-arid region where potential evaporation is almost double the annual precipitation (Winter, 1989). Wetlands in this region are commonly categorized into 1) recharge wetlands that recharge the groundwater from the depression-focused overland flow and snow-catch, 2) throughflow wetlands that receive and yield water from and to the groundwater system, and 3) discharge wetlands that have a small overland flow component and receive the majority of their water from groundwater discharge (Richardson et al., 1994). Like many arid and semi-arid regions of the world, salinization is a crucial issue affecting the productivity and degradation of agricultural lands in the PPR (Clearwater et al., 2016).

In the PPR landscape, the primary reason for high groundwater tables and salinization is due to three groundwater conditions: artesian discharge, evaporative rings, and hillside seeps (Henry, 2003, Henry et al., 1987, LaBaugh et al., 2018). Wetlands occupying higher landscape positions are generally non-saline due to recharging surface waters flushing salts into underlying fractured tills (Berthold et al., 2004, Hayashi et al., 1998a, b, Parsons et al., 2004), and/or filling-and-spilling of water into adjacent lower elevation wetlands (Shaw et al., 2012). In contrast, salts accumulate in low-lying areas where elevated groundwater tables create a hydraulic barrier

against flushing of solutes from the closed-basin wetlands that are enclosed by surrounding hummocks and lack surface drainage (van der Kamp & Hayashi, 2009).

Wet-dry cycles drive the complex hydrological processes that control both surface and sub-surface water dynamics (Valk, 2005) and salinity in the closed-basin PPR landscape (Heagle et al., 2013). Seasonally, mainly in the summer months, an evaporative ring of solute rich porewater can be created due to the out-seepage around the periphery of small ephemeral wetlands by a transient drawdown of the water table by transpiration of fringing phreatophytes (e.g., willow) (Meyboom, 1966b, Nachshon et al., 2013, Stolte et al., 1992, Winter & Rosenberry, 1995). There is a direct relationship between soil salinity and groundwater discharge (Arndt & Richardson, 1989). Typically, the low-lying areas where they are located below or with the same surrounding water table level are saline and known as discharge sloughs (Pennock et al., 2014). Also, the impermeable layers underneath can limit drainage and result in a discharge saline ring as the groundwater mounds around the wetlands (Stolte, 1997, Stolte et al., 1992).

Processes that accumulate soluble salts can contribute to soil salinity in susceptible areas of the landscape and hence, to the degradation of soil. Soil salinization is a natural process in the semi-arid Prairies where a soil water deficit is common, and the soil and groundwater commonly have a higher amount of mineral salts, including sodium, calcium, and magnesium sulfate (Nachshon et al., 2013). Within the Prairie region, salinity begins in a landscape where the water requirements of the existing land-use practice are lower than the snowmelt and precipitation (Heagle et al., 2013). Notably, salinization can occur rapidly when water tables rise due to wetter years. Subsequently, rapid evaporation and transpiration can remove soil water, and soluble salts become concentrated near the soil surface (LaBaugh et al., 1998).

Soil salinization can make a productive agricultural crop non-productive and reduce crop yield, thus reducing the economic returns to the farmers. The estimated annual income loss was \$257

million to Canadian farmers due to the soil salinity (Forge, 1998). However, the risk of salinization in the Canadian Prairies decreased between 1981 and 2011 primarily due to the decrease in summer fallow (7 million ha, 78 % reduction) and increased in the area of permanent cover (4.8 million ha, 14 % increase) with a most substantial portion of the change in Saskatchewan (over 3 million ha) (Bock, 2016). Nonetheless, across the Canadian Prairies, there is 4 million ha of salt-affected abandoned marginal degraded land, which is unsuitable for arable crop production, including 1.6 million ha in Saskatchewan, which has potential for salt-tolerant SRW plantation (Amichev et al., 2014b).

Reducing the risk of salinization and improving the condition of saline soil demands proper soil-water management. The primary requirement for saline soil rehabilitation and management is the leaching of soluble salts beyond the active root zone, which can be achieved by chemical amendments and engineering approaches such as surface and sub-surface drainage to control salt and water balance (Henry et al., 1987). In addition to the salty drainage water disposal, this approach requires high maintenance and capital investment. However, an alternative solution to this can be 'biodrainage,' i.e., the use of plant species with high growth capacity and deep root systems (Minhas & Dagar, 2016, Stirzaker et al., 1999). According to Miller et al. (1981), the possible best solution is to better utilize the soil water through several successful management practices such as 1) growing deep-rooted plants (e.g., perennial crops), 2) flexible intensive cropping systems (e.g., reducing summer fallow), and 3) draining selected upland, freshwater wetlands. Hence, the process of soil salinization associated with high water tables might be better managed using deep-rooted phreatophytic SRW agroforestry land-use practice (Dagar & Minhas, 2016). However, it is imperative to assess how SRW water use varies under specific site conditions, considering evaporative demands, soil type, depth to the groundwater table, and linked salinity (Dimitriou et al., 2009b, Minhas & Dagar, 2016).

Reducing the risk of soil salinization requires a spatial and temporal assessment of risks and further development and implementation of beneficial management practices (BMPs) (Bock, 2016). The inclusion of a new land-use practice, e.g., SRW – with high biomass production and deep rooting phreatophytic nature – in the wetland riparian zones requires precise knowledge of the consequence on spatial and temporal variation of the shallow groundwater table and salinity. The objectives of this multi-year field study were to assess 1) whether the introduction of SRW affected the fluctuation of shallow groundwater table temporally and spatially, and 2) whether associated groundwater table fluctuations had any impact on the temporal and spatial distribution of soil salinity, compared to adjacent annual cropping (AC) and pasture (PA) land-use in the riparian zones of two PPR wetland sites. It has been hypothesized that under SRW: 1) water table drawdown would be higher (i.e., greater depth to groundwater table), 2) soil salinity and salt accumulation would be lower at the upper soil layer (0-30 cm) compared to the deeper (30-60 cm) soil layer, and 3) soil salinity would gradually decrease over time.

3.4 Materials and Methods

3.4.1 Study Site

The experimental field sites were established in two adjacent PPR wetlands (distance between the boundary of the two sites is approximately 200 m) in Indian Head, Saskatchewan, Canada (N 50° 30.605' and W 103° 43.011') (Figure 3.2). The approximate extent of the area (estimated from FlySask2.ca) of site A was 1.8 ha, and site B was 1.3 ha. Saskatchewan Soil Survey Staff (1986) described the soil of both sites as non-calcareous Black Chernozems of the Oxbow association, poorly drained in depressions, with level to gentle rolling (0-10 % slopes) topography formed on loamy glacial till. In 2013, the soil salinity (0-60 cm depth) at site A (mean = 0.92, maximum = 2.11, and minimum = 0.41 mS cm⁻¹) was approximately half of that at site B (mean = 2.07, maximum = 3.39, and minimum = 0.44 mS cm⁻¹); salinity at both sites was within the range of previously reported values suitable for the adaptation and growth of SRW variety in

the Canadian Prairie (Hangs et al., 2011). Site A can be categorized as non-saline, and site B as weakly saline (Saskatchewan Soil Survey Staff, 1986). The 30-year climate norms (1981 to 2010) were 428.4 mm for annual total precipitation (321.7 mm rainfall and 110.5 mm snowfall), and +2.7°C for average annual temperature, with minimum and maximum values of -20.1 and +25.0 °C respectively at the Indian Head, Saskatchewan, Environment Canada Climate Station (Environment Canada, 2020).

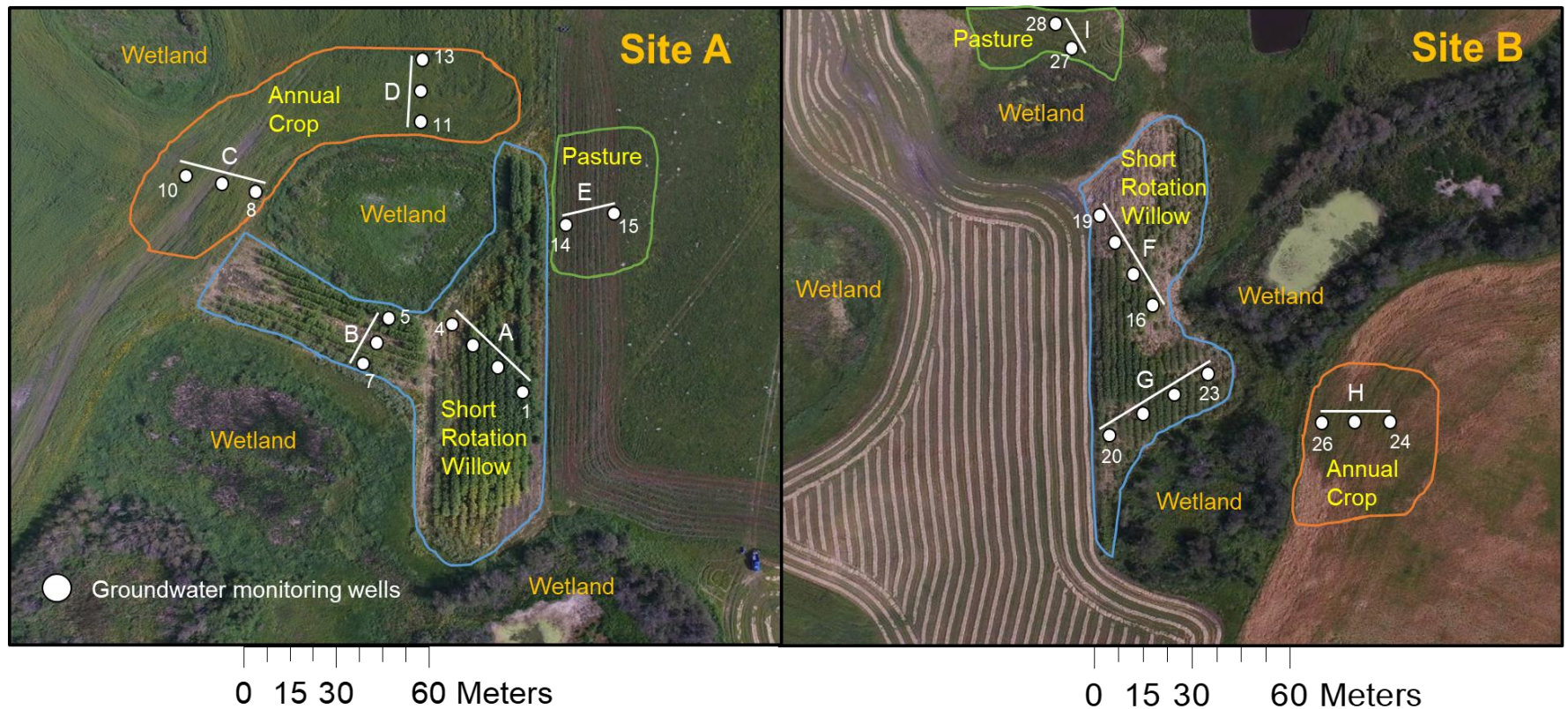


Figure 3.2 Unmanned aerial vehicle (UAV) photograph taken in the Fall of 2015, showing two experimental sites in Indian Head, Saskatchewan, Canada.

^a White dots with black circles indicate the position of shallow groundwater monitoring wells, and capital letters along straight lines indicate transects of soil and groundwater sampling.

The short rotation willow variety *Salix dasyclados* Wimm. (cultivar's popular name is 'India') was planted adjacent to pasture (PA) and annual crop (AC) in the riparian zones (Hayashi & Rosenberry, 2002) of both wetland sites on 10th and 11th June of 2013. The estimated planting area of SRW was 0.46 ha in site A, and 0.52 ha in site B (Figure 3.2). The dormant hardwood cuttings of SRW were approximately 25 cm long and planted by inserting into the rotovated soil under black plastic mulch in a double row design (13,300 plants ha⁻¹) (Figure A-S13). The distance between double rows was 2 m, and the rows were 30 cm apart. No fertilizer was applied to the sites after SRW planting occurred; however, Glyphosate (StartUp®; 540 grams acid equivalent per liter, present as potassium salt) was used two to three times during the growing season to inhibit weed growth between the rows. In the past, at both SRW sites, only barley and oats had been grown, and the land may have been under fallow once or twice during the ten years prior to SRW planting. Typical crops in the study sites are barley, oats, or flax, as the land area of both sites are slightly saline (Mirck & Schroeder, 2013, 2018).

The unmanaged PA had been established 10-12 years prior with an alfalfa (*Medicago sativa*) and brome grass (*Bromus madritensis*) mixture and typically only subjected to light grazing in early May for three to four weeks. During the study period (2013-2015), the AC was seeded to oats (*Avena sativa*); during the previous ten years, the AC land had been cultivated with barley, oats, and flax. The oats received 100 lbs of 50-20-0 fertilizer at seeding. Glyphosate (StartUp®; 540 grams acid equivalent per liter, present as potassium salt) was applied before sowing, and Prestige™ was used on the crop in June for broadleaf (and volunteer flax) weed control at recommended rates. The oats were seeded between May 7-15 and harvested during September 5-20 each year.

The SRW aboveground biomass components were manually harvested after three consecutive growing seasons in October 2015 and considered as a 3-year rotation cycle (including the

plantation year, i.e., 2013). Harvesting of SRW biomass was completed using brush saws in a 2-m by 2-m grid in each site with three replications.

3.4.2 Groundwater Table and Electrical Conductivity Monitoring

A total of 28 shallow groundwater table monitoring wells (15 in site A, and 13 in site B) were installed along transects across the sites (Figure 3.2). Each transect either extended from the field boundary to the wetland edge and/or from one wetland edge to another to cover all landforms and was parallel to the groundwater flow direction. The wells were constructed with PVC tubing (6-cm diameter, 2-m length) screened with 0.5 cm holes equally distributed throughout and sealed with a bottom cap (Figure A-S11). The entire PVC well was wrapped with porous woven fabric (nylon) to prevent intrusion of soil sediments into the wells during groundwater monitoring. Wells were installed in July 2013 by drilling a 7-cm diameter borehole to 2-m depth with a Giddings soil corer (Giddings Machine Company, Windsor, Colorado, USA) mounted onto a tractor (Model # 3120; John Deere, Moline, Illinois, USA). The PVC monitoring wells were then inserted into the borehole and sealed with 20 cm of pelletized bentonite and soil mixture around the well stock. The opening of the well was covered with a loosely fitted detachable PCV cap.

Depth to groundwater was monitored using a Mini-Diver data logger (Schlumberger Water Services, Kitchener, Ontario, Canada), installed into each groundwater monitoring well (Figure 3.2 and A-S11). Each of the data loggers was attached with a fishing line to the cap of the monitoring well. Groundwater table data were collected continuously (at 30-min intervals) throughout two consecutive growing seasons (May to September 2014 and 2015). A Baro-Diver (Schlumberger Water Services, Kitchener, Ontario, Canada) was placed at the soil surface to correct barometric measurements using the water column above the mini-divers. For construction and installation of shallow groundwater wells and water table monitoring, procedures were followed as described in USACE (2005) and Sprecher (2008).

Groundwater electrical conductivity (EC_{gw}) was monitored after collecting groundwater samples (please see the sample collection procedure described below) from each monitoring well and measured in the laboratory using a PC700 pH/mV/conductivity meter (Oakton, Vernon Hills, IL, USA). Contour maps (Figure A-S1, S2, S3, and S4) were developed to reveal the spatial distribution of soil EC_{soil} in contrast to the depth of groundwater table and groundwater EC_{gw} for each site and sampling dates during 2014 and 2015 by ordinary kriging using ArcGIS v10.5 (ESRI, Redlands, California, USA).

An on-site weather station (Campbell Scientific Canada, Edmonton, Canada) was installed to measure climatic variables during the growing seasons of 2014 and 2015. Air temperature and relative humidity were recorded on an hourly basis. Monthly total precipitation and average air temperature were calculated from the obtained climatic data.

3.4.3 EM38 Survey for Soil Electrical Conductivity and Salinity Mapping

In September of 2014 and 2015, soil apparent electrical conductivity (EC_a) across both sites was measured with an electromagnetic induction meter (EM38-MK2; Geonics Limited, Mississauga, ON, Canada) attached to an All-Terrain Vehicle (ATV) (Figure A-S12). The EM38 is specially developed to measure EC_a with two modes: vertical (response comes from 1 m soil depth) and horizontal (response comes from 0.5 m soil depth) (McNeill, 1980). A vertical (up to 1 m) survey was conducted to capture EC_a of the root zone under all land-uses except PA in 2014 at both sites. Between-row EM38 measurements were taken under SRW and AC; however, for PA, 1-m spacing was maintained. All measurements were taken following standard calibration (McNeill, 1984). Data were calibrated through the correlation developed between temperature adjusted EC_a and salinity (EC) measured from the collected soil samples in the laboratory (soil to water ratio of 1:2) to improve the reliability. Obtained EM38 data were used to create soil salinity maps using ArcGIS v10.5 (ESRI, Redlands, California, USA) ordinary kriging (Figure 3.6 and 3.7) (Cassel, 2007).

3.4.4 Elevation Survey and Mapping

Universal Transverse Mercator (UTM) coordinates and corresponding elevations were taken from the field experimental sites in a 1-m by 1-m grid using a Leica GS15 (Leica Geosystems, Heerbrugg, Switzerland) real-time kinetic Global Positioning System (rtkGPS). The elevation data were collected from three different land-use practices from both sites during the Fall of 2015. Based on the rtkGPS surveys, digital elevation models (DEMs) were created in ArcGIS v10.5 (ESRI, Redlands, California, USA) using ordinary kriging (Figure 3.6 and 3.7).

3.4.5 Groundwater and Soil Sample Collection

Groundwater samples were collected (Figure A-S12) monthly using a Masterflex E/S Portable Sampler – 115 VAC (Cole-Parmer® Instrument Company, Montreal, QC Canada) as procedures described in Vail et al. (2013). Immediately before sampling, each groundwater monitoring well was purged using the peristaltic pump, after which it was allowed to recharge to a representative part of the groundwater. The groundwater samples were collected in 250-mL polypropylene sample bottles and placed in coolers for transport to the laboratory, where they were refrigerated at 4°C until analyzed (laboratory analyses details below).

Soil samples were collected in May, July, and September of 2014 and 2015 within 1-m radius of each monitoring well (Figure 3.2 and A-S12); background samples were collected in 2013 (the SRW plantation year) from the same locations (prior to monitoring well installation). At each point, an auger was used to collect soil samples from three depth increments: 0-15 cm, 15-30 cm, and 30-60 cm. Samples were immediately transferred into a Ziploc® bag, labeled, and stored temporarily in a cooler for transport to the laboratory, where they were refrigerated at 4°C until analyzed.

3.4.6 Laboratory Analyses of Groundwater and Soil Samples

Groundwater and soil samples were analyzed for pH, electrical conductivity (EC), sodium (Na^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), and sulfate (SO_4^{2-}). Additionally, volumetric soil water

content, soil texture, and cation-exchange capacity were measured for each soil sampling point, and bulk density (please see materials and methods section of Chapter 5 for details analysis procedure) was measured at each site under three different land-use practices. All collected soil samples were subdivided into two portions. The first portion was kept intact (i.e., field moist sample); gravimetric water content was calculated from the weight loss of approximately 20-g soil sample oven-dried for 24 hours at 105°C in an aluminum tin (Topp et al., 2008). The volumetric soil water content (VSWC) was calculated from gravimetric water content and soil bulk density. The second portion was air-dried, ground, and passed through a 2-mm sieve for particle size distribution, cation exchange capacity (CEC), pH, electrical conductivity (EC), and ammonium acetate extractable Na^+ , Ca^{2+} , Mg^{2+} , and SO_4^{2-} . The particle size distribution was determined by the modified pipette method (Kroetsch & Wang, 2008). CEC was measured by the ammonium acetate methods at pH 7 (Hendershot et al., 2008a). Soil pH was determined in 20 mL of deionized water with 10 g of air-dried soil samples (2 : 1 ratio) by digital pH meter (PC700 pH/mV/conductivity, Oakton, Vernon Hills, IL, USA) (Hendershot et al., 2008b). Soil EC determined from the same extract used for pH measurement after 1 hour shaking with an end-over-end shaker, then filtered through the highly retentive filter (No. 42, Whatman Inc., Piscataway, NJ), and measured using a digital EC meter (PC700 pH/mV/conductivity, Oakton, Vernon Hills, IL, USA) (Miller & Curtin, 2008). The Na^+ , Ca^{2+} , Mg^{2+} , and SO_4^{2-} were measured using a 1M ammonium acetate (buffered at pH 7) extraction (Hendershot et al., 2008a) and analyzed by atomic emission (Na^+) and by atomic absorption (Ca^{2+} and Mg^{2+}) spectroscopy (Varian Spectra 220 Atomic Absorption Spectrometer; Varian Inc., Palo Alto, CA, USA). The SO_4^{2-} analysis was done via microwave plasma-atomic emission spectrometry (4100 MP-AES; Agilent Technologies, Melbourne, Australia). Total dissolved salts (TDS) were calculated from the measured EC with a conversion factor of 640 for EC values between 0.1 to 5 mS cm^{-1} , and 800 for EC > 5 mS cm^{-1} (Bresler et al., 2012). Soil exchangeable sodium percentage (ESP) and

sodium adsorption ratio (SAR) were calculated by following equations 3.1 and 3.2 (Abrol et al., 1988), respectively, as follows:

$$ESP = \frac{\text{Exchangeable Na}^+}{\text{CEC}} \times 100 \dots\dots\dots (3.1)$$

$$SAR = \text{Na}^+ / \sqrt{\frac{\text{Ca}^{2+} + \text{Mg}^{2+}}{2}} \dots\dots\dots (3.2)$$

3.4.7 Statistical Analysis

Statistical analyses and data visualization were completed using R version 3.4.4 for Windows (R Core Team, 2018). Data visualization was performed through bar plots, box plots, and line graphs using the "ggplot2" package. Data were tested for normality by the Shapiro-Wilk test and histogram. Homogeneity of variances or homoscedasticity was tested by Levene's test using the "car" package. Pearson correlation among the measured groundwater and soil variables were performed using the "psych" package. When required, the square root transformation was performed to improve the assumption of normality and homoscedasticity of the data. Analysis of variance (ANOVA) with linear mixed-effects models was used to test for significant differences across land-use practices, years, and depths (for soil only) in measured variables using the "lmerTest". The mixed approach was selected due to its suitability for unequal variances, nestedness, and unbalanced design. Mean comparisons of quantified variables were accomplished using Tukey Honest Significant Differences (TukeyHSD) test via "TukeyC". A principal component analysis was performed using "FactoMineR" and "factoextra" to investigate the relationship among relevant groundwater and soil characteristics measured under different land-use practices, years, and depths (for soil only). In the PCA biplot, the angle (cosine) of the between-variable vectors indicated the correlation strength among variables, and arbitrary groupings were based on an angle of 30°. Vectors of directly correlated variables point in the same direction, and indirectly correlated variables point in the opposite direction, while vectors at right angles to each other are uncorrelated.

3.5 Results

3.5.1 Variation in Groundwater Table and Salinity

In terms of GWT, no consistent land-use patterns were observed between sites. The depth to GWT significantly varied among the land-use practices within site B ($p < 0.001$) but was not significant within site A ($p = 0.325$) (Table 3.1), indicating other factors were controlling the observed variability. The depth to GWT was significantly higher ($p < 0.001$) in 2014 than 2015 under all land-use practices in both sites, suggesting that the rise in the GWT was possibly caused due to the higher precipitation events throughout the wet year (i.e., during 2014) (Figure 3.3 and 3.4). The spatial variation of depth to GWT was affected by wet versus dry years (i.e., wet = 2014, and dry = 2015), with the corresponding groundwater and soil EC in both sites shown in Figure A-S1, S2, S3, and S4. In both years, the most significant fluctuations in GWT depth were observed between June and August, when the precipitation was highest (Figure 3.3 and 3.4), revealing that groundwater responded to precipitation patterns.

Table 3.1 Analysis of variance (ANOVA) with a linear mixed-effects model for measured depth to groundwater table and groundwater electrical conductivity from three land-use practices at two sites during the growing season of two consecutive years (2014 and 2015).

Response variable	Sources of variation	Site A			Site B	
		df	F - value	p - value	F - value	p - value
Depth to GWT	Land-use	2	1.232	0.325 ^{ns}	10.281	<0.001 ^{***}
	Year	1	44.091	<0.001 ^{***}	43.737	<0.001 ^{***}
	Month	4	2.305	0.061 ^{ns}	1.133	0.345 ^{ns}
EC _{gw}	Land-use	2	16.859	<0.001 ^{***}	24.353	<0.001 ^{***}
	Year	1	0.607	0.437 ^{ns}	5.167	0.025 [*]
	Month	4	2.062	0.089 ^{ns}	3.637	0.008 ^{**}
TDS	Land-use	2	16.912	<0.001 ^{***}	10.281	<0.001 ^{***}
	Year	1	0.540	0.464 ^{ns}	43.737	<0.001 ^{***}
	Month	4	2.195	0.073 ^{ns}	1.133	0.345 ^{ns}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b Data was transformed to square root.

^c EC_{gw} = groundwater electrical conductivity, GWT = depth to groundwater table, TDS = total dissolved salts.

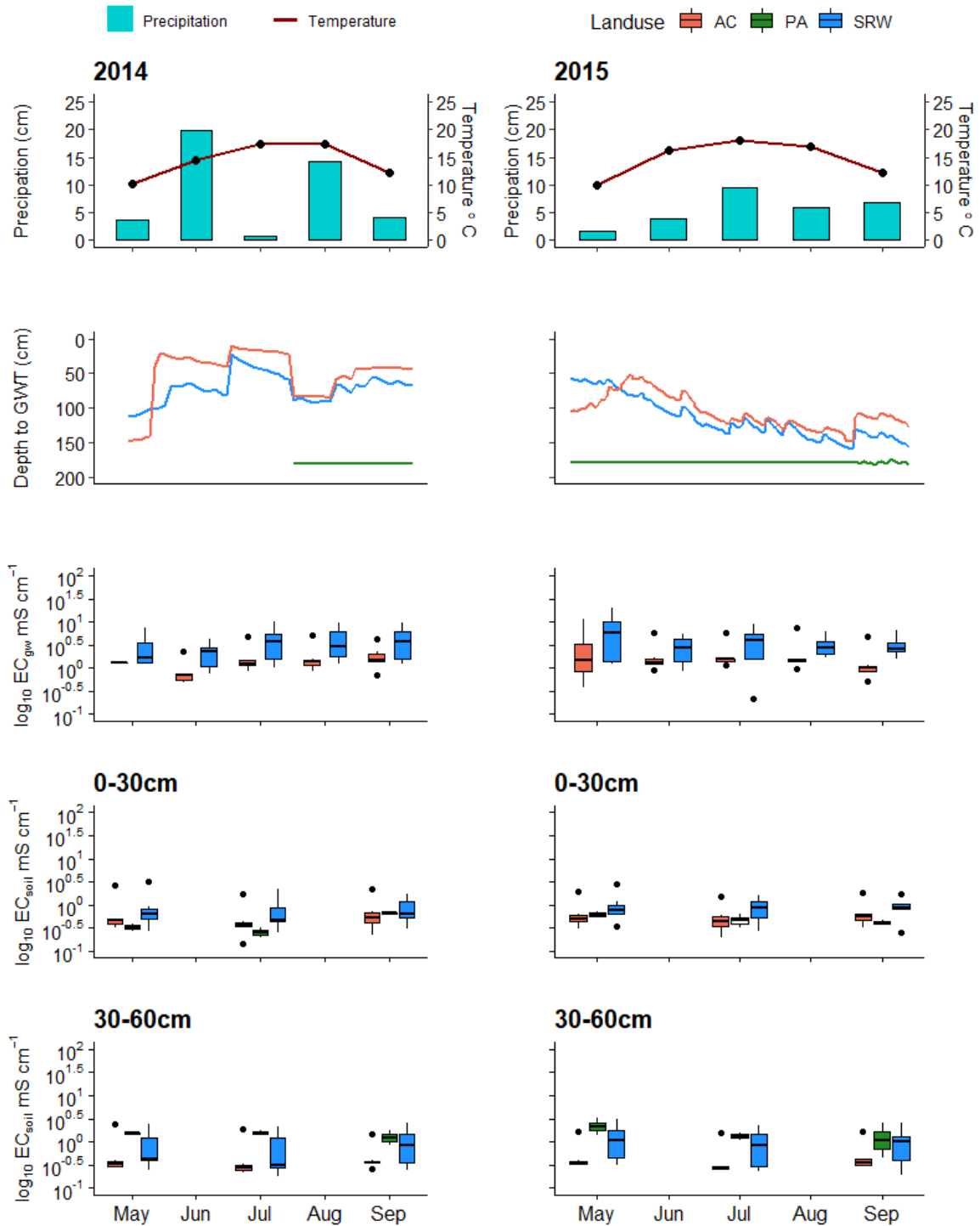


Figure 3.3 Temporal variation of precipitation, temperature, depth to the groundwater table, groundwater and soil electrical conductivity under three different land-use practices in two consecutive growing seasons of 2014 and 2015 from site A.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, GWT = depth to groundwater table, EC_{gw} = groundwater electrical conductivity, EC_{soil} = soil electrical conductivity.

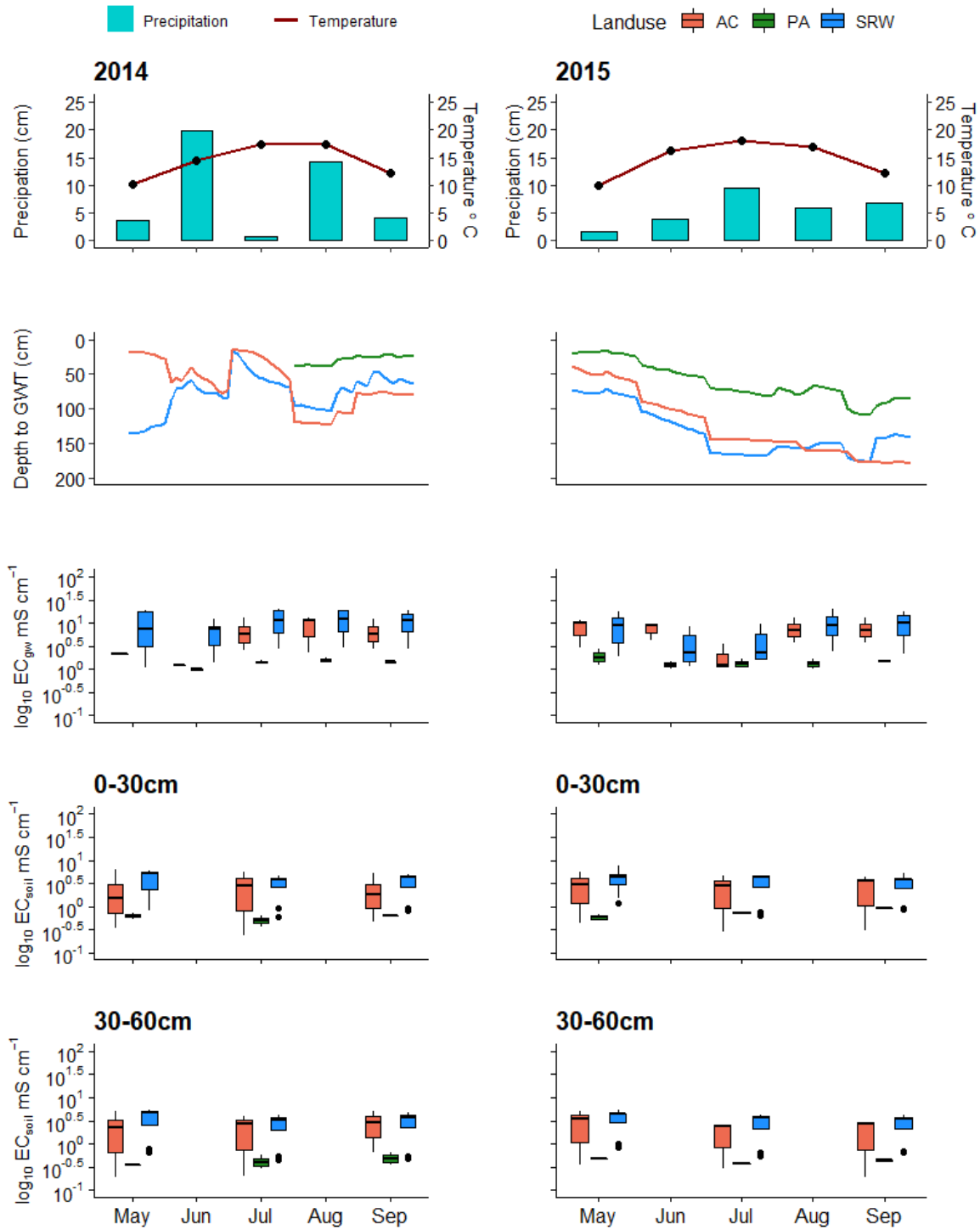


Figure 3.4 Temporal variation of precipitation, temperature, depth to the groundwater table, groundwater and soil electrical conductivity under three different land-use practices in two consecutive growing seasons of 2014 and 2015 from site B.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, GWT = depth to groundwater table, EC_{gw} = groundwater electrical conductivity, EC_{soil} = soil electrical conductivity.

Like depth to GWT, the variations in EC_{gw} among land-use practices were significant ($p < 0.001$) (Table 3.1), but the effect was not consistent between sites, suggesting that other underlying soil factors controlled the observed variability. Site B showed higher EC_{gw} (two-fold) than site A. The EC_{gw} varied significantly ($p < 0.05$) between years and among months in site B, but not significantly ($p > 0.05$) in site A (Table 3.1).

A significant positive correlation ($r = 0.40$, $p < 0.001$) was observed between depth to GWT and EC_{gw} in site A, whereas the correlation was positive but non-significant ($r = 0.14$, $p > 0.05$) in site B, suggesting that the EC_{gw} and TDS increased with the increase in depth to GWT (Figure A-S5). The correlations between the depth to GWT and groundwater Na^+ , Ca^{2+} , Mg^{2+} , and SO_4^{2-} were positive and significant ($p < 0.05$) in both sites (Figure A-S5). Significant positive correlations ($r = 0.63$, $p < 0.001$ in site A, and $r = 0.18$, $p < 0.05$ in site B) between the depth to GWT and elevation indicated that higher elevation resulted in the lowered GWT depth (i.e., higher depth to GWT) (Figure A-S5).

No significant differences were observed between sites ($p = 0.430$) and among landforms ($p = 0.056$) in terms of SRW biomass (Figure A-S6A and B). The relationships were variable between SRW biomass with depth to GWT and EC_{gw} (Figure A-S7A and B).

Principal component analyses (PCA) based on land-uses, years, and contributions of measured variables are shown in Figure 3.5. The PA land-use showed a distinct clustering in both sites but had a different pattern in each site (Figure 3.5A and D), whereas the years did not show any clustering (Figure 3.5B and E). Figure 3.5C and F represents the grouping and contribution (in percentage) of different measured variables for groundwater corresponding to the principal components. The highest contribution to both PC1 and PC2 in site A was from soil Mg^{2+} followed by Na^+ , EC, Ca^{2+} , SO_4^{2-} , clay, elevation, bulk density, pH, whereas in site B, Mg^{2+} followed by Ca^{2+} , EC, Na^+ , elevation, bulk density, clay, pH, SO_4^{2-} , and GWT, respectively (3C and F).

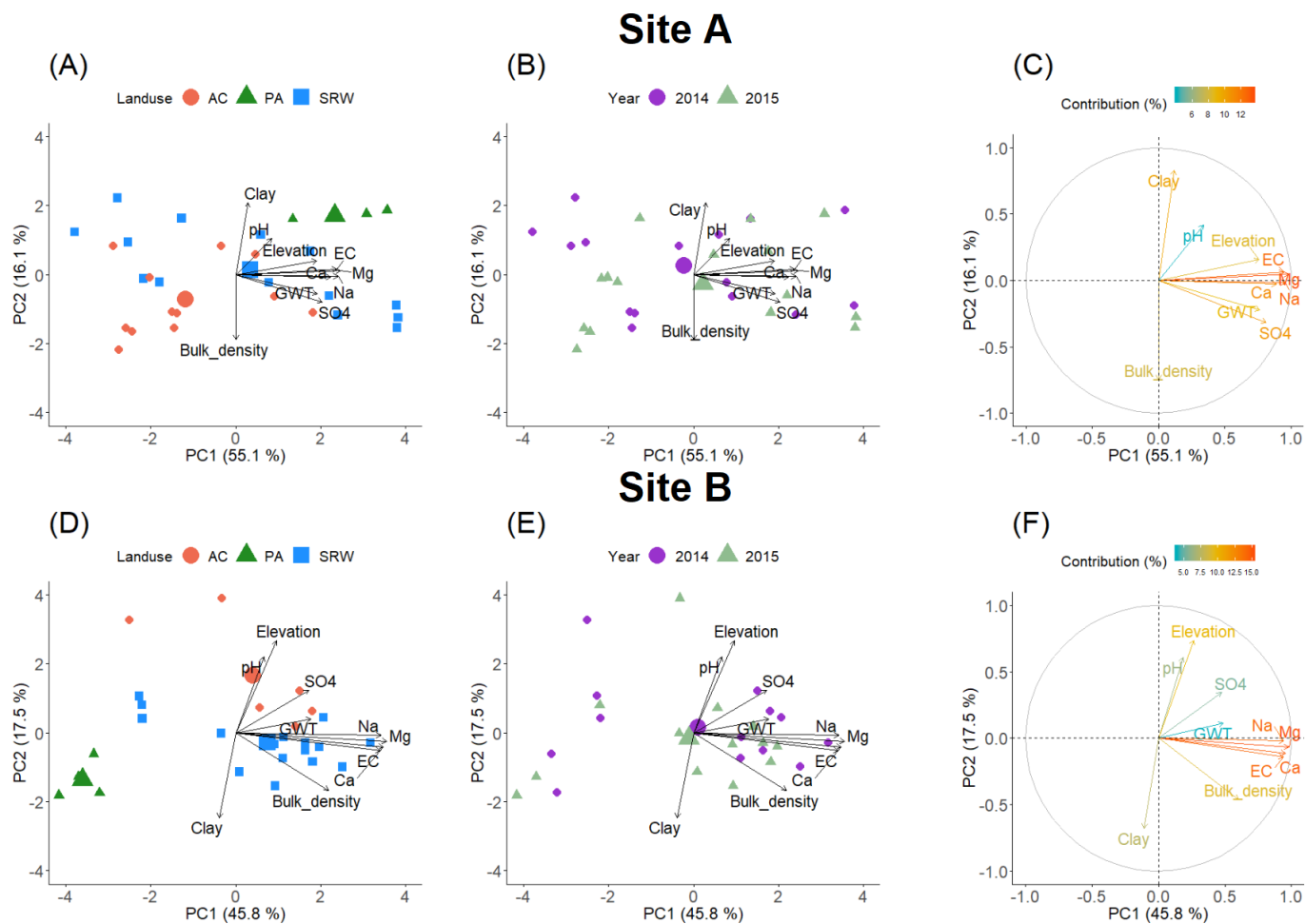


Figure 3.5 Principal component analysis (PCA) of measured groundwater table depth, EC, pH, sodium, calcium, magnesium, sulfate, clay content, elevation, and soil bulk density as observed variables based on land-use practices, years as factor variable, and contribution of variables, respectively.

^a Data was transformed to square root.

^b Contribution (in percentage) of the variables to the principal components.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, EC = electrical conductivity, GWT = depth to groundwater table.

3.5.2 Variation in Soil Salinity

Soil EC significantly differed ($p < 0.001$) among land-use practices (Table 3.2), but no consistent patterns were observed between sites (Table A-S2). No significant differences ($p > 0.05$) were observed between depths, years, and months in both sites (Table 3.2). Site B consistently showed higher EC (two-fold) than site A. Total dissolved salts (TDS) followed the identical patterns as measured soil EC (Table 3.2 and Table A-S2). The correlations between EC and SO_4^{2-} were positive ($r = 0.74$ in site A, and $r = 0.77$ in site B) and significant ($p < 0.001$) at both sites (Figure A-S8).

Table 3.2 Analysis of variance (ANOVA) with a linear mixed-effects model for measured volumetric soil water content, electrical conductivity, and exchangeable sodium percentage in soils from different land-use practices from two sites during the growing season of two consecutive years (2014 and 2015).

Response variable	Sources of variation	Site A		Site B		
		df	F - value	p - value	F - value	p - value
VSWC	Land-use	2	2.419	0.092 ^{ns}	3.901	0.022 [*]
	Depth	1	19.040	<0.001 ^{***}	26.384	<0.001 ^{***}
	Year	1	4.023	0.046 [*]	3.489	0.064 ^{ns}
	Month	1	5.411	0.005 ^{**}	1.059	0.349 ^{ns}
EC _{soil}	Land-use	2	8.961	<0.001 ^{***}	63.357	<0.001 ^{***}
	Depth	1	0.997	0.320 ^{ns}	2.519	0.115 ^{ns}
	Year	1	0.776	0.380 ^{ns}	0.001	0.978 ^{ns}
	Month	2	1.218	0.298 ^{ns}	1.718	0.183 ^{ns}
TDS	Land-use	2	8.961	<0.001 ^{***}	63.357	<0.001 ^{***}
	Depth	1	0.997	0.320 ^{ns}	2.519	0.115 ^{ns}
	Year	1	0.776	0.380 ^{ns}	0.001	0.978 ^{ns}
	Month	2	1.218	0.298 ^{ns}	1.718	0.183 ^{ns}
ESP	Land-use	2	14.945	<0.001 ^{***}	26.152	<0.001 ^{***}
	Depth	1	1.385	0.241 ^{ns}	9.328	0.003 ^{**}
	Year	1	9.267	0.003 ^{**}	0.134	0.715 ^{ns}
	Month	2	9.018	<0.001 ^{***}	2.029	0.135 ^{ns}
SAR	Land-use	2	14.069	<0.001 ^{***}	26.152	<0.001 ^{***}
	Depth	1	0.402	0.527 ^{ns}	9.328	0.003 ^{**}
	Year	1	12.364	<0.001 ^{***}	0.134	0.715 ^{ns}
	Month	2	14.960	<0.001 ^{**}	2.029	0.135 ^{ns}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b Data was transformed to square root.

^c VSWC = volumetric soil water content, EC_{soil} = soil electrical conductivity, TDS = total dissolved salts, ESP = exchangeable sodium percentage, SAR = sodium adsorption ratio.

A decreasing trend was observed in soil EC with increasing SRW biomass at the depth of 0-30 cm ($R^2 = 0.56$, $p = 0.053$ from site A, and $R^2 = 0.81$, $p = 0.002$ from site B) and 30-60 cm ($R^2 = 0.02$, $p = 0.635$ from site A, and $R^2 = 0.64$, $p = 0.017$ from site B) from both sites (Figure A-S9C and D). However, the opposite situation might be possible, i.e., the SRW biomass production was lower under high soil salinity.

The ESP significantly ($p < 0.001$) differed among land-use practices in both sites (Table 3.2) and followed similar patterns to soil EC (Table A-S2). However, none of the ESP values from this field study surpassed the critical soil sodicity value, i.e., $ESP > 15$ to be considered as sodic soil.

The spatial distribution of apparent soil electrical conductivity (EC_a) from EM38 survey showed higher soil EC_a in site B, which confirm higher overall soil salinity (i.e., EC_{soil}) compared to site A (Figure 3.6 and 3.7). However, correlations ($r = 0.03$ in site A, and $r = 0.01$ in site B) between site elevations and EC_{soil} were not significant ($p > 0.05$), suggesting that the soil salinity was not spatially dependant on the elevation at either of the experimental sites (Figure A-S10A and B).

Principal component analyses (PCA) did not show apparent clustering of soil variables based on land-use practices or year in site A (Figure 3.8A and B). However, in site B, PA land-use practice showed a slight clustering, whereas there was no clustering based on the year (Figure 3.8D and E). The highest contribution (in percentage) to both PC1 and PC2 in site A was from soil Na^+ followed by EC, Mg^{2+} , VSWC, SO_4^{2-} , ESP, elevation, Ca^{2+} , pH, clay. Whereas, in site B, EC had the highest contribution, followed by Na^+ , ESP, SO_4^{2-} , elevation, pH, bulk density, Ca^{2+} , Mg^{2+} , and clay, respectively (Figure 3.8C and F).

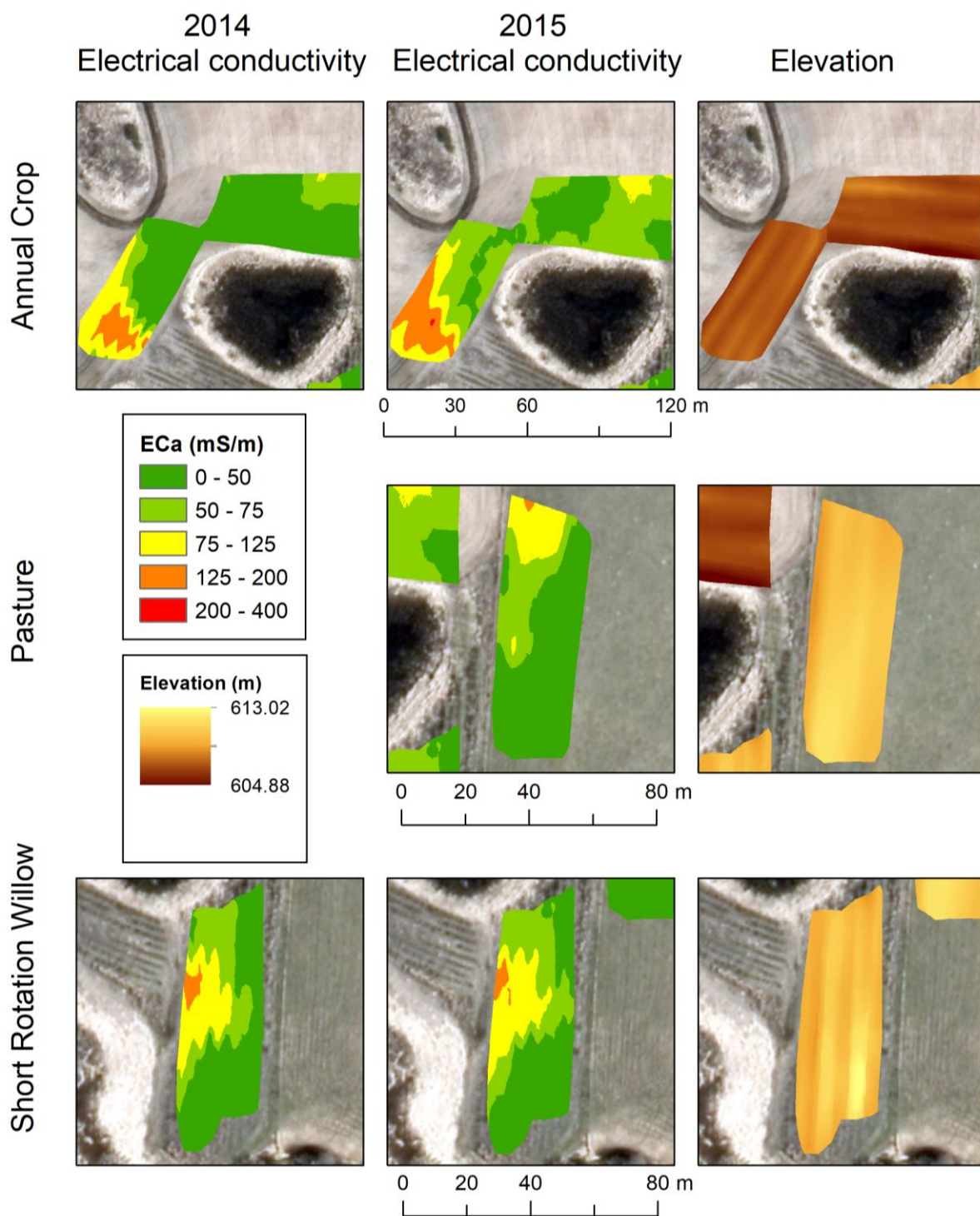


Figure 3.6 Spatial distribution of soil EC_a measured at 1m depth by EM38 at the end of the growing seasons of 2014 and 2015 from three different land-use practices from site A.

^a EC_a = apparent soil electrical conductivity.

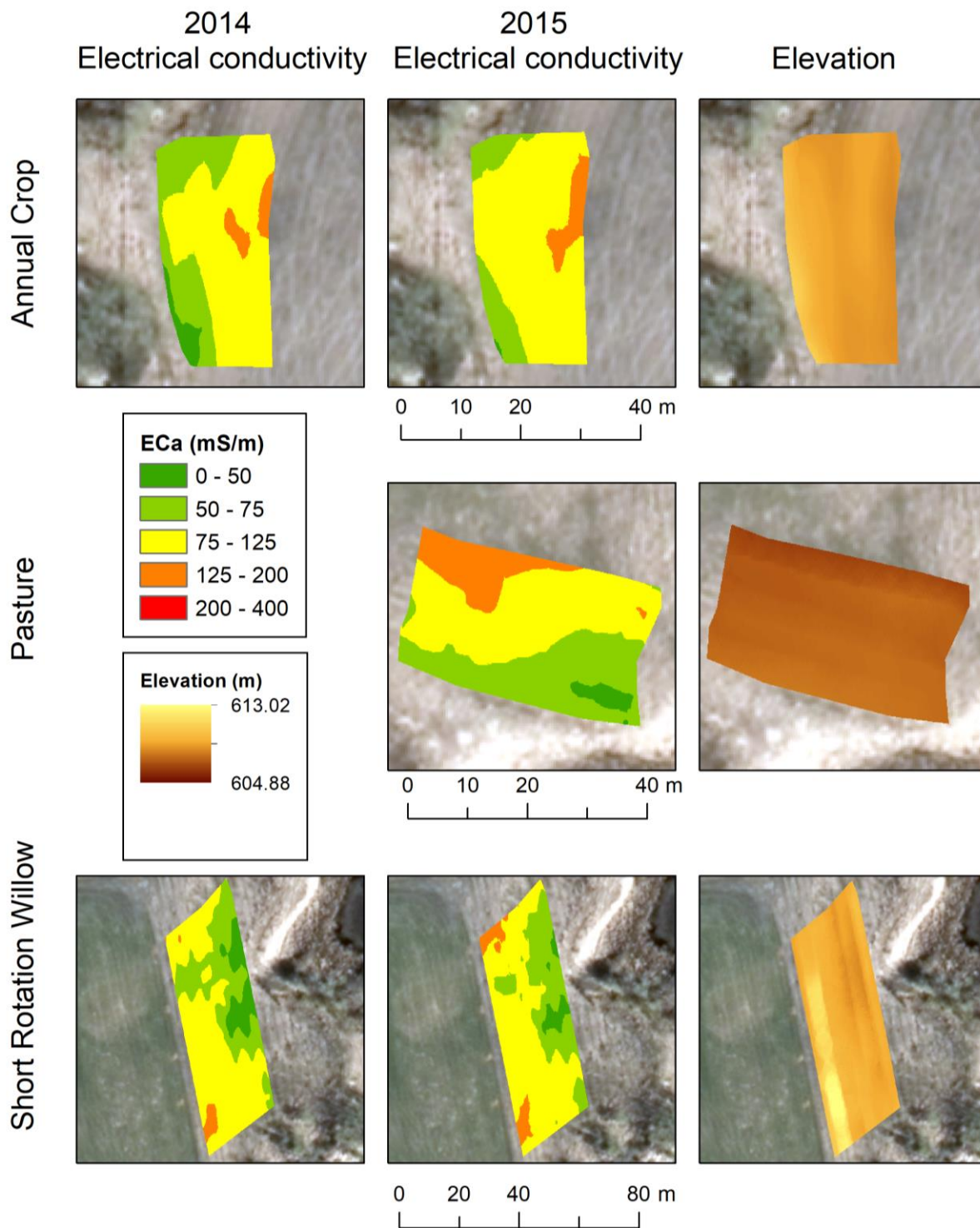


Figure 3.7 Spatial distribution of soil EC_a measured at 1m depth by EM38 at the end of the growing seasons of 2014 and 2015 from three different land-use practices from site B.

^a EC_a = apparent soil electrical conductivity.

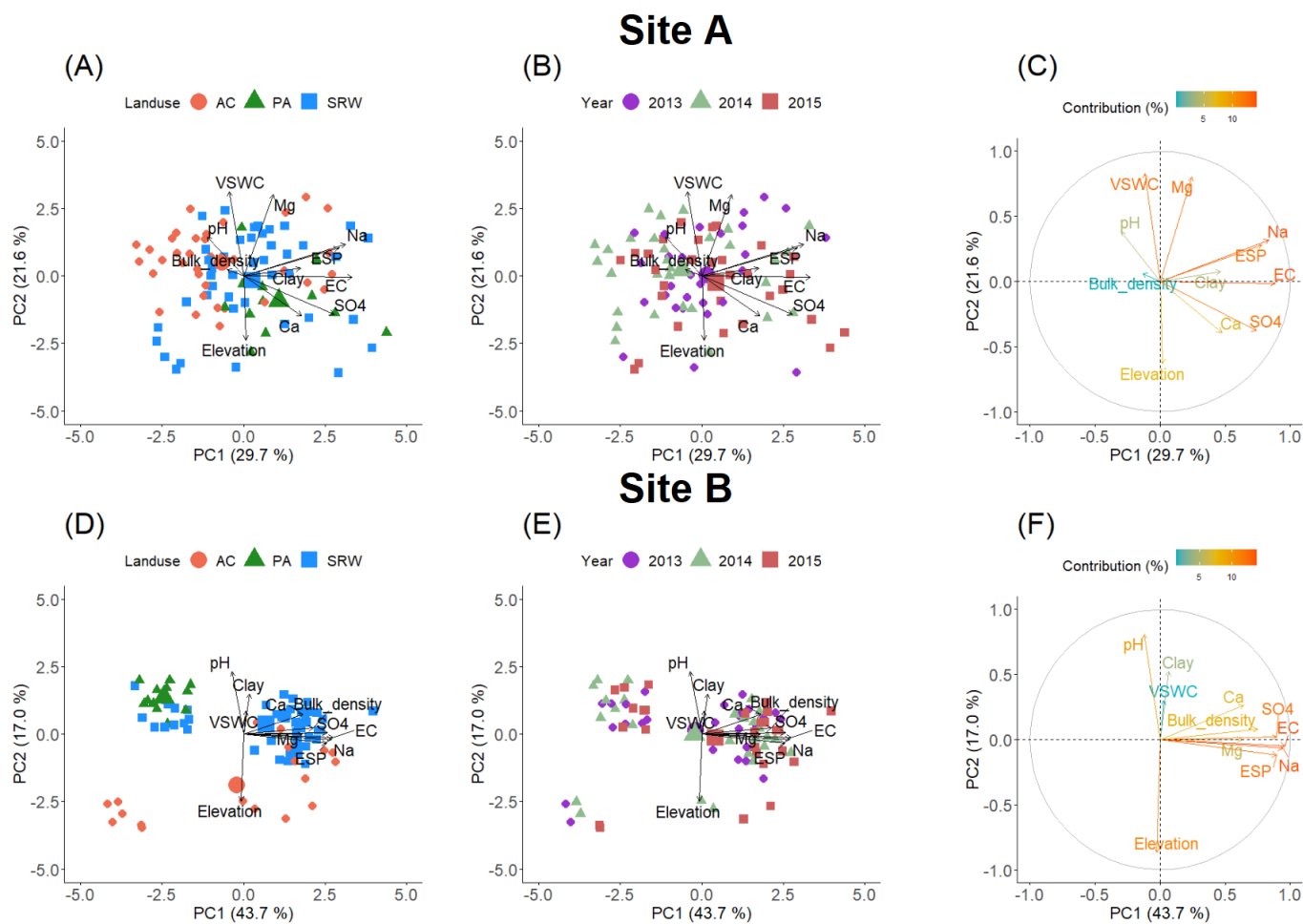


Figure 3.8 Principal component analysis (PCA) of measured soil EC, ESP, pH, volumetric water content, clay content, bulk density, sodium, calcium, magnesium, and sulfate as observed variables based on land-use practices, years as a factor variable, and contribution of variables, respectively.

^a Data was transformed to square root.

^b Contribution (in percentage) of the variables to the principal components.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, VSWC = volumetric soil water content, EC = electrical conductivity, ESP = exchangeable sodium percentage.

The soil texture (averaged for the depth up to 0-60 cm) under all three land-use practices from both experimental sites was clay loam, except under SRW from site A which was silty clay loam (SCL) (textural class not shown here). Clay content was significantly higher ($p < 0.001$) under PA land-use practice, followed by SRW and AC (Table A-S2, and S4). The upper soil layer (0-30 cm) contained a significantly lower ($p < 0.001$) clay content compared to 30-60 cm depth (Table A-S2, and S4). Positive correlations were observed between soil clay contents and EC_{soil} , however, significant in site A ($r = 0.25$, $p < 0.01$) and not significant in site B ($r = 0.04$, $p > 0.05$) (Figure A-S8A and B).

Significant ($p < 0.001$) negative correlations ($r = -0.60$ in site A, and $r = -0.47$ in site B) were observed between depth to GWT and VSWC in both sites (Figure A-S10A and B), suggesting shallower GWT depth significantly increased the VSWC. The correlations between depth to GWT with EC_{soil} and TDS were positive and nonsignificant ($r = 0.10$, $p > 0.05$) in site A, but positive and significant ($r = 0.34$, $p < 0.01$) in site B (Figure A-S10A and B), suggested that increased depth to GWT can lead to higher soil salinity. Whereas, the observed positive and statistically significant ($r = 0.57$, $p < 0.001$ in site A, and $r = 0.32$, $p < 0.01$ in site B) correlations between groundwater and soil EC indicate that the raised EC_{gw} may cause an increased EC_{soil} .

3.6 Discussion

3.6.1 The Implication of SRW on Groundwater Table Fluctuations

Studies around the world have shown a substantial lowering of the groundwater table with high rates of ET under agroforestry plantation compared to adjacent grassland (Jobbágy & Jackson, 2004), grassland and cropland (Nosetto et al., 2012), and the presence of riparian willows (Doody & Benyon, 2011, Doody et al., 2007). However, this study showed no significant difference between SRW and AC at either site, except for PA in site B.

Studies from wetlands within landscapes under annual crop production in North and South Dakota, USA, have demonstrated a more significant water table fluctuation compared to

grassland settings with phreatophytic nature (Euliss & Mushet, 1996). The authors suggested that this increase in water table fluctuations is mainly due to tillage, which can modify groundwater hydrology of wetlands within the agricultural landscapes. In this study, the soil underneath the SRW was rotavated just before the plantation establishment, which left a fallow band between the rows. Fallow lands are susceptible to greater water loss to runoff, percolation, and evaporation from the bare soil surface (Wiebe et al., 2007, 2005). The between-row hydrology may, therefore, have been more similar to an AC system than a native willow stand.

Within the PPR, phreatophytic willows are commonly found in the discharge areas (Meyboom, 1966b). The root system of SRW is relatively shallow (around 50 cm from the surface) and more concentrated near the soil surface, and the average transpiration rate of SRW stays between 300 to 450 mm per year (Dimitriou et al., 2009b). The average water table fluctuation caused by native willow rings in Saskatchewan has been reported as 3 cm day⁻¹ in the south-central region (Meyboom, 1966a), and 5 – 10 cm day⁻¹ in the south (Mirck & Schroeder (2018). In this study, the SRW stands had not yet achieved full potential biomass during their first rotation cycle; therefore, the within-row SRW also would not have had a significant impact on the depth of GWT as it would not yet have achieved its full ET potential. Instead, the site-specific factors such as topography, soil characteristics, and local climate dominated over the land-use effects. In an evaluation of the potential effects of SRW plantation on the environment, Dimitriou et al. (2009b) observed a higher typical rate of ET in SRW than arable crops; however, the rate can vary considerably based on site-specific factors as well as the variety and age of the plantation.

In long-term monitoring of water table and soil salinity build-up at the St. Denis National Wildlife Area from southern Saskatchewan, Canada, conversion from annual crops to deep-rooted perennials *Bromus inermis* Leyss. (brome grass) and *Medicago sativa* L. (alfalfa) caused the PPR wetlands to dry up within four to six growing seasons (van der Kamp et al., 1999). However, the neighboring wetlands with natural willow ring in the riparian zone and the

cultivated area did not alter the water table levels. Furthermore, the deep-rooted perennial grasses in the permanent cover transpired most of the water, which is mainly trapped from snow, infiltration of snowmelt and rainwater (during summer months) into the soil (van der Kamp et al., 2003). In contrast, in this field experiment, the depth to GWT was significantly shallower under PA land-use compared to AC and SRW; however, this was true only in site B but not in site A. Perhaps, the groundwater wells installed under PA land-use in site A did not capture the actual GWT situation as they were installed later than that of other land-use practices and showed a flat time-series curve.

3.6.2 The Implication of Spatial and Temporal Variation of Salinity

Generally, climate, topography, and site-specific factors are crucial parameters that control salinization. Soil texture, particularly clay content and porosity, plays a significant role in the variation of soil electrical conductivity (Heil & Schmidhalter, 2017, McNeill, 1980). In this study, both experimental field sites are mostly clayey and contained more clay in the deeper layer. Fine-textured soil has been reported to have less variability in EC compared to coarse-textured soil (Arndt & Richardson, 1993), which could produce less observed spatial and temporal variability.

The soil salinity may increase in the near-surface (upper soil layer) if deep-rooted vegetation (e.g., natural willow ring in the Prairie) is replaced by shallow-rooted crops, especially in the areas that contain natural deposits of salts (Henry et al., 1987). However, in this study, no consistent land-use pattern was observed in both experimental sites. Salinization is most likely to occur during the time of the year when evaporation exceeds the infiltration and percolation (Henry et al., 1987). This may have been the case in 2015 (relatively dryer year with higher groundwater EC) in both experimental field sites.

Geochemistry of the PPR wetland system is related to the evapotranspiration rate, recharge hydrology, ionic mobility, and exchange (Arndt & Richardson, 1988). Hence, the management of

groundwater discharge in the low lying areas in the PPR is crucial for limiting soil salinity (Henry et al., 1987). The relationships between both the groundwater EC and soil EC with TDS was 1:1 in this study, which is an indication of elevated SO_4^{2-} concentrations in throughflow and discharge wetlands (Arndt & Richardson, 1989). In contrast, recharge wetlands are free of calcite (CaCO_3) and gypsum ($\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$) and do not show similar geochemical properties. Accordingly, the experimental soils from this study contained a high amount of SO_4^{2-} in both sites (although site B contained 3 to 4 times more than site A); this indicated a discharge or throughflow wetland system.

Within the PPR, the development of soil salinity is mostly due to the transport of naturally occurring salts in the near-surface by the capillary movement of soil moisture (Richardson et al., 1994, van der Kamp & Hayashi, 2009). In this study, the groundwater salinity at the two experimental sites ranged from 0.47 to 16.66 mS cm^{-1} , suggesting a naturally high salt load. However, the exact location and extent of soil salinity is highly dependent on the aquifer characteristics, including thickness and permeability, as well as the distribution of evapotranspiration potential along with the slope position (Stolte et al., 1992).

Land-use driven vegetation changes, especially shifts between agroforestry plantation and grassland can alter water balances and soluble salt fluxes. In a study with phreatophytic plantations and adjacent native grasslands in Pampas, Argentina, Jobbágy & Jackson (2007) found that the phreatophytic discharge could control solute transfers from groundwater via 1) affecting solute transport to the rooting zone by altered groundwater flow within the aquifer, and 2) concentrating solutes in the rooting zone by water uptake plus solute exclusion. Noretto et al. (2013) similarly observed solute exclusion as a dominant salinization mechanism under tree plantations. However, the processes of soil salinization linked with enhanced groundwater consumption from agroforestry plantations discussed here from the literature are mainly associated with tree age that were > 12 years old. In contrast, in our study, the age of the SRW

establishment was within the first rotation (i.e., 3-year rotation cycle) and may not have fully developed root systems, hence less impact on soil salinization.

In the long run, the surplus water from adjacent cropland could maintain the sustained supply of groundwater where the agroforestry plantation trees are spread to maximize the capture of excess water from the surrounding landscape (Heuperman et al., 2002). Establishing deep-rooted plants under dry climatic conditions can significantly lower the groundwater table and thus can reverse the natural process of soil salinization (Nosetto et al., 2008, Schofield, 1992). Consequently, the fundamental intention for saline soil management is to move salt downward by lowering the water table (removing excess water) via the establishment of suitable land-use practice (deep-rooted salt-tolerant perennial vegetation), which has higher transpiration rate, e.g., SRW (Mirck & Schroeder, 2018, Mirck & Zalesny, 2015). In this way, soil EC, as well as the salts (TDS), can be reduced in the 0-30 cm rooting zone. Hence, the most cost-effective and environmentally sustainable option to manage salts and excess water in the discharge area in a wetland system could be the 'biodrainage' (Minhas & Dagar, 2016). Salinity management in the discharge area of wetland is challenging, particularly in the semi-arid climate and with glacial geology that results in unique hydrological conditions and distributions of naturally accumulated salts in the subsurface (Nachshon et al., 2013). As a supplement to the growing of deep-rooted perennial plant species in a field, having SRW in the fringes of the wetland could be part of the beneficial management practices due to the phreatophytic nature and high biomass production capacity that can better utilize the excess water in riparian areas to reduce salinity in the semi-arid PPR (Eilers et al., 1997, Hangs et al., 2011, Mirck & Schroeder, 2018, Stolte et al., 1992). However, the findings of this study suggest that the full potential of this management strategy would only be realized over the longer term, as SRW stands mature.

3.7 Conclusions

The land-use practices significantly impacted the GWT depth in site B, but not in site A, indicating inconsistent effects of land-use patterns between sites. Overall, depth to GWT followed the precipitation patterns. Intense fluctuations in GWT depth occurred between June to August when the precipitation was higher, whereas the fluctuations were smaller during May and September due to the reduced amount of precipitation. The higher precipitation events throughout the wet year (i.e., during 2014) resulted in a shallower depth to GWT across land-use practices in both sites. Significant variation was observed in EC_{gw} among land-use practices; however, variations between years and among months were inconsistent between sites. The EC_{soil} varied among land-use practices but was inconsistent between sites, whereas there were no observed variations between depths, years, and among months in both sites. Field observation exhibited a reduction in soil EC with increasing SRW biomass at both depths (i.e., 0-30 and 30-60 cm) from both sites; however, given the lack of consistent patterns, it is more likely that the SRW biomass production was lower in locations with inherently higher soil salinity.

Despite the prediction of high water consumption, the establishment of SRW in the riparian zones had minimal drawdown impact on the groundwater table within the timeframe of this study. However, over the longer term, SRW plantations could be used to reduce and manage salinity at the surface soil (0-60 cm) in the areas of low to moderate salinity (EC_{soil} 1-6 $mS\ cm^{-1}$) as a part of the best management practices, particularly on the degraded riparian marginal lands. Hence, SRW variety *Salix dasyclados* Wimm. ('India') can be a potential candidate for the management of salinity within the PPR.

4 EFFECTS OF RIPARIAN LAND-USE PRACTICES ON SEASONAL AND ANNUAL VARIATION OF NUTRIENTS IN GROUNDWATER AND SOIL

4.1 Preface

In addition to high water requirement, short rotation willow (SRW) is also known to have a high nutrient demand, especially when planted at high densities. In combination with high biomass production and nutrient export, SRW plantation may substantially deplete soil fertility and diminish soil productivity for agricultural crop production within the prairie pothole region (PPR) agroecosystems. Hence, to sustain the capability of wetlands delivering ecosystem services, it is vital to assess the impacts of this high nutrient demanding nature of SRW plantation on the status of wetland soil nutrient pools. This brief chapter summarizes the effects of SRW as a marginal riparian land-use practice on various soil nutrient pools compared to the adjacent pasture (PA) and annual crop (AC) production. Here the attention was given to a seasonal and annual variation of soil nutrients in conjunction with the discrete effects on soil hydrology and salinity under the SRW plantation in the riparian zones of the PPR wetland systems discussed in Chapter 3. The conceptual model (Figure 4.1) shows the variables that were assessed in Chapter 4 and that can potentially be impacted by land-use practices.

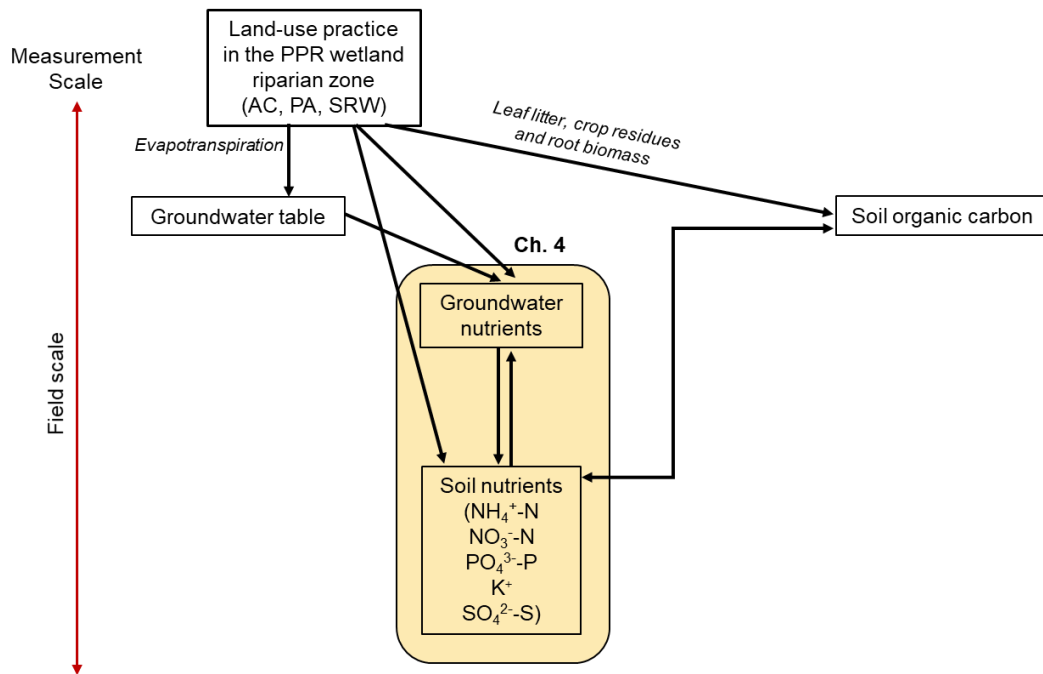


Figure 4.1 Conceptual model showing the potential effects of land-use practices on groundwater and soil nutrients in the riparian zones of the PPR wetland system.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, PPR = prairie pothole region.

4.2 Abstract

Short rotation willow (SRW) is a source of biomass energy currently being explored in the semi-arid PPR with high nutrient demand in conjunction with high growth rate and water use via prospective evapotranspiration. However, high biomass production capability and the input of organic matter through SRW leaf litter can also enhance nutrient cycling if the biomass is not removed from the system and thus can increase nutrient availability in soil. Consequently, it is uncertain whether the introduction of SRW (*Salix* Spp.) in the riparian zones of the PPR wetlands will enhance or diminish the soil nutrient pools. Hence, in a multi-year field experiment, the effects of SRW land-use practice was examined on the seasonal and annual variation of soil macronutrients (N, P, K, and S) compared to adjacent PA and AC production during the first SRW rotation (3-year cycle). The mean soil $\text{NH}_4^+\text{-N}$ and K^+ contents were lower under SRW compared to adjacent land-use practices, whereas $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, and $\text{SO}_4^{2-}\text{-S}$ contents were

higher under SRW compared to AC or PA. The depletion of soil $\text{NH}_4^+\text{-N}$ and K^+ under SRW plantation may have been intensified by higher growth rate and biomass production. Overall, the groundwater $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, K^+ , and $\text{SO}_4^{2-}\text{-S}$ concentrations were significantly higher ($p < 0.05$) under SRW plantation, whereas $\text{PO}_4^{3-}\text{-P}$ did not differ significantly ($p > 0.05$) across land-use practices from both sites. Hence, this study revealed that during their first 3-year rotation cycle, the establishment of SRW did not severely exhaust soil nutrient pools in the riparian zones of PPR wetland systems. However, to maintain future agricultural productivity and sustain the ecological functions of wetlands within the PPR, the impacts of SRW plantations on the variation and dynamics of soil nutrient pools need to be examined regularly.

4.3 Introduction

Soil nutrient supply and variability can be significantly affected by short rotation willow (SRW) due to its fast growth rate, high biomass production, and vegetative regrowth capacity (Amichev et al., 2014b). Plantations under agroforestry land-use practice, such as SRW (*Salix* Spp.), can consume and remove a higher amount of soil nutrients annually (Ledin, 1998). Soil macronutrient (N, P, K, and S) availability can affect the growth performance of SRW (Mirck & Schroeder, 2018), and for good growth, the optimum ratio should be N:P:K of 100:13:65 (Ericsson, 1981a, b). However, the use efficiency and removal rate vary by variety, planting density, and harvest cycle (Adegbidi et al., 2001).

Generally, trees under intensively managed plantation can grow faster and are most resourceful in nutrient acquisition (Fox, 2000). Hence, with extensive root systems, trees can exploit soil nutrients pools and also can recycle nutrients more efficiently (Bauhus & Messier, 1999). The introduction of tree plantations has significant effects on soil nutrient pools through 1) extracting nutrients from deeper layers of the soil profile; 2) changing soil hydrological conditions by increasing evapotranspiration and decreasing leaching; 3) altering pH by producing acidic root exudates that enhance mineral weathering and act as chelates; 4) removal and storage of soil

nutrients within their biomass (i.e., leaves, stems, and roots), and subsequent return via litterfalls or other pathways (e.g., root exudates); and 5) curtailing competition (Attiwill & Adams, 1993, Binkley & Giardina, 1998).

Rapid plant growth does not always result in loss of nutrients as some of the biologically facilitated processes (e.g., leaf litter cycling, plant root exudates, etc.) may also enhance nutrient sequestration and availability in the soil (Bélanger et al., 2004). Some of the nutrients could be returned to the soil through leaf litter cycling, but may also be lost from the soil system through the removal of a higher amount of biomass produced by the SRW when harvested (Cornelissen et al., 1997, Kopinga & Van den Burg, 1995, Simon et al., 1990). In fact, within the PPR, SRW plantations have shown high efficiency in nutrient cycling, even within the first rotation (4-year rotation cycle): they were relatively low nutrient demanding and had minimal removal with harvested biomass (Hangs et al., 2014b). In another study, Hangs et al. (2014a) indicated that the leaf litter decomposition under SRW land-use practice is capable of increasing nutrient availability and organic carbon sequestration within the first rotation of the 4-year cycle. However, SRW plantation establishment at nine sites across the prairies and southern Ontario led to reduced C, N, K, and P in the upper soil layers (0-20 cm) compared to the paired reference sites after a 3-year rotation cycle (Ens et al., 2013a).

The reported average annual yield of SRW ranges between 10-15 Mg ha⁻¹ (dry basis); however, it could be high as 27-30 Mg ha⁻¹ if plantations are fertilized and irrigated (Adegbidi et al., 2003, Adegbidi et al., 2001, Kopp et al., 2001, Labrecque et al., 1998, Volk et al., 2011). It has been estimated that with a 15-22 Mg ha⁻¹ biomass production, SRW could remove 75-86, 10-11, 27-32, 52-79, and 4-5 kg ha⁻¹ year⁻¹ of nitrogen (N), phosphorous (P), potassium (K), calcium (Ca), and magnesium (Mg), respectively, depending on the variety, plant density and harvest cycle (Adegbidi et al., 2001). For example, production with irrigated and fertilized SRW variety of "India" (*Salix dasyclados*) on a 3-year rotation showed the highest biomass production and

nutrient use efficiency, but the lowest rates of nutrient removal from the soil (Adegbidi et al., 2001).

The establishment of SRW can affect the dynamics of soil nutrients in various ways, besides high biomass production. With its extended root systems and high water-use efficiency, in combination with higher evapotranspiration requirements, SRW is capable of better utilizing nutrient-rich percolating water leaving agricultural systems (Aronsson & Perttu, 2001, Aronsson & Bergström, 2001, Aronsson et al., 2000). Therefore, SRW can enhance water quality by reducing N and P losses through leaching and surface runoff (Dimitriou et al., 2012a), and increase the potential of recycling nutrients in the riparian zones of wetlands.

Despite these potential benefits, it is unknown whether the establishment of SRW plantations in the marginal riparian zones will enhance or reduce the soil nutrient availability after the first rotation in contrast to adjacent pasture (PA) and annual crop (AC) production. Furthermore, SRW is one of the prevalent bioenergy producing plants of growing interest in North America (Kopp et al., 2001, Thevathasan et al., 2012, Volk et al., 2006). It is essential to assess the effects of fast-growing SRW on nutrient use in this region (Cornelissen et al., 1997, Simon et al., 1990), particularly within the PPR (Amichev et al., 2014a). Therefore, the objective of this multi-year field study was to assess the effect of SRW land-use practice on 1) the *in-situ* impacts of land-use practice on nutrient quantity, and 2) the seasonal and annual variation of groundwater and soil nutrients (N, P, K, and S). It has been hypothesized that the establishment of SRW land-use practice in the riparian zones of wetlands in the PPR would preserve the capacity of soil nutrient pools (i.e., will not be depleted gradually) over time and will maintain the seasonal variability.

4.4 Materials and Methods

For site descriptions, experimental design, and groundwater and soil sample collection methods, please see the Material and Methods section from Chapter 3.

4.4.1 Soil and Groundwater Samples Collection

Soil and groundwater samples collected for the measurements in Chapter 3 were used for the analysis of soil $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, K^+ , $\text{PO}_4^{3-}\text{-P}$, and $\text{SO}_4^{2-}\text{-S}$ reported in this chapter.

4.4.2 Analyses of Soil and Groundwater Samples

Soil inorganic N ($\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$) was extracted using 2.0 M KCl (Maynard et al., 2008), and soil K^+ , $\text{PO}_4^{3-}\text{-P}$, and $\text{SO}_4^{2-}\text{-S}$ were extracted using 1M ammonium acetate, buffered at pH 7 (Hendershot et al., 2008a). Soil extract and groundwater nutrient concentrations were measured as follows: $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$ and $\text{PO}_4^{3-}\text{-P}$ by colorimetric methods using a Technicon Auto-Analyzer (Technicon Industrial Systems, Tarrytown, NY, USA); K^+ by atomic emission spectroscopy (Varian Spectra 220 Atomic Absorption Spectrometer; Varian Inc., Palo Alto, CA, USA); and SO_4^{2-} by microwave plasma-atomic Emission spectrometry (4100 MP-AES; Agilent technologies, Melbourne, Australia).

4.4.3 Statistical Analyses

Statistical analyses were performed using R version 3.4.4 for Windows (R Core Team, 2018). Obtained data were tested for normality by the Shapiro-Wilk test and histogram. Homogeneity of variances or homoscedasticity was tested by Levene's test using the "car" package. When required, the square root transformation was performed to improve the assumption of normality and homoscedasticity of the data. Data visualization was performed through box plots and line graphs using the "ggplot2" package. Analysis of variance (ANOVA) with linear mixed-effects models was used to test for significant differences across land-use practice, years, months, and depths (for soil samples) in terms of measured nutrients using the "lmerTest". The mixed approach was chosen due to its suitability for unequal variances, nested, and unbalanced design. Mean comparisons of measured variables were performed using Tukey Honest Significant Differences (TukeyHSD) test using the "TukeyC". Pearson correlations were

performed using the “psych”, and principal component analysis (PCA) using “FactoMineR” and “factoextra” package.

4.5 Results

4.5.1 Variation in Soil Nutrients

Among land-uses, soil $\text{NH}_4^+\text{-N}$ content differed significantly in site A ($p = 0.002$), but not in site B ($p = 0.289$), whereas $\text{NO}_3^-\text{-N}$ differed significantly in site B ($p < 0.001$) but not in site A ($p = 0.091$; Table 4.1). The mean soil $\text{NH}_4^+\text{-N}$ content across land-use practices was in the order of $\text{PA} > \text{SRW} > \text{AC}$ in site A, and $\text{AC} > \text{PA} > \text{SRW}$ in site B. For $\text{NO}_3^-\text{-N}$ the order was $\text{SRW} > \text{PA} > \text{AC}$ in site A, and $\text{SRW} > \text{PA} > \text{AC}$ in site B (Table B-S1). Soil $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ contents were higher in 2015 compared to 2014 in both sites (Figure 4.2 and 4.3). For soil $\text{NH}_4^+\text{-N}$, the difference was statistically significant in both sites ($p < 0.001$), whereas soil $\text{NO}_3^-\text{-N}$ was significant only in site A ($p = 0.001$; Table 4.1). Soil $\text{NH}_4^+\text{-N}$ was higher in May than September in both sites ($p < 0.01$), whereas soil $\text{NO}_3^-\text{-N}$ content was higher in September than May (site A: $p < 0.001$; site B: $p = 0.134$) (Figure 4.2 and 4.3; Table 4.1).

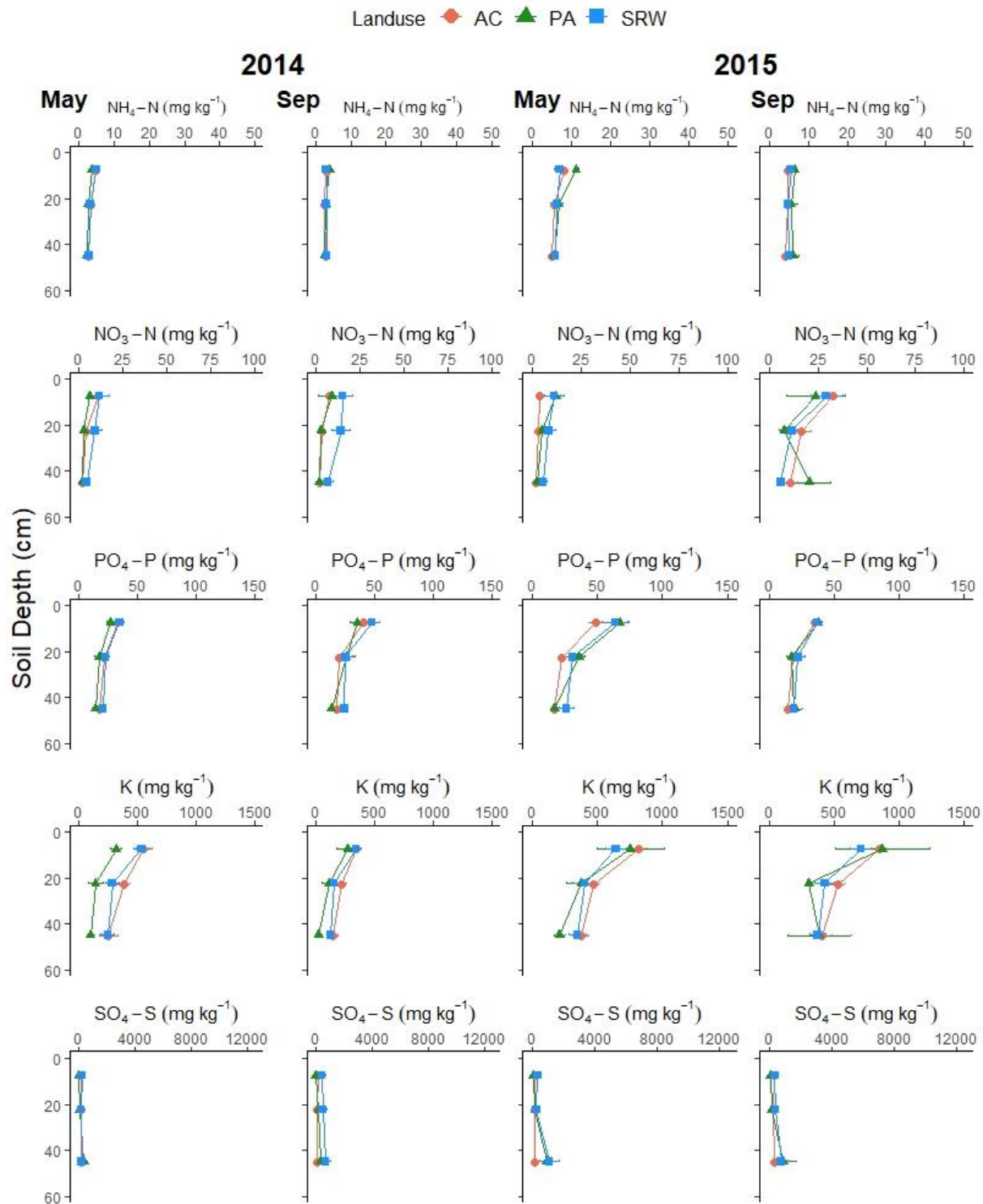


Figure 4.2 Seasonal and annual variation of $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, K^+ , and $\text{SO}_4^{2-}\text{-S}$ concentration in soil from different depths under three land-use practices during the growing seasons of 2014 and 2015 from site A.

^a AC = annual crop, PA = pasture, SRW = short rotation willow

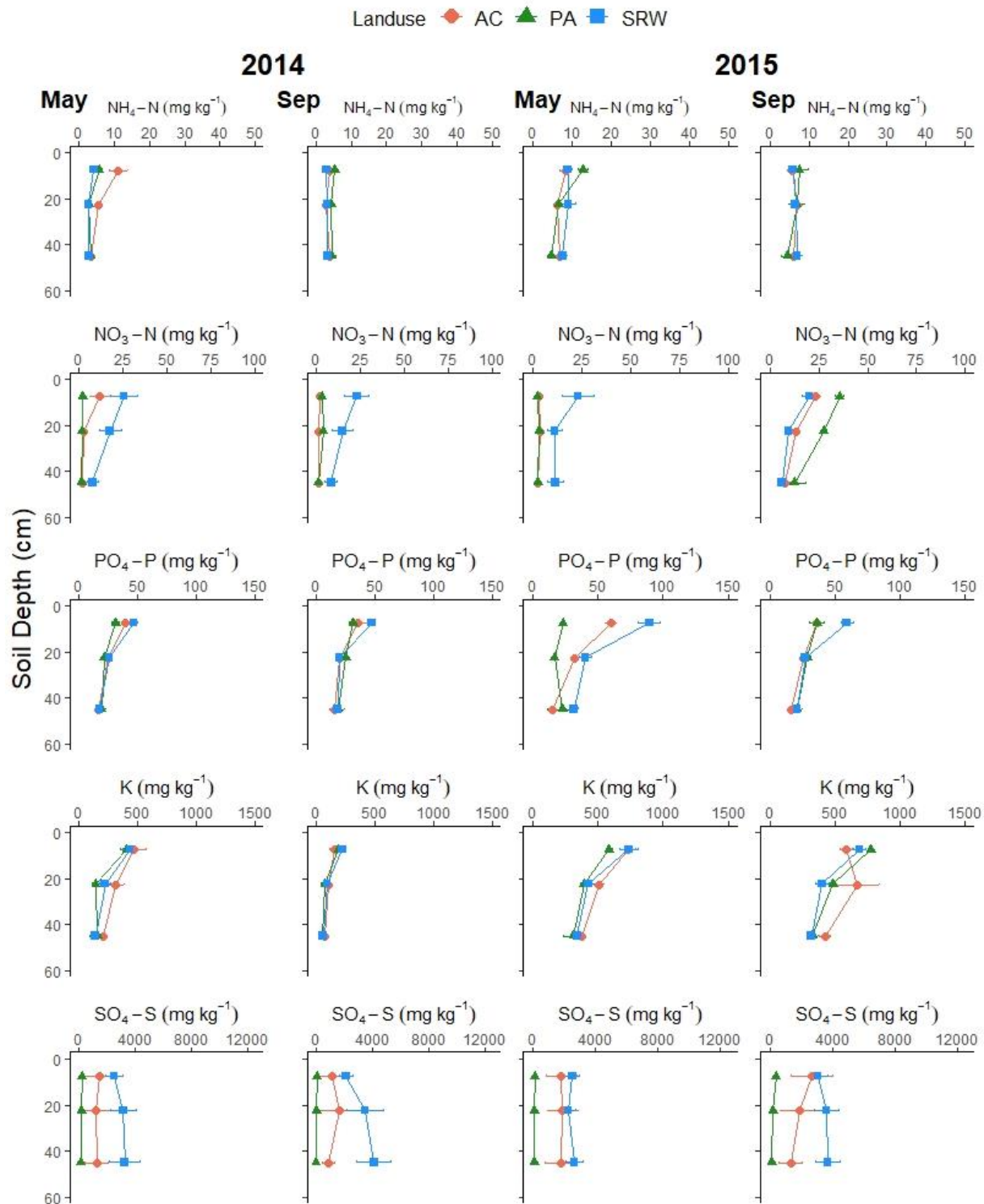


Figure 4.3 Seasonal and annual variation of $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, K^+ , and $\text{SO}_4^{2-}\text{-S}$ concentration in soil from different depths under three land-use practices during the growing seasons of 2014 and 2015 from site B.

^a AC = annual crop, PA = pasture, SRW = short rotation willow

Table 4.1 Analysis of variance (ANOVA) with a linear mixed-effects model for measured soil nutrients under three land-use practices from two sites during the growing season of two consecutive years.

Response variable	Sources of variation	Site A			Site B	
		df	F - value	p - value	F - value	p - value
NH ₄ ⁺ -N	Land-use	2	6.307	0.002 **	1.252	0.289 ^{ns}
	Depth	2	13.359	<0.001 ***	4.664	0.011*
	Year	1	147.932	<0.001 ***	112.772	<0.001 ***
	Month	1	19.847	<0.001 ***	8.228	0.005 **
NO ₃ ⁻ -N	Land-use	2	2.435	0.091 ^{ns}	10.988	<0.001 ***
	Depth	2	18.685	<0.001 ***	14.4502	<0.001 ***
	Year	1	11.056	0.001 **	2.551	0.112 ^{ns}
	Month	1	28.940	<0.001 ***	2.266	0.134 ^{ns}
PO ₄ ³⁻ -P	Land-use	2	5.058	0.007 **	16.075	<0.001 ***
	Depth	2	83.565	<0.001 ***	126.215	<0.001 ***
	Year	1	2.155	0.144 ^{ns}	34.254	<0.001 ***
	Month	1	3.211	0.075 ^{ns}	12.189	<0.001 ***
K ⁺	Land-use	2	4.095	0.018 *	2.476	0.088 ^{ns}
	Depth	2	79.767	<0.001 ***	70.002	<0.001 ***
	Year	1	130.986	<0.001 ***	263.253	<0.001 ***
	Month	1	8.426	0.004 **	24.256	<0.001 ***
SO ₄ ²⁻ -S	Land-use	2	7.598	<0.001 ***	59.887	<0.001 ***
	Depth	2	2.672	0.072 ^{ns}	0.129	0.878 ^{ns}
	Year	1	4.527	0.035 *	1.015	0.316 ^{ns}
	Month	1	6.001	0.015 *	0.546	0.461 ^{ns}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

Soil PO₄³⁻-P content was significantly different among land-use practices in both sites ($p < 0.01$; Table 4.1). The mean PO₄³⁻-P concentration across land-use practices was in the order of SRW > PA > AC in site A, and SRW > AC > PA in site B (Table B-S1). The mean soil PO₄³⁻-P content was higher in 2015 than 2014, and in May than September in both sites (Figure 4.2 and 4.3); however, the temporal variation was highly significant only in site B (Table 4.1).

The mean K⁺ concentration across land-use practices was in the order of AC > SRW > PA in both sites (Table B-S1). However, K⁺ content among land-use practices differed more in site A ($p = 0.018$) than site B ($p = 0.088$; Table 4.1). The mean soil K⁺ content was significantly higher

($p < 0.01$) in 2015 than 2014, and in May than September in both sites (Figure 4.2, 4.3, and Table 4.1).

The mean soil SO_4^{2-} -S content was significantly different ($p < 0.001$ in site A, and $p < 0.001$ in site B) among land-use practices from both sites (Table 4.1). The mean value of soil SO_4^{2-} -S concentration in the order of land-use practices was $\text{SRW} > \text{PA} > \text{AC}$ in site A, $\text{SRW} > \text{AC} > \text{PA}$ in site B (Table B-S1). Soil SO_4^{2-} -S content was higher in 2015 than 2014, and in September compared to May in both sites (Figure 4.2, 4.3 and Table B-S1); however, statistically significant in site A ($p = 0.035$ for years, and $p = 0.015$ for months), but not in site B ($p = 0.316$ for years and $p = 0.461$ for months) (Table 4.1).

Concentrations of all soil nutrients (NH_4^+ -N, NO_3^- -N, PO_4^{3-} -P, and K^+) significantly differed ($p < 0.05$) among depths across land-use practices in both sites except SO_4^{2-} -S which was not significant ($p < 0.05$) (Table 4.1). The mean nutrient contents of NH_4^+ -N, NO_3^- -N, PO_4^{3-} -P, and K^+ were in the order of 0-15 cm $>$ 15-30 cm $>$ 30-60 cm depths in both sites except SO_4^{2-} -S which was higher at lower depth (30-60 cm) (Figure 4.2 and 4.3).

4.5.2 Variation in Groundwater Nutrient Concentrations

There were no consistent temporal patterns in groundwater N concentrations across sites. The groundwater NH_4^+ -N and NO_3^- -N concentration differed significantly ($p < 0.01$) among land-use practices; however, the land-use patterns were not consistent between sites (Table 4.2 and Table B-S2). The mean NH_4^+ -N concentration was significantly higher ($p < 0.01$) in 2014 than in 2015 from both sites. Among the months, the mean concentration was in the order of May $>$ Sep $>$ Jul for NH_4^+ -N ($p = 0.183$ in site A, and $p = 0.018$ in site B), whereas May $>$ Jul $>$ Sep for NO_3^- -N ($p = 0.038$ in site A, and $p = 0.554$ in site B) across land-use practices in both sites (Table 4.2, Table B-S2 and Figure B-S1 and S2).

Table 4.2 Mean (\pm SD) groundwater nutrients under different land-use practices from both sites during 2014 and 2015.

Variable	Year	Site A			Site B		
		AC	PA	SRW	AC	PA	SRW
NH ₄ ⁺ -N (mg L ⁻¹)	2014	0.14 \pm 0.12bA	0.18 \pm 0.02aA	0.17 \pm 0.13aA	0.21 \pm 0.14aA	0.09 \pm 0.07bA	0.47 \pm 0.79aA
	2015	0.07 \pm 0.08bB	0.18 \pm 0.02aB	0.10 \pm 0.08aB	0.12 \pm 0.09aB	0.05 \pm 0.02bB	0.16 \pm 0.10aB
NO ₃ ⁻ -N (mg L ⁻¹)	2014	4.39 \pm 5.67bA	11.43 \pm 1.79aA	3.84 \pm 4.37aA	1.90 \pm 2.44bA	0.16 \pm 0.12bA	32.80 \pm 59.56aA
	2015	4.06 \pm 4.01bA	10.27 \pm 1.68aA	11.01 \pm 12.19aA	8.76 \pm 4.48bA	16.70 \pm 13.67bA	46.16 \pm 56.58aA
PO ₄ ³⁻ -P (mg L ⁻¹)	2014	0.13 \pm 0.03aA	0.11 \pm 0.01aA	0.15 \pm 0.10aA	0.13 \pm 0.02aA	0.13 \pm 0.01aA	0.14 \pm 0.04aA
	2015	0.14 \pm 0.34aB	0.12 \pm 0.01aB	0.07 \pm 0.07aB	0.06 \pm 0.02aB	0.06 \pm 0.04aB	0.07 \pm 0.05aB
K ⁺ (mg L ⁻¹)	2014	13.80 \pm 5.14aB	5.40 \pm 2.67bB	9.31 \pm 6.59aB	2.70 \pm 3.06bB	3.06 \pm 2.14bB	11.49 \pm 8.72aB
	2015	19.61 \pm 10.08aA	5.78 \pm 2.43bA	20.79 \pm 15.40aA	9.71 \pm 7.43bA	9.59 \pm 2.42bA	27.21 \pm 10.41aA
SO ₄ ²⁻ -S (mg L ⁻¹)	2014	157.73 \pm 123.05bA	289.59 \pm 22.32aA	252.46 \pm 186.67aA	306.11 \pm 113.72aA	72.72 \pm 12.56bA	304.76 \pm 131.95aA
	2015	170.46 \pm 157.13bA	203.90 \pm 123.23aA	378.75 \pm 161.28aA	343.33 \pm 105.71aA	143.96 \pm 46.47bA	240.86 \pm 157.88aA

^a Values represent mean \pm standard deviations (\pm SD). Means were calculated from the values of groundwater nutrients measured from May, July, and September every year.

^b Means within a row for land-use followed by the same small letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c Means within a column for year supported by the same capital letter are not significantly different ($p > 0.05$) using Tukey HSD.

^d AC = annual crop, PA = pasture, SRW = short rotation willow

The groundwater $\text{PO}_4^{3-}\text{-P}$ concentration differed significantly among months ($p < 0.001$ in site A, and $p < 0.001$ in site B) and years ($p < 0.001$ in site A, and $p = 0.007$ in site B); however, not significantly across land-use practices ($p = 0.781$ in site A, and $p = 0.383$ in site B) in both sites (Table 4.2 and Table B-S2). The mean $\text{PO}_4^{3-}\text{-P}$ concentration was significantly higher ($p < 0.01$) in 2014 than 2015 in both sites (Table 4.2). Among months the mean $\text{PO}_4^{3-}\text{-P}$ concentration were Sep > May > July in site A, and Sep > Jul > May in site B (Table 4.2 and Figure B-S1 and S2).

The groundwater K^+ concentration differed significantly among land-use practices ($p < 0.001$ in site A, and $p < 0.001$ in site B), and between years ($p < 0.001$ in site A, and $p < 0.001$ in site B), however, not significantly among months ($p = 0.997$ in site A, and $p = 0.497$ in site B) (Table 4.2 and Table B-S2). In both sites, the mean K^+ concentration was significantly higher ($p < 0.01$) in 2015 than in 2014 (Table 4.2). Across land-use practices, the mean K^+ concentration was in the order of AC = SRW > PA in site A, and SRW > AC = PA in site B, whereas, among months were Jul > Sep > May in site A, and Sep > May > Jul in site B (Table 4.2 and Figure B-S1 and S2).

The groundwater $\text{SO}_4^{2-}\text{-S}$ concentration significantly differed among land-use practices ($p < 0.001$ in site A, and $p < 0.001$ in site B), however, not significantly between years ($p = 0.150$ in site A, and $p = 0.299$ in site B), and among months ($p = 0.335$ in site A, and $p = 0.546$ in site B) in both sites (Table 4.2). The variation in land-use practice in terms of groundwater mean $\text{SO}_4^{2-}\text{-S}$ concentration in the order of significance was SRW = PA > AC in site A and in AC = SRW > PA site B (Table 4.2 and Figure B-S1 and S2).

4.6 Discussion

Lower $\text{NH}_4^+\text{-N}$ concentration under SRW indicates the possibility of higher N uptake, although this was not consistent between sites. The SRW is known to have high N demand, and the higher growth rate consequently enhances available N (NH_4^+ and NO_3^-) uptake that leads to noticeable depletion of available N in soil (Simon et al., 1990). It was estimated that harvesting

of SRW after a 3-year cycle could remove approximately 3 to 4 kg N per ton of stem (dry weight basis) (Ericsson, 1994). However, within the PPR, it has been estimated that the leaf litter production and decomposition in soil owing to the higher biomass production and accumulation under SRW land-use practice can enhance N release of about 22 kg ha⁻¹ (Hangs et al., 2014b).

It has been suggested that the higher rate of N uptake – especially NO₃⁻-N – under SRW can improve water quality and decrease both NO₃⁻ leaching and nitrous oxide emission via denitrification from soil (Aronsson & Bergström, 2001, Dimitriou et al., 2012a). In contrast, this study found comparatively higher concentrations of groundwater and soil NO₃⁻-N under SRW in both sites. However, a study found that riparian buffers established on upslope position with 5-15 mg L⁻¹ groundwater NO₃⁻-N content was not able to significantly reduce the concentration transmitted from the agricultural field edge towards the wetlands (Johnson et al., 2013).

Therefore, the landscape position of the riparian plantation may be a crucial factor that can affect groundwater NO₃⁻-N leaching from the agricultural non-point source into the wetlands.

Like many studies cited in the review by Chen et al. (2008), available PO₄³⁻-P content under SRW soils were relatively higher than the PA or AC land-use practices. It has been suggested that agroforestry can enhance soil organic matter and thus mineralization, which consequently can increase the available P in the topsoil. This can be ascribed to a combination of other factors such as higher P demand and uptake, favorable soil moisture conditions, improved solubility of P from the root and microbial exudates (e.g., organic acids), and higher phosphatase activity associated with ectomycorrhizae in the root zone (Chen et al., 2008).

However, it has also been reported that site disturbance can decrease soil P (Bormann et al., 1974). The establishment phase of the SRW plantation could be associated with the initial P loss from the system; however, continuing mineralization of soil organic matter or the high biomass production and leaf litter degradation can increase the P balance in the later stage of the plantation compared to adjacent annual crop (Ellert & Gregorich, 1996).

Potassium is a readily soluble soil nutrient that can be affected by water use and site disturbance associated with land-use change (Likens et al., 1994), such as with the establishment of SRW plantation. Hence, the increase in the transfer of dissolved K^+ can be feasible from the initial site disturbance, and subsequent release of immobilized nutrients as higher groundwater K^+ concentration was observed under SRW land-use practice. Additionally, the increase in the transpiration and subsequent high water use under the SRW land-use practice may lower the groundwater table and thus translocate the mobile K^+ into the soil (Likens et al., 1994). Perhaps this mechanism could be the critical reason for higher extractable potassium during the growing season of 2015 (i.e., relatively dry year, please see the data presented in chapter 1) compared to 2014. However, there is some decline in soil K^+ concentration under SRW land-use practice due to the higher uptake along with the enhanced growth rates and higher biomass production compared to AC. Furthermore, it has been found that the stem and higher leaf growth rates are correlated with the K^+ concentration to maintain the long term productivity of SRW (Ens et al., 2013a).

The soil SO_4^{2-} -S concentration was relatively high under SRW land-use practice compared to AC or PA. The leaf litter decomposition after the first SRW rotation could provide 47% of the S demand for the next rotation, and the turnover of the fine roots might further supply approximately 18 kg ha^{-1} (Hangs et al., 2014b). This bio-cycling, coupled with high background soil SO_4^{2-} -S, and relatively low S-demand of SRW may result in surplus S. The semi-arid glaciated plains of the North American PPR are generally rich in subsurface sulfate salts (Nachshon et al., 2013), which is further exacerbated by evapotranspiration and subsurface flow pathways. Therefore, hydrological changes caused by the specific land-use practice may result in the mobilization of SO_4^{2-} salts and concentration in the shallow subsurface (Nachshon et al., 2013), particularly under SRW, where the concentration in groundwater was significantly higher than other land-use practices. However, these involve multiple interacting processes, and the

changes due to land-use practices are sometimes difficult to predict due to the overriding effects of subsurface hydrogeological processes.

Generally, trees are efficient in nutrient recycling under natural conditions (Ericsson, 1994).

However, in this experiment, under a first-rotation SRW plantation, the sites were disrupted by physical disturbance during the establishment phase (i.e., tillage operations, and plantation of willow cuttings by machine). Similarly, in a 3-year rotation cycle under SRW plantation, Ens et al. (2013a) found decreased inorganic N, available P, and exchangeable K across nine sites in the Canadian prairie and southern Ontario region. Nonetheless, the productivity of nutrient recycling might improve in the future rotation under SRW plantation as disturbance will be eliminated or greatly reduced. For example, the average nutrients released from SRW leaf litter decomposition for N, P, K, S, Ca, and Mg, have been found to be 22, 4, 47, 10, 112, and 18 kg ha⁻¹, respectively (Hangs et al., 2014a), which indicates that SRW is capable of enhancing soil nutrients over time. However, there were calculated nutrient budget shortcomings (i.e., nutrient outputs > inputs + transfers) for N, P, K, Ca, and Mg of 17, 39, 112, 271, and 74 kg ha⁻¹, respectively, while S was surplus (60 kg ha⁻¹) with approximately 40 Mg ha⁻¹ of total below- and above-ground biomass production (Hangs et al., 2014b). This suggests that the removal of a higher amount of SRW biomass from the system will perhaps negatively impact the nutrient cycling through nutrient mining from the soil in the future rotation. Furthermore, in the following rotation, the SRW growth rate will be under full potential with a more robust root system, which possibly will demand more nutrient uptake and use from the soil.

4.7 Conclusions

The results from this study indicated that the cycling of different macronutrients (N, P, K, and S) are still in a fluctuating recovery and efficiency stage after the first rotation. Survival without any fertilizer application demonstrated that SRW land-use practice is perhaps low nutrient-demanding because the SRW is in its first rotation, and the root system is not fully established

yet. At this point, the SRW plantation in this study has not still accomplished the maximum potential growth rate and biomass production. However, the SRW willow growth rate will likely reach its full potential in the following rotation, and the root systems will continue to grow and mature, which will further increase nutrient uptake and use. Nonetheless, results from this field experiment revealed that the establishment of SRW in the riparian zones of the type of wetlands evaluated in this study would not severely diminish soil nutrient pools gradually during their first rotation.

5 SOIL ORGANIC CARBON BENEATH CONTRASTING RIPARIAN LAND-USES IN THE PRAIRIE POTHOLE REGION

5.1 Preface

Land-use practice changes have a substantial influence on the quantity as well as the chemical composition (i.e., chemical characteristics) of soil organic carbon (SOC). The dynamics of SOC are affected by its chemical stability, which is a crucial mechanism of SOC sequestration. Short rotation willow (SRW), known to be the high water and nutrient-demanding, can modify soil salinity in semi-arid PPR wetland systems, as discussed in Chapters 3 and 4 in the field experiment. Moreover, SRW land-use practice has a higher biomass production capacity compared to crop production, which could, in turn, add above- and below-ground biomass to the soil, potentially sequestering SOC. The SRW plantation in the form of agroforestry could thus mitigate climate change based on carbon sequestration in biomass and soil (i.e., positive soil organic carbon balance). It is crucial to quantify the SOC balance and assess the chemical stability to determine the sequestration potential under SRW plantation. Chapter 5 quantifies the sequestration capacity and characterizes the chemical compositions of SOC specifically under SRW plantation in the riparian zones of PPR wetlands compared to adjacent pasture (PA) and annual crop (AC) production in a field study. The conceptual model (Figure 5.1) shows the variables that were assessed in Chapter 5 and that could hypothetically be impacted by land-use practices.

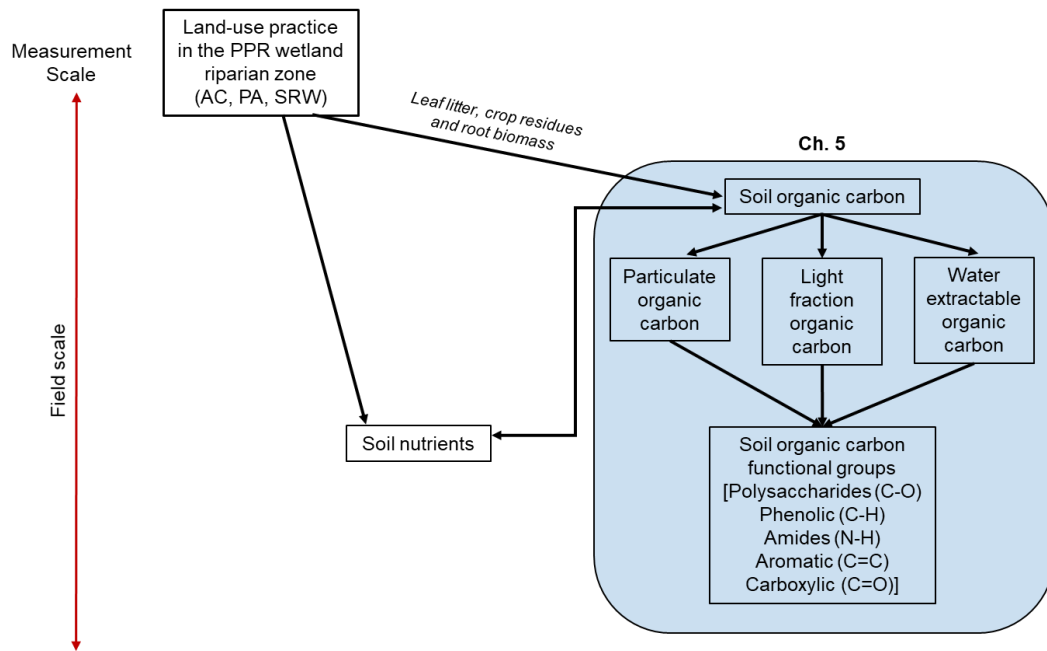


Figure 5.1 Conceptual model showing the potential effects of land-use practices on total SOC, POC, LFOC, WEOC, and SOC functional group in the riparian zones of the PPR wetland system.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, SOC = soil organic carbon, POC = particulate organic carbon, LFOC = light fraction organic carbon, WEOC = water extractable organic carbon, PPR = prairie pothole region.

5.2 Abstract

Short rotation willow (SRW) is a promising land-use practice in North America for providing a source of renewable energy and higher potential to sequester carbon while maintaining sustainable soil health. The impacts of SRW on soil organic carbon (SOC) quantity and characteristics are of concern for the prairie pothole region (PPR) agroecosystem due to the potential for mitigating atmospheric CO₂ emissions and impacting biogeochemical cycling. The SOC content, fractions, and composition were assessed at 0-30 cm depth under SRW plantation in the marginal riparian zones of PPR wetlands and compared to adjacent annual crop (AC) and pasture (PA) in a field experiment at two sites (A and B). Total (SOC), water extractable (WEOC), light fraction (LFOC), and particulate organic carbon (POC) were used to evaluate the content and its fractions, whereas Fourier Transform Infrared (FTIR) spectroscopy

was used to characterize the chemical composition in the whole soil. Total SOC was comparatively higher in PA in both sites; however, significant ($p < 0.05$) only in site B. Overall, the land-use effects on SOC was in the order of PA > SRW = AC. The LFOC and POC followed a similar land-use pattern in both sites. The WEOC was relatively higher in soils under SRW. Total SOC and WEOC content were significantly higher ($p < 0.05$) in the upper 15 cm across all land-use practices. Soil depth had more influence on SOC composition than land-use practices. The recalcitrant (RC) to labile (LC) ratios of phenolic and amides to polysaccharides were significantly higher ($p < 0.05$) in site A, while aromatic and carboxylic to polysaccharides were non-significantly lower ($p > 0.05$) under SRW in both sites. SOC functional groups were higher in the subsoil, which indicated altered spectral properties with depths. The higher alkyl-C to O-alkyl-C ratio suggested a higher degree of decomposition and better stability of SOC in the subsoil. Our results indicated that the SRW as a marginal riparian land-use practice in the PPR has beneficial impacts on SOC.

5.3 Introduction

Soil organic carbon (SOC) is a critical component of soil health, plays an essential role in the global carbon cycle (Janzen, 2004), and supports ecosystem services (i.e., nutrient cycling, production, habitat, biodiversity) through a variety of soil functions (Adhikari & Hartemink, 2016). Globally, the soil is the largest terrestrial pool of organic carbon (OC), which is about 2.3 times greater than atmospheric CO₂ and 3.5 times higher than the C in all living terrestrial plants (Lal, 2004). Terrestrial wetlands can store more than 20-30% of total earth's SOC (Mitsch & Gosselink, 2015), which is higher than the 0.5–2% C generally found in agricultural soils (Vepraskas & Craft, 2016), and 54% stored in the upper one meter (Jobbágy & Jackson, 2000). Soil organic carbon is derived directly from the plant inputs (or indirectly from animal residues) after microbial decomposition into various complex (bound as organo-mineral complex) (Paul, 2016) and physically uncomplexed (not attached to soil mineral particles) carbon particles

(Gregorich et al., 2006). Dynamics of SOC are affected by its stability, which is a crucial mechanism of C sequestration (Davidson & Janssens, 2006, Paul, 2016). The chemical stability of OC depends on whether it is protected from microbial decomposition by physical (aggregates), chemical (organo-mineral complexes and/or absorbed by soil particles), and/or biochemical (chemical composition of SOC) mechanisms (Han et al., 2016). The stability of SOC can be characterized as labile (LC), slow (SC), or recalcitrant (RC) based on the turnover rate (Six et al., 2002b), and into light fraction (LF) or heavy fraction (HF) based on density (Golchin et al., 1994). Labile C pools tend to react disproportionately to changes affecting the balance of SOC over a short period (Lal, 2006, Lucas & Weil, 2012). Compared to total SOC, LC pools such as particulate organic carbon (POC) (53-2000 μm), light fraction organic carbon (LFOC) (density $< 2.0 \text{ g cm}^{-3}$), and water extractable organic carbon (WEOC) (or dissolved organic carbon) are relatively small, have a fast turnover rate, and are easily affected by management systems (Mirsky et al., 2008, Six et al., 2002b). Hence, LC pools can be used as preliminary indicators to assess whether a particular land-use practice will act as a C sink or source (Gregorich et al., 1994, Sainepo et al., 2018).

Accumulation of SOC depends on the organic matter input and decomposition rates in soil (Janzen, 2006). The rate of SOC decomposition is usually controlled by the molecular structure, biological and environmental factors (Schmidt et al., 2011), such as litter quality and microbial community, which is closely related to C dynamics (Schimel & Schaeffer, 2012), and biogeochemical cycling in the agroecosystems (Ondrasek et al., 2019). The quality, function, and biochemical stability of SOC depend on the relative abundance of soil LC versus RC functional groups (Beer et al., 2008). As the soil organic matter (SOM) under different vegetation types from contrasting land-use practices decomposes, the chemical composition of SOC changes (Deng et al., 2019, Lafleur et al., 2015). Fourier Transform Infrared (FTIR) spectroscopy can reveal the biochemical characteristics and relative abundance of C

compounds (i.e., polysaccharides, phenolic, amides, aromatic, carboxylic) via the ratios of RC and LC fractions (Calderón et al., 2013).

Land-use practice change impacts the quantity and chemical composition (i.e., quality) of SOC (Ramesh et al., 2019). Changes in plant species, primary productivity, litter quantity, and quality under different land-use practices can have direct and indirect effects on SOC pools (Schoeneberger, 2008). A meta-analysis reported an increase in SOC with a land-use change from crop to pasture (+19 %) or forest (plantation forest +18 %; secondary forest +54 %) (Guo & Gifford, 2002). Likewise, another meta-analysis observed a significant increase in SOC stock due to land-use conversion to agroforestry from both agriculture and pasture (De Stefano & Jacobson, 2017). However, the changes in SOC content are greatly influenced by the climatic/environmental condition at the global scale and the natural variations in edaphic conditions and management at the local level (Stockmann et al., 2013). Changes in soil C content often occur gradually and can be challenging to detect in the short term, especially if there are high background levels already present (Gregorich et al., 1994).

The prairie pothole region (PPR) is a distinctive landscape in North America commonly known for its millions of small wetlands, large areas of native prairie (Winter, 1989), and highly productive agricultural lands (Gleason et al., 2011). Wetlands of the PPR provide various ecological benefits, including carbon sequestration (Badiou et al., 2011, Euliss et al., 2006). Under the anoxic soil environments, slow decomposition of PPR freshwater mineral wetlands leads to the accumulation of SOC, which accounts for 9% of the global wetland C pool (Bridgham et al., 2006). However, cultivation within the PPR has disrupted the native vegetation and led to the loss of SOC (Janzen et al., 1998, VandenBygaart et al., 2003). Natural solutions include land-use centered management interventions (Fargione et al., 2018), which typically allocate a significant fraction of their biomass and/or reduce the decomposition rates that can enhance SOC (Paustian et al., 2016). Across the PPR, there are millions of hectares of

marginal lands, including wetland riparian zones, which are not suitable for cultivation (Amichev et al., 2014b). In Saskatchewan, Canada, there is more than 2 million ha of marginal degraded lands that have the potential for C sequestration through short rotation willow (SRW) biomass production (Amichev et al., 2012). Hence, the establishment of SRW within the PPR could promote soil biological activity and SOC compared to annual crops; however, in the short term, it is usually lower than the pasture (Stauffer et al., 2014).

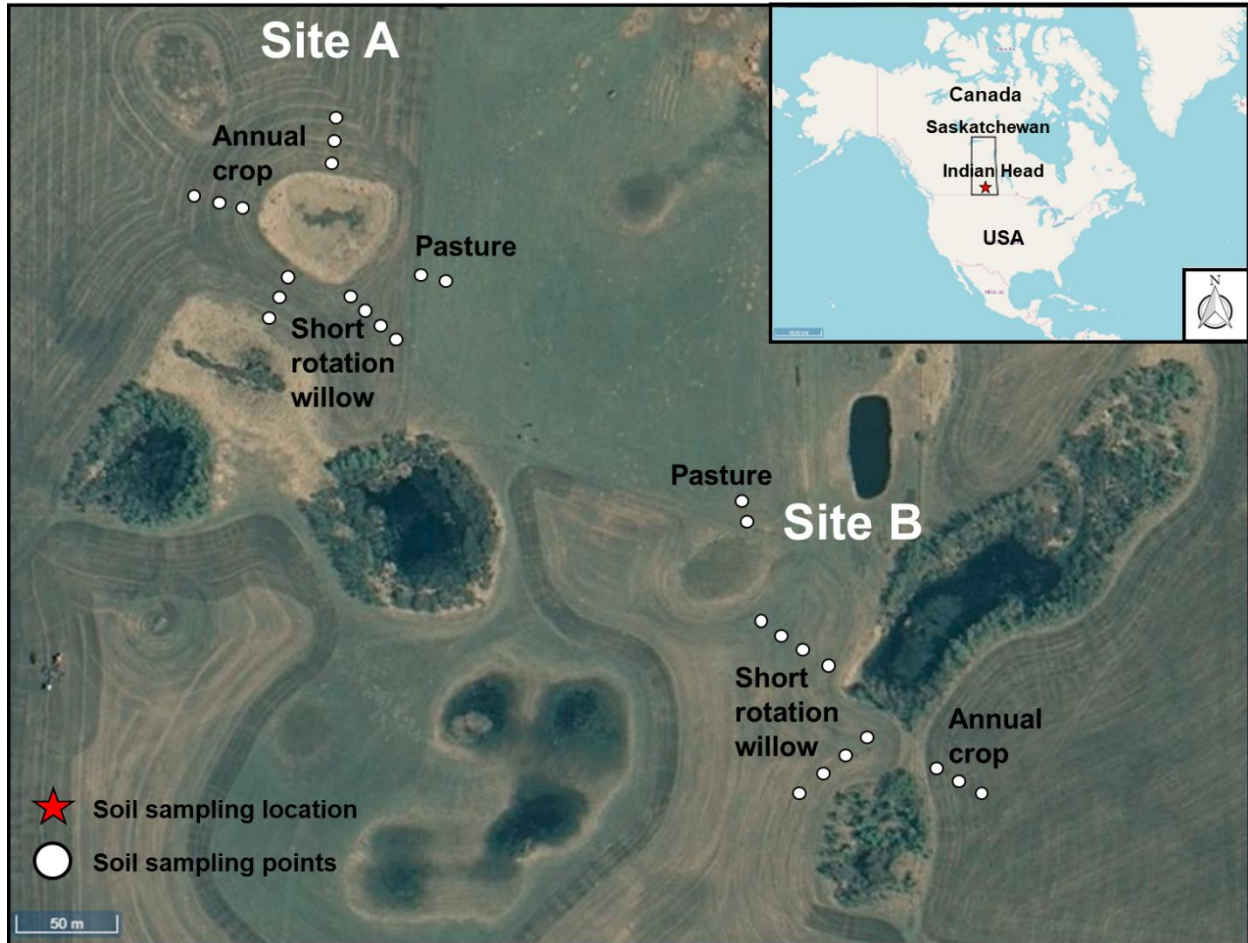
Short rotation willow (SRW) is a high-density, fast-growing, biomass-producing crop harvested under short rotation cycles for bioenergy purposes, which is of increasing interest in North America (Amichev et al., 2014b). It is also increasingly used in ecological restoration, nutrient buffering, and riparian erosion control (Kuzovkina & Quigley, 2005, Schoeneberger et al., 2012). It has already been explored as a source of bioenergy feedstock on marginal lands in the PPR, including wetland riparian zones in Saskatchewan, Canada (Amichev et al., 2014a, Hangs et al., 2014b). Research found that changing land-use practices to SRW from arable crops is likely to result in a net increase or neutral effect on soil C (Dimitriou et al., 2012b, Don et al., 2012, Qin et al., 2016); however, changing from grassland to SRW can be broadly neutral or decrease soil C (Harris et al., 2017, Harris et al., 2015). However, uncertainty remains about the SRW in grassland as the situation significantly depends on grassland type and management differences (Harris et al., 2015, Qin et al., 2016). For example, SRW can act as a net sink for C and reduce greenhouse gas emissions compared to marginally managed pasture (Harris et al., 2017). Hence, SRW land-use practice has the potential to sequester C while maintaining sustainable productivity of the soil (Lemus & Lal, 2005, Nair et al., 2010, Oelbermann et al., 2004). Short rotation willow can represent a carbon sink over a long period (Pacaldo et al., 2013); however, in the short term, LC pools can be used as a tool to assess C dynamics as it is sensitive to changes in land-use practices (Lockwell et al., 2012). The research question addressed is whether changes in quantity and quality of SOC will result in a sink or source under the first

rotation of SRW in the riparian zone of the PPR wetland system? We hypothesized that 1) the total SOC content will be higher, 2) physically uncomplexed (i.e., POC and LFOC) and dissolved (i.e., WEOC) organic carbon will be higher, and 3) the ratio of RC:LC will be lower under SRW compared to AC and PA land-use practices.

5.4 Materials and Methods

5.4.1 Study Site and Soil Sampling

The field study was conducted in two neighboring PPR wetland sites (sites A and B) in Indian Head, Saskatchewan, Canada (Figure 5.2) during 2013-2015. Both field sites consist of a level to gently-rolling (0-10 % slopes) topography formed on loamy glacial till with non-calcareous Black Chernozemic soils of the Oxbow Association (Saskatchewan Soil Survey Staff, 1986). At both sites, short rotation willow (SRW) variety *Salix dasyclados* Wimm. (cultivar 'India') was planted in the riparian zones of the wetlands in June of 2013. The SRW was planted adjacent to a 10-year-old stand of pasture (PA; the mixture of *Medicago sativa* and *Bromus madritensis*) and an area of the annual crop (AC; planted to *Avena sativa*). In 2013, 2014, and 2015, soil samples were collected each fall along transects at both sites (Figure 5.2) in each of the three different land-use practices. At each sampling point, soil samples were collected from two depths (0-15 and 15-30 cm) by an auger. All soil samples were transported into the laboratory and frozen until analysis. Each sample was divided into two sub-samples. The major portion of each field-moist sample was air-dried and sieved through 2 mm sieve (for soil physical and chemical properties, total SOC, POC, LFOC, and FTIR analysis for the chemical composition of SOC). Another portion (for WEOC) was stored field-moist at -20°C. For soil bulk density, three replicate samples were collected at 0-15 and 15-30 cm depths by using a core sampler (3-cm tall × 5.4-cm i.d.) from each soil sampling point.



Source: SGIC (2020)

Figure 5.2 Location of the field study sites and soil sampling points at Indian Head, SK, Canada (N 50° 30.605'; W 103° 43.011', elevation 579 m above sea level).

^a The white dots represent points of soil samples collection used for soil carbon measurements.

5.4.2 Physically Uncomplexed, Dissolved and Total Organic Carbon in Soil

For total soil organic carbon (SOC), representative sub-samples were ground with a roller-mill and analyzed using an automated combustion technique using a Leco-2000 CNS analyzer (Leco Corporation, St. Joseph, MI, USA) (Skjemstad & Baldock, 2008). Analysis of SOC involved pre-treating the soil with hydrochloric acid to remove carbonates in the ceramic crucible after weighing, then drying the sample for 24 h at 80°C before analysis for C. Equivalent mass calculations (Ellert & Bettany, 1995) were used to correct all SOC values. For particulate organic carbon (POC), 25 g of air-dried soil was separated by dispersing soil with 100 mL

sodium hexametaphosphate, shaking for 16 h, and passing the dispersed samples through a 53- μm sieve (Cambardella & Elliott, 1992). Materials remaining on the sieve consisted of sand and POC, dried at 60°C, weighed, and the obtained materials were analyzed for SOC using the automated combustion technique using a Leco-2000 CNS analyzer (Leco Corporation, St. Joseph, MI, USA) after treating the soil with hydrochloric acid to remove carbonates (Skjemstad & Baldock, 2008). For light fraction organic carbon (LFOC), 25 g of air-dried soil was suspended in 50 mL of NaI solution (specific gravity 1.7 g cm⁻³) and dispersed for 60 min using a reciprocating shaker. After a settling period of 48 h, the suspended material was transferred by suction to a filtration unit (Gregorich & Beare, 2008). The composited light fraction materials were washed in 0.01 M CaCl₂ solution followed by distilled water, dried at 60°C, and weighed. The LF materials were analyzed for C using the automated combustion technique using Leco C632 CNS analyzer (Leco Corporation, St. Joseph, MI, USA). For water extractable organic carbon (WEOC), 20 g of moist field soil was extracted with 30 mL of 5 mM CaCl₂ solution (Chantigny et al., 2008). The resulting slurry was filtered through a vacuum filter unit equipped with a 0.45 μm polycarbonate filter. The filtered solutions were analyzed for dissolved organic carbon by automated combustion procedures using a TOC-VCPN analyzer (Shimadzu Scientific Instruments, Kyoto, Japan).

5.4.3 Soil Physical and Chemical Characteristics

Soil bulk density was calculated from the ratio of the mass of oven-dried soil (at 105°C) to the bulk volume of cylindrical core (68.71 cm³) soil collected from the desired soil layer (i.e., 0-15 and 15-30 cm) in the field (Hao et al., 2008). Soil volumetric water content was calculated from the bulk density and gravimetric water content measured by the weight loss of 5 g of fresh soil samples after they were dried at 105°C (Topp et al., 2008). Soil particle size distribution was calculated using the modified pipette method (Kroetsch & Wang, 2008). Total soil carbon (TSC) was determined by dry combustion using a Leco-2000 CNS analyzer (Leco Corporation, St.

Joseph, MI, USA) (Skjemstad & Baldock, 2008). Total nitrogen (TN) was determined using dry combustion with a Leco C632 CNS analyzer (Leco Corporation, St. Joseph, MI, USA) (Rutherford et al., 2008). Light fraction C and N were obtained during LFOC analysis (Gregorich & Beare, 2008) were analyzed for C and N by the automated combustion technique using Leco C632 CNS analyzer (Leco Corporation, St. Joseph, MI, USA) to obtain light fraction organic nitrogen (LFON). The solution obtained during WEOC measurement (Chantigny et al., 2008) was also analyzed for total dissolved nitrogen (TDN) using automated combustion procedures using a TOC-VCPN analyzer (Shimadzu Scientific Instruments, Kyoto, Japan).

5.4.4 Fourier Transform Infrared Spectroscopic Analyses of Soil Organic Carbon Composition

The structural composition of labile and recalcitrant SOC chemical functional groups were investigated by FTIR spectroscopy. Representative soil sub-samples were air-dried and finely roller-milled before analyses, and then measured in Attenuated Total Reflectance mode by pressing the powder sample on a diamond-coated ZnSe crystal. Spectral data were recorded using an FTIR spectrometer (Bruker Optics Equinox 55, Ettlingen, Germany) equipped with an N₂(l)-cooled MCT detector over the range of 4000 to 400 cm⁻¹. For all samples, ambient air was used as a background. Spectral wavenumber from 950 to 1750 cm⁻¹ was used for SOC characterization, as all the important chemical functional groups, including biochemical labile carbon (LC) and recalcitrant carbon (RC), fall within these values (Calderón et al., 2013). Absorption bands represented polysaccharides (i.e., LC fraction) at 950 to 1170 cm⁻¹ (~1030 cm⁻¹: C-O stretching and O-H deformation). Biochemically recalcitrant carbon fractions were between 1400 to 1750 cm⁻¹; specifically, phenolic and aliphatic groups (~1420 cm⁻¹: C-H deformation of CH₂ or CH₃), amide groups (~1510 cm⁻¹: aromatic C=C or CO of amide groups), lignin and other aromatics and aromatic or aliphatic carboxylates (~1630 cm⁻¹: aromatic C=C and asymmetric COO⁻), and carboxylic acids and aromatic esters (~1720 cm⁻¹: C=O stretch of

COOH or COOR) (Broder et al., 2012, Gillespie et al., 2015). The relative abundance (ratio) of recalcitrant C to labile C (RC:LC) chemical functional groups present in soils from different land-use practices were calculated from the relative peak ratios of 1420/1030, 1510/1030, 1630/1030, and 1720/1030 (Niemeyer et al., 1992).

5.4.5 Data Presentation and Statistical Analyses

All data were visualized and statistically analyzed using the R version 3.4.4 for Windows (R Core Team, 2018). The normality of data was assessed by the Shapiro-Wilk test and histogram. Homogeneity of variances or homoscedasticity of the data were assessed by Levene's test using "car" package. Data were square root transformed whenever necessary to meet the assumptions of normality. Univariate analysis of variance (ANOVA) with generalized linear mixed-effects models (Bolker et al., 2009, Zuur et al., 2009) through "lmerTest" was used to see a significant difference (hypothesis testing) among different land-use practices, depths and year for soil organic carbon and its fractions. When significant effects were found in ANOVA, multiple comparisons of means among land-use practices were compared by Tukey Honest Significant Difference test (Tukey HSD) using the "TukeyC" package. Permutation multivariate ANOVA (PERMANOVA) was used to assess significant difference (hypothesis testing) of total soil organic carbon and its fractions combinedly using the "vegan" package. The relationship among SOC, POC, LFOC, WEOC, and soil physiochemical characteristics were assessed through Spearman's rank-order correlation test using "corrplot" package. Non-metric multidimensional scaling (NMDS) with Bray-Curtis matrix of dissimilarities was used to plot the original position in multidimensional space to visualize the difference among land-use practices and years along with soil organic carbon fractions using "vegan" package. The linear relationship among soil characteristics (physical and chemical) and soil organic carbon fractions were analyzed by redundancy analysis (RDA) through the development of multiple linear regression to reflect variables in the same Cartesian coordinate system using "vegan" package.

Spectral data obtained through FTIR were first treated for baseline correction and normalization using “ChemoSpec” package. The FTIR spectra from each land-use practices and depths were quantitatively compared by integrating normalized data across regions assigned to soil organic carbon chemical functional groups. The significant difference in the RC:LC ratios of SOC chemical functional groups among land-use practices in different depth classes were determined through ANOVA. The FTIR absorption band intensities were analyzed using PERMANOVA through “vegan” package to see differences among land-use practices and between depths. Similarity percentage (SIMPER) was used to determine which FTIR absorption band was responsible for the contrast among land-use practices and depths. Bray-Curtis distance measurement was used both for PERMANOVA and SIMPER. The heatmap representation along with hierarchical cluster analysis (HCA) of both LC and RC chemical functional groups revealed by FTIR was constructed to identify clustering in different land-use practices of depth classes using “ChemoSpec” package. The linkage of both LC and RC with total soil organic carbon and their fractions as well as the soil physical and chemical characteristics under different land-use practices were evaluated by constrained analysis of principal coordinates (CAP) by “vegan” package.

5.5 Results

5.5.1 Soil Organic Labile Carbon Fractions under Different Land-use Practices

Soil organic carbon content was relatively higher under PA land-use practices compared to AC and SRW in both sites at both depths (Figure 5.3A, B, C, J, K, L): however, differences were only significant ($p < 0.05$) in site B, and there were no differences between AC and SRW (Table 5.1). There were no differences in SOC among measurement years, and 0-15 cm had significantly ($p < 0.001$) higher SOC than 15-30 cm across all land-uses at both sites (Table 5.1).



Figure 5.3 Distribution of total organic carbon and labile carbon fractions under different land-use practices. Total soil organic carbon at 0-15cm and 15-30cm depths from site A (A, B, C) and site B (J, K, L) in 2013, -14, -15, respectively. Water extractable organic carbon at 0-15cm and 15-30cm depths from site A (D, E) and site B (M, N) in 2014, -15, respectively. Particulate organic carbon from site A (F, G) and site (O, P) in 2014, -15, respectively. Light fraction organic carbon from site A (H, I) and site (Q, R) in 2014, -15, respectively.

^a Error bar represents standard deviations.

^b AC = annual crop, PA = pasture, SRW = short rotation willow, SOC = soil organic carbon, WEOC = water extractable organic carbon, POC = particulate organic carbon, LFOC = light fraction organic carbon.

Table 5.1 Significance levels from ANOVA of the effects of land-use practices, depths, and year on soil labile and recalcitrant carbon fractions.

Sources of variation		TSC			SOC		WEOC			POC		LFOC	
		df	F stat	p-value	F stat	p-value	df	F stat	p-value	F stat	p-value	F stat	p-value
Site A	Land-use	2	4.92	0.009 **	1.37	0.260 ^{ns}	2	1.55	0.219 ^{ns}	8.84	<0.001 ***	13.95	<0.001 ***
	Depth	1	0.94	0.332 ^{ns}	39.53	<0.001 ***	1	12.54	<0.001 ***	-	-	-	-
	Year	2	6.61	0.002 **	0.25	0.779 ^{ns}	1	0.02	0.895 ^{ns}	1.90	0.178 ^{ns}	0.46	0.501 ^{ns}
Site B	Land-use	2	4.09	0.020 *	6.52	0.002 **	2	1.49	0.233 ^{ns}	9.19	<0.001 ***	6.95	0.004 **
	Depth	1	0.33	0.562 ^{ns}	31.55	<0.001 ***	1	10.10	0.002 **	-	-	-	-
	Year	2	0.85	0.432 ^{ns}	1.79	0.173 ^{ns}	1	6.60	0.013 *	0.14	0.712 ^{ns}	1.01	0.325 ^{ns}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b TSC = total soil carbon, SOC = soil organic carbon, WEOC = water extractable organic carbon, POC = particulate organic carbon, LFOC = light fraction organic carbon.

In terms of WEOC, no significant difference ($p > 0.05$) was noted among land-use practices in both sites (Figure 5.3; Table 5.1). Water extractable organic carbon content was significantly different ($p < 0.01$) between depths and was significantly higher ($p < 0.05$) at the top 0-15 than 15-30 cm depth. There was no significant difference ($p > 0.05$) in site A but significant ($p < 0.05$) in site B; however, 2014 had a relatively higher WEOC concentration than 2015.

Both particulate organic carbon content and LFOC (measured in 0-15 cm only) were significantly different ($p < 0.01$) among land-use practices (Table 5.1). The pasture had significantly higher ($p < 0.05$) POC and LFOC content compared to other land-use practices; however, there were no significant differences ($p > 0.05$) found between AC and SRW for POC or LFOC (Figure 5.3).

Multivariate analysis of variance (PERMANOVA) was used to assess the combined effects of land-use practices, depths, and time (i.e., years) on the total soil organic and carbon fractions (Table 5.2). A significant impact of depth ($p < 0.001$) was observed in site A, whereas, in site B the difference among land-use practices, depths, and years was statistically significant ($p < 0.05$). However, in site B, the variance it explained as indicated by the R^2 value from the PERMANOVA test (R^2 for land-use is 0.084, and for the year is 0.032) was relatively low (Table 5.2). Similarity percent (SIMPER) results (not shown here) showed that the contrast between the year 2014 and 2015 was significant ($p < 0.05$) for POC and LFOC in site A, whereas for POC and WEOC in site B. Significantly higher ($p < 0.05$) contrast was observed between depths for SOC and WEOC content.

Table 5.2 Significance levels from PERMANOVA of the effects of land-use practices, depths, and year on labile and recalcitrant carbon fractions in the soil.

	Sources of variation	df	F-Model	R ²	Pr (>F)
Site A	Land-use	2	1.331	0.022	0.247 ^{ns}
	Depth	1	35.848	0.291	0.001 ^{***}
	Year	2	0.309	0.005	0.760 ^{ns}
	Residuals	84		0.682	
Site B	Land-use	2	4.635	0.084	0.006 ^{**}
	Depth	1	24.003	0.216	0.001 ^{***}
	Year	2	2.840	0.051	0.032 [*]
	Residuals	72		0.649	

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively; ^{ns}, is not significantly different ($p > 0.05$).

Non-metric multidimensional scaling (NMDS) of total soil organic and labile carbon fractions did not differ among land-use practices in both sites (Figure 5.4A, B), suggesting that land-use was not a key factor driving the variability. The NMDS ordination tends to show a clustering along with PA land-use practice in both sites, with stress values less than 0.10, which provides a good representation of data in reduced dimension. From the NMDS plot, the association of soil LFOC with PA land-use practices is evident in both sites (Figure 5.4A, B). Also, a relatively wide dispersion was noted for SRW and AC along axis 1 in both sites. The NMDS axis 1 was positively correlated with SOC and WEOC, whereas it was negatively correlated with POC and LFOC in both sites. The NMDS axis 2 was positive with POC and negatively with SOC, LFOC, WEOC in site A, and positively with SOC and POC, and negatively with LFOC and WEOC in site B (Figure 5.4A, B).

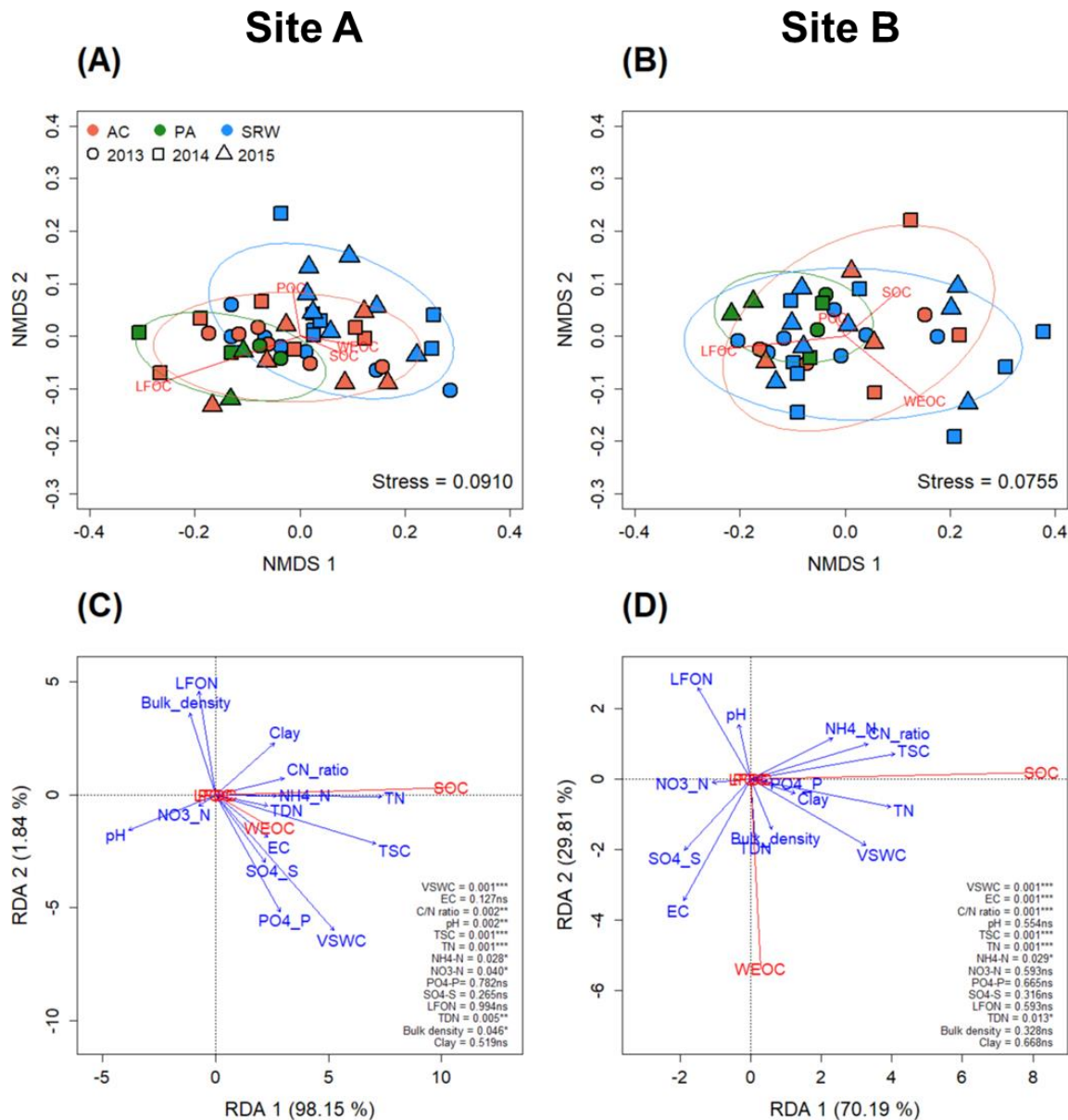


Figure 5.4 Non-metric multidimensional scaling (NMDS) for the comparison of total organic and labile carbon fraction and redundancy analysis (RDA) for revealing the effects of measured soil physical and chemical characteristics visualized from site A (A, C), and site B (B, D) for different land-use practices in 2013, -14, and -15.

^a Blue vectors ($r > x$) indicate linear correlations between the ordination and soil physical and chemical properties. Directions and lengths of the vectors indicate the strength of correlations between variables. The angles between vectors reflect their correlations (i.e., a vector pair with an angle of 20° have strong positive correlation as $\cos(20^\circ) = 0.94$, and with an angle of 90° are uncorrelated as $\cos(90^\circ) = 0$).

^b *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^c AC = annual crop, PA = pasture, SRW = short rotation willow, SOC = soil organic carbon, WEOC = water extractable organic carbon, POC = particulate organic carbon, LFOC = light fraction organic carbon, TN = total nitrogen, LFON = light fraction organic nitrogen, TDN = total dissolved nitrogen, EC = electrical conductivity, TSC = total soil carbon, VSWC = volumetric soil water content.

Redundancy analysis of the total soil organic and labile carbon fractions was performed to explore the relationship with the measured soil physical and chemical characteristics (Figure 5.4C, D). The first two-component explained 98.15 % and 1.84 % of site A (Figure 5.4C), and 70.19 % and 29.81 % of site B, respectively (Figure 5.4D). The results illustrated that the vector line of bulk density, VSWC, C/N ratio, pH, TSC, TN, $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, and TDN in site A, whereas VSWC, EC, C/N ratio, TSC, TN, $\text{NH}_4^+\text{-N}$, TDN in site B were statistically significant ($p < 0.05$).

5.5.2 Soil Organic Carbon Chemical Composition under Different Land-use Practices

The normalized and average FTIR spectra from different land-use practices of both sites were used to distinguish the soil organic carbon chemical functional groups (Figure 5.5A, B). Overall, the highest intensity absorbance peak was observed at 1030 cm^{-1} (absorbance range 950 to 1170 cm^{-1}) due to C-O stretching and O-H deformation, attributed mostly to polysaccharides, which suggested a large portion of LC content under all land-use practices. Across the RC range (1400 to 1750 cm^{-1}), higher absorbance peaks were observed for phenolic and amide groups (1420 and 1510 cm^{-1}) both under SRW and AC in site A and under SRW and PA in site B (Figure 5.5A, B), suggesting different relative concentrations among contrasting land-use practices in both sites. There was a smaller absorbance peak noted for aromatic and carboxylic groups (1630 and 1720 cm^{-1}).

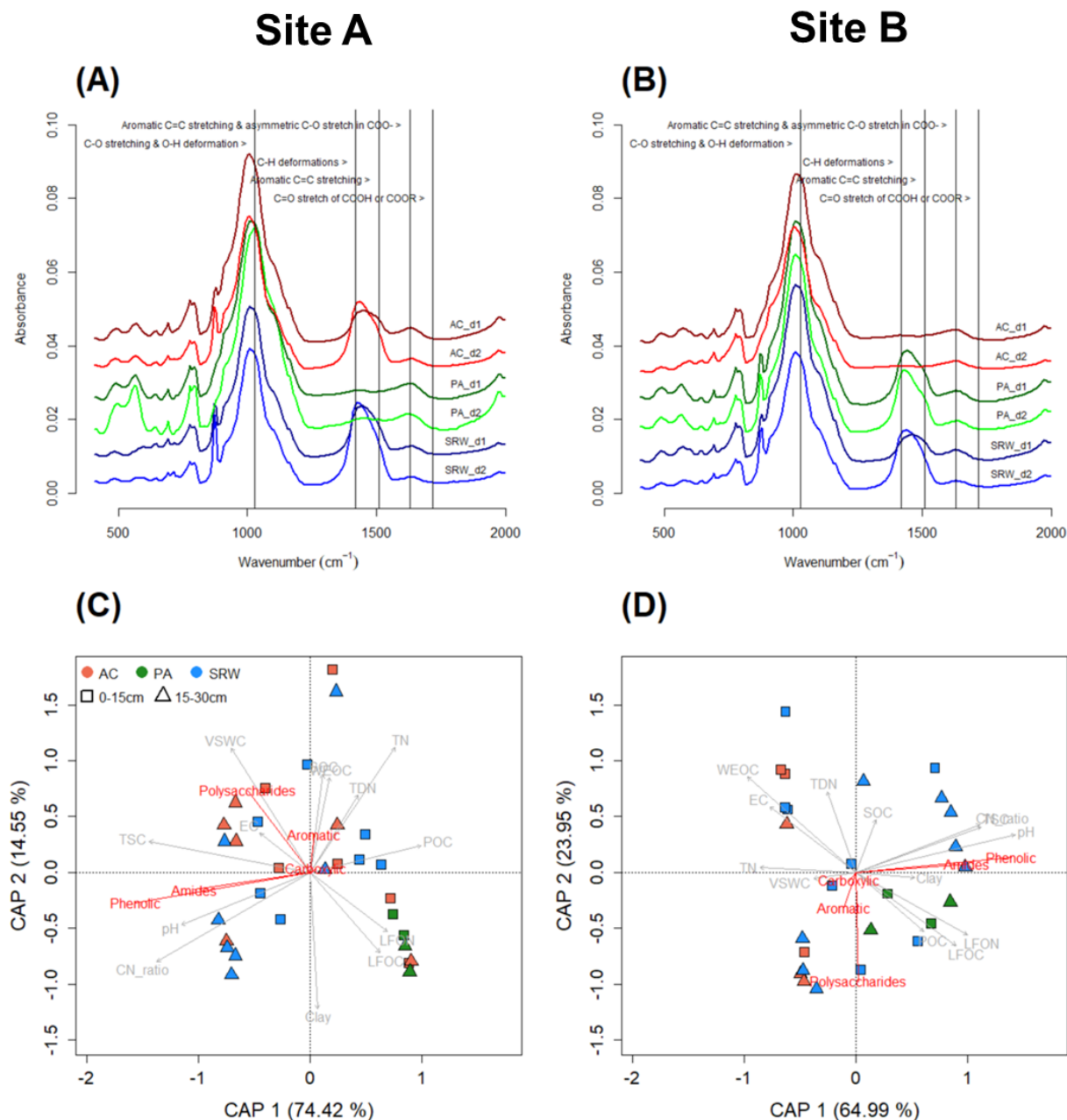


Figure 5.5 Fourier-transform infrared spectra (normalized and averaged), and constrained analysis of principal coordinates (CAP) of soil organic carbon chemical functional groups based on labile and recalcitrant carbon fractions under different land-use practices from site A (A, C), and site B (B, D).

^a Data were square root transformed for CAP analysis.

^b AC = annual crop, PA = pasture, SRW = short rotation willow, d1 = 0-15 cm, d2 = 15-30 cm, SOC = soil organic carbon, WEOC = water extractable organic carbon, POC = particulate organic carbon, LFOC = light fraction organic carbon, TSC = total soil carbon, TN = total nitrogen, LFN = light fraction organic nitrogen, TDN = total dissolved nitrogen, EC = electrical conductivity, VSWC = volumetric soil water content.

In site A, the CAP analysis based on the relative abundance of chemical functional groups by land-use accounted for 88.97 % (74.42 % for CAP 1 and 14.55 % for CAP 2), compared to 88.94% of the total variance in site B (64.92 % for CAP 1 and 23.95 % for CAP 2; Figure 5.5C and D, respectively). Axis 1 from CAP analysis for both sites covered a significant variation ($p = 0.002$ in site A, and $p = 0.001$ in site B); the ordination plots tend to separate the PA from other land-use practices along CAP 1. The chemical functional groups of SOC were distinctly separated in quadrants in both sites (Figure 5.5C, D). In site A, aromatic and carboxylic were in the first quadrant, amides and phenolic in the third, and polysaccharides groups in the fourth. In site B, amides and phenolic were in the first quadrant, polysaccharides in the second, and aromatic and carboxylic groups in the fourth.

The mean \pm SD (standard deviations) ratios of different RC:LC soil organic carbon chemical functional groups with the Tukey HSD test are presented in Table 5.3. Statistically significant ($p < 0.01$) variation was observed in phenolic to polysaccharides (1420/1030) and amides to polysaccharides (1510/1030) ratios among the land-use practices in site A (Table 5.4).

However, none of the SOC chemical functional groups ratios were significant in terms of land-use practices at either site (Table 5.4). Tukey results showed that the mean ratio for SRW was significantly ($p < 0.05$) different; however, AC and PA land-use practices were not statistically significant from each other ($p > 0.05$). Overall, all the RC:LC ratios were higher at lower 15-30 compared to top 0-15 cm depth, except the ratios of aromatic to polysaccharides (1630/1030), which was opposite in both sites (Table 5.3). The mean ratio of 1420/1030 (phenolic to polysaccharides) and 1510/1030 (amides to polysaccharides) were significantly higher ($p < 0.05$) at 15-30 cm than at 0-15 cm in site A; however, none of the other differences between depths was statistically significant. In site B, none of the differences between depths were statistically significant ($p > 0.05$), except for 1630/1030 (aromatic to polysaccharides), the top 0-

15 cm was significantly ($p < 0.05$) higher compared to 15-30 cm depth (Table 5.3 and Table 5.4).

Table 5.3 Mean (\pm SD) ratios of the soil RC:LC chemical functional groups revealed by FTIR spectra under different land-use practices and depths in both sites.

			Ratio	Ratio	Ratio	Ratio
			1420/1030	1510/1030	1630/1030	1720/1030
Site A	Land-use	AC	0.268 \pm 0.240ab	0.175 \pm 0.130ab	0.058 \pm 0.017a	0.006 \pm 0.006a
		PA	0.028 \pm 0.007b	0.031 \pm 0.011b	0.073 \pm 0.008a	0.010 \pm 0.007a
		SRW	0.467 \pm 0.320a	0.267 \pm 0.151a	0.051 \pm 0.028a	0.004 \pm 0.002a
Depth	0-15 cm	0.208 \pm 0.188b	0.153 \pm 0.127b	0.063 \pm 0.021a	0.005 \pm 0.003a	
	15-30 cm	0.450 \pm 0.349a	0.245 \pm 0.164a	0.050 \pm 0.023a	0.006 \pm 0.007a	
Site B	Land-use	AC	0.014 \pm 0.004a	0.014 \pm 0.005a	0.054 \pm 0.005a	0.006 \pm 0.002a
		PA	0.289 \pm 0.178a	0.172 \pm 0.070a	0.057 \pm 0.011a	0.005 \pm 0.002a
		SRW	0.289 \pm 0.416a	0.193 \pm 0.227a	0.051 \pm 0.019a	0.005 \pm 0.004a
Depth	0-15 cm	0.125 \pm 0.159a	0.110 \pm 0.157a	0.059 \pm 0.009a	0.006 \pm 0.003a	
	15-30 cm	0.325 \pm 0.455a	0.187 \pm 0.223a	0.046 \pm 0.018b	0.005 \pm 0.003a	

^a Values represent mean \pm standard deviations (\pm SD).

^b Means within a column for land-use and depth followed by the same letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, LC = labile carbon, RC = recalcitrant carbon.

^d Representative FTIR spectra absorbance band 1030 = polysaccharides (C-O), 1420 = phenolic (C-H), 1510 = amides (N-H), 1630 = aromatic (C=C), 1720 = carboxylic (C=O).

Table 5.4 Significance levels from ANOVA of the effects of land-use practices and depths on the ratio of the soil RC:LC chemical functional groups.

		Ratio		Ratio		Ratio		Ratio		
		1420/1030		1510/1030		1630/1030		1720/1030		
		df	F stat	p-value	F stat	p-value	F stat	p-value	F stat	p-value
Site A	Land-use	2	6.625	0.004 **	6.613	0.004 **	1.967	0.158 ^{ns}	2.882	0.072 ^{ns}
	Depth	1	8.688	0.006 **	4.504	0.042 *	3.389	0.076 ^{ns}	0.089	0.767 ^{ns}
Site B	Land-use	2	1.861	0.230 ^{ns}	2.461	0.105 ^{ns}	0.408	0.669 ^{ns}	0.215	0.808 ^{ns}
	Depth	1	2.783	0.139 ^{ns}	1.333	0.259 ^{ns}	6.077	0.021 *	1.542	0.225 ^{ns}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b LC = labile carbon, RC = recalcitrant carbon.

^c Representative FTIR spectra absorbance band 1030 = polysaccharides (C-O), 1420 = phenolic (C-H), 1510 = amides (N-H), 1630 = aromatic (C=C), 1720 = carboxylic (C=O).

The PERMANOVA indicated that FTIR band absorbances were not significantly ($p > 0.05$) different among land-use practices; however, they differed significantly ($p < 0.05$) between depth classes in both sites (Table 5.5). The SIMPER results (not shown here) showed that there were significantly higher phenolic ($p = 0.03$) and amide ($p = 0.01$) groups present in PA than SRW land-use practice, and significantly higher phenolic ($p = 0.01$) and amide ($p = 0.01$) groups at 0-15 than 15-30 cm in site A. No significant difference ($p > 0.05$) were observed among land-use practices or depths in site B.

Table 5.5 Significance levels from PERMANOVA of labile and recalcitrant soil organic carbon chemical functional groups revealed by FTIR spectra.

	Sources of variation	df	F-Model	R ²	Pr (>F)
Site A	Land-use	2	2.509	0.141	0.070 ^{ns}
	Depth	1	4.579	0.129	0.019 [*]
	Residuals	26		0.730	
Site B	Land-use	2	1.048	0.078	0.403 ^{ns}
	Depth	1	2.818	0.105	0.040 [*]
	Residuals	72		0.817	

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively; ^{ns}, is not significantly different ($p > 0.05$).

The heatmap with hierarchical cluster analysis revealed that, based on the land-use practices, SOC chemical functional groups could be divided into two clustering groups at both depths in both sites (Figure 5.6). The AC and SRW formed an identical clustering group distinct from PA at both 0-15 and 15-30 cm depth in site A, and 0-15 cm depth in site B (Figure 5.6A, B, C). However, in site B, at 15-30 cm depth, PA and SRW formed an identical clustering group apart from AC land-use practice (Figure 5.6D).

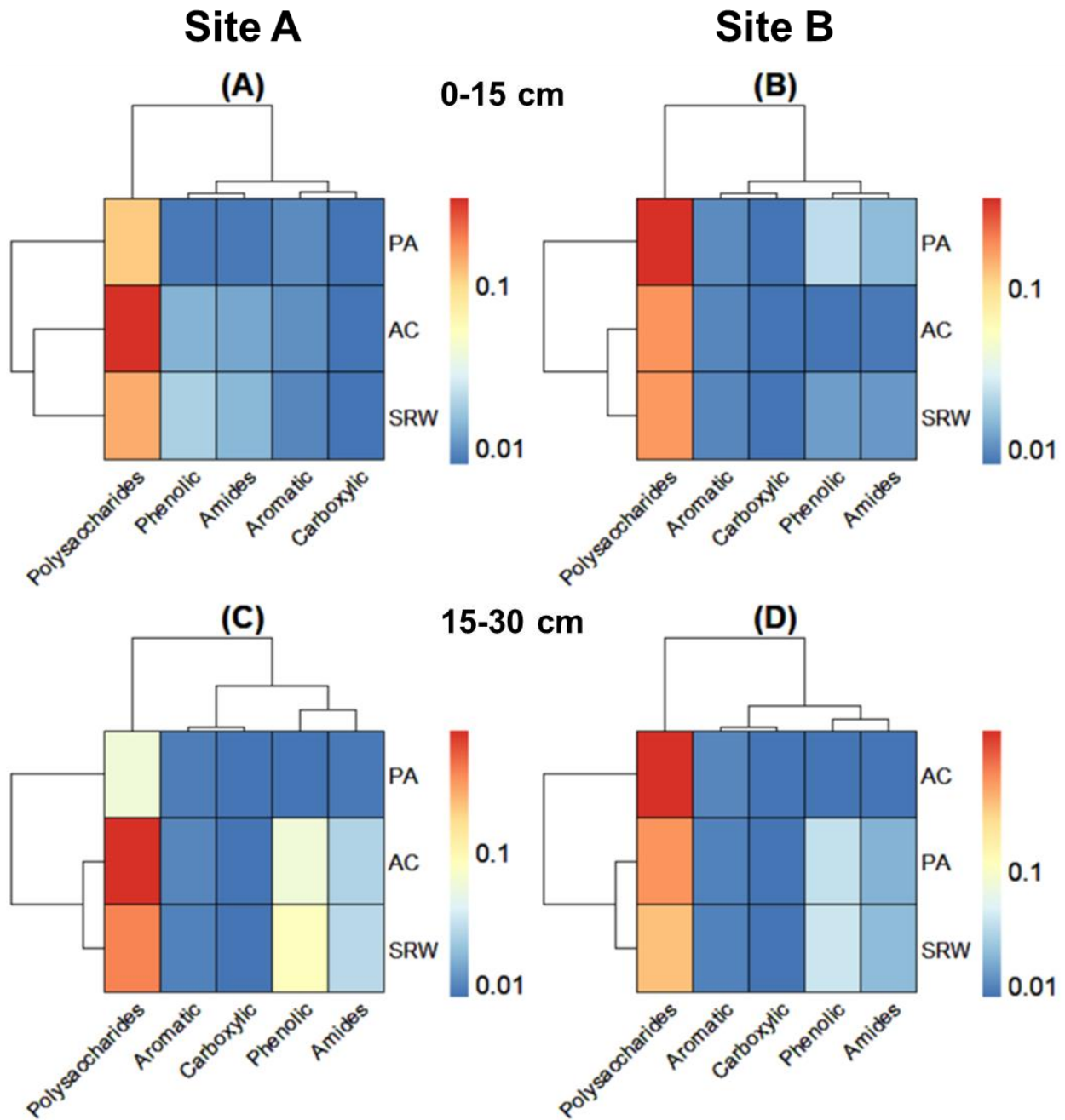


Figure 5.5 Heatmap with hierarchical cluster analysis of soil organic carbon chemical functional groups under different land-use practices at 0-15 and 15-30 cm depths from site A (A, C) and site B (B, D).

^a AC = annual crop, PA = pasture, SRW = short rotation willow.

5.5.3 Soil Physical and Chemical Characteristics

Soil physical and chemical characteristics under different land-use practices at 0-15 and 15-30 cm depths from both sites are presented in Table C-S1. No significant differences ($p > 0.05$) were observed among land-use practices in terms of VSWC, EC, pH, TSC, TN, C/N ratio, NH_4^+ -N, NO_3^- -N, PO_4^{3-} -P, and TDN except for SO_4^{2-} -S, LFON, and clay content in site A, and pH, TSC, NH_4^+ -N, PO_4^{3-} -P, SO_4^{2-} -S, TDN, and bulk density in site B, which were significantly different ($p < 0.05$). Significantly ($p < 0.05$) higher clay content was observed at 15-30 cm compared to 0-15 cm depth in both sites. The relationship between SOC and TSC, C/N ratio, VSWC, TN were significantly ($p < 0.05$) positive, whereas with LFOC, LFON and pH were significantly ($p > 0.05$) negative (Figure C-S1). However, none of the other relationships was statistically significant ($p > 0.05$).

5.6 Discussion

5.6.1 Land-use Effects on SOC Content

Total SOC was higher in PA compared to other land-use practices from both sites in our experiment. This is consistent with a meta-analysis on the effects of different land-use practices on soil C stocks that found higher SOC concentration in PA compared to AC and plantation forests (Guo & Gifford, 2002) and the findings of Harris et al. (2017) where 0-30 cm SOC stocks were substantially higher under PA than SRW. Similarly, Stauffer et al. (2014) observed lower SOC under SRW plantation compared to PA. These differences may be attributable to differences in the nature of the belowground biomass: Guo et al. (2007) observed 36 % higher annual inputs from the fine roots (diameter < 2 mm) biomass under the PA land-use practice than adjacent plantation forests. Moreover, compared to PA, the roots under plantation forests were mostly alive and died more slowly.

Lower SOC content under AC compared to PA land-use practice in this experiment is probably due to the removal of a large portion of C sequestered by the grain harvest, straw removal, and

repeated mechanical disturbance during the crop production every year (Grigal & Berguson, 1998). Cultivation is the most important factor driving SOC loss from the agriculture soil, and our results are consistent with the literature (Lal, 2006, Lal et al., 2004). Moreover, increased microbial decomposition of SOC is likely due to the turning and breakup of soil aggregates by tillage under AC land-use practice (Six et al., 2002a).

No significant year effects were observed in our study. A study with the first harvest of SRW (*Salix* Spp.) across a wide range of soils and land-use in Canada found an initial loss of soil C at plantation establishment regardless of initial SOC concentrations (Ens et al., 2013a). However, high-biomass SRW is expected to add a substantial amount of both above- and below-ground C to the soil in the subsequent years (Nair et al., 2010, Oelbermann et al., 2004). Within 2-3 years after SRW planting, the one-time effect of cultivation disturbance is diminished, decomposition will slow down, and the leaf litter input will contribute to further accumulation of SOC (Grigal & Berguson, 1998, Paul et al., 2002). After SRW establishment, SOC contents have been found to significantly decrease during the first five years at the upper 0-15 cm depth, followed by recovery from this point on to age 19 years (Pacaldo et al., 2013). We did not find any significant decrease or increase in SOC under SRW during the first rotation. However, a meta-analysis indicated that SOC under SRW establishment might not fully recover within the 20-30 years (Guo & Gifford, 2002, Laganière et al., 2010). Ultimately, the C recovery process will vary based on the management practices in the subsequent rotation under SRW, as it may receive more frequent disturbance than other land-use practices.

The LC fractions often comprise a substantial portion of the total SOC and can respond to change in input (Gregorich et al., 2006). Light fraction and particulate OC mainly originate from fresh plant residues and are significant to SOC turnover in agricultural soils because they provide a readily decomposable substrate to microorganisms (Gregorich et al., 1994). We observed significant land-use effects on the POC and LFOC content in the 0-15 cm increment.

Both POC and LFOC were significantly higher under PA compared to SRW or AC land-use practices. Similar to our experiment, Akinsete & Nortcliff (2014) observed higher LC content under pasture compared to agricultural land-use practices. However, other researchers have found that labile forms of C were lower in pasture soils (Paul et al., 2008) and agricultural soils (Purakayastha et al., 2007) compared to secondary or plantation forests and that POC was not different (Sainepo et al., 2018) between annual crop and pasture land-use practices. Six et al. (2002a) suggested that SOC within the macroaggregates of afforested soils was stabilized for a relatively long period. In our experiment, the differences in SOC among the PA, SRW, and AC land-use practices are mostly due to differences in residue inputs, which affect the C availability for decomposition. As highlighted above, the higher POC and LFOC in soils from PA land-use practice in our field experiment is likely due to high annual inputs of root litter (Guo et al., 2007).

In our experiment, the WEOC was higher under SRW, followed by AC and PA in both sites. The WEOC content in soils has been correlated with total SOC and found to be relatively lower under agriculture compared to a forest (Ćirić et al., 2016); hence a similar situation perhaps occurred in this study. In contrast to agricultural land-use, grassland ecosystems have been shown to have three times higher WEOC in the 0-10 cm layer (Pabst et al., 2013). It has also been observed that the content of WEOC in cropped soils can be 4-5 times lower than pasture because of continuous cultivation (Ghani et al., 2003).

We observed significant depth effects under all land-use practices in both sites. Soil OC content at different depths significantly differed between different land-use practices. Similar to our experiment, Wang et al. (2016) found higher OC concentration in the surface (0-20 cm) compared to deeper soil depth under different land-use practices (i.e., cropland, shrubland, and forest), and indicates that the ground litter and live biomass were the major influential factors. Also, the allocation of biomass strongly affects the distribution of SOC with depth through plant production and patterns (Jobbágy & Jackson, 2000). In our experiment, a decreased WEOC

concentration was observed in the subsoil (20-30 cm), indicating the reduction of LC pools under SRW after two years of establishment (Lockwell et al., 2012). Hence, the WEOC concentration can significantly decrease with increasing soil depth (Zhang et al., 2006). The total OC and WEOC contents in soil were significantly higher at 0-15 cm compared to 15-30 cm depths in our experiment, proposing that land-use effects remained confined to the surface layers of the soils (Young et al., 2005, Zhang et al., 2006).

5.6.2 Land-use Effects on SOC Chemical Functional Groups

Contrasting land-use practices significantly affected the RC:LC ratio of phenolic to polysaccharide (alkyl-C/O-alkyl-C) and amide to polysaccharide (amide/O-alkyl-C) only in site A. None of the other (RC:LC) ratios were significantly different among land-use practices. Research has suggested that altered leaf litter input due to the difference in vegetation can control the chemical composition and structure of SOC (Guo et al., 2016). The RC:LC ratio can better explain the degree of decomposition and the stability of SOC via its enrichment in phenolic, aromatic, and carboxylic groups relative to polysaccharides (Deng et al., 2019). A higher value of the RC:LC ratio represents a higher degree of decomposition and better stability of SOC (Baldock et al., 1997). The relative abundance of alkyl-C (i.e., phenolic) and amide groups were higher, and O-alkyl-C (i.e., polysaccharides) was lower, whereas the ratios of phenolic and amides to polysaccharides were higher under SRW compared to AC and PA land-use practice. Lafleur et al. (2015) found substantially higher alkyl-C and O-alkyl-C in soil under SRW after two to six years compared to an adjacent agricultural field. Like our experiment, the authors observed considerably higher alkyl-C to O-alkyl-C ratio under SRW compared to the agricultural field. The presence of a higher alkyl-C chemical functional group under SRW indicates the presence of compounds that are resistant to decomposition, such as fatty acids, waxes, and resins compared to other land-use practices. It has been indicated that the leaf litters or roots or both under SRW are more resistant to decay compared to litter produced by

agricultural land-use practices (Lafleur et al., 2015). Hence, the authors indicated that the decomposition leaf litter of willow initially occurred quickly; however, with the progress of decay, the byproducts accumulate in the topsoil, and thus the SOC becomes more stable.

On the other hand, the O-alkyl-C, amides, and aromatic groups such as carbohydrates, amino acids, amino sugars, and lignin are less resistant to decay than the alkyl-C (Baldock et al., 1997, Golchin et al., 1994). In our study, the relative abundance of polysaccharides was higher under AC, while the alkyl-C and amides were lower; however, aromatic and carboxylic functional groups were higher in both sites. Helfrich et al. (2006) observed a decrease in O-alkyl-C with an increase in alkyl-C abundance via the decomposition of forest litters. In contrast, the O-alkyl-C was highest, and alkyl-C, aryl-C, and carbonyl-C content were lowest under the forest and grassland compared to agricultural soil. Therefore, indicated that both grassland and agricultural soils contained a higher proportion of aryl-C and carbonyl-C dominated by the mineral associated SOC. The increased relative abundance of aromatic-C groups under agricultural soils induced by the cultivation with the decrease of SOC content (Helfrich et al., 2006). Solomon et al. (2005) further supported that the aromatic C groups are dominant in the soils under plantations and continuously cultivated soils.

The grassland soil of the Prairie has been shown to have higher absorbance at C-H (phenolic and aliphatic groups) at 0-15cm, indicating the presence of higher SOC content and different spectral properties compared to the adjacent cropland under wheat (Calderón et al., 2011). The O-alkyl-C (i.e., polysaccharides) is more substantial in long-term PA than a cultivated agricultural soil, and mostly present as a POC fraction occluded within the stable soil aggregate (Golchin et al., 1995). In our experiment, the PA land-use has not been cultivated for at least ten years, SRW was cultivated once three years before establishment, and AC has been cultivated every year. Also, we observed a significantly higher content of POC and LFOC under PA land-use practice than SRW and AC. Perhaps, in an uncultivated soil like PA, an stable soil C pool

developed within the soil aggregates during the decades of root growth, and the relatively higher O-alkyl-C abundance is associated with SOC in the occluded light fraction, which is usually lost by cultivation (Golchin et al., 1994).

The FTIR analysis of soil revealed that depth has more influence on SOC functional groups than land-use practice. The relative abundance of FTIR bands of all SOC chemical functional groups and the RC:LC ratios of phenolic, amide and carboxylic to polysaccharides were lower, whereas aromatic to polysaccharide ratio was higher at 0-15 cm depth in both sites. Comparably, Dhillon et al. (2017) observed decreased polysaccharides and carboxylic groups with depth, whereas the aromatics increased with depth under both shelterbelt agroforestry and adjacent agricultural land-use practices in Saskatchewan, Canada. Our results support the findings that SOC chemical composition shifts with depth and labile C molecules are more common in the subsoil (Vancampenhout et al., 2012). The increased amount of alkyl-C, aromatic-C, and carbonyl-C accumulation at depth may reflect reduced decomposition or microbial synthesis of these materials in soil (Gregorich et al., 1996). Compared to topsoil OC, a higher amount of microbial-derived C compounds and the decreased proportions of energy-rich plant-derived materials in the subsoil is very common (Rumpel & Kögel-Knabner, 2010). Hence, the aromatic group (i.e., lignin) decreased, whereas aliphatic compounds increased with depth in soil (Feng & Simpson, 2007). Stronger absorbance of polysaccharides (e.g., carbohydrates), carboxylic, and ester bands at the surface (0-5 cm) compared to higher (5-15 cm) soil depth in the Prairie indicates different spectral properties with soil depth (Calderón et al., 2011).

5.7 Conclusions

Land-use practices can affect SOC content, fractions, and chemical composition considerably. Overall, the LFOC, POC, and total SOC followed a similar land-use pattern in both sites in the order of PA > SRW = AC. The PA had significantly higher total SOC, whereas comparatively higher WEOC content was observed under SRW land-use practice.

FTIR analysis of soil revealed that the depth has more influence on SOC functional groups than land-use practice. A relatively lower abundance of FTIR bands of all SOC chemical functional groups was observed in the 0-15 cm layer. Likewise, the RC:LC ratio of phenolic, amide and carboxylic to polysaccharides were relatively lower in 0-15 cm, while aromatic to polysaccharides was higher in both sites. The relative abundance of FTIR absorbance bands and the ratio of RC:LC was consistent and followed a similar pattern with depths across the land-use practices. The relatively higher abundance of SOC functional groups in the subsoil indicated altered spectral properties with depths. A higher value of the alkyl-C to O-alkyl-C ratio suggested a higher degree of decomposition and better stability of SOC in the subsoil.

Our study indicates that the establishment of SRW plantation (during the first rotation) in the degraded marginal riparian wetland soils in the PPR has a limited impact on SOC content compared to AC. However, in the successive rotations, fast-growing and high biomass producing SRW could be a substantial C sink. However, the absence of cultivation could slow down the decomposition, and leaf litter addition will further help to maintain SOC.

6 ELEVATED SALINITY AND WATER TABLE DRAWDOWN SIGNIFICANTLY AFFECT GREENHOUSE GAS EMISSIONS IN SOILS FROM CONTRASTING LAND-USE PRACTICES IN THE PRAIRIE POTHOLE REGION

6.1 Preface

Land-use practice change has an impact on greenhouse gas (GHG) emissions. Short rotation willow (SRW) land-use practice offers a means to mitigate carbon dioxide (CO₂) emission in addition to biomass production for carbon-neutral bioenergy. Additionally, with improved soil organic carbon balance through high biomass production, SRW land-use practice can affect GHG emissions by further controlling many soil factors such as organic carbon, water, and nutrient availability, as well as through altered salinity. Hence, the effects of these altered soil factors due to the establishment of new land-use practices on GHG emissions and balance need to be addressed. Chapters 3, 4, and 5 examined the *in-situ* effects of SRW plantation in a field-scale experiment on soil hydrology and salinity, nutrients status, and carbon balances, respectively. Chapter 6 covers the effects of SRW and contrasting adjacent land-use practices (i.e., pasture = PA, and annual crop = AC) on the soil factors in a microcosm study on the GHG (CO₂, N₂O, CH₄) emissions as affected by the variation of the groundwater table and salinity. The conceptual model (Figure 6.1) shows the variables that were assessed in Chapter 6 and can hypothetically be impacted by the soils from past land-use practices with the variation of groundwater table and salinity.

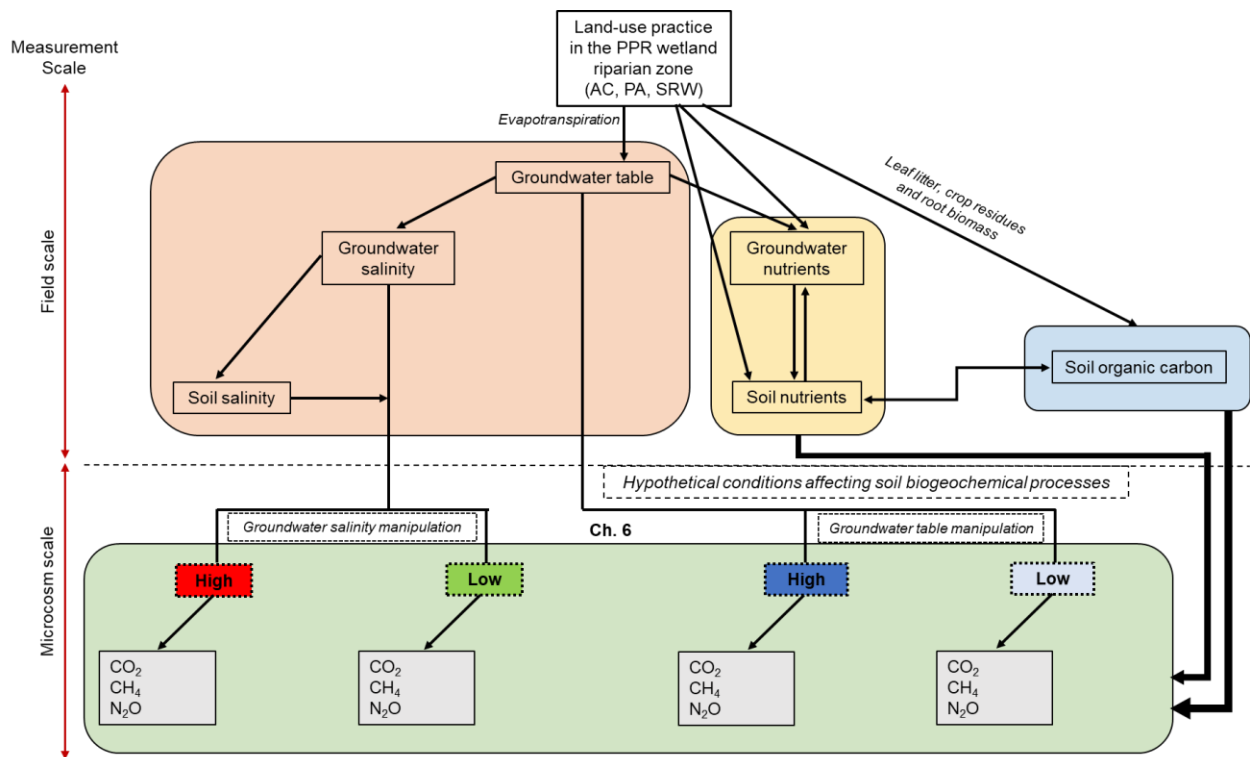


Figure 6.1 Conceptual model showing the potential land-use effects on GHG (CO₂, CH₄, N₂O) emissions from the soil as affected by the variation of groundwater table and salinity.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, GHG = greenhouse gas.

6.2 Abstract

Land-use practices within an agroecosystem have a profound influence on greenhouse gases (GHG) emissions: therefore, identifying the factors driving emissions from contrasting land-use practices in the prairie pothole region (PPR) is vital. Land-use practices can alter groundwater and salinity, which can further impact GHG emissions, particularly in the hydrologically dynamic riparian zones. Therefore, GHG emissions (CO₂, CH₄, and N₂O) were estimated in soils collected from two PPR sites with three adjacent land-use practices (i.e., annual crop = AC, pasture = PA, and short rotation willow = SRW), and exposed to declining water table depths (2 to 26 cm depth), and different groundwater salinity levels (S0 = control, S1 = 6 mS cm⁻¹, and S2 = 12 mS cm⁻¹) in a microcosm experiment. Land-use practices significantly affected ($p < 0.001$) GHG emissions in soils from both sites in the order of PA > AC = SRW. Compared to the

control, emissions of CO₂ and CH₄ were significantly lower ($p < 0.05$), while N₂O emission was significantly higher ($p < 0.05$) under higher salinity treatments (i.e., S1 and S2). Emissions were significantly ($p < 0.001$) affected by depth to groundwater table and specific to each gas. Overall, the CO₂ and CH₄ emission increased up to week four and then decreased with declining water table depths, whereas N₂O emission increased up to week six and reached a maximum. Our results indicate that the soils from SRW have significantly lower ($p < 0.01$) CO_{2e} emissions (i.e., global warming potential) compared to AC and PA land-use practices. Groundwater salinity in the PPR under contrasting land-use has significant impacts on the increased GHG emissions and crucial climate feedbacks; however, the magnitude and the direction are highly dependent on wetland hydrology.

6.3 Introduction

Agroecosystems can be significant contributors of carbon dioxide (CO₂), methane (CH₄), and nitrous oxide (N₂O) to the atmosphere—and thus contribute to global warming (IPCC, 2018)—as a result of C and N cycling in the soil (Smith et al., 2008). The North American prairie pothole region (PPR) is characterized by relatively small and highly productive wetlands embedded within the agriculture-dominated landscape. The PPR delivers essential ecosystem services such as improving soil and water quality, storing water, reducing soil erosion, and providing habitat for wildlife, especially waterfowl (Gleason et al., 2008). Salt dynamics within the PPR wetlands are driven by hydrology, which cycles seasonally and also responds to changes in land-use practice (Nachshon et al., 2013). Vegetation in the riparian zone pulls water from the soil and transfers it to the atmosphere via transpiration (Millar, 1971), which can result in a gradual decline in the water table. In turn, declining groundwater can increase soil salinity and deposit soluble salts at the surface of the soil (Arndt & Richardson, 1989). The land-use practice affects both the production and consumption of greenhouse gas (GHG) through its influence on wetland soil hydrology. For instance, wetland-riparian zones that are drained and

cropped likely would have minimal CH₄ production because this practice promotes aerobic conditions that do not favor methanogenesis (Smith et al., 2003). Conversely, the same catchment would have a higher likelihood of emitting N₂O due to a combination of N-fertilizer additions and less saturated soil moisture conditions (Davidson et al., 2000). Hence, changes in land-use practice can increase the potential for GHG emissions and diminish the capacity of PPR wetlands to deliver ecosystem services (Gleason et al., 2009).

Land-use practice can strongly influence soil-derived GHG emissions (Liebig et al., 2005, Schaufler et al., 2010, Tangen et al., 2015). In general, wetlands have a greater GHG emission potential than forestlands, croplands, and grasslands (Oertel et al., 2016); however, the amount of CO₂, CH₄, and N₂O emitted vary depending on the type of vegetation and environmental conditions (Kayranli et al., 2009). The production of GHG in wetlands is controlled by a variety of highly variable abiotic factors that are affected by land-use; these include groundwater table (GWT)/soil moisture regime, the period of inundation, redox conditions, and groundwater salinity (Marton et al., 2012). The land-use practice also affects soil biological processes that regulate GHG emissions by influencing the makeup of the soil microbial and plant communities, and the availability of organic substrates (Tangen et al., 2015). Moreover, riparian land-use practice affects the microclimate and soil properties that can influence the production/consumption and emission of GHG (Moore et al., 2017). Consequently, land-use practices affecting wetland riparian zones have the potential to significantly alter the amount of GHG released into the atmosphere (Vidon, 2010).

Land-use practice can alter soil organic carbon (SOC) dynamics and, in turn, affect GHG emissions (Kooch et al., 2016, Lang et al., 2010, Merino et al., 2004). For example, agroforestry is a promising land-use practice that can increase above- and below-ground C stocks (Paul et al., 2003), mitigate N₂O and CO₂ emissions, and increase the sink potential of the soil for CH₄ when compared to cropland (Mutuo et al., 2005). One study (Baah-Acheamfour et al., 2016)

recommended that agroforestry and grassland cover be incorporated into agricultural lands to reduce emissions of CH₄ and N₂O. Parmar et al. (2015) also found that “short rotation” forestry can contribute to GHG savings via reduced soil respiration losses. Thus, establishing perennial agroforestry systems such as Short Rotation Willow (SRW) in the riparian zones of PPR wetlands may deliver GHG mitigation benefits. However, the effects of agroforestry practices on soil N₂O and CH₄ emissions are only poorly understood (Albrecht & Kandji, 2003). It has also been suggested that SOC could be sequestered in PPR wetlands by re-establishing permanent vegetation (i.e., grass) (Bedard-Haughn et al., 2006a). However, it is unclear how the establishment of perennial SRW vegetation in the marginal riparian zones of the semi-arid PPR wetlands affect GHG emissions under dynamic soil hydrology (e.g., GWT) and salinity.

The effects of a fluctuating GWT on GHG emissions from peatlands (Berglund & Berglund, 2011, Blodau et al., 2004, Updegraff et al., 2001) and riparian mineral wetlands (Mander et al., 2015) have been studied previously. Also, the effects of salinity on GHG emissions associated with a change in land-use (Martin & Moseman-Valtierra, 2015, Sheng et al., 2015) or depth to the GWT (Ardón et al., 2018, Mander et al., 2011) has been studied in coastal wetlands. However, studies available have variable results. For instance, in a microcosm experiment, artificial salinity treatments suppressed CO₂ emission under both droughts and flooded conditions, whereas CH₄ emission increased in flooded compared to drought conditions; but the impacts of salinity were conditional on hydrologic treatments for N₂O (Ardón et al., 2018). In contrast, salinity inhibited CH₄ production but increased CO₂ and N₂O emissions in a tidal forest soil (Marton et al., 2012). In another microcosm study, salinity increased N₂O emissions and reduced CO₂ and CH₄ emissions, whereas, increasing soil moisture from 25 to 100% water holding capacity (% WHC) of the soil amplified CO₂, CH₄ increased up to 50% and then decreased at 75% WHC, and did not affect N₂O emissions in semi-arid Australian soil from a cropped field (Maucieri et al., 2017). Whereas, in the riparian zones of mineral wetlands,

flooding increased CH₄ emission, and CO₂ and N₂O emissions increased as the depth to GWT decreases (Mander et al., 2015). In constructed wetlands, CH₄ emissions were reduced, and N₂O emissions amplified at high salinity (>10 ‰), whereas the CO₂ emissions were highest at intermediate salinity, i.e., ~5 ‰ (Sheng et al., 2015). Nevertheless, studies on the combined effects of GWT and salinity on GHG emissions under contrasting land-use practices within mineral wetlands in the PPR are scarce.

Depending on a variety of factors, wetland soils can act as either a source or sink for GHG (Badiou et al., 2011, Beetz et al., 2013). Mitigating GHG emissions and developing best management practices for agroecosystems in the PPR will require a better understanding of how land-use practice affects the source/sink potential of these wetlands. Also, examining the GHG emissions under the combined effects of fluctuating water table and salinity in the context of contrasting land-use practices will improve our ability to address the issues and mitigation actions while advancing agricultural sustainability in the PPR. Therefore, the present study was conducted to examine the impact of a declining groundwater water table—with different groundwater salinity levels—on GHG emissions from riparian zone soils collected from different land-use practices. We hypothesized that 1) regardless of salinity and land-use practice, CO₂ emissions will increase and CH₄ emissions will decrease as the soil water table level is lowered; 2) N₂O emission will initially increase as the water table is lowered, and then decrease; and 3) regardless of soil water table level and land-use practice, emissions of CO₂, CH₄, and N₂O will decrease as the salinity level increases.

6.4 Materials and Methods

6.4.1 Site Description and Collection of Intact Soil Cores

A controlled microcosm experiment was conducted to determine the influence of groundwater salinity and declining water table level on soil-derived emissions of CO₂, CH₄, and N₂O. Soils were collected from sites managed under three different land-use practices at two sites in the

PPR. Both sites (Site A and Site B) were located near the Agriculture and Agri-Food Canada Indian Head Agroforestry Development Centre at Indian Head, Saskatchewan, Canada (N 50° 30.605'; W 103° 43.011') (Figure D-S1). Soils at both sites were classified as Oxbow Association, non-calcareous Black Chernozems developed on loamy glacial till in a landscape with level to gentle rolling (0–10% slope) topography (Saskatchewan Soil Survey Staff, 1986). At both sites, the SRW treatments (*Salix dasyclados* Wimm, popularly known as 'India') were established in June 2013 in the marginal fallow riparian zones. The section of pasture (PA) comprised of a mix of alfalfa (*Medicago sativa*) and bromegrass (*Bromus madritensis*) that had been established in 2001–2003. Both SRW and PA areas were located (Figure D-S1) adjacent to the cropped area that was seeded with oat (*Avena sativa*).

The soils at Site A were non-saline, with ECs ranging from 0.6 to 1.9 mS cm⁻¹; soils at Site B were non- to slightly saline, with ECs ranging from 1.0 to 2.6 mS cm⁻¹ (see Table D-S1). Intact soil cores (n = 3) were collected from each of the three land-use treatments at Sites A and B (i.e., annual crop [AC], pasture [PA], and short rotation willow [SRW]) in mid-August 2015 (Figure D-S9). Intact soil cores were used to avoid the disturbance produced by sieving (Reichstein et al., 2005). The soil cores were collected during using a truck-mounted hydraulic punch (Giddings Machine Company Ltd., Windsor, CO, USA) fitted with cylindrical (30-cm tall × 9-cm i.d.) PVC sleeves. Cores were collected three years after the SRW plantation establishment. The overlying litter-fibric-humic layer and grasses were removed prior to collecting the soil cores from the field. All soil cores were collected from the riparian zones. For the SRW, all soil cores were collected from the middle of two planted rows, i.e., 1-m apart from each planted row (the distance between rows was 2 m). In total, 54 soil cores (2 sites × 3 land-use practices × 9 reps) were collected and transported back to the laboratory at the University of Saskatchewan in coolers where they were preserved frozen (at -20°C) until the start of the incubation study. An additional soil core (0–30 cm depth; 9-cm i.d.) was collected from the same

sampling location under three different land-use practices from both sites and used to analyze soil physical and chemical properties (see Table D-S1). Samples for bulk density were collected at 0-15 and 15-30 cm using a hand-held core sampler (3-cm tall × 5.4-cm i.d.).

6.4.2 Initial Soil Characterization

Soil physiochemical properties were determined prior to the start of the microcosm experiment. Each soil was divided into three subsamples, which were processed as follows: 1) one subsample was air-dried, ground, passed through a 2-mm sieve, and analyzed for particle size distribution, cation exchange capacity (CEC), pH, electrical conductivity (EC), and ammonium acetate extractable N and P; 2) the second subsample was air-dried, finely ground with a ball mill grinder, and analyzed for organic- and total-C and total-N; and 3) the third subsample was frozen until it was analyzed for water extractable organic carbon (WEOC) and water extractable organic nitrogen (WEON). Samples collected for bulk density measurement were weighed, oven-dried at 105°C for 24 h, cooled to room temperature in a desiccator, and reweighed. Bulk density was determined by dividing the oven-dry weight of the soil by the volume (68.71 cm³) of the core sampler.

Soil physiochemical analyses were carried out using the procedures described in *Soil Sampling and Methods of Analysis* (Carter & Gregorich (2008)). The modified pipette method (Kroetsch & Wang, 2008) was used to determine soil particle size distribution. Cation exchange capacity was determined using ammonium acetate at pH 7, followed by colorimetric analysis using a Technicon Auto-Analyzer (Technicon Industrial Systems; Tarrytown, NY, USA) (Hendershot et al., 2008a). Soil pH and EC were determined in a 1:2 (w/v) soil:deionized-water suspension using a digital pH meter (Oakton™ PC700 pH/mV/conductivity meter; Oakton Instruments, Vernon Hills, IL, USA) (Hendershot et al., 2008b), and EC was determined in the same extract after 1 hour shaking with an end-over-end shaker, then filtered through the highly retentive filter (No. 42, Whatman Inc., Piscataway, NJ), and measured using digital EC meter (PC700

pH/mV/conductivity, Oakton, Vernon Hills, IL, USA) (Miller & Curtin, 2008). Ammonium (NH_4^+ -N), nitrate (NO_3^- -N), phosphate (PO_4^{3-} -P), and sulfate (SO_4^{2-} -S) were measured using a 1M ammonium acetate (buffered at pH 7) extraction followed by colorimetric analysis for NH_4^+ -N, NO_3^- -N, PO_4^{3-} -P using the Technicon Auto-Analyzer (Technicon Industrial Systems, Tarrytown, NY, USA), and SO_4^{2-} -S analyzed using Microwave Plasma-Atomic Emission Spectrometer (Model 4100, Agilent Technologies, Santa Clara, CA, USA) (Simard, 1993). Total soil carbon (TSC) and soil organic carbon (SOC) were determined by dry combustion—following HCl fumigation to remove carbonates—using a Leco-2000 CNS analyzer (Leco Corporation, St. Joseph, MI, USA) (Skjemstad & Baldock, 2008). Total nitrogen (TN) was determined using dry combustion with a Leco C632 CNS analyzer (Leco Corporation, St. Joseph, MI, USA) (Rutherford et al., 2008). Water extractable organic C and WEON were determined by gently mixing defrosted soil (20 ± 1 g) with 30-mL of 5 mM CaCl_2 , filtering the suspension through a 0.45- μm polycarbonate membrane filter (Whatman Inc., Piscataway, NJ, USA), and measuring total C and N in the filtrate using a TOC-VCPN analyzer (Shimadzu Scientific Instruments, Kyoto, Japan) (Chantigny et al., 2008).

6.4.3 Experimental Design

The microcosm incubation experiment was set up in the greenhouse at the University of Saskatchewan using a nested experimental design (Krzywinski et al., 2014, Schielzeth et al., 2013); the experiment was conducted over nine weeks. The 54 soil cores were used for treatment and arranged according to the following: 2 sites \times 3 land-use practices \times 3 groundwater salinity treatments (control = 0.3 mS cm^{-1} , S1 = 6 mS cm^{-1} , and S2 = 16 mS cm^{-1}) \times 3 replicates (Figure D-S2 and S10). Each experimental unit consisted of a 19-L plastic (PVC) bucket (38.1 cm tall \times 30.48 cm i.d.) containing a 2.5-cm thick layer of gravel, 17-L of synthetic groundwater, and a single intact soil core—the bottom of which was wrapped in 1-mm mesh fiberglass screen to hold the soil securely (Figure 6.2). The PVC cylinders housing the soil cores

were drilled with a uniform series of 3-mm holes and to allow for movement of the synthetic groundwater into, and out of, the soil core.

Initially, the synthetic groundwater was maintained level with the surface of the soil cores (Figure 6.2), with subsequent GWT drawdown achieved by manually lowering the water level by 2 cm at the end of the first week, and then by 3 cm at the end of each of the next nine weeks (Figure 6.2). The dominant salts present in the soil and groundwater in the Prairie region of Canada and the northern United States are Na_2SO_4 , KCl, CaCl_2 , and MgSO_4 (Last & Ginn, 2005). Thus, the synthetic groundwater treatments were prepared using a 5:2:12:14 mix of Na_2SO_4 :KCl: CaCl_2 : MgSO_4 salts (by weight) in distilled water, but the quantity (g) of salts was double in S2 from that of the S1 treatment. The control (no added salts) salinity treatment consisted of distilled water alone. The EC of the synthetic groundwater was checked weekly to ensure that salinity remained constant. The volumetric soil water content (VSWC) and EC of the experimental soil cores were also measured by a digital soil moisture meter (HydroSense II, Campbell Scientific Inc., Logan, UT, USA) at the time of GHG samples collection. The temperature of the greenhouse chamber in which the incubation experiment was conducted was maintained at $20 \pm 1^\circ\text{C}$; relative humidity in the greenhouse ranged from 37.73% to 67.05% (average 50.33%) during the first seven weeks of the experiment, and then from 16.05% to 43.26% (average 29.53%) during the last three weeks of the experiment (Figure D-S3).

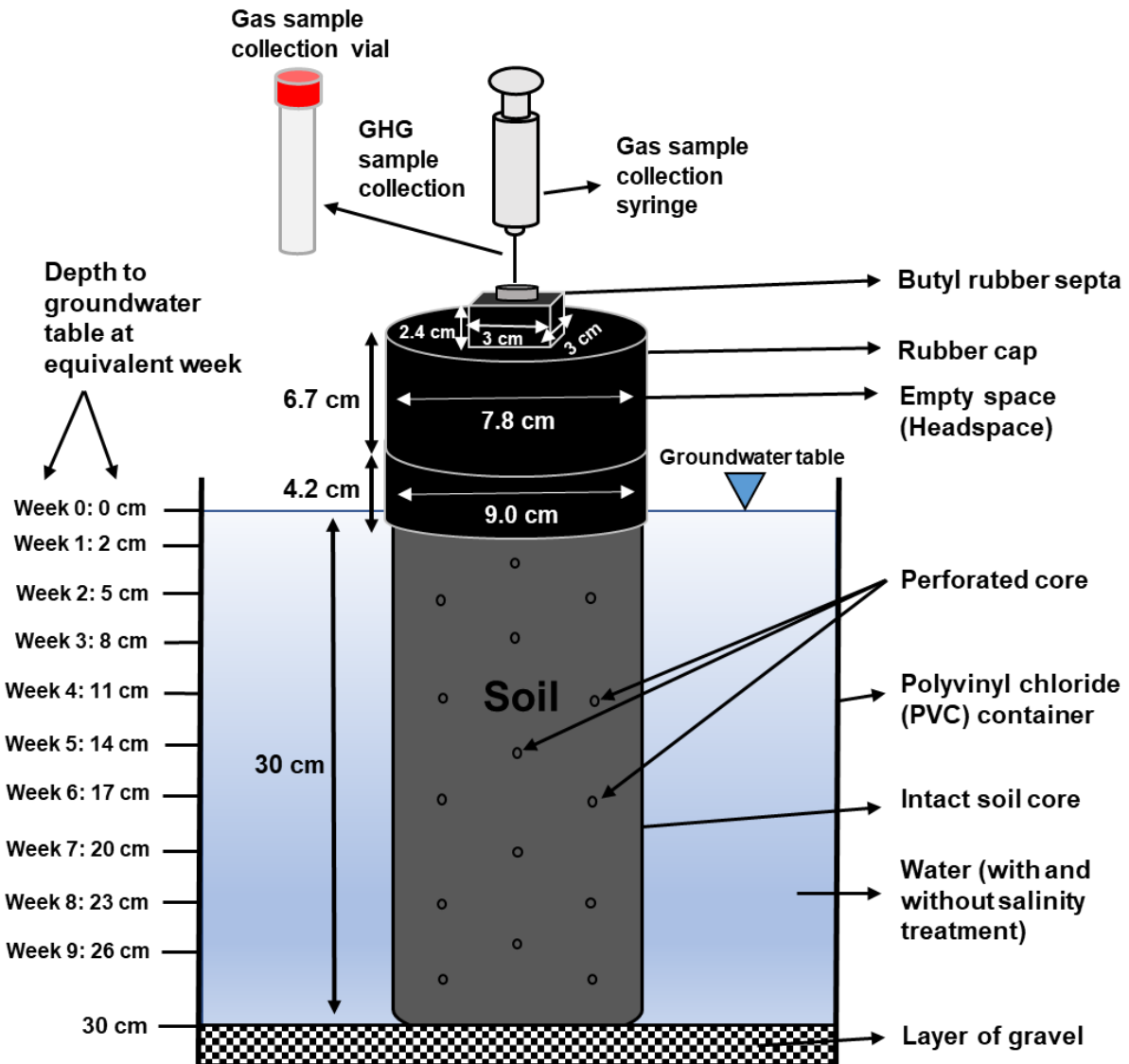


Figure 6.2 An individual experimental unit with intact soil core and greenhouse gas chamber used for microcosm experiment (**Note:** diagram is not to scale).

6.4.4 GHG Flux Measurements

Greenhouse gas flux measurements were done using non-vented, static (i.e., non-steady state) chambers (Collier et al., 2014, Rochette & Bertrand, 2008) constructed using an ABS cleanout adapter (model # RLN105-030) and male plug (model # RLN106R-030) fitted with a sampling port sealed using a gas-impermeable, grey butyl rubber septum (Supelco, USA) (see Figure 6.2). Gas flux measurements were made seven days after each GWT adjustment by attaching

the sampling chamber to the top of the cores using a flexible coupling (model # FC-33) and sampling the headspace atmosphere immediately after the chamber was attached (t_0) and again after 30 min (t_{30}). The cores remained open to the atmosphere during the period between GWT adjustments.

Headspace gas samples were collected at t_0 and t_{30} using a 20-mL polypropylene syringe (Monoject™, Luer lock fitting) fitted with a 25-gauge needle and were immediately injected into pre-evacuated 12-mL Exetainer® vials (LabCo Inc., High Wycombe, UK). Ambient air samples—used as a check on the t_0 samples— were also collected on each sampling day. The gas samples were then brought to the Prairie Environmental Agronomy Laboratory in the Department of Soil Science at the University of Saskatchewan for analysis. The concentrations of CO₂, CH₄, and N₂O in each gas sample were determined using gas chromatography (Farrell & Elliott, 2008). Sample analyses were performed using a Bruker 450 GC (Bruker Biosciences Corporation, USA) equipped with a thermal conductivity detector (TCD), flame ionization detector (FID), and electron capture detector (ECD) for the detection and quantification of CO₂, CH₄, and N₂O, respectively. Samples were introduced into the GC using a CombiPAL auto-sampler (CTC Analytics AG, Switzerland); data processing was completed using Varian Star Chromatography Workstation (ver. 6.2) software. The GHG fluxes were calculated from the change in concentration measured during the 30-min chamber deployment using Equation 6.1:

$$F = \Delta C \frac{V \cdot k_t}{A} \dots\dots\dots (6.1)$$

where F is the GHG flux at time zero (mg m⁻² d⁻¹); ΔC is the change in concentration (mg CO₂, CH₄, or N₂O L⁻¹ min⁻¹) measured during the 30-min deployment period; V is the volume of the chamber headspace (0.6089 L); A is the surface area of the soil cores (0.0064 m²); and k_t is the time constant (1440 min d⁻¹). Net GHG fluxes were calculated by subtracting the respective blank (sample from ambient air collected at the time of GHG sampling) values from the values for the soil cores to correct for potential gas losses through leaks and sampling removal.

Cumulative GHG emissions for each land-use were calculated using linear interpolation (Equation 6.2) as described in (Pennock et al., 2010). This assumes that emissions were constant throughout the day of the measurement and during the seven days since the previous water table adjustment.

$$CF = (F_{w1} \times 1) + (F_{w2} \times 7) + (F_{w3} \times 7) + (F_{w4} \times 7) + (F_{w5} \times 7) + (F_{w6} \times 7) + (F_{w7} \times 7) + (F_{w8} \times 7) + (F_{w9} \times 7) \dots\dots\dots (6.2)$$

Where CF is the cumulative GHG flux (mg m^{-2}); F_w is the daily flux rate measured at the end of each week (weeks 1 through 9; total 57 days of incubation); 7 is the number of days in a week. However, this calculation only accounts for GHG emissions for the last day (sampling day) of the first week. Global warming potentials (GWP) for each land-use practice were calculated for a 100-year time scale using conversion factors for $\text{CO}_2 = 1$, $\text{CH}_4 = 25$, $\text{N}_2\text{O} = 298$ after adjusting by mass to obtain carbon flux ($\text{CO}_2\text{-C}$) (Myhre et al., 2013, Wang et al., 2017a).

6.4.5 Statistical Analyses

Soil GHG emissions data were statistically analyzed and visualized using R version 3.4.4 for Windows (R Core Team, 2018). Shapiro-Wilk test and histogram were used to assess the normality and Levene's test to check the homogeneity of variances using "car" package. Soil GHG emissions were compared non-parametrically by plotting soil CO_2 , CH_4 , and N_2O fluxes against measured VSWC and EC in each soil core against declining GWT depths in each week of measurements. Cumulative soil GHG emissions were also compared non-parametrically by plotting CO_2 , CH_4 , and N_2O fluxes land-use practices and groundwater salinity treatments from both sites using "ggplot2". The relationships between CO_2 , CH_4 , and N_2O emissions, VSWC, EC, and initial soil parameters were measured non-parametrically by Spearman rank-order correlation and visualized using "corrplot" package. Assumptions of both univariate and multivariate analysis of variance (ANOVA) normality were fulfilled by adding a positive constant number (+2) during the transformation (Logarithmic with base 10) to manage negativity (for CH_4 and N_2O) in data. Significant differences (i.e., hypothesis testing) among land-use practices,

groundwater salinity treatments, and water table depths were compared parametrically by univariate ANOVA with nested design and linear mixed-effects models (Zuur et al., 2009) using 'lmerTest'. Pairwise multiple comparison procedures (Tukey's HSD method) were used as a post-hoc test to isolate the groups that differed from the others in ANOVA. The permutation multivariate ANOVA (PERMANOVA) and analysis of similarities (ANOSIM) were used to assess significant differences (multivariate hypothesis testing) in GHG emissions among land-use practices, groundwater salinity, and GWT depths. ANOSIM was also used to calculate a matrix of dissimilarity ranks after converting the scores to find the ratio between within-group and between-group similarities. Unconstrained ordination with a non-metric multidimensional scale (NMDS) was used to plot the position in multidimensional space with a reduced number of dimensions to visualize the difference among groundwater salinity treatments, GWT depths, and land-use practices. The variation partitioning analysis (VPA) was used to determine the proportional contribution of land-use practices, groundwater salinity, and water table depth in the variation of GHG emissions. Constrained ordination with redundancy analysis (RDA) was performed to summarize the variation explained by measured soil physiochemical characteristics. The PERMANOVA, ANOSIM, NMDS, VPA, and RDA analyses were performed using "vegan" package (Oksanen et al., 2017). All the differences were considered significant at p -values ≤ 0.05 (95% confidence interval or alpha level = 0.05).

6.5 Results

6.5.1 Emissions of GHG in Soils from Contrasting Land-use Practices, Elevated Groundwater Salinity, and Declining Groundwater Table

The soils from PA land-use showed a significantly ($p < 0.001$) higher CO₂ emission followed by AC and SRW in both sites (Table 6.1 and 6.2). The emission of CO₂ from soils followed consistent land-use patterns in both sites in the order of PA > AC = SRW (Table 6.1).

Cumulative CO₂ emission was higher in soils from site A than site B (Figure 6.3). The CO₂

emission was significantly ($p < 0.05$) higher in control (S0) compared to the manipulated groundwater salinity treatments (i.e., S1 and S2) in soils across all land-use practices from both sites (Table 6.1 and 6.2). A significant ($p < 0.001$) difference in CO₂ emission was observed among the depth to GWT (Table 6.2). The CO₂ flux initially showed an increasing trend with the decline in GWT depths (i.e., weeks of measurements) in soils across all land-use practices from both sites and showed a decreasing trend afterward (Table 6.1 and Figure D-S4 and S5). The mean emission of CO₂ was significantly ($p > 0.05$) higher at the depth to GWT treatment of 11-cm (week 4) in both sites, and lowest at 26-cm (week 9) in site A, and 20-cm (week 7) in site B (Table 6.1 and 6.2).

Table 6.1 Mean (\pm SE) GHG emissions, EC, and VSWC measured weekly with their equivalent groundwater table depths and salinity treatments from soil cores collected from different land-use practices from two sites.

	Site A					Site B				
	CO ₂	CH ₄	N ₂ O	VSWC	EC	CO ₂	CH ₄	N ₂ O	VSWC	EC
	(mg m ⁻² d ⁻¹)			(%)	(mS cm ⁻¹)	(mg m ⁻² d ⁻¹)			(%)	(mS cm ⁻¹)
Land-use										
AC	1016 (\pm 43) b	2.57 (\pm 0.83) b	0.70 (\pm 0.09) b	48 (\pm 0.42) a	3.3 (\pm 0.04) a	784 (\pm 41) b	1.08 (\pm 0.81) b	1.20 (\pm 0.15) b	47 (\pm 0.43) c	3.2 (\pm 0.04) b
PA	2348 (\pm 123) a	22.67 (\pm 5.96) a	5.35 (\pm 0.76) a	47 (\pm 0.39) a	3.3 (\pm 0.04) a	1756 (\pm 83) a	2.36 (\pm 0.76) a	1.82 (\pm 0.20) a	49 (\pm 0.33) b	3.4 (\pm 0.04) a
SRW	595 (\pm 24) c	0.43 (\pm 0.18) b	0.68 (\pm 0.10) b	47 (\pm 0.48) a	3.3 (\pm 0.05) a	560 (\pm 13) c	0.01 (\pm 0.01) c	0.27 (\pm 0.04) c	50 (\pm 0.18) a	3.5 (\pm 0.04) a
Salinity										
S0	1499 (\pm 114) a	19.2 (\pm 5.91) a	0.35 (\pm 0.06) b	43 (\pm 0.29) b	2.8 (\pm 0.01) c	1180 (\pm 89) a	1.79 (\pm 0.85) a	0.32 (\pm 0.06) c	46 (\pm 0.42) c	3.0 (\pm 0.02) c
S1	1333 (\pm 123) b	5.0 (\pm 1.64) b	3.27 (\pm 0.68) a	50 (\pm 0.13) a	3.4 (\pm 0.01) b	1016 (\pm 72) a	0.45 (\pm 0.15) a	1.27 (\pm 0.19) b	49 (\pm 0.17) b	3.4 (\pm 0.02) b
S2	878 (\pm 98) c	1.4 (\pm 0.44) c	3.11 (\pm 0.49) a	50 (\pm 0.04) a	3.7 (\pm 0.01) a	858 (\pm 72) b	1.21 (\pm 0.71) a	1.69 (\pm 0.18) a	50 (\pm 0.06) a	3.8 (\pm 0.01) a
Depth to GWT (week of measurement)										
2-cm (W1)	1057 (\pm 206) cd	0.20 (\pm 0.04) d	0.28 (\pm 0.08) d	49 (\pm 0.53) a	3.3 (\pm 0.07) a	1042 (\pm 149) cd	0.19 (\pm 0.06) d	0.14 (\pm 0.07) d	51 (\pm 0.29) a	3.4 (\pm 0.05) a
5-cm (W2)	1522 (\pm 203) b	2.07 (\pm 0.56) cd	0.47 (\pm 0.09) cd	49 (\pm 0.49) a	3.4 (\pm 0.07) a	1154 (\pm 160) b	1.67 (\pm 0.65) b	0.21 (\pm 0.07) cd	50 (\pm 0.29) a	3.5 (\pm 0.06) a
8-cm (W3)	1489 (\pm 196) ab	15.07 (\pm 4.14) b	1.28 (\pm 0.43) bcd	49 (\pm 0.55) b	3.3 (\pm 0.07) a	1286 (\pm 194) ab	6.96 (\pm 3.08) a	0.29 (\pm 0.11) bcd	50 (\pm 0.33) b	3.5 (\pm 0.06) a
11-cm (W4)	1981 (\pm 291) a	36.82 (\pm 15.77) a	2.56 (\pm 0.94) abc	48 (\pm 0.61) c	3.3 (\pm 0.07) a	1339 (\pm 190) a	0.99 (\pm 0.38) bc	0.82 (\pm 0.23) abc	49 (\pm 0.41) c	3.4 (\pm 0.07) a
14-cm (W5)	1375 (\pm 191) bc	14.94 (\pm 7.15) bc	2.91 (\pm 1.04) ab	47 (\pm 0.72) d	3.3 (\pm 0.08) b	1095 (\pm 120) bc	0.27 (\pm 0.08) cd	1.11 (\pm 0.26) ab	48 (\pm 0.52) d	3.4 (\pm 0.07) b
17-cm (W6)	1401 (\pm 192) bc	5.43 (\pm 2.38) cd	3.90 (\pm 1.35) a	47 (\pm 0.84) e	3.3 (\pm 0.08) b	1105 (\pm 113) bc	0.17 (\pm 0.04) cd	1.69 (\pm 0.34) a	48 (\pm 0.62) e	3.4 (\pm 0.08) b
20-cm (W7)	1007 (\pm 133) d	2.04 (\pm 0.78) d	3.04 (\pm 0.95) a	46 (\pm 0.87) f	3.2 (\pm 0.08) c	678 (\pm 56) d	0.11 (\pm 0.03) d	1.83 (\pm 0.32) a	47 (\pm 0.71) f	3.3 (\pm 0.08) c
23-cm (W8)	1093 (\pm 125) d	0.31 (\pm 0.12) d	2.88 (\pm 0.77) a	46 (\pm 0.85) f	3.2 (\pm 0.08) c	776 (\pm 64) d	-0.01 (\pm 0.02) d	2.00 (\pm 0.36) a	47 (\pm 0.75) f	3.3 (\pm 0.08) c
26-cm (W9)	950 (\pm 90) d	0.14 (\pm 0.05) d	2.88 (\pm 1.08) a	46 (\pm 0.91) f	3.3 (\pm 0.09) b	822 (\pm 50) d	0.01 (\pm 0.01) d	1.77 (\pm 0.31) a	47 (\pm 0.78) f	3.4 (\pm 0.08) b

^a Means within a column for sites, land-use, salinity, and depth to groundwater table followed by the same letter are not significantly different ($p > 0.05$) using Tukey HSD.

^b SE = standard error, GHG = greenhouse gas, EC = electrical conductivity, VSWC = volumetric soil water content, AC = annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm⁻¹, S2 = 12 mS cm⁻¹, GWT = groundwater table.

Table 6.2 Analysis of variance (ANOVA) with nested design and linear mixed-effects models for soil GHG emissions, VSWC, and EC measured weekly with their equivalent groundwater table depths and salinity treatments in core soils from three land-use practices from two sites.

Sources of variation	CO ₂		CH ₄		N ₂ O		VSWC		EC		
	df	F stat	p - value	F stat	p - value	F stat	p - value	F stat	p - value	F stat	p - value
Site A											
Land-use	2	249.03	<0.001 ***	25.47	<0.001 ***	25.65	<0.001 ***	0.98	0.411 ^{ns}	1.57	0.260 ^{ns}
Salinity	2	11.10	0.004 **	7.63	0.012 *	13.88	0.002 **	306.99	<0.001 ***	646.84	<0.001 ***
Depth to GWT	8	17.43	<0.001 ***	16.39	<0.001 ***	12.22	<0.001 ***	27.49	<0.001 ***	12.44	<0.001 ***
Site B											
Land-use	2	55.67	<0.001 ***	28.38	<0.001 ***	54.08	<0.001 ***	5.66	0.026 *	58.46	<0.001 ***
Salinity	2	5.59	0.026 *	2.71	0.069 ^{ns}	46.69	<0.001 ***	18.78	<0.001 ***	427.49	<0.001 ***
Depth to GWT	8	9.23	<0.001 ***	10.78	<0.001 ***	25.29	<0.001 ***	29.44	<0.001 ***	20.52	<0.001 ***

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b GHG = greenhouse gas, VSWC = volumetric soil water content, EC = electrical conductivity, GWT = groundwater table.

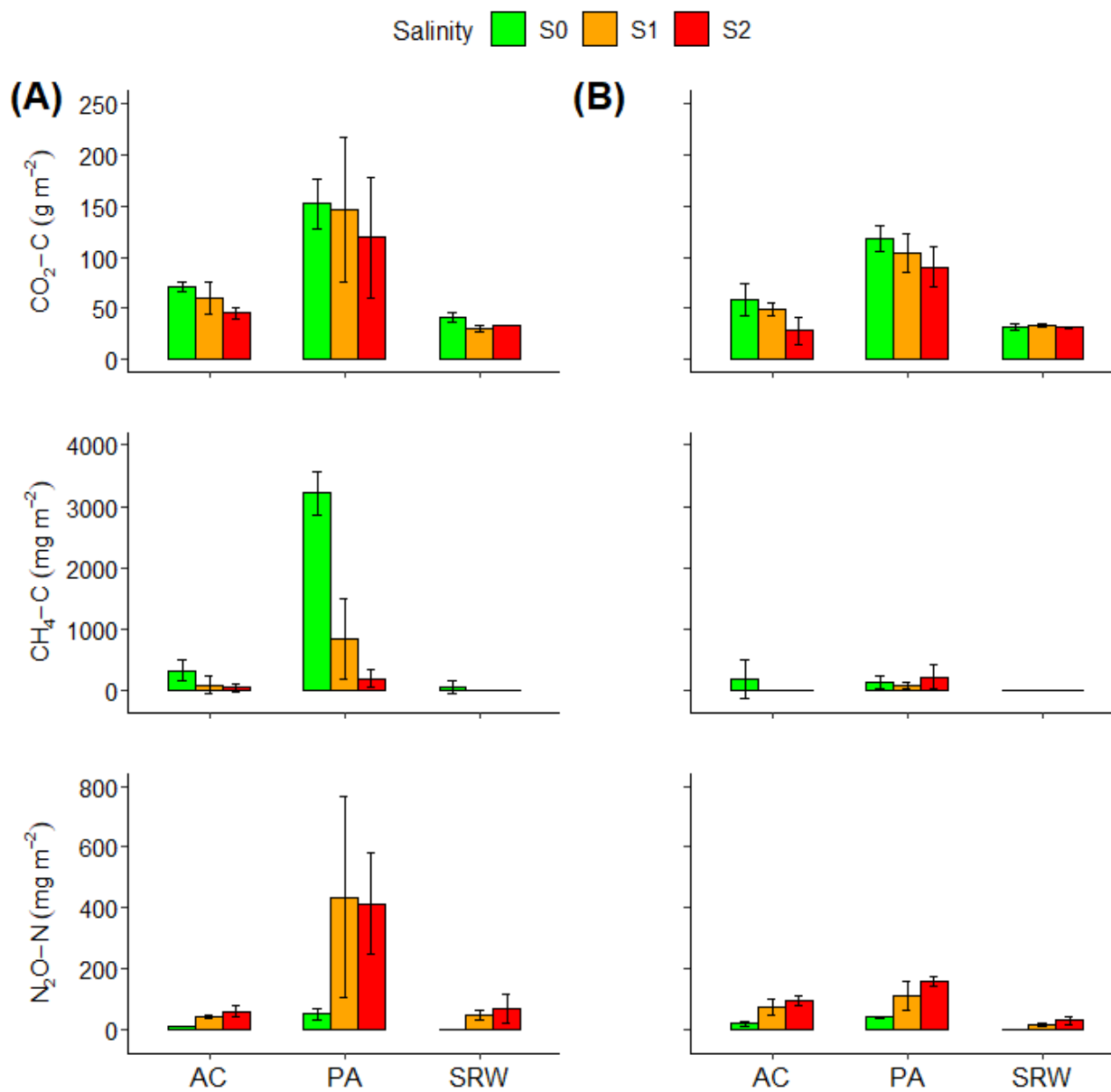


Figure 6.3 Cumulative GHG emissions from core soils with different groundwater salinity treatments from soils collected from three land-use practices from A) site A, and B) site B.

^a Error bar stands for standard deviations (\pm SD).

^b AC = annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm⁻¹, S2 = 12 mS cm⁻¹, GHG = greenhouse gas.

The CH₄ emission significantly ($p < 0.001$) differed among the soil from all land-use practices in both sites (Table 6.2). The mean CH₄ emission was significantly higher ($p < 0.001$) in soils from PA (PA > AC = SRW) and showed a consistent pattern among land-use practices from both sites (Table 6.1). Cumulative CH₄ emission was quite low, and variable between sites, being negligible in site B compared to site A (Figure 6.3). Groundwater salinity treatments (both S1 and S2) significantly ($p = 0.012$) reduced CH₄ emission compared to control (i.e., S0) in site A, however, not significantly ($p = 0.069$) in site B (Table 6.2). The CH₄ flux in soils from all land-use practices from both sites showed a slightly increasing trend up to week four and then decreased with a further decline in depths to GWT (Table 6.1 and Figure D-S4 and S5). The highest mean CH₄ emission was observed at the depth to GWT of 11-cm (week 4) in site A and 8-cm (week 3) at site B, whereas lowest at 26-cm (week 9) depth to GWT treatment in both sites (Table 6.1 and 6.2).

The N₂O emission was significantly ($p < 0.001$) higher in soils from PA land-use practice followed by AC and SRW and followed similar land-use patterns in the order of PA > AC = SRW in both sites (Table 6.1 and 6.2). The cumulative N₂O emission was significantly ($p < 0.01$) increased under both manipulated groundwater salinity treatments (i.e., S1 and S2) in soils from both sites compared to control, which suggested that salinity increased N₂O emission (Table 6.1 and 6.2). Overall, the cumulative N₂O emission was relatively low and variable between sites, being higher in soils from site A compared to site B (Figure 6.3). The N₂O flux showed a slightly increasing trend with a peak at week seven and remained stable with the decline in depths to GWT, although somewhat variable in soils across all land-use practices from both sites, as the VSWC in the core reached an optimum condition from the saturated level (Table 6.1 and Figure D-S4 and S5). Lowest mean N₂O emission was observed at the depth to GWT of 2-cm (week 1) in both sites, whereas the highest emission was at 17-cm (week 6) in site A and 23-cm (week 8) in site B (Table 6.1 and 6.2).

Multivariate unconstrained ordination (NMDS analysis) of soil GHG emissions data (stress value for site A is 0.0670, and site B is 0.0724) differed considerably among land-use practices in both sites, indicating the land-use practice type was a key factor driving the variability (Figure 6.4A, B, D, and E). The NMDS ordination also showed a distinct clustering of GHG emissions based on land-use practices in both sites (stress values below 0.10 provide a fair representation of data in reduced dimension), indicating robust land-use effects from the PA soils.

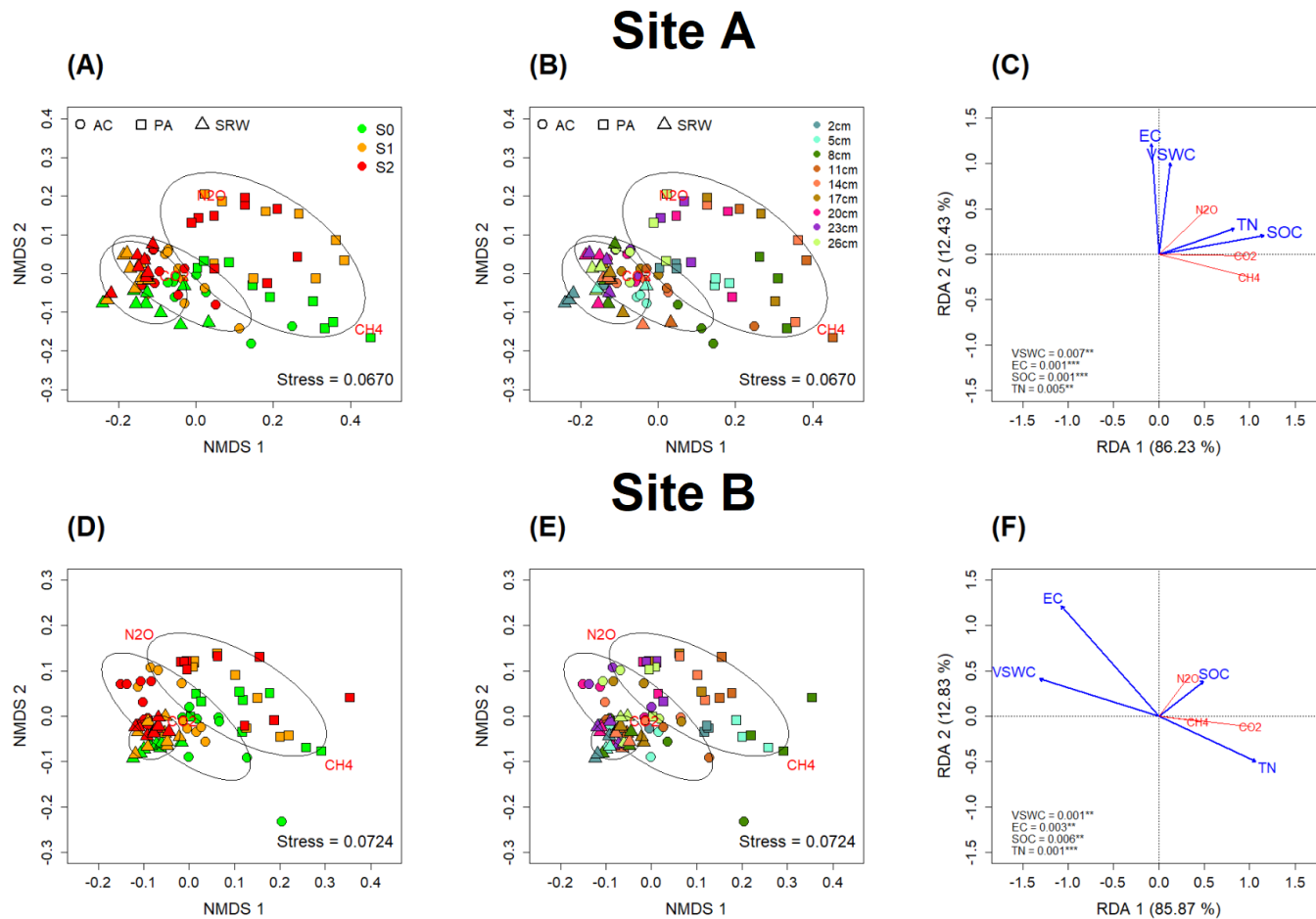


Figure 6.4 Non-metric multidimensional scaling (NMDS) test of soil GHG emissions visualized with land-use and groundwater salinity, land-use and groundwater table depth, and redundancy analysis (RDA) from site A (A, B, C) and site B (D, E, F).

^a Blue vectors indicate linear correlations between the ordination and soil physiochemical properties. Directions and lengths of the vectors indicate the strength of correlations between variables and the ordination. The angles between vectors reflect their correlations (i.e., a vector pair with an angle of 20° have strong positive correlation as $\cos(20) = 0.94$, and with an angle of 90° are uncorrelated as $\cos(90) = 0$).

^b *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$ level of significance, respectively; ns, is not significantly different ($p > 0.05$).

^c AC = annual crop, PA = pasture, SRW = short rotation willow, EC = electrical conductivity, SOC = soil organic carbon, VSWC = volumetric soil water content, TN = total nitrogen, GHG = greenhouse gas.

The multivariate permutation analysis of variance (PERMANOVA) test confirmed the significant difference in GHG emissions among the land-use practices ($p = 0.001$), salinity ($p = 0.001$), and depth to GWT ($p = 0.001$) in both sites (Table 6.3). The VPA test exhibited that the land-use practice alone has the highest contribution to the variation of soil GHG emissions in both sites (site A = 79.3 % and site B = 69.6 %), followed by depth to GWT (i.e., measurement week) and salinity treatments (Figure D-S6).

Table 6.3 Permutation multivariate analysis of variance (PERMANOVA) test for GHG emissions under different groundwater salinity and water table levels in soil cores collected from three different land-use practices at two field sites.

Sources of variation	Site A				Site B		
	df	Pseudo- F	R^2	Pr ($>F$)	Pseudo- F	R^2	Pr ($>F$)
Land-use	2	35.91	0.23	0.001 ***	13.37	0.10	0.001 ***
Salinity	2	24.94	0.17	0.001 ***	25.11	0.17	0.001 ***
Groundwater table	8	8.02	0.22	0.001 ***	13.41	0.31	0.001 ***

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b GHG = greenhouse gas.

6.5.2 Soil Physiochemical Characteristics and their Relationships with GHG

6.5.2.1 Physiochemical Characteristics of Experimental Soil

The physiochemical properties of soils used for the microcosm experiment are presented in Table 6.4. No significant differences were observed in soil physiochemical properties among land-use practices or between sites except SOC, TN, SO_4^{2-} content (ANOVA results are not shown here). The SOC and TN were significantly ($p < 0.05$) higher in soils from PA compared to other land-use practices in the order of PA > SRW = AC in both sites (Table 6.4). However, no significant difference ($p > 0.05$) were found in SOC and TN content between sites. The SO_4^{2-} content was approximately eight times higher in soils from site B than site A (Table D-S1). In the

order of land-use practices, the soil SO_4^{2-} contents were $\text{SRW} > \text{PA} = \text{AC}$ in site A, and $\text{SRW} = \text{AC} > \text{PA}$ in site B, suggesting no consistent land-use patterns were observed between sites.

6.5.2.2 Relationships of GHG with Soil Physiochemical Characteristics

Overall, the relationship between soil GHG (CO_2 , CH_4 , and N_2O) and soil clay content, SOC, TN, and C/N ratio were positive, whereas bulk density, initial EC, WEOC, and $\text{SO}_4^{2-}\text{-S}$ were negative (Figure 6.5). Significant positive relationships ($p < 0.05$) between soil GHG and clay content, SOC, and C/N ratio were observed but were non-significant ($p > 0.05$) between SOC with CH_4 , and C/N ratio with N_2O . The relationships between all GHG emissions and bulk density were significantly negative ($p < 0.05$) (Figure 6.5). Correlations between soil $\text{PO}_4^{3-}\text{-P}$, $\text{SO}_4^{2-}\text{-S}$, and WEOC content were negative and significant ($p < 0.05$) with CO_2 and CH_4 emissions except with N_2O . None of the correlations between other initial soil physiochemical properties and soil GHG emissions were statistically significant ($p > 0.05$) (Figure 6.5).

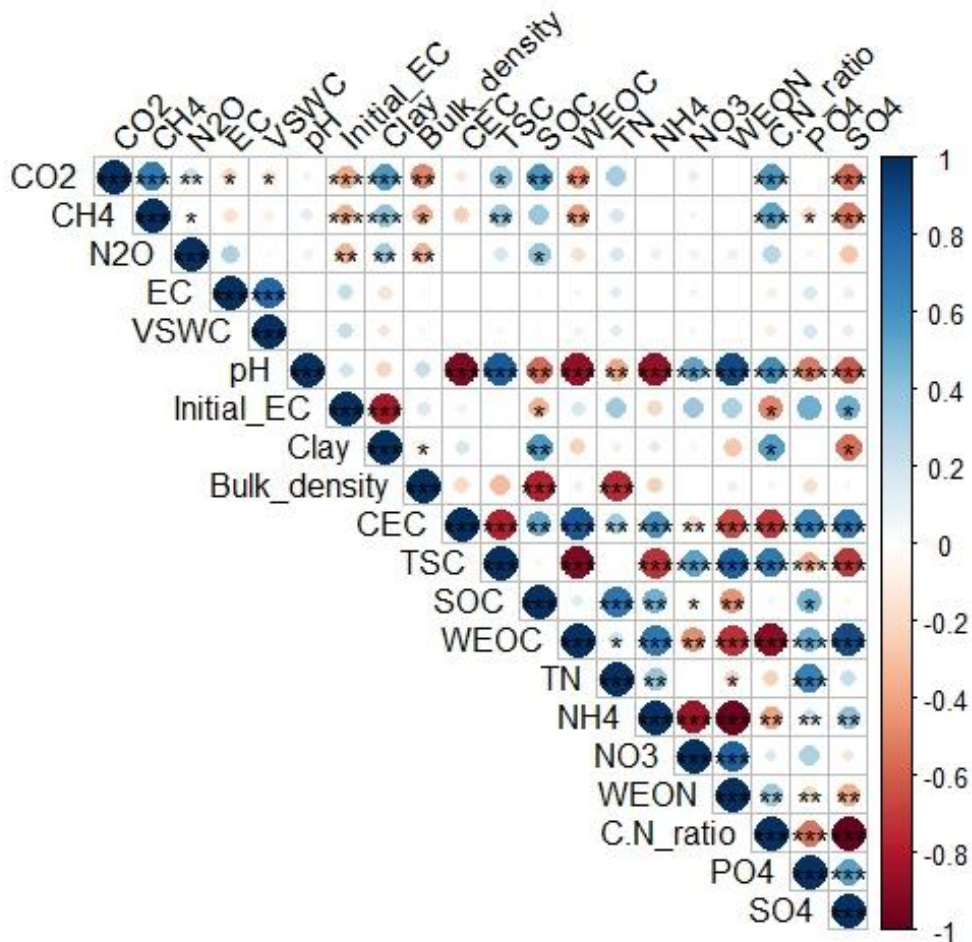


Figure 6.5 Relationship (Spearman rank-order correlation) among GHG, VSWC, EC, and physiochemical characteristics of the experimental soils.

^a Blue circles indicate positive and red circles indicate a negative relationship. Larger circles and deeper colors indicate stronger relationships.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$ level of significance, respectively; and the remainder are not significant ($p > 0.05$).

^c EC = electrical conductivity, VSWC = soil water content, CEC = cation exchange capacity, TSC = total soil carbon, SOC = soil organic carbon, WEOC = water extractable organic carbon, TN = total nitrogen, WEON = water extractable organic nitrogen, C/N ratio = carbon and nitrogen ratio.

6.5.2.3 Redundancy Analysis (RDA) between Soil Physiochemical Characteristics and GHG

Redundancy analysis (RDA) was performed to find out the relation and the effects of soil physiochemical properties and GHG emissions shown in Figure 6.4C and F. The first two component axes explained 86.23 % and 12.43 % of site A (Figure 6.4C), and 85.87 % and 12.83 % (Figure 6.4F) of site B of soil GHG, respectively. The vector lines of SOC, TN, VSWC, EC from site A and site B were statistically significant ($p < 0.05$), showing that SOC and TN played a most crucial role in explaining soil GHG emissions in both sites. There was a significant positive correlation ($p < 0.05$) between SOC, TN, and soil GHG emissions in both sites A and B (Figure 6.4C and F).

6.5.2.4 Relationships Amongst Manipulated Water Table and Salinity with VSWC and EC Measured During the Microcosm Experiment

Groundwater salinity manipulation (Paragraph 7.5.2.3, page 165) resulted in a statistically significant difference ($p < 0.05$) in soil EC among different salinity treatment levels (in S1 and S2 compared to control) in both sites (Table 6.1 and 6.2). Similarly, water table manipulation resulted in a significant difference ($p < 0.05$) in observed VSWC among groundwater table depths in both sites. We did not find any significant difference ($p > 0.05$) in VSWC or EC among land-use practices from site A ($p > 0.05$); however, there was a significant difference ($p < 0.05$) in site B because of groundwater salinity and water table manipulation (Table 6.1 and 6.2). We also observed a significant ($p < 0.05$) positive relationship between soil EC and VSWC in both sites (Figure 6.5 and Figure D-S8) during the incubation experiment.

6.5.3 Global Warming Potential

The effects of different land-use practices from two sites on the global warming potential (GWP) of CO₂, CH₄, and N₂O during the incubation period were calculated (Table 6.4) based on CO₂e. The GWP was significantly affected ($p < 0.05$) by the origin of the soil from three different land-

use practices and two sites. The GWP was significantly high in soils from PA, followed by AC and SRW land-use practices in both sites, whereas site A showed significantly higher GWP than site B.

Table 6.4 Global warming potential (GWP) of CO₂, CH₄, and N₂O (equivalent to CO₂) of three different land-use practices from two sites.

	Land-use	GWP (mg CO ₂ -e m ⁻² d ⁻¹)
Site A	AC	1288.88 b
	PA	4509.89 a
	SRW	808.47 c
Site B	AC	1167.85 b
	PA	2358.27 a
	SRW	639.59 c
<i>F</i> = 15.19		<i>F</i> = 112.73
<i>p</i> = 0.002 **		<i>p</i> < 0.001 ***

^a Means within a column for land-use followed by the same letter are not significantly different ($p > 0.05$) using Tukey HSD.

^b *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^c GWP = global warming potential, AC = annual crop, PA = pasture, SRW = short rotation willow.

6.6 Discussion

6.6.1 Effects of Land-use, Salinity and Groundwater Table on GHG Emissions

6.6.1.1 Land-use Effects

In our study, CO₂ emission was significantly affected by the soils from contrasting land-use practices of both sites, suggesting that land-use was a significant driver of CO₂ emission via influencing the heterotrophic respiration of SOC (Oertel et al., 2016). The highest mean and cumulative CO₂ emission in our experiment was seen from soils from PA, followed by AC and SRW land-use practices, respectively. Enrichment of SOC can trigger microbial activities, which in turn cause a considerable amount of C and N to be released or immobilized that can lead to the emission of CO₂, CH₄, and N₂O, subject to various proximal and distal drivers in soil (Oertel

et al., 2016). In particular, land-use practices control SOC accumulation; therefore, any changes in land-use practices can change the source or sink characteristics for GHG (Liebig et al., 2005). We observed a significant positive relationship with background SOC, and with both SOC and TN from the RDA test in both sites, suggested as a substantial driver of GHG emissions that attributed to the different land-use practices. Hence, in our experiment, elevated CO₂ emissions from both sites were perhaps triggered by higher SOC content and turnover rates from root biomass from the soils from PA land-use practice.

It has been found that PA soil can be a significant source or sink of C and N and crucial for mitigating GHG emission in the US Great Plains region (Follett et al., 2012). Similar to this study, the C/N ratio was positively correlated with CO₂ and CH₄ emissions (Shi et al., 2014, Weslien et al., 2009). Lang et al. (2010) found that the SOC and C/N ratio are the dominant factors that control CO₂ and N₂O emissions from soil. According to Tangen et al. (2015), SOC is usually lost when undisturbed native Prairie catchments are converted to agricultural land and restoring to PA from AC increased SOC in wetland soils within the PPR. However, the SOC quantification associated with land-use could be difficult, especially with a high degree of variability of both biotic and abiotic factors along with a combination of slow SOC sequestration over time (Tangen et al., 2015). Similar to our experiment, Parmar et al. (2015) found reduced GHG emissions in soil cores collected from short rotation forestry. In contrast, Lang et al. (2010) found significantly higher CO₂ emission in the forest compared to PA soils, whereas the N₂O emission was the opposite.

There could be several factors or combinations of factors that contributed to the contrasting variabilities in CH₄ emissions in our experimental soils. For example, Lang et al. (2010) found that PA soils were a weak source of CH₄ emission, whereas forest soils were a weak sink of CH₄. Methane emission from the soil is generally related to the moist environment where methanogenesis can occur (Bridgham et al., 2013, Levy et al., 2012). The CH₄ emission can be

influenced by C/N ratio; however, it is significantly dependent on the soil water content relative to other factors (Gundersen et al., 2012). Using stable C isotope, Wu et al. (2018) found that increased SOC and microbial biomass carbon, and lower C/N ratio and inorganic N following afforestation were a critical factor for high CH₄ uptake. In a meta-analysis of 5000 chamber measurements collected from a range of land-use types, Levy et al. (2012) observed small emissions or a lower rate of net uptake in mineral soils and high emission from organic soils, while SOC, VSWC, and pH were the best sub-set of explanatory variables. Hence, higher SOC content in our experimental soils from PA land-use practice from both sites perhaps caused a significant difference in CH₄ emissions compared to AC and SRW. Moreover, we found higher background SO₄²⁻ content in site B than site A, and higher SO₄²⁻ content under SRW compared to PA land-use practices. Also, SO₄²⁻ content was negatively correlated with CO₂ and CH₂ fluxes in soil. Conceivably the high SO₄²⁻ content inhibited the CH₄ emission under wetter conditions (Ardón et al., 2018); therefore, the CH₄ emission from SRW was lower compared to the PA land-use practice.

In a controlled laboratory experiment to assess the effects of land-use and climate (particularly soil temperature and moisture) on the potential GHG emission from intact soil cores collected from 13 European sites, Schaufler et al. (2010) found higher CO₂ and N₂O emission from grasslands compared to croplands, forests, and wetlands. We found significantly higher N₂O emissions from PA soil, followed by AC and SRW in both sites. Similar to our experiment, Follett et al. (2012) observed high C and N content in the PA soils, which were the most probable cause for simulated microbial activities and both high CO₂ and N₂O emission. Additionally, the optimum C/N ratio ranged between 11 to ≥ 30 and was negatively correlated with N₂O emission (Gundersen et al., 2012). Research has shown that heterotrophic nitrifying bacteria are often able to denitrify with low NO₃-N but high SOC content under aerobic conditions (Wrage-Mönning et al., 2018).

6.6.1.2 Salinity Effects

Salinity treatments significantly decreased CO₂ and CH₄ emissions except for the CH₄ emission from site B ($p = 0.069$), which were already low; however, it significantly increased the N₂O emission in soils from both sites. A microcosm experiment performed on the coastal forested wetlands (Ardón et al., 2018) found that salinity can suppress CO₂ emission under both flooded and drought conditions, and increase CH₄ emission (300 to 1200 %) under the flooded condition and almost no release under drought condition; however, the magnitude and direction were reliant on the hydrology. Similar to our experiment, an incubation study by Maucieri et al. (2017) examined short-term effects of irrigation water salinity on soil GHG emissions from semi-arid Australian soil; CO₂ emission was reduced by 19 % at 5-mS cm⁻¹ and 28 % at 10-mS cm⁻¹, whereas N₂O emission increased 60 %, and CH₄ emission was not affected by increased salinity apart from soil water holding capacity. Setia et al. (2011) also found a significant decrease in CO₂ emission with increasing salinity ranged from 1 to 5 mS cm⁻¹.

The salt concentration and water content regulate the osmotic potential in the soil; thus, at both high salinity and low water content, soil microorganisms can tolerate the high osmotic potentials by synthesizing osmolytes, which lets them continue metabolism (Yan & Marschner, 2013). Consequently, perhaps both the distinct and linked effects of groundwater salinity and hydrology controlled the N₂O emission during our experiment. Fluctuating aerobic-anaerobic conditions and environments low in oxygen can promote N₂O production by nitrifier denitrification; furthermore, high concentration of NO₂⁻ following the application of ammonium (or urea) has been linked to high pH (Wrage-Mönnig et al., 2018). Dang et al. (2017) likewise observed enhanced N₂O production in an incubation experiment under higher soil salinity and suggested that the addition of available carbon (glucose) and nitrogen (nitrate) created favorable conditions for denitrification. Comparably, in our experimental soil, SOC and TN attributed from contrasting land-use practices act as a significant driver that controlled the GHG emissions. Hence,

increased N₂O emission through denitrification with increased salinity is likely (Marton et al., 2012).

We observed very low emissions or some cases uptake of CH₄ in site B, as well as in site A (except under PA land-use practice). It is evident that higher salinity increases the SO₄²⁻ availability that acts as an alternative terminal electron acceptor under anaerobic condition and can shift microbial metabolism towards more energetically favorable processes (Bridgham et al., 2013). A significant inverse correlation has been observed between the CH₄ emission and SO₄²⁻ content in wetland soil in the PPR (Pennock et al., 2010). Our experimental soil from site B has very high SO₄²⁻ content as well as we found a robust negative correlation with CH₄. Therefore, the low CH₄ emission was observed from those soils that have high SO₄²⁻ content in the soil. Similarly, the presence of high SO₄²⁻ in soil inhibited the CH₄ emission in an incubation experiment (Ardón et al., 2018) and riparian areas of PPR wetlands (Dunmola et al., 2010).

6.6.1.3 Water Table Effects

The GWT depth significantly controlled the soil GHG emissions from our experimental soil cores. In our manipulative experiment, we found a significant positive relationship between the declined GWT depths and VSWC. Soil water content controls microbial activity and all related processes and is the single most crucial soil parameter that regulates GHG emissions (Oertel et al., 2016). Overall, higher CO₂ emission can occur from the rapid decomposition of C in well-drained areas (Freeman et al., 2001), N₂O emissions are most likely between strict aerobic and anaerobic conditions (Davidson et al., 2000), and CH₄ is produced via the reduction of CO₂ in a strictly anaerobic microbial process known as methanogenesis (Bridgham et al., 2013). Conversely, soils can also be a sink of atmospheric CH₄ through microbial oxidation under aerobic conditions (Thangarajan et al., 2013).

In our experiment, the CO₂ emission rate was variable and to some extent dependent on VSWC in both sites; CH₄ emissions decreased with declining water table depths as the VSWC

decreased. The N₂O emissions were low under the higher water table level as the VSWC in the experimental soil cores were near-saturated; however, emission increased once the moisture condition became ideal (at the midpoint of groundwater table treatments). Similarly, a laboratory incubation study found high N₂O emission with improved availability of soil water but not under saturated condition, hence indicated that water-filled pore spaces and C availability primarily controlled the denitrification process and thus N₂O emission (Gillam et al., 2008). Research with Canadian prairie and forest soils under short term flooding observed significant CH₄ emission, while 80-90 % of the CH₄ absorbed within a week under non-flooded conditions (Wang & Bettany, 1997). Moreover, an incubation study with soils from agroforestry land-use showed significantly lower cumulative CO₂ and N₂O emissions compared to the riparian forest under optimum soil moisture condition, whereas lower N₂O emission compared to agricultural crop production under fluctuating soil moisture condition (Moore et al., 2017).

In contrast, an incubation experiment with peat cores from central and eastern Canada (Blodau et al., 2004) found a deeper water table increased CO₂ production through soil respiration and microbial biomass, whereas CH₄ production and emissions decreased. Additionally, in a lysimeter experiment using undisturbed peat soil columns, higher CO₂ emissions were observed at lower water table depth (40 cm below surface) compared to the greater depths (80 cm below surface), CH₄ emissions were very low or negative, whereas peak N₂O emissions were in intermediate depth (Berglund & Berglund, 2011). In a field-scale study to assess the impact of short-term effects of changing GWT depth (i.e., flooding) on soil GHG emissions, Mander et al. (2015) observed a significant increase in CH₄ emission and decreases in both CO₂ and N₂O emission under riparian old grey alder stands with the shallower water table.

A study within the PPR agricultural landscape observed that the hotspots of GHG are predominantly driven by soil moisture and SOC availability (Dunmola et al., 2010). For our microcosm study, we collected intact soil cores from the annual crop, pasture, and short rotation

willow plantation; however, we did not measure GHG emissions directly in the field. Therefore, we might not accurately compare our results with that of field-scale study; however, we observed that the VSWC in our experimental cores largely controlled the GHG emissions. When soils dry out, the substrate supply becomes increasingly limited for microbes as the water drains out from soil pores and water films around the soil aggregates become thinner and disconnected (Yan et al., 2015). However, we should consider that lowering of the water table may perhaps expose new layers in soil containing varied substrate composition and availability for microbial decomposition as well as soil physical properties that might have different impacts on the emissions rate (Berglund & Berglund, 2011).

6.6.2 Global Warming Potential and Contrasting Land-use Practices

We observed significantly lower GWP (CO₂-e) in soils from SRW land-use compared to AC and PA land-use practices. Likewise, in a field-scale study in the Canadian prairie region, Baah-Acheamfour et al. (2016) observed that the establishment of agroforestry systems could reduce emissions of CH₄ and N₂O, compared to grassland and thus can provide benefit to mitigate climate change within the agroecosystem. Amadi et al. (2016) found that agroforestry in the form of shelterbelt has the potential to mitigate GHG by reduced N₂O emission and storing CH₄ as a sink through enhanced SOC storage compared to adjacent cropped fields within the Prairie Ecozone of Saskatchewan, Canada. Another field-scale study in a humid temperate region of southern Europe Merino et al. (2004) observed that afforestation could significantly increase SOC content as a consequence of the transformation of the crop field, decrease N₂O emission, and increase CH₄ uptake. Thus, SRW can be a promising land-use practice in the fallow marginal riparian zones of PPR agroecosystem (Amichev et al., 2014b). However, it is often challenging to generalize the impact of agroforestry on the GHG budget without a better understanding of the plant types as well as soil and climatic drives that control the GHG emissions (Benanti et al., 2014).

6.7 Conclusions

Our results showed that adjacent contrasting riparian land-use practices had a significant influence on GHG emissions within the PPR agroecosystems. We observed significantly higher CO₂, CH₄, and N₂O emissions from PA land-use practice. Changes in soil properties, mainly organic C and N, because of contrasting land-use practices helped to explain the observed difference in soil GHG emissions. Conceivably, high background SO₄²⁻ concentration in soils collected from SRW land-use practices decreased the CH₄ emission and subsequently contributed less towards the GWP.

In support of our hypothesis, we saw that lowering the water table decreased CH₄ emission with the reduction of VSWC but resulted in higher N₂O emission under an intermediate water table position when VSWC in the cores reached suitable conditions for denitrification. In contrast to our hypothesis, we noticed a variable CO₂ emission, i.e., first increased with lowering the water table and then started to decrease as the VSWC diminished. We observed a decrease in CO₂ and CH₄ emissions, but a significant escalation in N₂O emission with elevated groundwater salinity, partially supporting our hypothesis.

The GWP of SRW was significantly lower compared to AC and PA, which recommended a potentially promising land-use practice in the fallow marginal riparian zones that are not suitable for crop production due to the higher salinity or unsuitable for agricultural crop production within the PPR agroecosystem. Overall, our experiment showed a decrease in GHG emissions with increasing salinity and varying responses to GWT based on GWT depth and the GHG in question.

7 SOIL ENZYME ACTIVITY AS AFFECTED BY LAND-USE, SALINITY, AND GROUNDWATER FLUCTUATIONS IN WETLAND SOILS OF THE PRAIRIE POTHOLE REGION

7.1 Preface

Soil enzymes are the primary mediators of biogeochemical processes and thus the mineralization of C, N, P through soil organic matter decomposition in which microbes play a crucial role. Extracellular enzymes in the soil are sensitive to disturbance and land-use practice changes. In addition to high biomass production for carbon-neutral bioenergy, SRW land-use practice has the capability of improved soil organic carbon balance. Land-use practice shifts can affect GHG emissions by further controlling many soil factors such as organic carbon, water, and nutrients availability, as well as the altered salinity. However, the question remains that the soil condition affected by the land-use practices can also alter the soil extracellular enzyme activities (EEAs). Chapters 5 and 6 examined the potential C sequestration and GHG emissions, respectively, under SRW plantation compared to adjacent annual crop (AC) production and pasture (PA). Therefore, this chapter assesses the change in the soil EEAs related to C, N, and P cycling as affected by the groundwater table and salinity variation in the soils from different land-use practices on a microcosm scale. The conceptual model (Figure 7.1) outlines the variables that were assessed in Chapter 7 and can hypothetically be impacted by the soils from past land-use practices with the variation of groundwater table and salinity.

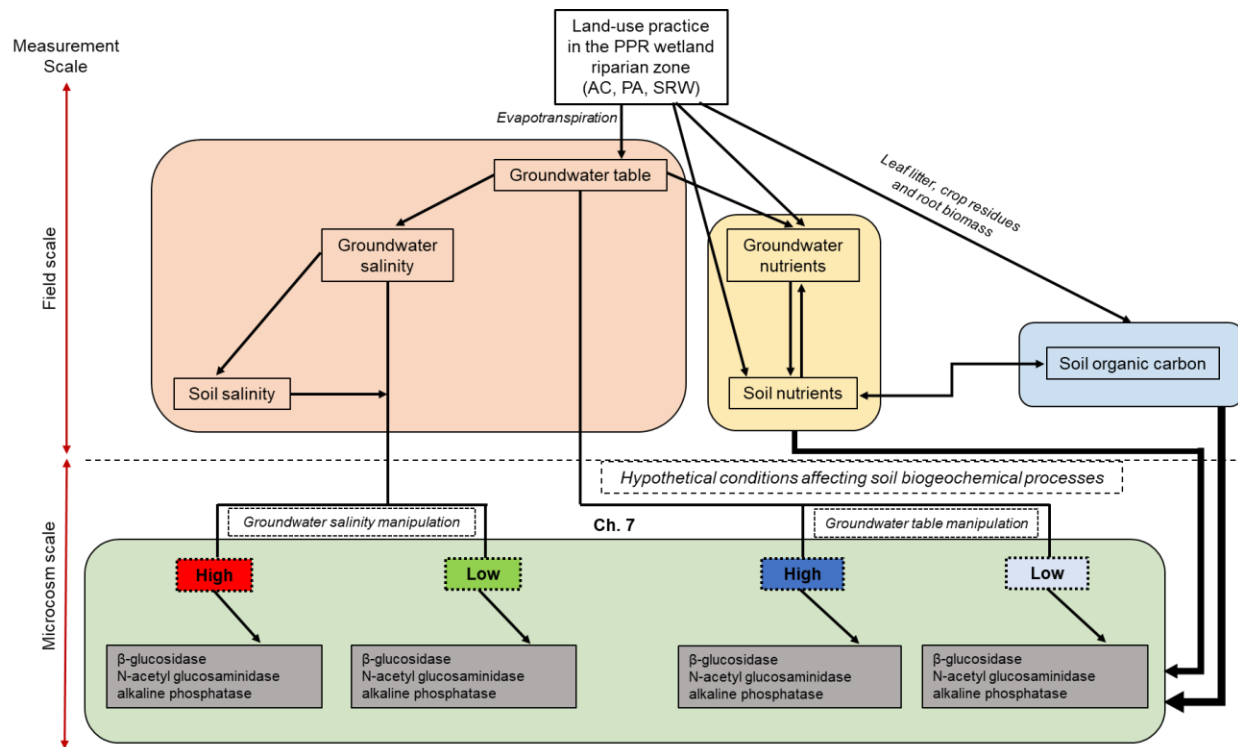


Figure 7.1 Conceptual model showing the potential land-use effects on soil EEAs (BG, NAG, AP) as affected by the variation of groundwater table and salinity.

^a AC = annual crop, PA = pasture, SRW = short rotation willow, EEAs = extracellular enzyme activities, BG = β -glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase.

7.2 Abstract

Land-use change and climatic variability are significant drivers for the loss of ecosystem services and soil quality in the prairie pothole region (PPR). Shifting land-use practices can significantly alter the groundwater table, soil salinity, and the quality and quantity of soil organic matter (SOM). In wetland soils, these land-use induced changes may influence biogeochemical processes facilitated by extracellular enzymes (EEs) involved in C, N, and P cycling through SOM decomposition. The objective of this study was to assess the effect of groundwater and salinity on β -glucosidase (BG), N-acetyl glucosaminidase (NAG), and alkaline phosphatase (AP) activities in PPR soils collected from three different adjacent riparian land-use practices. In a microcosm study conducted over ten weeks, soils were treated with groundwater salinity levels

(control, 6 mS cm⁻¹, and 12 mS cm⁻¹) and declining groundwater table depths. Both univariate ANOVA and multivariate PERMANOVA tests showed that soil extracellular enzyme activities (EEAs) differed significantly ($p < 0.05$) among land-use practices and between groundwater table depths. Conversely, it showed a non-significant ($p > 0.05$) impact of groundwater salinity on soil EEAs. Land-use effects on EEAs were in the order of PA > AC = SRW, suggesting that the effects resulted from background SOC and TN as well as soil nutrient availability. Soil EEAs significantly ($p < 0.05$) reduced under a deeper simulated groundwater table depth, except reverse for BG in site B, indicated that the lowered groundwater table could lead to transitory drought stress for SOM decomposers.

7.3 Introduction

Extracellular enzymes (EEs), facilitated by microorganisms, are the primary mediators of C, N, and P cycling through soil organic matter (SOM) decomposition in soil (Burns et al., 2013, Luo et al., 2017, Sinsabaugh et al., 2008). Soil microorganisms produce a variety of soil EEs to acquire energy and nutrients (Sinsabaugh et al., 2009, Wallenstein & Burns, 2011). Soil hydrolytic enzymes play a vital role in biogeochemical cycling and mediate SOM decomposition, transformation, and mineralization (Shi et al., 2018). Among the hydrolases, β -glucosidase (BG) catalyzes the final step of cellulose decomposition and release glucose; N-acetyl glucosaminidase (NAG) degrades chitin and other β -1, 4-linked glucosamine polymers to release assimilative C and N; and alkaline phosphatase (AP) hydrolyze phosphomonoesters, and some cases phosphodiester, releasing phosphate for microbes and plants in the soil (Luo et al., 2017). Hence, soil extracellular enzyme activities (EEAs) can be used as indicators of change in soil function due to land-use practices (Trasar-Cepeda et al., 2008) and climate-linked environmental stresses (Henry, 2012, Schimel et al., 2007); their activity can detect changes sooner than other soil analyses (Acosta-Martínez et al., 2007). Numerous studies have shown that EEAs were sensitive to changes in soil characteristics due to change in land-use

practices (Acosta-Martínez et al., 2003a, Acosta-Martínez et al., 2003b, Bandick & Dick, 1999, Stauffer et al., 2014, Tischer et al., 2015, Wallenius et al., 2011), fluctuating groundwater table depths (Freeman et al., 1996, Pulford & Tabatabai, 1988, Wiedermann et al., 2017), and variations in salinity (Frankenberger & Bingham, 1982, García & Hernández, 1996, García et al., 1994, Pan et al., 2013, Shi et al., 2019). Therefore, EEAs are potential indicators of soil quality, useful for understanding soil ecosystem functioning (Bandick & Dick, 1999).

Wetland soils can serve as a reservoir of water, carbon, and nutrients, with fluctuations in water levels influencing the type and intensity of biogeochemical processes. The prairie pothole region (PPR) retains millions of small wetlands that support prairie grasses, habitat for migratory birds, productive agricultural land, and many other ecosystem services (Mitsch & Gosselink, 2000, Richardson & Arndt, 1989). With its semi-arid climate, the PPR is composed of a hydrologically distinct and highly sensitive wetland ecosystem that are vulnerable to land-use and climate change (Johnson et al., 2005, Johnson et al., 2010, Werner et al., 2013). Soils of this region experience both drought and deluge (Johnson et al., 2004, Winter & Rosenberry, 1998). There is potential for future drier climatic conditions (Millett et al., 2009), jeopardizing the ecosystem services provided by the PPR due to the alteration of shallow groundwater induced by rapid evaporation and increased transpiration through wetland riparian zone land-use practices (Poiani & Johnson, 1991, Poiani & Johnson, 1993). Intensive agricultural land-use practices, including wetland drainage, have disturbed the native vegetation and soils throughout the PPR (Bartzen et al., 2010), (Guntenspergen et al., 2002, McCauley et al., 2015). Hence, stresses related to land-use can diminish the functionality and capability of wetland ecosystems to sustain soil health and environmental quality (Rosen et al., 1995). Furthermore, hydrology research in Prairie wetlands suggests that surrounding land-use changes can significantly affect the water balance due to higher potential evapotranspiration vs. precipitation (Conly & van der Kamp, 2001).

Short rotation willow (SRW) is a high biomass producing crop that was introduced in Canada during the early 1990's as an environmentally sustainable land-use practice fulfilling multiple ecological benefits including the sustainable supply of bioenergy feedstock (Amichev et al., 2014b). However, this practice is relatively new to the PPR of Saskatchewan (Amichev et al., 2014a). Establishing fast-growing SRW within the riparian zones can impact groundwater hydrology and the soil water balance (Caldwell et al., 2018, Mercau et al., 2016). During the growing season, groundwater can be depleted by rapid evapotranspiration from the wetland vegetation in the riparian zones (Hayashi et al., 2016), which might become critical for agricultural production and wetland management in this region and globally (Fan et al., 2013).

Land-use practices that supply elevated levels of crop residues can significantly increase the soil EEAs (Bandick & Dick, 1999, Jordan et al., 1995). In cultivated soil systems, EEAs were higher where organic residues were added as compared to treatments without organic amendments (Bandick & Dick, 1999); and showed that the soil β -glucosidase activities best reflect the management effects on soil quality. Soil EEAs (except α - and β -glucosidase, and α - and β -galactosidase) remained higher in the adjacent pasture (PA) compared to annual crop (AC) production (Bandick & Dick, 1999). In a study with SRW compared to forestry, pasture, and agroecosystem, high laccase and phosphatase activities were observed in the forest soil compared to the other land-uses and did not significantly differ between the SRW and the other land-uses (Stauffer et al., 2014).

Salinization is a pressing environmental challenge globally (Rengasamy, 2006) and a significant threat to agricultural productivity across the PPR (Eilers et al., 1997, Nachshon et al., 2014).

Precipitation events contribute to groundwater fluctuations and dilution of soil salinity (LaBaugh et al., 1995), whereas drought periods can concentrate salts in riparian soils (Levy et al., 2018). This oscillation in salinity (Euliss & Mushet, 1996, LaBaugh et al., 2018) can also potentially

affect soil biogeochemical cycling (Evenson et al., 2018, Holloway et al., 2011) through nutrient imbalances and the lower osmotic potential of the soil solution.

Elevated soil salinity can reduce EEAs directly via the effects of osmotic potential and specific ions on enzymes and indirectly by lowering microbial biomass (Rath & Rousk, 2015). For example, in a laboratory experiment, Frankenberger & Bingham (1982) found that soil β -glucosidase, phosphatase, sulfatase, amylase, and dehydrogenase activity decreased with increasing electrical conductivity (EC); however, the degree of inhibition varied among the EEAs, and the nature and amount of salts added. Egamberdieva et al. (2011) observed that soil glucosidase, alkaline phosphatase, phosphodiesterase, urease, and protease activity were inhibited by higher soil salinity treatments (5.6 and 7.1 mS cm⁻¹) compared to non-saline soil (1.3 mS cm⁻¹). Additionally, high salt concentrations are often combined with low availability of soil water and have different effects on microorganisms (Kakumanu & Williams, 2014).

During drought conditions, water is held more tightly to soil aggregates as matric potential decreases (Kakumanu et al., 2013). Drought conditions created due to greater water table depth have shown to affect soil EEAs. For instance, β -glucosidase and phenol oxidase activities decreased with declining water table depth in a mesocosm experiment with alpine wetland (Wang et al., 2017b). In a mesocosm experiment with peat monoliths in Michigan, USA, Wiedermann et al. (2017) measured hydrolytic EEAs at intermediate depth and found the reduced activity of β -glucosidase, N-acetyl glucosaminidase, alkaline phosphatase, and sulfatase except for cellulase. However, there are also conflicting results in literature with the water table drawdown experiment. In a field-based experiment in Welsh peatland Freeman et al. (1996) found a 31 to 67% increase in β -glucosidase, phosphatase, and sulphatase activities upon water table drawdown, suggesting that drought condition increased mineralization rate through direct stimulation of enzymes.

Understanding the interactions among climatic conditions, groundwater hydrology, salinity, and biogeochemical cycling associated with prevailing land-use practices within the PPR is complex. Individual effects of land-use, salinity, and groundwater table variation due to climatic variability on soil EEAs have been well documented in the literature. However, their *combined* effects on soil hydrolytic EEAs, especially in mineral wetlands, has not been studied before. We conducted a microcosm experiment with controlled groundwater table levels at two levels of salinity with intact soil cores collected from three adjacent riparian land-use practices from the PPR to evaluate the effects on three hydrolytic soil EEAs, i.e., β -glucosidase, N-acetyl glucosaminidase, and alkaline phosphatase. We hypothesized that soil EEAs would be: 1) higher in soils from pasture compared to annual crop and short rotation willow land-use practices, irrespective of groundwater table depths or salinity levels; 2) lower under higher salinity (groundwater EC = 12 mS cm⁻¹) due to microbial stress from increased osmotic potential; 3) lower under a deeper groundwater table level because of reduced SOM decomposition resulting from a decrease in soil moisture.

7.4 Materials and Methods

7.4.1 Site Description and Collection of Undisturbed Soil Cores

For the microcosm incubation experiment, 54 undisturbed soil cores (9 cores per land-use \times 3 land-uses \times 2 sites; 0 to 30 cm depth) were collected from riparian zones of PPR wetlands within three different adjacent land-use areas at each of two neighboring PPR wetland sites (site A and site B) at Indian Head, Saskatchewan, Canada (N 50° 30.605'; W 103° 43.011'; 605 m in elevation). The three contrasting land-uses included: short rotation willow = SRW, annual crop = AC, and pasture = PA. The soils of both sites are non-calcareous Black Chernozems of the Oxbow Association, with level to gentle rolling (0-10 % slopes) topography formed on loamy glacial till (Saskatchewan Soil Survey Staff, 1986). At both sites, short rotation willow variety *Salix dasyclados* Wimm. (cultivar 'India') was planted in June 2013 side-by-side with pasture

(established 10-12 years before with alfalfa (*Medicago sativa*) and bromegrass (*Bromus madritensis*) mixture, and annual crop (cultivated oats *Avena sativa*). All the soil cores were collected in mid-August of 2015 following sufficient natural warming with peak microbial activities. The collection time represented the end of the 3-year rotation cycle of SRW plantation. Intact soil cores were collected in order to avoid disturbance effects in soils produced by sieving (Reichstein et al., 2005). Cores were made using a transparent PVC-cylinder of 9 cm diameter and 30 cm height with a sharpened bottom and capped on both ends. Cores were collected with a truck-mounted with hydraulic corer (Giddings Machine Company Ltd., Windsor, CO, USA) based on the landscape position and distance from the wetland basin (i.e., riparian zones under each land-use practices) (See Chapter 6). Collected soil cores were transported back to Saskatoon in coolers and preserved frozen at -20°C until the experiment. One additional soil core from the same micro-location under each land-use practice from both sites was also collected for the measurements of soil physiochemical properties and divided into three subsamples according to analyses requirements.

7.4.2 Soil Physiochemical Properties

The first subsample was air-dried, ground, and passed through 2 mm sieve for particle size distribution, cation exchange capacity (CEC), pH, electrical conductivity, ammonium acetate extractable N, and P. The second was air-dried and ground finely with a ball mill grinder for organic C, total C, and N. The third was frozen until analysis for water extractable organic carbon (WEOC) and water extractable organic nitrogen (WEON). During core sampling, soil samples for bulk densities were also collected in the field using a hand-held core sampler (diameter = 5.4 cm, height = 3 cm), which were then weighed and oven-dried at 105°C for 24 hours (Hao et al., 2008). Particle size distribution was determined by the modified pipette method (Kroetsch & Wang, 2008). CEC was measured by the ammonium acetate methods at pH 7 (Hendershot et al., 2008a). Soil pH was determined in 20 mL deionized water with 10 g air-

dried soil samples (2 : 1 ratio) by digital pH meter (PC700 pH/mV/conductivity, Oakton, Vernon Hills, IL, USA) (Hendershot et al., 2008b). Soil EC determined in the same extract used for pH measurement after 1 hour shaking with an end-over-end shaker, then filtered (No. 42, Whatman Inc., Piscataway, NJ), and measured using a digital EC meter (PC700 pH/mV/conductivity, Oakton, Vernon Hills, IL, USA) (Miller & Curtin, 2008). Ammonium ($\text{NH}_4^+\text{-N}$) and nitrate ($\text{NO}_3^-\text{-N}$) were determined using 2.0M KCl (Maynard et al., 2008), and phosphate ($\text{PO}_4^{3-}\text{-P}$) were measured using a 1M ammonium acetate (buffered at pH 7) extraction and colorimetric analysis using a Technicon Auto-Analyzer (Technicon Industrial Systems, Tarrytown, NY, USA) (Simard, 1993). Total soil carbon (TSC) and soil organic carbon (SOC) were measured by dry combustion (model Leco-2000, Leco Corporation, St. Joseph, MI) (Skjemstad & Baldock, 2008); carbonates were removed by HCl acid fumigation in a desiccator (Bisutti et al., 2004) for SOC measurements. Total nitrogen (TN) was determined by dry combustion with a CNS analyzer (model C632, Leco Corporation, St. Joseph, MI) (Rutherford et al., 2008). WEOC and WEON determined with fresh soil (20 ± 1 g) gently mixed with 30 mL 5mM CaCl_2 , then filtered through 0.45 μm polycarbonate membrane filter (Whatman Inc., Piscataway, NJ), and measured using a TOC-VCPN analyzer (Shimadzu Scientific Instruments, Kyoto, Japan) (Chantigny et al., 2008).

7.4.3 Microcosm Experimental Setup

A microcosm incubation experiment following a nested design (Krzywinski et al., 2014, Schielzeth et al., 2013) was carried out for ten weeks in the agricultural greenhouse of the University of Saskatchewan, Canada, to understand the effect of preceding land-use practices and its associated change in groundwater table depths and salinity on soil EEAs related to C, N, and P cycles. For this purpose total 54 (2 sites \times 3 land-use \times 3 salinity levels \times 3 replicates) intact soil cores were used from two PPR wetland sites under three land-use practices (SRW, AC, and PA), with three groundwater salinity levels (control = 0.3 mS cm^{-1} , S1 = 6 mS cm^{-1} , and S2 = 16 mS cm^{-1}) with three replicates ($n = 3$).

7.4.4 Groundwater Table Manipulation and Salinity Treatments

Each experimental unit (Figure 7.2) consisted of a PVC bucket (height 38.1 cm and width 30.48 cm, with 19 L capacity) to house a single intact soil core. The bottom of each bucket was layered with approximately 2.5 cm of gravel. The bottom portion of each soil core was wrapped with a piece of fiberglass screen of 1 mm (mesh size 18) opening to hold the soil securely. The top portion of the cores was kept open. The PVC cylinders casing the soil cores were punched with a uniform series of holes (with 3 cm horizontal by 7 cm vertical distance using a 3 mm diameter needle) to facilitate water movement into the core. Dynamic (declining) groundwater table depths were maintained from 0 cm (soil surface = high or shallow water table) to 29 cm (water table dropdown to the bottom of the intact soil cores = low or deep water table). The groundwater table was dropped by stepping down the water table level from the surface level (at 0 cm in week 0) to a depth of 2 cm in the first week, and then 3 cm each week for 10 weeks when it reached at the bottom of the cores (at 29 cm in week 10). The water table dropdown was controlled manually by removing saline water from the container to a pre-specified depth for each week.

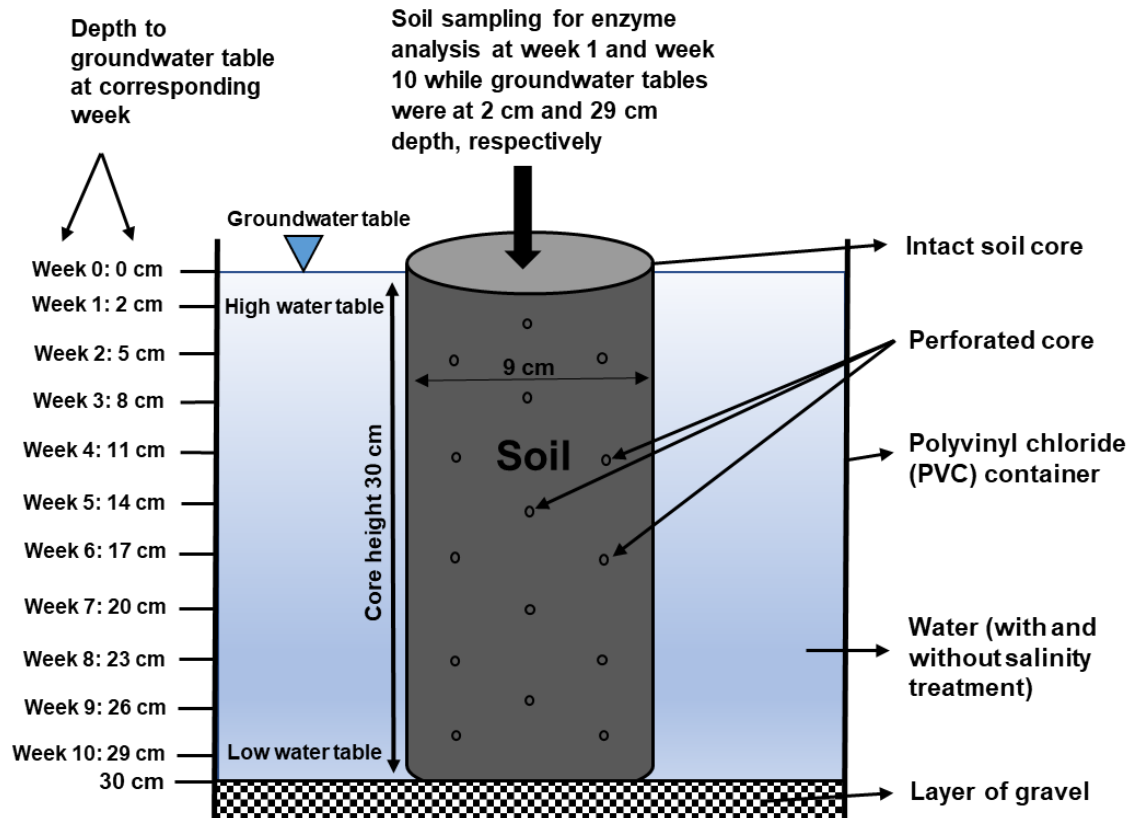


Figure 7.2 Illustration of an individual experimental unit with intact soil core used for incubation study (**Note:** the diagram is not to scale).

Groundwater salinity levels for the treatments of S1 and S2 were attained mixing Na_2SO_4 : KCl : CaCl_2 : MgSO_4 salts at the ratio of 5:2:12:14 with distilled water. All these salts were chosen because they composed of dominant cations and anions that are commonly present in the soil and groundwater in the Prairie region of Canada and the northern United States (Last & Ginn, 2005). The control salinity treatment was distilled water without any salts. EC of the water was monitored weekly to maintain the groundwater salinity at the desired level. In-core soil volumetric soil water content (VSWC) and EC were also measured at the time of soil samples collection for enzyme activity analyses using a digital soil moisture meter (HydroSense II, Campbell Scientific Inc., Logan, UT, USA). The temperature and relative humidity of the greenhouse chamber used for microcosm incubation experiment were controlled at $20 \pm 1^\circ\text{C}$ and average 44%, respectively (Figure E-S1 and S2).

7.4.5 Collection of Soils for Enzyme Activity

Soils for EEAs were collected from each intact soil core at week-1 and week-10 (Figure 7.2). Surface soil samples (0-10 cm depth) were collected without disturbing the soil cores as much as possible using a small stainless-steel tube. Soil samples were transferred immediately in plastic Ziploc bags to preserve the field moisture conditions. Samples were kept on ice for transport and stored in a -20°C freezer until analyses. Gravimetric water content was calculated from a 5-g sub-sample (Topp & Ferre, 2002).

7.4.6 Extracellular Soil Enzyme Activity Analyses

The activities of three soil EEs involved in the hydrolysis of organic compounds (Hydrolase group) important to soil C, N, and P cycling were carried out: β -glucosidase (EC 3.2.1.21), N-acetyl glucosaminidase (EC 3.2.1.30), and alkaline phosphatase (EC 3.1.3.1) using a MUB (4-methylumbelliferone) based high-throughput fluorometric microplate assay (Bell et al., 2013, Hargreaves & Hofmockel, 2015). EEAs were determined in triplicate on 1 g of field moist/fresh soil, mixed with 125 mL Tris buffer (also known as modified universal buffer) adjusted to pH 8 (Deng et al., 2013). Mixing was done in a blender at high speed for 30 s to make a homogenous slurry. While stirring on a magnetic stirrer, 1800 μ L of soil slurry was taken into a 5mL centrifuge tube where it received one of the three synthetic C-, N-, and P- rich substrates bound with 4-Methylumbelliferyl (MUF) fluorescence dye solutions (4-Methylumbelliferyl β -D-glycopyranose for BG, 4-Methylumbelliferyl N-acetyl- β -D-glucosaminide for NAG, and 4-Methylumbelliferyl phosphate for AP). Substrate amended soils were incubated for 3 hours at 24°C at 140 rpm on a mechanical shaker. All the samples were centrifuged for 5 minutes at 2000 rpm, and 10 μ L 1 M NaOH were added before centrifugation to enhance fluorescence. With an electronic pipette, 250 μ L of the centrifuged solution was dispensed into each well of a black 96 well microplate. Fluorescence was determined using a FilterMax F5 Microplate Reader (Molecular Devices, USA) at the wavelengths for excitation light of 360 nm and emission light of 465 nm. The

fluorescence readings of the substrate wells were converted to MUF units using a series of 4-methylumbelliferone standards in concentrations ranging from 0-100 μM (0, 2.5, 5, 10, 25, 50, 100 μM). For the correction of different quenching and autofluorescence properties of the samples, all standards were prepared with three analytical replicates in each soil suspension. Specific enzyme activity refers to the enzyme activity divided by the SOC content for each of the soils.

7.4.7 Statistical Analyses and Data Visualization

Visualization of soil enzyme data and statistical analyses were performed using R version 3.4.4 for Windows (R Core Team, 2018) and the following packages “car” (Fox et al., 2018), “corrplot” (Wei et al., 2017), “ggplot2” (Wickham, 2016, Wickham et al., 2018), “lmerTest” (Kuznetsova et al., 2017), “TukeyC” (Faria et al., 2018), “vegan” (Oksanen et al., 2017), and “HH” (Heiberger, 2017). Linear mixed-effects models (Bolker et al., 2009, Schielzeth et al., 2013, Zuur et al., 2009) for nested design (Krzywinski et al., 2014) were used to find the difference among treatments or factors. The Shapiro-Wilk test and histogram were used to test the normality of obtained EEAs data. Homogeneity of variances or homoscedasticity was tested by Levene’s test using “car” package. For both univariate and multivariate analyses, the square root transformation was performed to improve the assumption of normality and homoscedasticity. The relationship among soil enzymes and physiochemical properties were assessed by Spearman’s rank-order correlation test and visualized using “corrplot” package. A linear regression model was used to recognize the general relationship between applied groundwater table depths and groundwater salinity treatments using “ggplot2” package. Analysis of variance (ANOVA) with a nested design and linear mixed-effects models was used from “lmerTest” to assess significant difference (hypothesis testing) of individual EEA. The permutation multivariate ANOVA (PERMANOVA) was used to assess significant difference of EEAs combinedly. Tukey Honest Significant test (Tukey HSD) used for univariate multiple comparisons of means among

treatments in case of significant effects found in ANOVA using the “TukeyC” package. Unconstrained ordination (with Bray-Curtis matrix of dissimilarities), non-metric multidimensional scale (NMDS) was used to plot the original position in multidimensional space with a reduced number of dimensions to visualize the difference between sites, among groundwater table depths, among land-use practices and among groundwater salinity treatments along with soil EEAs. The linear relationship between soil physiochemical properties and EEAs were analysed by redundancy analysis (RDA) through the development of multiple linear regression to reflect variables in the same Cartesian coordinate system. The proportional contribution of land-use practices, groundwater salinity, and water table depth to variation in soil EEAs were determined by variation partitioning analysis (VPA). Analysis of similarities (ANOSIM) was used to assess significant differences (hypothesis testing) among land-use practices, groundwater salinity treatments, and between groundwater table depths, along with soil EEAs. ANOSIM was used to calculate a matrix of dissimilarity ranks after converting the scores to find the ratio between within-group and between-group. The NMDS, RDA, VPA, and ANOSIM analyses were carried out using “vegan” package. All statistical tests were statistically significant at p -values ≤ 0.05 .

7.5 Results

7.5.1 Soil EEAs

7.5.1.1 Differences in Soil EEAs Following the Effects of Land-use Practices

The mean values of soil EEAs (BG, NAG, AP), and post-hoc multiple comparison procedure (Tukey HSD test) results are presented in Table 7.1. All EEAs were significantly higher ($p < 0.001$) from PA compared to the soils from AC and SRW land-use practices in both sites (Table 7.1 and 7.2). Soil BG, NAG, and AP activities across the soils from different land-use practices were in the order of PA > SRW = AC. Compared to AC and SRW, the soil from PA are twice as high for BG, five times greater for NAG, and three times greater for AP (Table 7.1).

Table 7.1 Mean (\pm SE) soil EEAs (BG, NAG, AP), VSWC, and soil EC measured under different groundwater table levels and salinity treatments in the soil cores collected from different land-use practices at two field sites A and B.

	Site A					Site B				
	BG	NAG	AP	VSWC	EC	BG	NAG	AP	VSWC	EC
	(nmol activity g ⁻¹ C h ⁻¹)			(%)	(mS cm ⁻¹)	(nmol activity g ⁻¹ C h ⁻¹)			(%)	(mS cm ⁻¹)
Land-use										
AC	69 \pm 3.0 b	5.20 \pm 0.38 b	27 \pm 1.2 b	47 \pm 1.11 a	3.3 \pm 0.10 a	78 \pm 3.1 b	5.7 \pm 0.73 b	37 \pm 2.8 b	47 \pm 1.01 b	3.2 \pm 0.08 b
PA	171 \pm 15.0 a	31.10 \pm 3.20 a	96 \pm 10.8 a	47 \pm 0.87 a	3.2 \pm 0.08 a	190 \pm 5.8 a	41.0 \pm 3.58 a	113 \pm 5.5 a	49 \pm 0.90 a	3.4 \pm 0.08 a
SRW	55 \pm 5.0 b	4.20 \pm 0.53 b	26 \pm 2.7 b	47 \pm 1.18 a	3.3 \pm 0.11 a	98 \pm 7.8 b	6.1 \pm 0.52 b	33 \pm 1.9 b	50 \pm 0.54 a	3.5 \pm 0.07 a
Salinity										
S0	92 \pm 15.0 a	12.00 \pm 3.00 b	53 \pm 11.4 a	42 \pm 0.89 c	2.8 \pm 0.04 c	114 \pm 13 a	20.0 \pm 5.0 a	71 \pm 11.4 a	46 \pm 1.10 c	3.0 \pm 0.05 c
S1	107 \pm 17.0 a	18.00 \pm 4.60 a	51 \pm 10.2 a	49 \pm 0.40 b	3.3 \pm 0.03 b	134 \pm 16 a	13.0 \pm 2.0 a	61 \pm 9.5 a	49 \pm 0.48 b	3.4 \pm 0.03 b
S2	97 \pm 15.0 a	10.00 \pm 2.30 b	47 \pm 8.9 a	50 \pm 0.11 a	3.7 \pm 0.04 a	118 \pm 10 a	20.0 \pm 5.6 a	51 \pm 7.2 a	51 \pm 0.20 a	3.7 \pm 0.03 a
Groundwater table										
High	127 \pm 14.6 a	16.00 \pm 3.50 a	69 \pm 9.9 a	49 \pm 0.53 a	3.3 \pm 0.07 a	112 \pm 11.5 b	19.0 \pm 4.0 a	69 \pm 4.0 a	51 \pm 0.29 a	3.4 \pm 0.05 a
Low	70 \pm 6.6 b	11.00 \pm 1.90 b	31 \pm 3.4 b	46 \pm 0.99 b	3.2 \pm 0.09 b	132 \pm 9.3 a	16.0 \pm 3.3 b	53 \pm 3.3 b	47 \pm 0.79 b	3.3 \pm 0.07 b

^a Means within a column for land-use, salinity, and groundwater table followed by the same letter are not significantly different ($p > 0.05$) using Tukey HSD.

^b SE = standard error, EEAs = extracellular enzyme activities, BG = β -glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase, EC = electrical conductivity, VSWC = volumetric soil water content, AC = annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm⁻¹, S2 = 12 mS cm⁻¹.

Table 7.2 Analysis of variance (ANOVA) for EEAs, VSWC, and EC under different groundwater salinity and water table levels in soil cores collected from three different land-use practices at two field sites.

Sources of variation	BG		NAG		AP		VSWC		EC		
	df	F stat	p - value	F stat	p - value	F stat	p - value	F stat	p - value	F stat	p - value
Site A											
Land-use	2	147.35	<0.001 ***	80.65	<0.001 ***	148.25	<0.001 ***	0.08	0.930 ns	1.67	0.541 ns
Salinity	2	3.09	0.095 ns	4.93	0.036 *	0.58	0.563 ns	122.74	<0.001 ***	250.62	<0.001 ***
Groundwater table	1	122.70	<0.001 ***	11.61	0.002 **	103.42	<0.001 ***	49.42	<0.001 ***	12.96	<0.001 ***
Site B											
Land-use	2	71.63	<0.001 ***	41.52	<0.001 ***	90.37	<0.001 ***	7.41	0.013 *	24.63	<0.001 ***
Salinity	2	1.78	0.223 ns	0.22	0.809 ns	3.98	0.058 ns	30.27	<0.001 ***	240.51	<0.001 ***
Groundwater table	1	21.53	<0.001 ***	4.15	0.047 *	117.92	<0.001 ***	62.52	<0.001 ***	33.39	<0.001 ***

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ns, is not significantly different ($p > 0.05$).

^b EEAs = extracellular enzyme activities, VSWC = volumetric soil water content, EC = electrical conductivity, BG = β -glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase.

The unconstrained NMDS ordination showed a robust clustering of soil EEAs based on PA land-use practice in both sites, but not for AC and SRW (Figure 7.3A, B, D, and E). The stress values for NMDS from both sites were less than 0.05, which provides an excellent representation of data in a reduced dimension. The NMDS analysis of soil EEAs differed considerably among land-use practices in both sites, suggesting that land-use was a key factor driving variability (Figure 7.3A, B, D, and E). The multivariate permutation analysis of variance (PERMANOVA) test confirmed the significant difference among the land-use practices ($p = 0.001$) in both sites (Table 7.3). The VPA test showed that the land-use practice alone has the greatest contribution to the variation of soil EEAs in both sites (site A = 66.7 %, and site B = 85.9 %) (Figure E-S3).

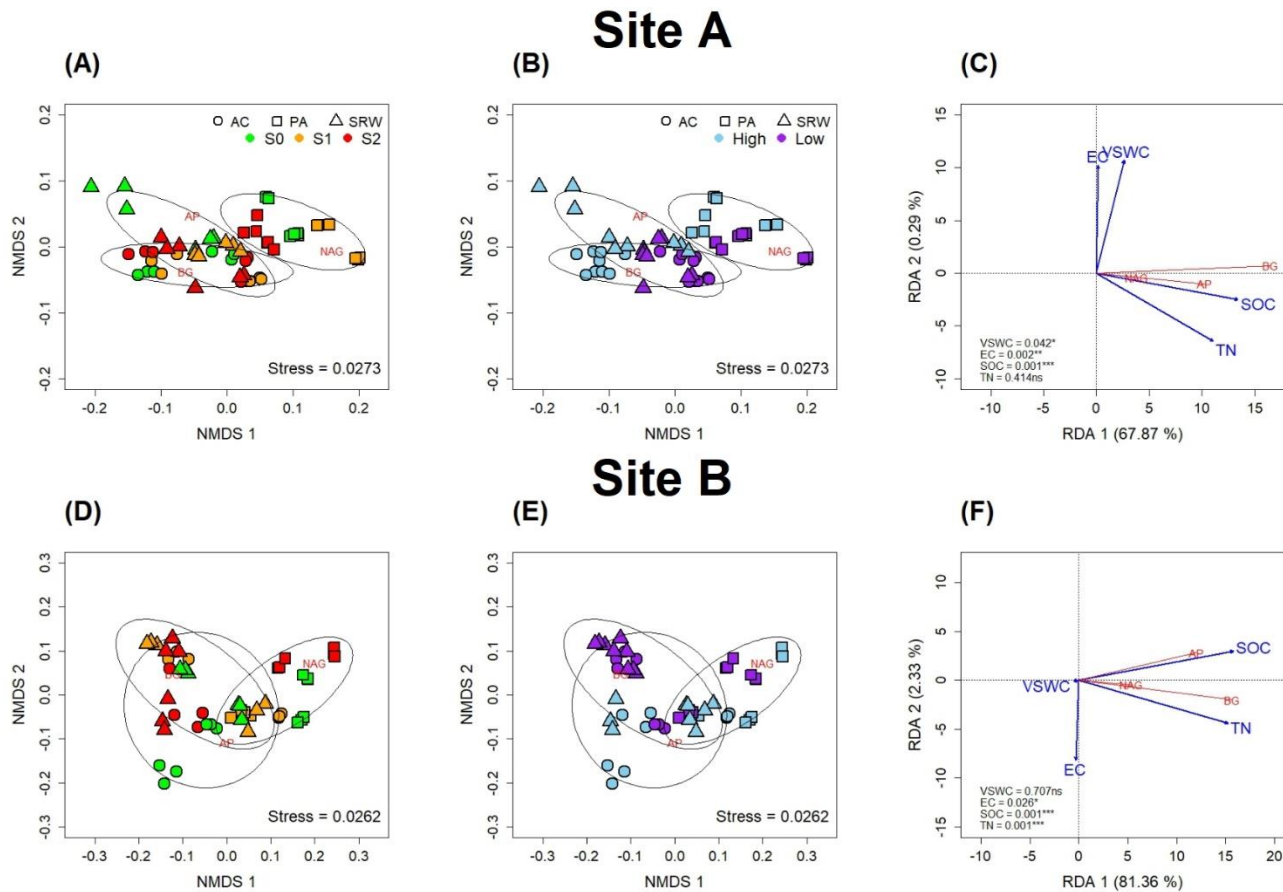


Figure 7.3 Non-metric multidimensional scaling (NMDS) test of soil EEAs visualized with land-use and groundwater salinity, land-use and groundwater table depth, and redundancy analysis (RDA) from site A (A, B, C) and site B (D, E, F).

^a Blue vectors ($r > x$) indicate linear correlations between the ordination and soil physiochemical properties. Directions and lengths of the vectors indicate the strength of correlations between variables. The angles between vectors reflect their correlations (i.e., a vector pair with an angle of 20° have strong positive correlation as $\cos(20) = 0.94$, and with an angle of 90° are uncorrelated as $\cos(90) = 0$).

^b *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively; ns, is not significantly different ($p > 0.05$).

^c AC= annual crop, PA = pasture, SRW = short rotation willow, BG = β -glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase, EC = electrical conductivity, SOC = soil organic carbon, VSWC = volumetric soil water content, TN = total nitrogen, S0 = control, S1 = 6 mS cm^{-1} , S2 = 12 mS cm^{-1} .

Table 7.3 Permutation multivariate analysis of variance (PERMANOVA) test for EEAs under different groundwater salinity and water table levels in soil cores collected from three different land-use practices at two field sites.

Sources of variation	Site A				Site B		
	df	Pseudo- <i>F</i>	R ²	Pr (> <i>F</i>)	Pseudo- <i>F</i>	R ²	Pr (> <i>F</i>)
Land-use	2	28.13	0.53	0.001 ***	19.07	0.43	0.001 ***
Salinity	2	2.85	0.10	0.061 ns	1.17	0.04	0.317 ns
Groundwater table	1	11.27	0.18	0.001 ***	10.23	0.16	0.001 ***

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$ level of significance, respectively; ns, is not significantly different ($p > 0.05$).

^b EEAs = extracellular enzyme activities.

7.5.1.2 Differences in Soil EEAs Under Groundwater Salinity Levels

The univariate ANOVA test showed no significant difference ($p > 0.05$) in soil EEAs in both sites except for NAG ($p = 0.036$) activity in site A (Table 7.1 and 7.2). Similarly, both the NMDS and PERMANOVA tests did not show any distinct grouping or significant difference ($p > 0.05$) among the groundwater salinity treatments in both sites (Figure 7.3A and D; Table 7.3), suggesting the soil EEAs were not affected by the imposed elevated salinity levels.

Groundwater salinity only accounted for 0.4 % and 2.7 % contribution to the variation of soil EEAs in site A and site B, respectively (Figure E-S3).

7.5.1.3 Differences in Soil EEAs Under Groundwater Table Depths

Overall, significantly higher ($p < 0.05$) soil EEAs were observed under higher (at week 1 = 2 cm) vs. lower groundwater table depth (at week 10 = 29 cm), suggesting that the groundwater table depth was an influencing factor driving the variability in soil EEAs (Table 7.2). Higher mean values for all EEAs were observed under the high water table, except for soil BG activity in site B, where the response to the water table was the opposite (Table 7.1), indicating that the effect of water table drawdown affected each of the enzymes differently.

The NMDS ordination showed a notable group difference between high and low groundwater table treatments (Figure 7.3B and E). The PERMANOVA test indicated that the soil EEAs were significantly affected ($p = 0.001$) by groundwater table depths in both sites (Table 7.3). The VPA showed that the groundwater table depth contributed 20.9 % in site A and 3.4 % in site B, suggesting the water table contributed to the variation of soil EEAs after the land-use practices (Figure E-S3).

7.5.2 Physiochemical Properties of Experimental Soil

The physiochemical properties of soils used for the microcosm experiment are presented in Table 4. No significant differences were observed in soil physiochemical properties among land-use practices, and between sites except SOC, TN, SO_4^{2-} content (ANOVA results are not shown here). The SOC and TN were significantly ($p < 0.05$) higher in soils from PA compared to other land-use practices in the order of $\text{PA} > \text{SRW} = \text{AC}$ in both sites (Table 7.4). However, no significant difference ($p > 0.05$) were found in SOC and TN content between sites. The SO_4^{2-} content was approximately eight times higher in soils from site B than site A (Table E-S1). In the order of land-use practices, the soil SO_4^{2-} contents were $\text{SRW} > \text{PA} = \text{AC}$ in site A, and $\text{SRW} = \text{AC} > \text{PA}$ in site B, suggesting no consistent land-use patterns were observed between sites.

Table 7.4 Physiochemical properties of soils used for microcosm study

Site	Land-use practice	pH	EC (mS cm ⁻¹)	Soil texture	Clay (%)	Bulk density (g cm ⁻³)	CEC (cmol _c kg ⁻¹)	TSC (%)	SOC (%)	WEOC (mg C kg ⁻¹)	TN (%)	NH ₄ ⁺ -N (mg kg ⁻¹)	NO ₃ ⁻ -N (mg kg ⁻¹)	WEON (mg N kg ⁻¹)	C/N ratio	PO ₄ ³⁻ -P (mg kg ⁻¹)	SO ₄ ²⁻ -S (mg kg ⁻¹)
Site A	AC	8.4	1.4	CL	33.0	1.4	36.7	4.1	2.2	3.5	0.2	4.3	25.1	8.1	11.3	24.9	82.1
	PA	7.8	0.6	CL	34.0	1.3	43.9	3.4	2.9	5.2	0.3	6.7	7.0	3.7	11.6	28.5	98.2
	SRW	8.0	1.9	SCL	27.0	1.3	37.4	3.7	2.2	5.2	0.2	6.0	12.5	5.1	10.8	21.3	641.5
Site B	AC	7.8	1.0	CL	33.0	1.3	48.7	3.1	2.5	5.9	0.2	6.0	17.0	4.5	9.8	30.0	2353.2
	PA	8.0	2.6	CL	32.0	1.2	42.5	4.9	2.7	4.6	0.3	5.2	33.4	8.1	10.8	31.2	274.4
	SRW	7.8	2.8	CL	30.0	1.4	45.9	2.8	2.4	6.2	0.3	6.3	13.9	4.3	9.4	37.8	3496.2

^a AC = annual crop, PA = pasture, SRW = short rotation willow, EC = electrical conductivity, CL = clay loam, SCL = sandy clay loam, CEC = cation exchange capacity, TSC = total soil carbon, SOC = soil organic carbon, WEOC = water extractable organic carbon, TN = total nitrogen, WEON = water extractable organic nitrogen, C/N ratio = carbon and nitrogen ratio.

7.5.2.1 Relationships of EEAs with Soil Physiochemical Properties

The relationships between soil EEAs (BG, NAG, and AP) with soil clay content, SOC, and TN were significant ($p < 0.05$) and positive, whereas for bulk density it was significant ($p < 0.05$) and negative (Figure 7.4). The soil EEAs were positively correlated with VSWC, EC (due to the salinity treatment), CEC, NH_4^+ , NO_3^- , C/N ratio, and PO_4^{3-} , whereas negatively correlated with pH, background EC, WEOC, WEON, and SO_4^{2-} , however, not significantly ($p > 0.05$) (Figure 7.4).

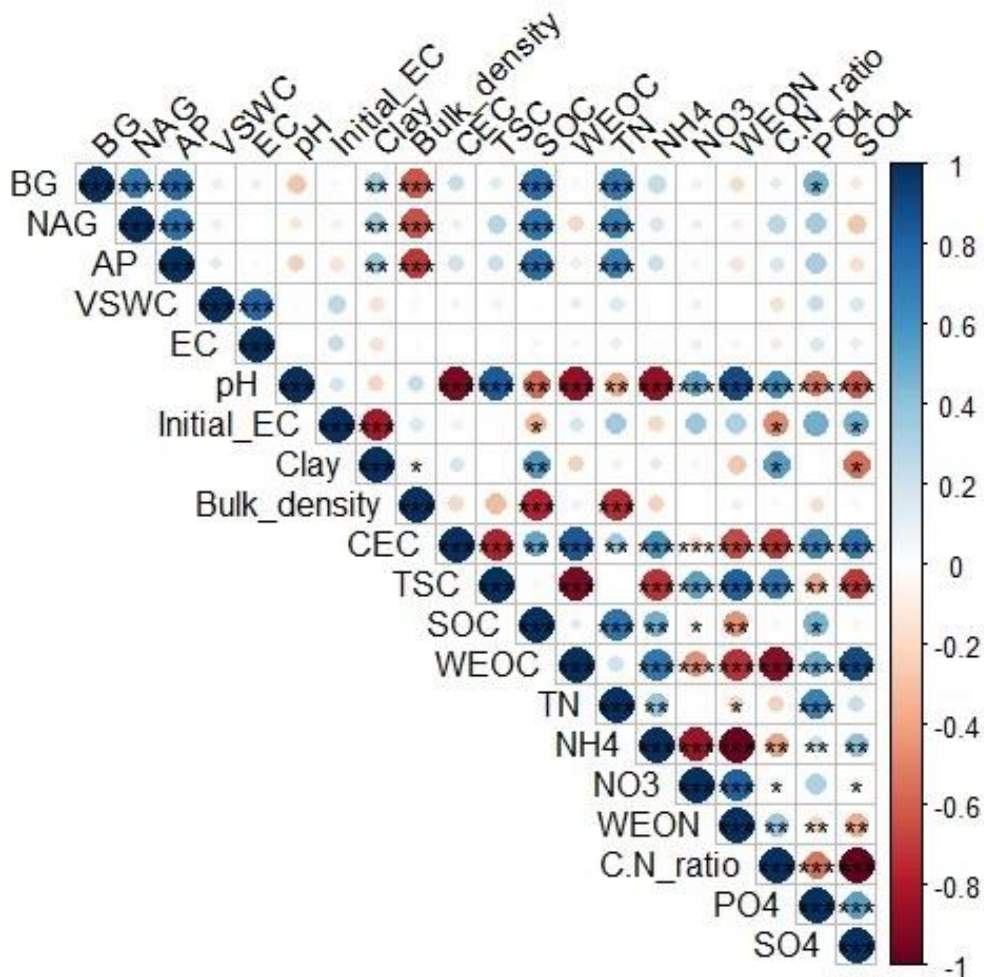


Figure 7.4 Spearman rank-order correlations among BG, NAG, AP and with initial soil physiochemical properties.

^a Negative correlations are depicted in red and positive correlations in blue. Increasing correlation strength is indicated by increasing circle diameter and deeper color.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$ level of significance, respectively, and remaining are not significant ($p > 0.05$).

^c BG = β -glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase, VSWC = volumetric soil water content, EC = electrical conductivity, CEC = cation exchange capacity, TSC = total soil carbon, SOC = soil organic carbon, WEOC = water extractable organic carbon, TN = total nitrogen, WEON = water extractable organic nitrogen, C/N ratio = carbon and nitrogen ratio.

7.5.2.2 Redundancy Analysis (RDA) Between Soil Physiochemical Properties and EEAs

Redundancy analysis (RDA) was performed to explore the relationship of soil physiochemical properties with EEAs shown (Figure 7.3C and F). The first two component axes explained 67.87 % and 0.29 % of site A (Figure 7.3C), and 81.36 % and 2.33 % (Figure 7.3F) of site B of soil EEAs, respectively. The vector lines of SOC, VSWC, EC from site A, and SOC, TN, EC from site B were statistically significant ($p < 0.05$), showing that SOC played a large role in explaining soil EEAs in both sites. Significant positive correlations ($p < 0.05$) observed between SOC and soil EEAs in both sites A and B (Figure 7.3C, and F). The relationship between TN and soil EEAs were positive and significant ($p < 0.05$) in site B, but not significant ($p > 0.05$) in site A.

7.5.2.3 Relationships Amongst Manipulated Water Table and Salinity with VSWC and EC Measured During the Microcosm Experiment

Groundwater salinity manipulation (Paragraph 6.5.2.4, page 133) resulted in a statistically significant difference ($p < 0.05$) in soil EC among different salinity treatment levels (in S1 and S2 compared to control) in both sites (Table 7.1 and 7.2). Similarly, water table manipulation resulted in a significant difference ($p < 0.05$) in observed VSWC between high and low groundwater table depths in both sites. We did not find any significant difference ($p > 0.05$) in VSWC or EC among land-use practices from site A ($p > 0.05$); however, there was a significant difference ($p < 0.05$) in site B because of groundwater salinity and water table manipulation (Table 7.1 and 7.2). We also observed a significant ($p < 0.05$) positive relationship between soil EC and VSWC in both sites (Figure 7.4 and Figure E-S5) during the incubation experiment.

7.6 Discussion

7.6.1 Land-use, Groundwater Salinity, and Water Table Effects on Soil EEAs

7.6.1.1 Land-use

In this experiment, the highest soil EEAs were observed in soils from PA land-use practice compared to AC and SRW, suggesting a likely relationship to higher microbial activity in

grassland soil. Soils from PA land-use practice had the highest SOC, TN, and overall $\text{PO}_4^{3-}\text{-P}$ contents, and reflected the highest soil EEAs in our study, perhaps due to the faster SOC alteration and balanced substrate availability in PA soils that differ from other land-use practices. We observed a clear grouping for PA from the NMDS ordination plot in both sites and significant linear relationships between EEAs with SOC and TN from RDA analysis. Wallenius et al. (2011) observed decreased EEAs activity in the order of forest organic layer \approx forest mineral layer > meadow grassland > crop field in a plot-scale study, which they related to SOC and TN. Likewise, microbial community structure is highly specific to land-use practices, and SOM content is the primary reason for the variation of both structural and functional properties of soil microorganisms (Wallenius et al., 2011). In a regional-scale study, Cenini et al. (2016) observed a positive relationship between SOC content and BG activity, and L-leucine amino-peptidase (LAP) + NAG activity with a soil N content of grassland soils. Kuramae et al. (2012) found that soil factors (SOC, TN, $\text{PO}_4^{3-}\text{-P}$, and pH) had a more robust impact on soil bacteria than land-use practices. At the metabolic scale, the proportionality constant that connects C:N:P stoichiometry of organic matter and enzymatic activities controls the elasticity of extracellular enzymatic reactions (Sinsabaugh et al., 2014). However, different land-uses have resulted from the difference in inherent soil characteristics over large geographic areas predictably have more influence on soil properties over land-use practices itself.

In general, the type of land-use practice can influence SOC content and thus affect soil EEAs through the breakdown process of SOM and the loss of labile organic carbon (Trasar-Cepeda et al., 2008). Acosta-Martínez et al. (2003a) found lower EEAs in continuous cropland than reserve grassland and native rangeland, and a strong relationship with SOC and TN. In a field experiment, Bandick & Dick (1999) found higher EEAs in the continuous grass field than in cultivated fields except for α - and β -glucosidase. In a global-scale meta-analysis in soils from 40 ecosystems, Sinsabaugh et al. (2008) observed increased activities of β -glucosidase, N-acetyl

glucosaminidase, and phosphatase with increased SOM content. This indicated that hydrolyzing capability of the SOM depends on enzymatic stoichiometry, which links the elemental stoichiometry of microbial biomass and detrital organic matter to microbial nutrient assimilation and growth (Sinsabaugh & Follstad Shah, 2012).

Soil EEAs were not significantly different between AC and SRW, and significantly lower compared to PA land-use practices from both sites in our experiment, most probably because of observed non-distinguishable variabilities in SOC and TN content. Moreover, in our field experiment, we did not observe any significant differences in SOC quantity or quality between AC and SRW. However, several studies on SRW suggested conflicting results in terms of SOC accumulation compared to other land-use practices. For example, the topsoil SOC increased under SRW plantation, on former agricultural land compared to conventional AC (Dimitriou et al., 2012b) or to adjoining agricultural fields (Lafleur et al., 2015); it did not increase significantly compared to grassland (Harris et al., 2017) or after re-conversion to arable land (Toenshoff et al., 2013); and no significant change after conversion to SRW compared to no-till alfalfa field and buckwheat field (Lockwell et al., 2012). Three years after the conversion of arable land to the SRW, fungal abundance was promoted; however, soil alkaline phosphatase, cellobiohydrolase, and phenoloxidase were higher than AC soils but lower than forest and PA soils (Stauffer et al., 2014).

Within the context of AC systems, tillage practices can further impact different biological attributes including soil microbial biomass, soil organic C, and N. We used intact soil cores from an AC area under conventional tillage, which might be the reason for relatively lower SOC content, and lower EEAs compared to pasture land-use. Gupta & Germida (1988) compared soil EEAs between cultivated and adjacent native PA soil in Canadian Prairie and found that tillage suppressed 49% of phosphatase and 65% of arylsulphatase activity. In semi-arid agricultural land of west Texas, Acosta-Martínez et al. (2003b) observed increased soil β -glucosidase, β -

glucosaminidase, alkaline phosphatase, and arylsulfatase activities under general crop rotation and conservation tillage compared to a single crop and conventional tillage. This suggests that the production of EEs and C turnover rapidly occur in particulate organic matter fractions, thus increased by physical disruption of soil structure associated with tillage (Allison & Jastrow, 2006).

7.6.1.2 Salinity

We did not find any significant effects of groundwater salinity treatments in our experiment. Previous studies have reported that salinity can suppress soil EEAs, and all enzymes are not equally sensitive to the salinity (Pan et al., 2013). For example, García & Hernández (1996) observed a higher degree of hydrolase (β -glucosidase and phosphatase) inhibition by salinity compared to oxidoreductases (dehydrogenase and catalase). The reduction of EEAs in saline soil is primarily due to the lower microbial biomass, osmotic potential, and specific ion effects of the salts present (Rath & Rousk, 2015). Shi et al. (2019) found that the addition of organic amendments can increase microbial biomass and EEAs in saline-alkaline soil, suggesting that SOM can improve SOC and nutrient conditions for microbial activity due to higher substrate availability under saline conditions. Likewise, the addition of readily decomposable substrate can improve microbial salt tolerance (Mavi & Marschner, 2013, Wong et al., 2008). We observed relatively high enzyme activity in site B, despite a slightly higher mean background soil salinity than site A; however, none of our sites can be classified as saline soil as the average EC was $< 4 \text{ mS cm}^{-1}$.

7.6.1.3 Water Table

Soil EEAs were significantly reduced by lowered groundwater table (i.e., deeper GWT depth) compared to shallower water tables, except for BG in site B, which was the opposite, suggesting that the lowered groundwater table can lead to transitory drought stress for SOM decomposers and reduced enzyme production. In a mesocosm experiment with peat soil, Wiedermann et al.

(2017) observed a similar result with the greatest groundwater table drawdown effect on phosphatase enzyme. In a mesocosm experiment with declining water table depth from 0 to 20 cm in Alpine wetland soil Wang et al. (2017b) found a significant decrease in β -glucosidase and phenol oxidase activities. In a field-based water table drawdown experiment with peat soil, Freeman et al. (1996) found that β -glucosidase and phosphatase activities were raised between 31 to 67 % with a water table drawdown without a corresponding increase in microbial respiration. Therefore, the authors suggested a direct stimulation of existing enzymes rather than stimulation of new enzyme synthesis as the cause. Henry (2012) proposed three hypothetical models to predict the variation of EEAs with soil moisture gradients (poorly-drained, a well-drained, arid) that suggested in a poorly-drained water-saturated (anaerobic) soil; initial water table drawdown can stimulate enzyme activity, whereas further drying can reduce EEAs through the restriction of water. Perhaps a similar response explained the quadratic response of EEAs to water table drawdown and consequent changes in moisture status in our experiment. Three roles of water have been suggested: 1) as a resource to maintain water potential, 2) as a solvent, and 3) as a transport medium. Thus depending on conditions, water as a resource might be the least important regulator of soil biogeochemical processes (Schimel, 2018). Roles as both a solvent and a transport medium are critical as the majority of organic substances are water-soluble, and the movement of chemicals in solution from sources to microorganisms regulate metabolism (Schimel & Schaeffer, 2012).

The effects of physiochemical properties of *in situ* soil are not discrete. Even under ideal conditions, one individual factor seldom solely drives soil biogeochemical processes because of interactions among soil properties. However, one individual factor might dominate the soil's ecological processes, such as SOM decomposition. Specific management practices within agroecosystems, such as the addition of plant residues, can also interact with soil moisture and affect EEA. Geisseler et al. (2011) found that the addition of crop residues in a combination of

higher soil moisture likely increase protease, β -glucosidase, glucosaminidase, and exocellulase activities, whereas EEAs were less affected by higher soil moisture when no residues were added. As a result, it was hypothesized that the presence of the substrate potential of EEAs might be decoupled from microbial biomass size and respiration under dry moisture conditions. The total soil water potential is the result of both osmotic and matric potential in soil (Kakumanu & Williams, 2014). Likewise, low soil water content with lower matric potential and low osmotic potential due to salinity in the soil is typical in semi-arid regions (Chowdhury et al., 2011). However, it is difficult to differentiate the distinct effect of water potential and osmotic potential (Chowdhury et al., 2011) as microbes have a similar mechanism to react to drought and for high salt concentrations in soil (Schimel et al., 2007).

7.6.2 Soil Physiochemical Properties and EEAs

The relationships were variable among all EEAs with physiochemical properties of the experimental soils. Soil EEAs were positively correlated with clay content, even though there was no significant variation among land-use practices or sites. Clay content has little explanatory power to reflect the SOM cycling compared to other physiochemical properties in the soil (Rasmussen et al., 2018). A significant negative correlation was observed between soil EEAs with bulk density, and a positive correlation with initial SOC and TN content. Similarly, findings of Dick et al. (1988) and Xie et al. (2017) indicated a direct and/or indirect links of these soil properties with microbial functions for continuing the soil enzymatic activities (Allison et al., 2011, Sinsabaugh et al., 2008). Soil pH can affect soil nutrient availability, decomposition of SOC, and activity and diversity of microorganisms that are involved in soil biochemical reactions, including soil EEAs (Dick et al., 2000). We observed a minor non-significant variation in initial experimental soil texture and pH. According to Sinsabaugh et al. (2008), soil hydrolytic enzymes are more stable under conditions of small pH variation compared to oxidative soil EEAs. None of the remaining soil properties were correlated significantly with soil EEAs except

soil $\text{NH}_4^+\text{-N}$ and $\text{PO}_4^{3-}\text{-P}$ with BG activity. The microbial economic theory suggests that microbes produce extracellular enzymes that target essential macronutrients only if they are deficient (Allison et al., 2011). It has been suggested that soil physiochemical properties, as well as microorganisms, are highly heterogeneous, which may vary significantly over temporal and spatial scales (Baker et al., 2009). However, contrasting land-use practices within a single PPR wetland system is more likely to influence soil physiochemical properties, especially C, N, and P, from similar soil characteristics.

7.7 Conclusions

The results of this study suggest that land-use practice had the most significant impact on soil EEAs. Significantly higher EEAs in soils from PA attributable to the higher SOC turnover from the past land-use practices, while no significant difference observed between AC and SRW consistent with non-distinguishable variabilities in background SOC and TN content of our experimental soil as well as in the field study. We found a significant effect of groundwater drawdown on soil EEAs. However, no significant effects were distinguished for salinity treatments recommended that EEAs (BG, NAG, and AP) in soil feasibly respond to the differences in resource availability attributable to land-use and metabolic limitation as a result of interacting effects of shifting groundwater tables and salinity.

The SOC content was the primary parameter that influences biological activity and, therefore, was highly correlated with all soil EEAs. Differences in SOC, TN, and nutrients possibly induced changes in soil microbial activities and could mediate the variation in EEAs among the soils from different land-use practices. However, using *in-situ* enzyme activity as an indicator of land-use practices can be challenging as they can vary between different soil and different enzymes as well as at a spatial scale. Therefore, interrelating effects of land-use practices in combination with fluctuating groundwater table and salinity on the structure and functioning of the soil

microbial community in a field setting is required to enhance our understanding of the PPR wetland soils.

8 SUMMARY, SYNTHESIS, AND FUTURE RESEARCH DIRECTION

8.1 Summary of Findings

The overall objective of this research was to address the impact of short rotation willow (SRW) establishment in the riparian zones of PPR wetland systems compared to adjacent pasture (PA) and annual crop (AC) on wetland soil hydrology and salinity, nutrient contents, organic carbon sequestration, greenhouse gas (GHG) emissions, and extracellular enzyme activities (EEAs) involved in biogeochemical cycling (Figure 8.1).

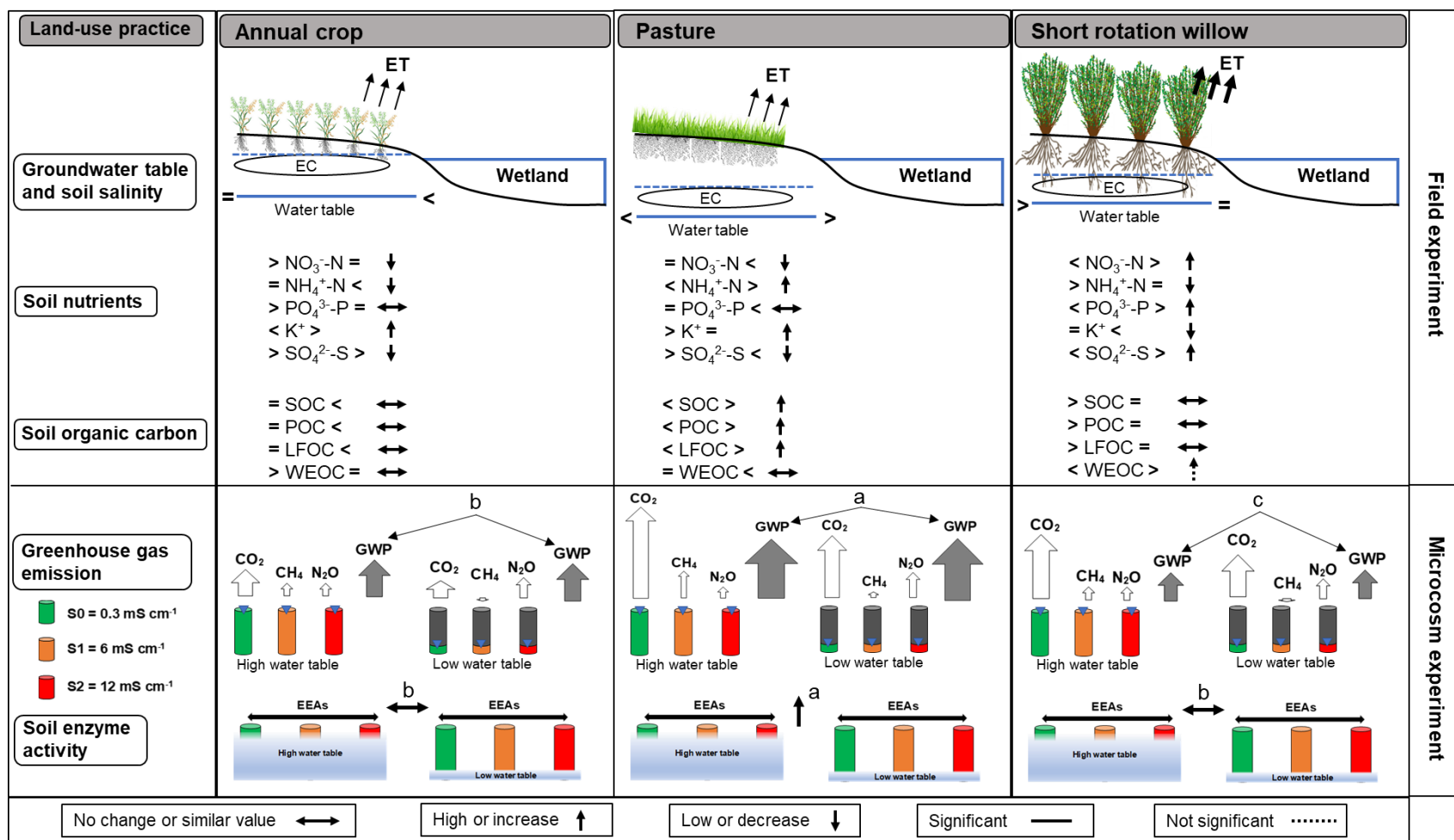


Figure 8.1 Summary of the status and relations among soil hydrology, salinity, nutrients, organic carbon, greenhouse gas emissions, and enzyme activities under different land-use practices of the PPR wetland system.

^a '>' indicates higher than; '<' indicates lower than; '=' indicates equal values.

^b Values within land-use practices followed by the same small letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c EC = electrical conductivity, ET = evapotranspiration, EEAs = extracellular enzyme activities, GWP = global warming potential, LFOC = light fraction organic carbon, POC = particulate organic carbon, SOC = soil organic carbon, WEOC = water extractable organic carbon.

The depth to groundwater table (GWT) was significantly affected by land-use practices in site B but not in site A, suggesting that the variations were not consistent across land-use practices between sites. Groundwater EC was significantly impacted by land-use practices but also did not follow consistent land-use patterns in both sites. Fluctuations in GWT depth were intense between June to August while the precipitation was higher, whereas lesser during May and September due to the decreased precipitation. However, the depth to GWT among the months did not differ significantly. A significant change in the GWT depth (i.e., lower depth to GWT from the surface) was observed due to higher precipitation events throughout the wet year (i.e., during 2014) compared to the dry (i.e., during 2015) across all land-use practices from both sites. The soil EC varied significantly among land-use practices; however, variation between depths, years, and months were not significant in both sites. Positive correlations were observed between depth to GWT vs. groundwater, and soil EC indicated that increased depth to GWT, perhaps, resulted in higher salinity and accumulation of salts under different land-use practices. Field observation exhibited a reducing tendency in soil EC with increasing SRW biomass at both depths (i.e., 0-30 and 30-60 cm) from both sites. However, the reverse situation might also be possible, therefore the underlying soil physical and chemical differences in where they were planted may reflect the obtained results. The biomass production from SRW was lower in site B (two-fold) compared to site A, possibly due to the higher background soil EC from site B; however, no significant difference was observed between sites.

Compared to adjacent land-use practices, soil $\text{NH}_4^+\text{-N}$ and K^+ contents were lower under SRW, which may reflect a higher demand in SRW plantation. In contrast, relatively high concentrations of groundwater and soil $\text{NO}_3^-\text{-N}$ were observed under SRW in both sites, indicated a low removal efficiency by SRW, possibly due to lower biomass production or the plantation discrimination among landform positions. Soil $\text{PO}_4^{3-}\text{-P}$ content was higher under SRW plantation, possibly due to an improved solubility from the root and microbial exudates, i.e.,

organic acids and favorable soil moisture conditions. Higher soil SO_4^{2-} -S concentration at the lower soil depth (30-60 cm) suggested the presence of high sulfate-rich salts in the subsurface of semi-arid glaciated PPR soil.

After the first rotation (3-year cycle), no significant increase in SOC was observed under SRW plantation compared to other land-use practices. The PA land-use practice had higher SOC, POC, and LFOC contents compared to SRW and AC, whereas no significant difference was observed between SRW and AC. There was a slight increase in the WEOC concentration under SRW than other land-use practices but not statistically significant. The upper 15 cm soil contained significantly higher total SOC compared to the lower soil layer (15-30 cm), confirmed that the soil depth influences the SOC accumulation. The abundance of FTIR bands of all SOC chemical functional groups were higher at lower soil depth (15-30 cm), indicated altered spectral properties with depth. In both sites, the RC:LC ratio of phenolic, amide, and carboxylic to polysaccharides were relatively lower in surface soils, whereas aromatic to polysaccharides was higher and followed a consistent land-use pattern. Higher alkyl-C to O-alkyl-C ratio in the subsoil suggested a higher degree of decomposition and better stability of SOC.

In the microcosm study, soils from contrasting land-use practices significantly affected the GHG emissions. The PA soils produced significantly higher GHG (CO_2 , CH_4 , and N_2O) emissions compared to AC and SRW, in the order of $\text{PA} > \text{AC} = \text{SRW}$. Changes in soil properties, mainly the higher background organic C and N content maintained by the PA land-use practices in the past, seemingly shaped the observed difference in soil GHG emissions. There were observed significant increases in N_2O emission and a cut in CO_2 and CH_4 emissions with raised groundwater salinity. Depth to GWT significantly affected the GHG emissions across the soils from different land-use practices from both sites. The CH_4 emission was comparatively minor and decreased with the reduction in volumetric soil water content (VSWC) in the core soil linked with a deeper water table. The N_2O emission was relatively small and observed highest under

an intermediate water table position from saturation to the declined depth to GWT (at 26 cm depth), perhaps due to suitable VSWC for denitrification. The CO₂ emission was variable, i.e., increased with as the water table was lowered up to week four, but then started to decrease as the depth to GWT declined further. The SRW had a lower global warming potential (GWP) compared to AC and PA, suggesting that it can be a potentially promising land-use practice in the PPR.

Soil extracellular enzyme activities (EEAs) were significantly impacted by land-use practice and groundwater table depths but not significantly by salinity. The land-use effects on the soil EEAs were in the order of PA > AC = SRW. The PA soil had significantly higher soil EEAs suggesting higher SOC turnover owing to the past land-use practice. No significant difference was observed between AC and SRW, perhaps because of non-distinguishable variabilities in background SOC and TN contents of the experimental soil from both sites. Soil EEAs significantly decreased with greater depth to GWT, which indicated transitory drought stress to the SOM decomposers.

8.2 Synthesis: Effects of Land-use Practices on Wetland Soil Hydrology, Salinity, and Biogeochemistry in the PPR

The findings of this study improve our understanding of land-use induced hydrological alteration and its implication on soil salinity and biogeochemical processes in the PPR wetland systems. In this experiment, land-use practices impacted the GWT depth significantly in site B but not significantly in site A and did not follow comparable land-use patterns in both sites. Hence, it can be said that the land-use practice was not the sole factor controlling the GWT, but other overriding factors such as climatic and site-specific factors, e.g., landscape position and soil characteristics may have an influence. Even though with the potential predicted high water consumption, this research demonstrated that SRW plantation in the field level did not have much effect on the GWT in the riparian zones of PPR wetland systems. This situation was

possibly attributable to the early stage of SRW plantation (i.e., first rotation cycle) compared to the previous studies reported (please see Chapter 3) that observed a declined depth to GWT under agroforestry after several years. On the other hand, the permanent PA land-use is comparatively mature and dense cover with deep-rooted perennial grasses that can transpire at higher rates (van der Kamp et al., 2003). Compared to PA, the observed increase in water table fluctuations under AC was mainly because of tillage operation that can modify groundwater hydrology of wetlands within the PPR agroecosystems (Euliss & Mushet, 1996).

It is evident from the literature that the phreatophytic SRW plantation typically has a higher water consumption rate through high ET (Dimitriou et al., 2009b) and connected with raised groundwater and soil salinity (Jobbágy & Jackson, 2004, 2007, Noretto et al., 2013). Both in terms of depth to GWT, groundwater and soil salinity, there were no consistent land-use patterns observed between sites, indicates other underlying soil factors might override the observed patterns of variability. Nevertheless, the establishment SRW could be used to reduce and manage salinity at the shallow soil (0-60 cm) layer, as the soil EC showed a decreasing trend with higher SRW biomass. Yet, the conflicting situation might be possible, i.e., the SRW biomass production was lower under higher background soil salinity. However, the most cost-effective and environmentally sustainable option to manage salts and excess water in the discharge area in the PPR wetland system could be the 'biodrainage' via plantation of SRW (Heuperman et al., 2002, Minhas & Dagar, 2016).

The SRW land-use practice likely has higher $\text{NH}_4^+\text{-N}$ and K^+ demand, which may result in the lower concentration N and K^+ under improved growth rate and biomass production. Previous studies from several authors have frequently categorized the SRW plants as 'nutrient demanding' because of high growth rate and biomass production (Cornelissen et al., 1997, Ens et al., 2013a, Simon et al., 1990); thus some of the soil nutrients feasibly lost from the soil by harvested biomass (Adegbidi et al., 2001). However, research demonstrated that the SRW (i.e.,

Salix Spp.), compared to AC, can produce higher biomass using a fraction of nutrients, as well as high nutrient recycling capability through leaf litter (Hangs et al., 2014a, Hangs et al., 2014b). Similarly, this research demonstrated that SRW land-use practice is perhaps low nutrient-demanding because of the survival and reasonable growth rate without any fertilizer application and moderate soil salinity.

At this point, SRW plantation has not yet accomplished the maximum potential growth rate and biomass production in its first rotation cycle. However, the SRW growth rate will likely reach full potential in the following rotation, and the root systems will continue to grow and mature, which might further increase the demand for nutrients uptake and use (Amichev et al., 2014b, Dimitriou & Mola-Yudego, 2017, Dimitriou et al., 2012a). Therefore, the higher groundwater and soil NO₃-N concentration under SRW may be due to lower removal efficiency during the first rotation with lower biomass production, the plantation discrepancy in landscape position (Johnson et al., 2013), moreover higher leaching from the adjacent agricultural non-point source (Richardson et al., 1994). Therefore, it could pose the possible risks of further nitrous oxide emission through denitrification from the soil (Aronsson & Bergström, 2001, Dimitriou et al., 2012a).

The SRW land-use practice has a higher potential to sequester SOC (Lemus & Lal, 2005, Nair et al., 2010, Oelbermann et al., 2004), but it requires a long period of over 20 years (Pacaldo et al., 2013). Furthermore, the land-use practice change to SRW from arable crops likely to neutral or net increase (Dimitriou et al., 2012b, Qin et al., 2016), whereas from grassland to SRW can generally be neutral or lower in SOC (Harris et al., 2017, Harris et al., 2015). Evidently, in a short period (i.e., 3-year rotation cycle), this study indicates that the SRW plantation has little impact on SOC content at these two sites. However, in a future rotation, it could be a considerable sink of SOC in the riparian wetland soils in the PPR compared to AC and PA land-use practices. Furthermore, a higher value of the alkyl-C to O-alkyl-C ratio under SRW land-use

practice suggested a higher degree of decomposition and better stability of SOC in the subsoil, thus improved sequestration potential. Hence, the fast-growing high biomass producing SRW could increase carbon capture and storage rates on degraded and abandoned marginal riparian lands, and at least not have any negative impact on the SOC storage.

This research showed significantly high GHG emissions and EEAs in soils from PA mainly related to higher SOC turnover because of the effects of the past land-use practice on increasing SOC and microbial activity. While there was no significant difference observed between AC and SRW in terms of GHG emissions and EEAs, possibly because of no difference in background SOC and TN contents of the experimental soil from both sites. Moreover, the land-use patterns of SOC content from the field experiment followed similar patterns with GHG emissions and EEAs, which was in the order of PA > AC = SRW. It has been suggested that the change in land-use practice can alter the dynamics of SOC and affect GHG emissions (Kooch et al., 2016). Agroforestry, which has the potential to increase SOC, can mitigate and increase the sink potential of the soil GHG compared to cropland (Mutuo et al., 2005). Consequently, establishing perennial agroforestry systems such as SRW in the riparian zones of PPR wetlands could deliver GHG mitigation benefits. The soils from SRW land-use practice from the microcosm experiments have significantly lower GWP compared to AC and PA. Moreover, the future sequestration potential under SRW can provide multiple benefits of improved soil health and can act as a mitigation pathway to reduce global warming. Furthermore, the establishment of SRW could reduce the per-capita GHG emission in the Saskatchewan, which is currently three times higher than the national average in Canada (Government of Canada, 2019).

The SRW land-use practice in the riparian zones of the PPR wetlands can impact the shallow groundwater hydrology and salinity because of vegetation linked water loss via evapotranspiration (Hayashi et al., 2016). Hence, the limitation of soil water and raised salinity could become critical for soil EEAs (Sinsabaugh et al., 2008). Furthermore, land-use practices

that supply improved levels of SOC via plant residues can significantly increase the soil EEAs (Bandick & Dick, 1999). In this study, soil EEAs and GHG emissions were reduced significantly under a declined groundwater table, indicated that the lowered water table could lead to a short-term water shortage and thus affect the SOM degradation (Schimel, 2018, Schimel & Schaeffer, 2012). The decline in the groundwater table in combination with elevated salinity under different land-use practices could also boost GHG emissions particularly the N₂O, which may lead to positive feedbacks to climate change. The results from this research indicated that the shallow groundwater salinity in the PPR under contrasting land-use would have essential impacts on the increased GHG emissions. However, the magnitude and direction are highly dependent on wetland soil hydrology (Dunmola et al., 2010). Also, soil moisture conditions (aerobic vs. anaerobic) can substantially affect the C, N, and P mineralization (Bridgham et al., 1998).

Wetland soils are also a vital supplier of SOM, hence fuels soil biogeochemical processes and permit denitrification (Bastviken et al., 2005) and methanogenesis (Bridgham et al., 2006). The research suggested that wetland soil can be a suitable environment for the sequestration of CO₂, yet they also are natural sources of GHG emissions (Mitsch & Gosselink, 2015), depending on soil hydrology and the environmental conditions (Euliss et al., 2006, Kayranli et al., 2009).

Soil organic matter decomposition mediated by EEs depends on various interconnecting factors. Soil EEAs, particularly hydrolases, are primary mediators in SOM degradation and nutrient cycling (Ondrasek et al., 2019). The SOC turnover generally was driven by EEAs (Feng et al., 2018), whereas soil moisture and nutrients such as N and P are the crucial edaphic factors for influencing soil enzymes (Wang et al., 2018). However, at the ecosystem level, a single factor might not direct the entire soil biogeochemical processes. Extreme variation in any factor may alter the soil ecological processes overall.

Nonetheless, this study highlights the potential to benefit from SRW plantation in the fallow marginal riparian zones – lands that are not suitable for crop production due to the higher salinity or unsuitable for agricultural crop production – within the PPR agroecosystem. Moreover, plantation of well-adapted salinity tolerant SRW cultivars in the riparian zone (e.g., degraded marginal lands that otherwise may produce very little) can contribute to the production of harvestable biomass that is capable of supply bioenergy feedstock sustainably (Amichev et al., 2014b). Therefore, the plantation of SRW (*Salix* Spp.) is recommended as a part of the best management practices on the degraded marginal riparian land in the riparian zones of the PPR agroecosystems.

8.3 Future Research Direction

In this dissertation, the presented research provides a comprehensive assessment of the effects of establishing SRW land-use practice in the riparian zones of two PPR wetland systems. The study attempted to answer several vital questions both in the field and microcosm scale in the area of wetland soil hydrology, salinity, nutrients, SOC sequestration, GHG emissions, and enzyme activities involved in the biogeochemical cycling. It is crucial to understand how SRW plantations might influence – particularly when plantations have matured – wetland hydrology (i.e., GWT depth), soil salinity, and contributions to carbon sequestration and GHG emissions with many different wetland types across a broader geographic region. Future recommended research that expands and builds on this work to provide further insight is covered in the following sections.

8.3.1 Simulation Models

The SRW has high water use potential through higher evapotranspiration (ET) rate compared to annual crops, because of the higher growth rate and biomass production capability (Dimitriou et al., 2009b). The increased groundwater consumption via ET by phreatophytic afforested plants has been linked with elevated groundwater and soil salinization (Jobbágy & Jackson, 2004).

However, the rate of ET markedly varies with local precipitation, atmospheric temperature, humidity, soil types, groundwater level, variety, and age of the plants. In this research project, we assumed that the SRW has a higher ET rate compared to adjacent contrasting land-use practices that would decrease salinity due to the lowering of the water table. However, the actual rate of SRW variety specified ET was not assessed while considering the atmospheric temperature and relative humidity of the experimental sites. Hence, future research should focus on the hydrological models including the variety-specific water use, local climate, soil hydrology as well as the solute transport considering the build-up of the salts in the soil (i.e., salinity). The use of simulation models such as SWAP (Soil Water Atmosphere Plant) or SWAT (Soil Water Assessment Tool) could serve a considerable role in the future field-scale water and salinity management in the semi-arid PPR region. Particularly the SWAP model, which can simulate the transport of water, solutes, and heat in the vadose or saturated zone in interaction with vegetation expansion in a landscape (Kroes et al., 2017, van Dam et al., 2008). It can thus evaluate various management options such as land-use practices while considering the local climate as it relates to field-scale water and solute movement.

Furthermore, SWAP is also capable of generating soil water fluxes for nutrients, e.g., the ANIMO model, which can be used to explore the uptake, fate, and transport of the nutrients in soil under different vegetation covers (Kersebaum et al., 2007). More precisely, the ANIMO model is a soil process-based simulation model for the evaluation of N and P loads on surface waters (Groenendijk et al., 2005), nitrate leaching to groundwater, and GHG emissions (Heinen, 2006, Hendriks et al., 2007).

8.3.2 Isotopic Tracer Technique

This research found that the SRW land-use practice can diminish soil N and K during their first rotation, which are essential nutrients for plant growth and biomass production. Besides, earlier studies have found the availability of soil N and P as an essential nutrient that needs

management across multiple rotations (Hangs et al., 2014a, Hangs et al., 2014b), and soil Ca and Mg strongly correlated with SRW productivity and consequent depletion (Ens et al., 2013b). However, it is still uncertain that the supply of the nutrients will remain persistent throughout the future rotation. Hence, long-term nutrient cycling, depletion, and fertilizer requirements related to the SRW variety and growth are needed. Fertilizer amendments can be of a supplement to the shortcoming of the N, P, and K in the soil. Thus, it is recommended that the dose and rate of depletion along with the biomass production over the multiple rotations should be evaluated in the riparian zones of the PPR wetland systems.

Additionally, in this study, a lower apparent NO_3^- -N and PO_4^{3-} -P utilization rate were observed by the SRW, which can be a significant concern for the future water quality for the wetlands in this region. If the SRW plantation in the riparian zones could not uptake NO_3^- -N and PO_4^{3-} -P from the soil and groundwater, these can be leached into the wetlands, and thus cause rapid eutrophication. Thus, the uptake and rate of nutrient cycling by the SRW should be further studied via labeled ^{14}N , ^{31}P , ^{39}K , ^{34}S nutrients application (i.e., isotopic tracer technique). Thus, understanding of nutrient dynamics will be improved, and the sinks can be better identified. Also, the study will help to optimize the fertilizer rate; the ability to utilize or capture excess nutrients by the SRW plantation can be achieved with desired economic and environmental benefits.

8.3.3 Synchrotron Techniques

The rate of SOC decomposition and sequestration is primarily controlled by the molecular structure and biochemical stability (Schmidt et al., 2011). Land-use has a crucial impact on the quantity and chemical composition of SOC (Ramesh et al., 2019). In this thesis, POC, LFOC, and WEOC were measured in addition to the belowground SOC content under different land-use practices. Moreover, FTIR analysis revealed that the depth has more influence on SOC functional groups than land-use practice. However, a similarity was observed in chemical

functional groups of SOC across a 46 km ecotonal climosequence in Saskatchewan (Purton et al., 2015). They revealed that the pedon-scale processes have more significant influence than landscape (e.g., land-use) or regional (e.g., climate) scales.

The SOC sequestration potential depends on the plant species, litter quality, and quantity (Stockmann et al., 2013). Additionally, higher root growth and root biomass production, in conjunction with the higher plant growth, have a connection with salt transportation and deposition linking the salinity in the soil. Moreover, root-derived organic compounds such as decaying roots are identified as a vital source of SOC. Therefore, identifying the processes that can maximize the SOC sequestration potential under SRW land-use practice is required. A critical next step is to use and compare novel approaches on a molecular level to determine the chemical composition of SOC and better understand its interactions with minerals, salts, and water (Hsu et al., 2018). Hence, modern state-of-the-art synchrotron technique, such as computed tomography (CT) scanner for the root architecture systems, scanning electron microscopy (SEM), and different soft X-ray spectroscopy beamline to image soil samples would be imminent research that can enhance the understanding of carbon sequestration mechanism and climate change mitigation strategies (Holman & Martin, 2006, Lehmann & Solomon, 2010, Lombi & Susini, 2009, Raab & Lipson, 2010).

One of the main constraints with the conventional method is, after all the processing, the chemical composition of SOC (i.e., what we want to study) may change in the soil. There are many advantages to using nondestructive synchrotron techniques. However, the biggest challenge that can be overcome would be able to see the exact what was naturally occurring in the soil, particularly at the molecular level (Lombi & Susini, 2009).

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APPENDIX A. SUPPLEMENTARY MATERIALS FOR CHAPTER 3

Table A-S 1 Mean (\pm SD) depth to the groundwater table, physiochemical properties, and elevation under different land-use practices from both sites.

Variable	Year	Site A			Site B		
		AC	PA	SRW	AC	PA	SRW
Depth to GWT (cm)	2014	72.82 \pm 33.32aB	107.42 \pm 50.25aB	70.78 \pm 47.29aB	88.47 \pm 39.41aB	51.02 \pm 31.08bB	76.33 \pm 37.33aB
	2015	100.51 \pm 34.55aA	128.90 \pm 52.89aA	116.50 \pm 48.78aA	113.92 \pm 33.66aA	64.56 \pm 29.67bA	131.83 \pm 39.14aA
EC _{gw} (mS cm ⁻¹)	2014	2.25 \pm 1.78cA	7.00 \pm 2.35aA	3.54 \pm 2.70bA	6.96 \pm 4.95bA	2.19 \pm 1.75cA	10.16 \pm 6.40aA
	2015	2.53 \pm 2.75cA	6.21 \pm 3.25aA	4.53 \pm 3.84bA	6.54 \pm 4.06bB	1.49 \pm 0.53cB	7.33 \pm 5.56aB
TDS (mg L ⁻¹)	2014	1604.53 \pm 1440.67cA	5601.60 \pm 1879.73aA	2598.08 \pm 2257.68bA	5449.39 \pm 4079.19bA	1574.72 \pm 1485.53cA	7996.80 \pm 5274.20aA
	2015	1830.51 \pm 2240.18cA	4913.92 \pm 2685.02aA	3390.40 \pm 3175.85bA	5016.00 \pm 3432.18bB	956.16 \pm 340.07cB	5696.40 \pm 4593.08aB
pH	2014	7.54 \pm 0.30aA	7.57 \pm 0.02aA	7.50 \pm 0.25aA	7.58 \pm 0.19aA	7.48 \pm 0.27bA	7.55 \pm 0.19aA
	2015	7.28 \pm 0.34aB	7.54 \pm 0.04aB	7.34 \pm 0.43aB	7.53 \pm 0.15aB	7.36 \pm 0.44bB	7.45 \pm 0.25aB
Na ⁺ (mg L ⁻¹)	2014	71.85 \pm 53.16cA	185.17 \pm 62.04aA	81.60 \pm 74.93bA	180.10 \pm 125.23aA	56.08 \pm 52.78bA	255.07 \pm 186.52aA
	2015	77.35 \pm 72.20cA	167.03 \pm 81.75aA	126.72 \pm 86.71bA	215.54 \pm 43.86aA	44.33 \pm 30.01bA	207.41 \pm 75.55aA
Ca ²⁺ (mg L ⁻¹)	2014	95.91 \pm 74.98cA	148.65 \pm 41.62aA	125.76 \pm 76.90bA	156.57 \pm 97.56aA	75.67 \pm 32.58bA	186.40 \pm 80.90aA
	2015	67.58 \pm 61.89cA	139.50 \pm 52.88aA	119.81 \pm 59.58bA	130.98 \pm 43.97aA	39.51 \pm 35.14bA	167.38 \pm 61.62aA
Mg ²⁺ (mg L ⁻¹)	2014	147.62 \pm 83.99cA	306.02 \pm 87.16aA	211.30 \pm 146.21bA	321.63 \pm 169.07aA	136.25 \pm 70.24bA	388.35 \pm 181.62aA
	2015	145.56 \pm 107.79cA	291.60 \pm 105.64aA	232.62 \pm 108.75bA	330.37 \pm 69.86aA	114.31 \pm 49.23bA	359.31 \pm 122.57aA
SO ₄ ²⁻ (mg L ⁻¹)	2014	187.81 \pm 106.13bB	233.66 \pm 57.59bB	240.57 \pm 155.09aB	277.13 \pm 98.51aA	140.58 \pm 68.67bA	279.32 \pm 112.09aA
	2015	205.53 \pm 131.13bA	206.26 \pm 98.01bA	337.70 \pm 147.34aA	315.17 \pm 101.61aA	157.82 \pm 44.01bA	263.19 \pm 131.49aA
Elevation (m)	2014	583.33 \pm 0.23b	584.18 \pm 0.16a	583.62 \pm 0.49b	585.38 \pm 0.56a	584.28 \pm 0.11b	584.66 \pm 0.22b

^a Values are mean \pm standard deviations (\pm SD) of the groundwater samples collection months. Means were calculated from the values of groundwater parameters measured from May, June, July, August, and September every year.

^b Means within a row for land-use followed by the same small letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c Means within a column for year supported by the same capital letter are not significantly different ($p > 0.05$) using Tukey HSD.

^d EC = electrical conductivity, TDS = total dissolved salts, GWT = depth to groundwater table, AC = annual crop, PA = pasture, SRW = short rotation willow.

Table A-S 2 Mean (\pm SD) soil physiochemical characteristics under different land-use practices from both sites.

Variable	Depth	Year	Site A			Site B		
			AC	PA	SRW	AC	PA	SRW
EC _{soil} (mS cm ⁻¹)	0-30cm	2014	0.70 \pm 0.70c	0.42 \pm 0.20a	0.88 \pm 0.72b	2.68 \pm 2.39b	0.60 \pm 0.12c	3.62 \pm 1.76a
		2015	0.67 \pm 0.50c	0.51 \pm 0.15a	0.96 \pm 0.60b	2.76 \pm 1.98b	0.75 \pm 0.16c	3.62 \pm 1.74a
	30-60cm	2014	0.58 \pm 0.64c	1.47 \pm 0.34a	0.93 \pm 0.82b	2.55 \pm 1.93b	0.43 \pm 0.15c	3.28 \pm 1.70a
		2015	0.54 \pm 0.51c	1.72 \pm 1.01a	1.12 \pm 0.93b	2.23 \pm 1.68b	0.44 \pm 0.05c	3.17 \pm 1.54a
TDS (mg L ⁻¹)	0-30cm	2014	449.07 \pm 449.58c	270.93 \pm 129.63a	562.90 \pm 460.98b	1713.07 \pm 1527.09b	381.87 \pm 79.59c	2318.13 1127.51a
		2015	429.16 \pm 322.26c	325.33 \pm 93.84a	612.57 \pm 380.98b	1767.82 \pm 1268.95b	480.00 \pm 105.47c	2313.60 \pm 1112.53a
	30-60cm	2014	369.42 \pm 409.18c	937.60 \pm 218.45a	598.25 \pm 525.46b	1634.84 \pm 1236.63b	275.20 \pm 96.81c	2099.47 \pm 1089.52a
		2015	344.89 \pm 327.13c	1097.60 \pm 647.45a	718.32 \pm 593.81b	1430.04 \pm 1076.88b	278.40 \pm 31.03c	2029.33 \pm 982.48a
pH	0-30cm	2014	8.30 \pm 0.40	7.90 \pm 0.16	8.24 \pm 0.23	7.61 \pm 0.19	8.41 \pm 0.28	8.16 \pm 0.26
		2015	8.20 \pm 0.34	7.82 \pm 0.10	8.10 \pm 0.17	7.48 \pm 0.20	8.22 \pm 0.28	8.04 \pm 0.24
	30-60cm	2014	8.56 \pm 0.40	8.43 \pm 0.24	8.43 \pm 0.34	7.76 \pm 0.28	8.36 \pm 0.19	8.37 \pm 0.30
		2015	8.41 \pm 0.34	8.11 \pm 0.15	8.21 \pm 0.25	7.57 \pm 0.22	8.37 \pm 0.20	8.21 \pm 0.22
Na ⁺ (mg kg ⁻¹)	0-30cm	2014	65.59 \pm 72.50	71.13 \pm 70.39	83.55 \pm 81.94	356.30 \pm 327.48	79.47 \pm 42.89	414.11 \pm 226.55
		2015	93.00 \pm 65.98	103.34 \pm 78.84	116.60 \pm 60.62	524.25 \pm 432.16	144.41 \pm 53.15	601.51 \pm 601.14
	30-60cm	2014	41.84 \pm 69.90	180.57 \pm 55.56	69.50 \pm 72.29	281.66 \pm 243.61	39.79 \pm 43.28	335.68 \pm 216.79
		2015	65.00 \pm 55.61	202.07 \pm 73.58	113.35 \pm 80.42	369.19 \pm 216.69	74.14 \pm 26.74	381.63 \pm 235.75
Ca ²⁺ (mg kg ⁻¹)	0-30cm	2014	3407.89 \pm 668.46	3624.26 \pm 1365.15	3791.58 \pm 645.18	3493.53 \pm 1394.28	3505.68 \pm 784.63	4219.54 \pm 1131.40
		2015	4666.87 \pm 1068.68	4805.02 \pm 578.55	5407.93 \pm 1076.31	4837.28 \pm 1132.07	4916.13 \pm 831.84	6279.89 \pm 2424.50
	30-60cm	2014	3632.71 \pm 791.83	3805.95 \pm 1475.70	3556.54 \pm 1073.31	3232.27 \pm 1627.64	3499.75 \pm 597.42	4812.62 \pm 1559.75
		2015	4765.48 \pm 1103.51	5209.61 \pm 731.87	5762.06 \pm 1704.55	3885.44 \pm 1508.94	4764.16 \pm 650.90	7737.34 \pm 3899.97
Mg ²⁺ (mg kg ⁻¹)	0-30cm	2014	1404.62 \pm 481.32	827.75 \pm 231.52	1570.70 \pm 611.00	1771.78 \pm 807.75	1477.84 \pm 452.16	2105.06 \pm 437.45
		2015	1446.14 \pm 462.98	1025.48 \pm 328.11	1519.87 \pm 538.00	1961.06 \pm 829.43	1643.03 \pm 598.69	2188.27 \pm 376.89
	30-60cm	2014	1070.15 \pm 430.93	1482.03 \pm 524.01	1252.95 \pm 508.49	1709.73 \pm 644.60	1141.43 \pm 536.67	1757.40 \pm 440.70
		2015	1085.14 \pm 408.88	1296.38 \pm 387.97	1282.74 \pm 547.33	4273.18 \pm 7791.77	1285.06 \pm 596.20	1846.41 \pm 485.62
SO ₄ ²⁺ -S (mg kg ⁻¹)	0-30cm	2014	262.21 \pm 400.52	485.59 \pm 680.07	383.12 \pm 429.94	1612.10 \pm 1543.57	308.36 \pm 305.44	2488.67 \pm 1800.17
		2015	330.88 \pm 457.23	514.57 \pm 650.70	486.91 \pm 420.72	2043.71 \pm 1465.50	399.66 \pm 329.99	2572.70 \pm 1525.60
	30-60cm	2014	275.95 \pm 465.25	799.67 \pm 720.80	556.33 \pm 667.94	1287.85 \pm 968.25	221.13 \pm 256.81	3024.17 \pm 2804.75
		2015	318.26 \pm 475.89	1208.94 \pm 782.22	976.38 \pm 1101.47	1642.13 \pm 1134.07	232.38 \pm 230.74	2736.29 \pm 1753.67
ESP	0-30cm	2014	1.15 \pm 1.13c	1.20 \pm 0.74a	1.33 \pm 1.15b	5.28 \pm 4.48a	1.43 \pm 0.77b	5.63 \pm 2.93a
		2015	1.36 \pm 0.79c	1.47 \pm 0.95a	1.50 \pm 0.70b	5.70 \pm 3.58a	1.94 \pm 0.71b	5.74 \pm 4.40a
	30-60cm	2014	0.56 \pm 0.91c	3.10 \pm 0.90a	1.13 \pm 1.11b	4.70 \pm 3.89a	0.73 \pm 0.63b	4.32 \pm 2.75a
		2015	0.96 \pm 0.68c	2.70 \pm 0.97a	1.50 \pm 0.99b	4.70 \pm 2.83a	1.17 \pm 0.32b	3.64 \pm 2.13a

Table A-S 2 – continued

SAR	0-30cm	2014	1.29± 1.36c	1.39± 1.17a	1.58± 1.50b	6.49± 5.77a	1.58± 0.85b	7.23± 3.82a
		2015	1.66± 1.14c	1.89± 1.39a	1.99± 1.03b	8.39± 6.18a	2.53± 0.93b	8.95± 8.61a
	30-60cm	2014	0.80± 1.22c	3.51± 0.94a	1.35± 1.34b	5.45± 4.66a	0.78± 0.79b	5.64± 3.61a
		2015	1.19± 0.95c	3.51± 1.22a	1.95± 1.41b	6.14± 3.44a	1.35± 0.48b	5.51± 3.30a
VSWC (%)	0-30cm	2014	41.85± 10.99a	24.88± 6.39b	43.68± 12.53a	37.69± 3.55b	36.92± 2.96a	38.60± 5.93b
		2015	37.66± 10.99a	30.50± 8.87b	37.46± 11.64a	36.76± 7.72b	39.04± 2.47a	36.77± 4.24b
	30-60cm	2014	35.14± 8.14a	24.66± 7.27b	35.79± 7.94a	31.52± 5.05b	40.17± 4.92a	34.35± 5.30b
		2015	36.85± 8.52a	22.76± 7.07b	33.70± 13.56a	28.45± 5.30b	38.85± 1.51a	32.81± 5.16b
CEC (cmol _c kg ⁻¹)	0-30cm	2014	36.85± 1.71bA	43.76± 0.10aA	35.57± 7.50bA	40.70± 10.35bA	42.92± 0.51bA	46.24± 9.13aA
	30-60cm		37.17± 3.04bA	40.52± 4.52aA	37.48± 13.66bA	40.52± 14.72bA	38.89± 2.29bA	49.07± 11.34aA
Clay (%)	0-30cm	2014	29.42± 2.35bB	38.50± 4.93aB	29.57± 5.79bB	31.00± 3.00cB	36.00± 4.38aB	32.12± 1.48bB
	30-60cm		31.33± 4.70bA	44.00± 3.29aA	35.00± 4.84bA	31.33± 1.80cA	36.00± 1.10aA	35.62± 4.73bA
Bulk density (g cm ⁻³)	0-30cm	2014	1.41± 0.13a	1.29± 0.03b	1.28± 0.08b	1.26± 0.11b	1.24± 0.04b	1.29± 0.07a

^a Values are mean ± standard deviations (±SD) of the soil samples collection months. Means were calculated from the values of soil parameters measured from May, July, and September every year.

^b Means within a row for land-use followed by the same small letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c Means within a column for year supported by the same capital letter are not significantly different ($p > 0.05$) using Tukey HSD.

^d EC = electrical conductivity, TDS = total dissolved salts, ESP = exchangeable sodium percentage, SAR = sodium adsorption ratio, VSWC = volumetric soil water content, CEC = cation exchange capacity.

Table A-S 3 Analysis of variance (ANOVA) with a linear mixed-effects model for measured groundwater pH, salts (Na⁺, Ca²⁺, Mg²⁺, SO₄²⁻), and elevation from three land-use practices from two sites during the growing season of two consecutive years.

Response variable	Sources of variation	Site A			Site B	
		df	F - value	p - value	F - value	p - value
pH	Land-use	2	0.216	0.806 ^{ns}	2.715	0.070 ^{ns}
	Year	1	16.495	<0.001 ^{***}	7.171	0.008 ^{**}
	Month	4	3.516	0.009 ^{**}	9.961	<0.001 ^{***}
Na ⁺	Land-use	2	9.195	<0.001 ^{***}	30.057	<0.001 ^{***}
	Year	1	3.463	0.065 ^{ns}	0.023	0.879 ^{ns}
	Month	4	2.593	0.039 [*]	1.446	0.223 ^{ns}
Ca ²⁺	Land-use	2	10.730	<0.001 ^{***}	37.456	<0.001 ^{***}
	Year	1	3.167	0.077 ^{ns}	4.391	0.038 [*]
	Month	4	0.903	0.464 ^{ns}	1.163	0.331 ^{ns}
Mg ²⁺	Land-use	2	12.929	<0.001 ^{***}	41.029	<0.001 ^{***}
	Year	1	0.163	0.687 ^{ns}	0.148	0.701 ^{ns}
	Month	4	2.316	0.060 ^{ns}	2.590	0.040 [*]
SO ₄ ²⁻	Land-use	2	7.235	0.001 ^{***}	12.987	<0.001 ^{***}
	Year	1	4.408	0.038 [*]	0.009	0.927 ^{ns}
	Month	4	1.463	0.217 ^{ns}	1.174	0.326 ^{ns}
Elevation	Land-use	2	55.828	<0.001 ^{***}	259.940	<0.001 ^{***}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

^b Data was transformed to square root.

Table A-S 4 Analysis of variance (ANOVA) with a linear mixed-effects model for measured soil pH, salts (Na⁺, Ca²⁺, Mg²⁺, SO₄²⁻), CEC, clay, and bulk density from different land-use practices from two sites during the growing season of two consecutive years.

Response variable	Sources of variation	Site A		Site B		
		df	F - value	p - value	F - value	p - value
pH	Land-use	2	26.749	<0.001 ***	110.566	<0.001 ***
	Depth	1	36.658	<0.001 ***	20.262	<0.001 ***
	Year	1	19.743	<0.001 ***	16.343	<0.001 ***
	Month	2	7.793	<0.001 ***	7.880	<0.001 ***
Na ⁺	Land-use	2	14.239	<0.001 ***	29.226	<0.001 ***
	Depth	1	0.410	0.523 ^{ns}	7.846	0.006 **
	Year	1	19.107	<0.001 ***	7.147	0.008 **
	Month	2	15.037	<0.001 ***	0.975	0.379 ^{ns}
Ca ²⁺	Land-use	2	3.905	0.022 *	17.786	<0.001 ***
	Depth	1	0.353	0.553 ^{ns}	1.362	0.245 ^{ns}
	Year	1	97.295	<0.001 ***	45.133	<0.001 ***
	Month	2	1.261	0.286 ^{ns}	1.361	0.260 ^{ns}
Mg ²⁺	Land-use	2	1.504	0.225 ^{ns}	6.699	0.002 **
	Depth	1	8.105	0.005 **	1.421	0.235 ^{ns}
	Year	1	0.043	0.836 ^{ns}	2.217	0.139 ^{ns}
	Month	2	0.412	0.663 ^{ns}	1.850	0.161 ^{ns}
SO ₄ ²⁻ -S	Land-use	2	11.267	<0.001 ***	46.470	<0.001 ***
	Depth	1	4.337	0.039 *	0.019	0.890 ^{ns}
	Year	1	5.380	0.022 *	0.7194	0.398 ^{ns}
	Month	2	21.446	<0.001 ***	0.445	0.642 ^{ns}
CEC	Land-use	2	3.138	0.046 *	11.788	<0.001 ***
	Depth	1	0.101	0.751 ^{ns}	0.277	0.599 ^{ns}
Clay	Land-use	2	70.909	<0.001 ***	17.170	<0.001 ***
	Depth	1	40.544	<0.001 ***	18.538	<0.001 ***
Bulk density	Land-use	2	88.696	<0.001 ***	32.693	<0.001 ***

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively;

^{ns}, is not significantly different ($p > 0.05$).

^b Data was transformed to square root.

^c CEC = cation exchange capacity.

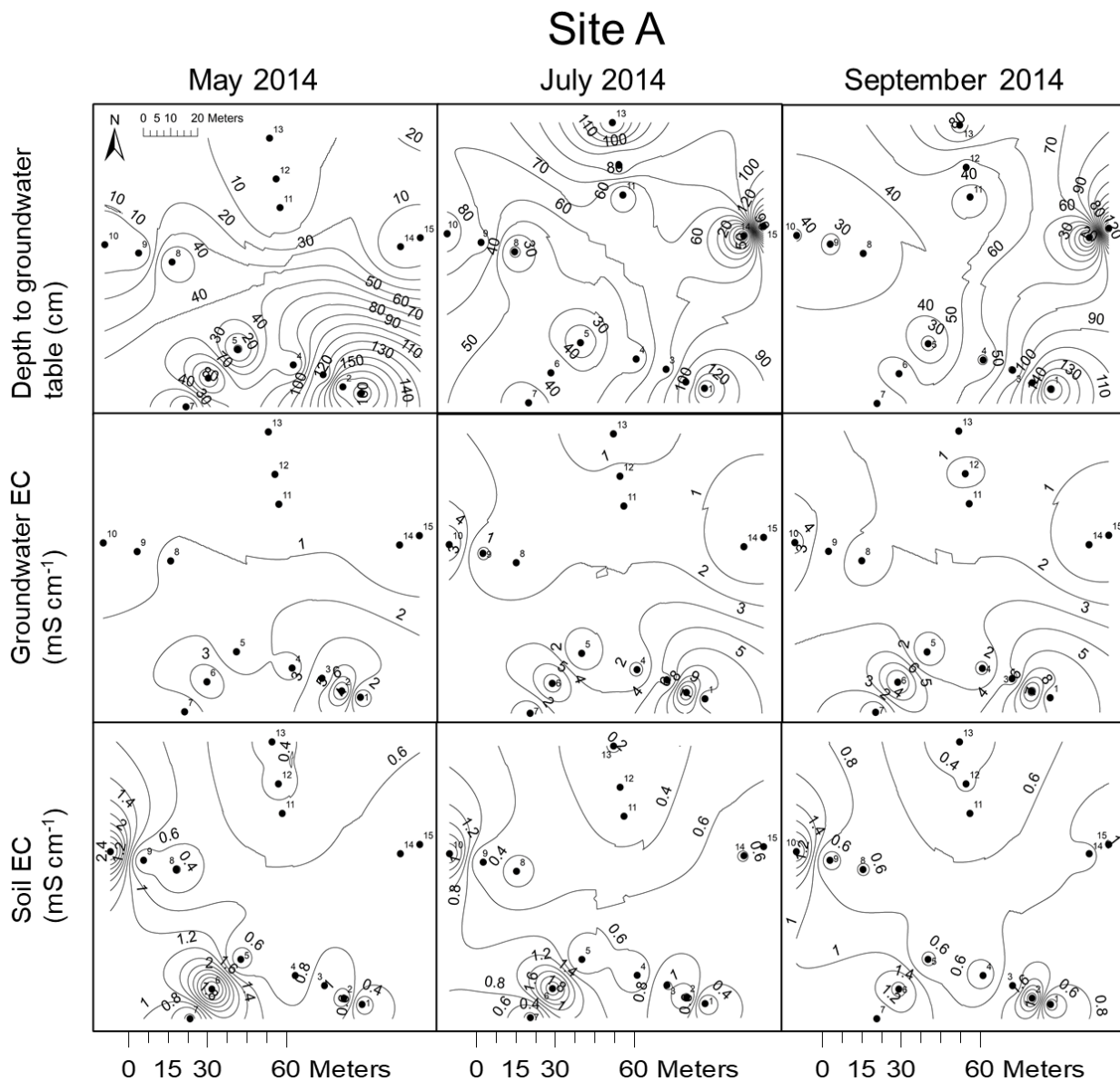


Figure A-S 1 Contour map of depth to the groundwater table, groundwater, and soil electrical conductivity derived from kriging on three sampling days during the growing seasons of 2014 from three different land-use practices from site A.

^a Black dots indicate the position of groundwater monitoring wells.

^b EC = electrical conductivity.

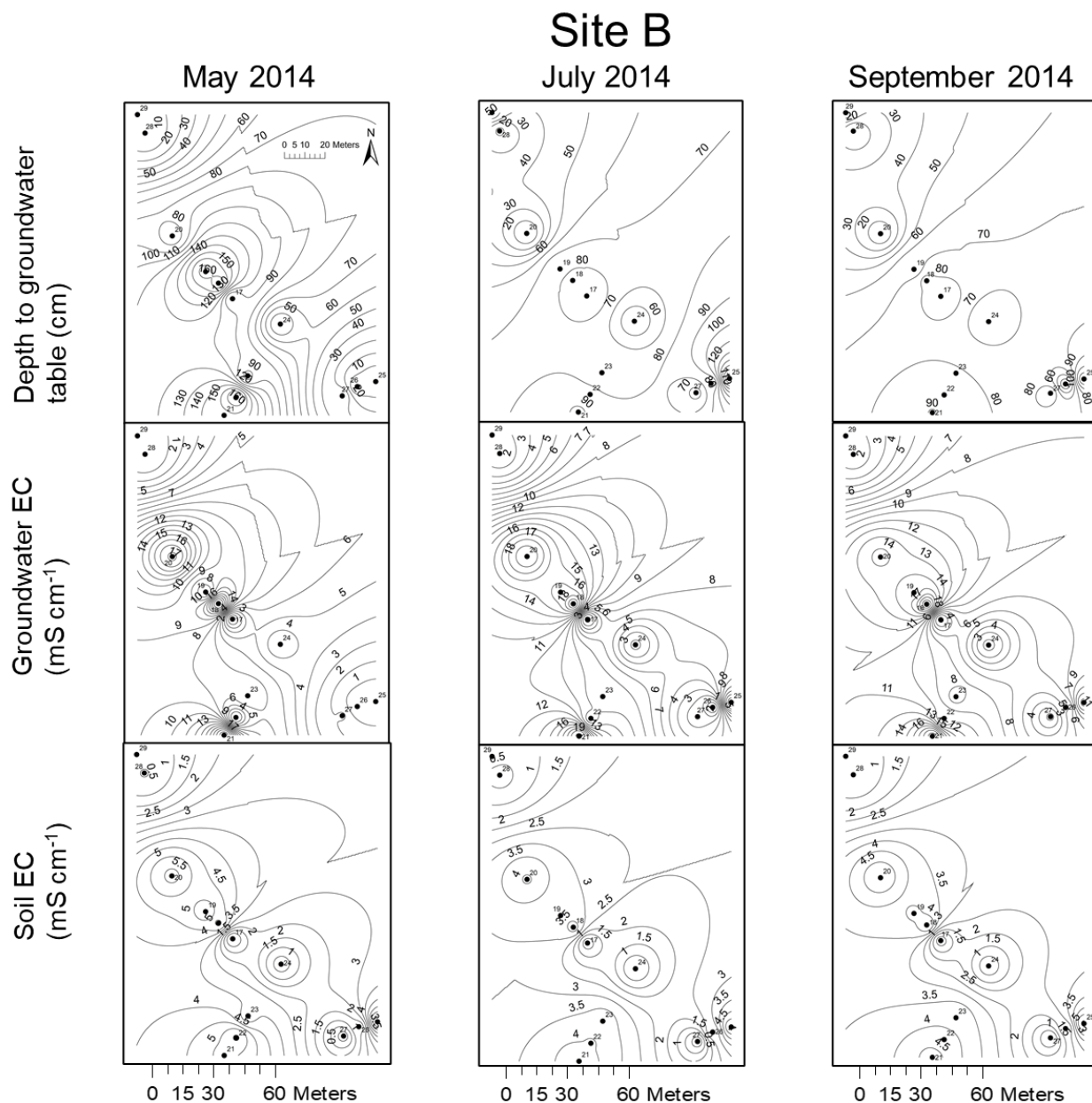


Figure A-S 2 Contour map of depth to the groundwater table, groundwater, and soil electrical conductivity derived from kriging on three sampling days during the growing seasons of 2014 from three different land-use practices from site B.

^a Black dots indicate the position of groundwater monitoring wells.

^b EC = electrical conductivity.

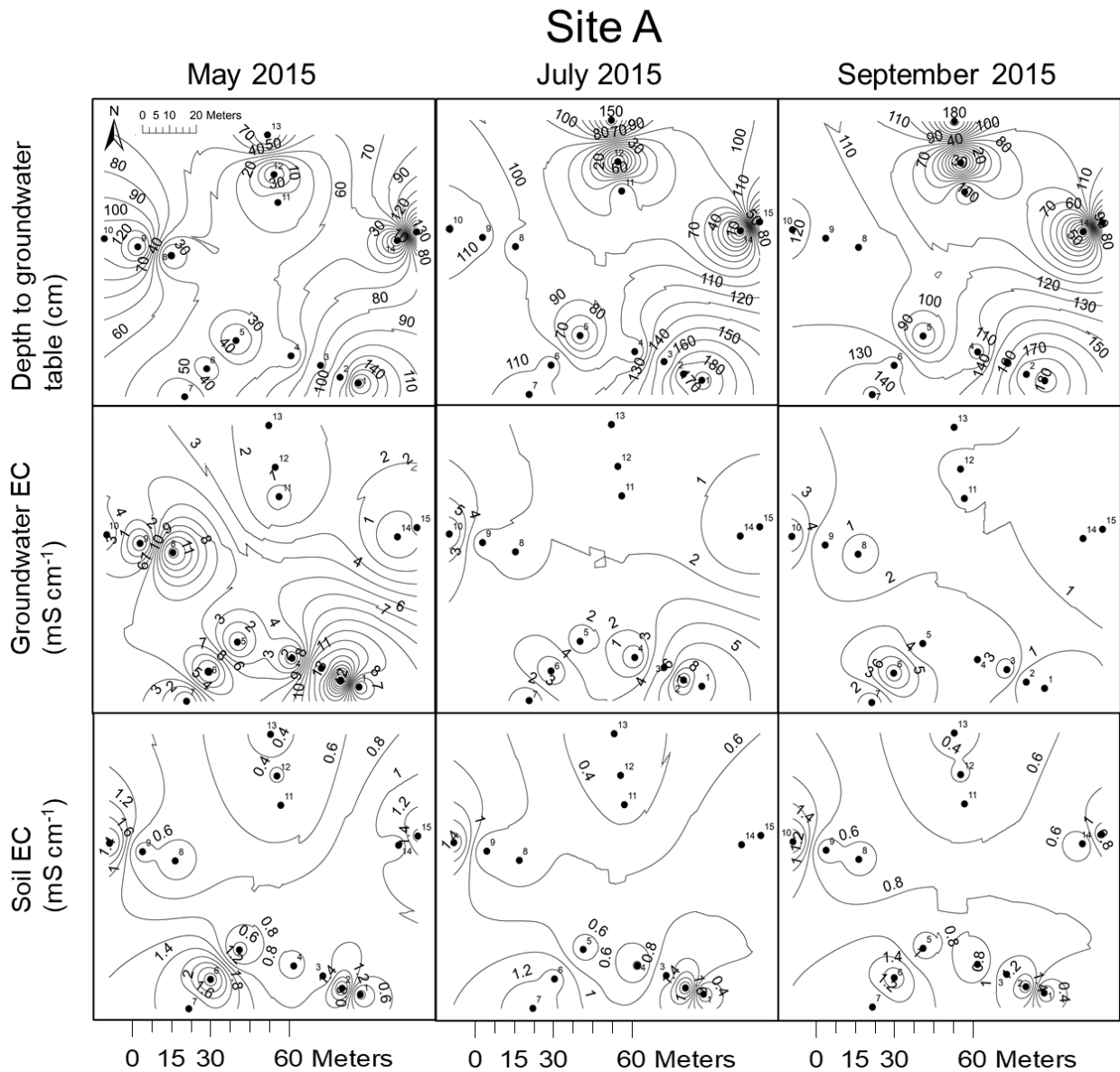


Figure A-S 3 Contour map of depth to the groundwater table, groundwater, and soil electrical conductivity derived from kriging on three sampling days during the growing seasons of 2015 from three different land-use practices from site A.

^a Black dots indicate the position of groundwater monitoring wells.

^b EC = electrical conductivity.

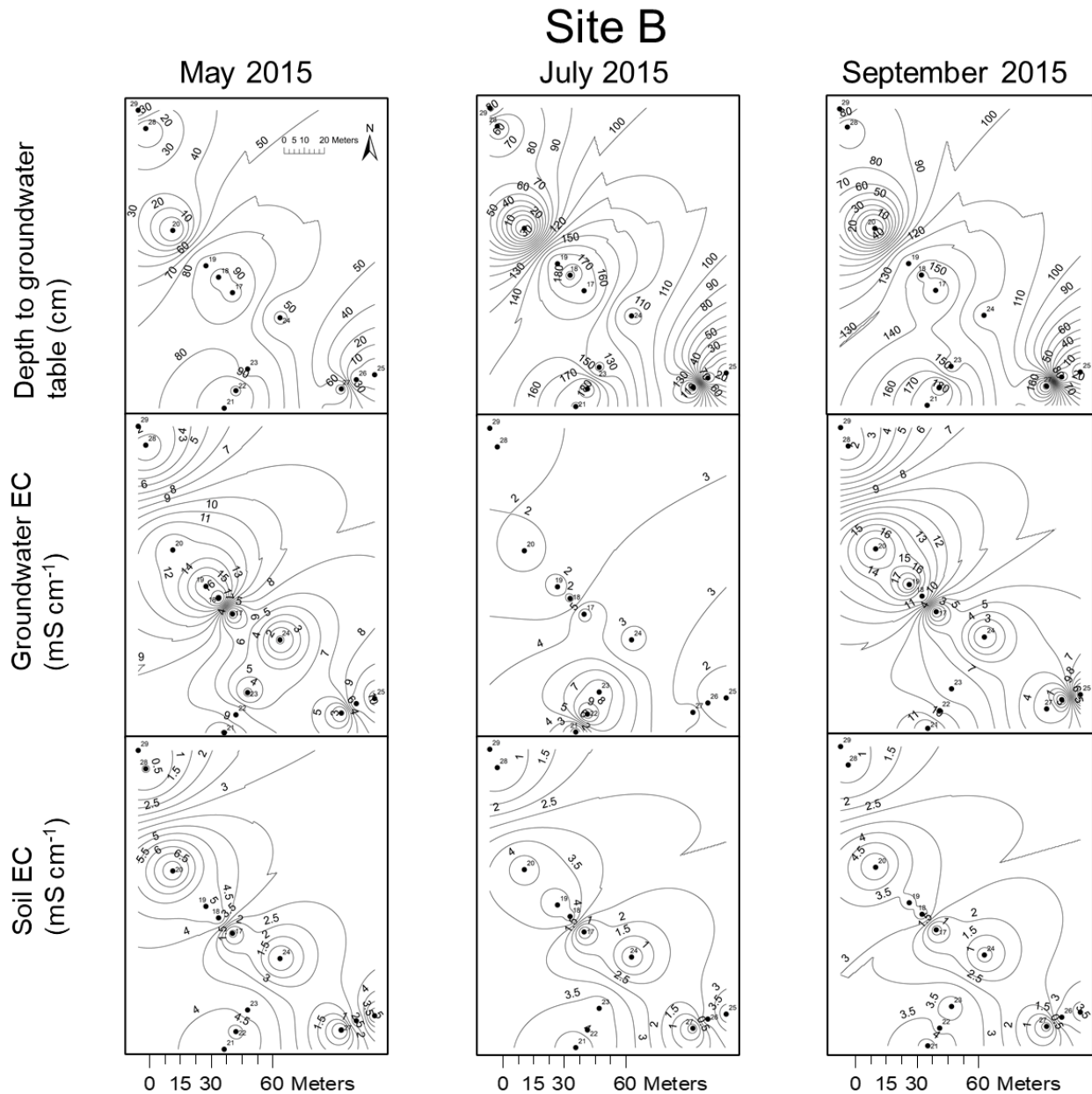


Figure A-S 4 Contour map of depth to the groundwater table, groundwater, and soil electrical conductivity derived from kriging on three sampling days during the growing seasons of 2015 from three different land-use practices from site B.

^a Black dots indicate the position of groundwater monitoring wells.

^b EC = electrical conductivity.

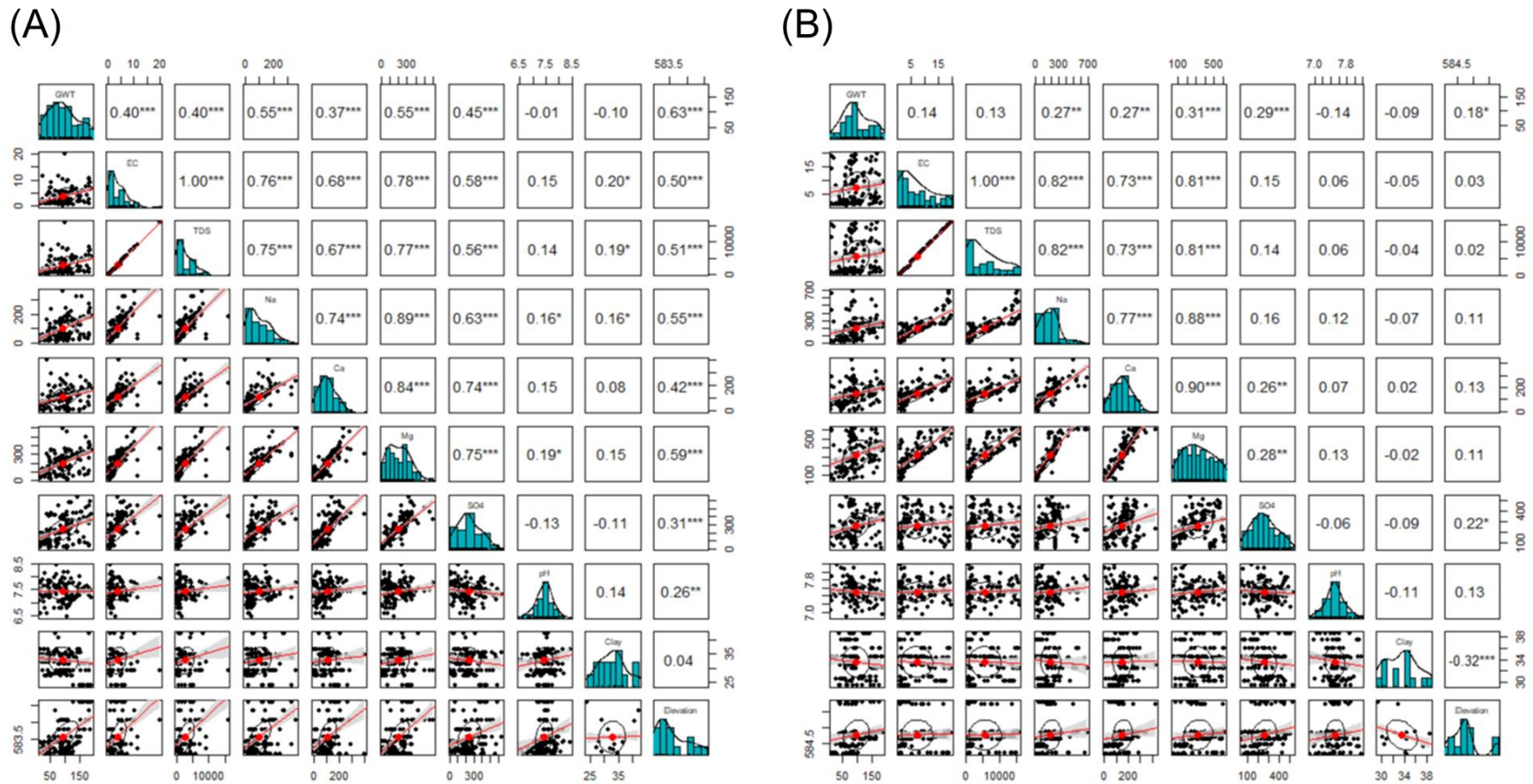


Figure A-S 5 Correlations (Pearson) among groundwater parameters measured during field experiment from A) site A, and B) site B.

^a The red line shows linear fit, the shaded area shows 95% of the confidence interval, values in the correspondent box indicates individual correlation coefficients (r), and p values indicate the level of significance.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; and rests are not significant ($p > 0.05$).

^c GWT = depth to groundwater table, EC = electrical conductivity, TDS = total dissolved salts.

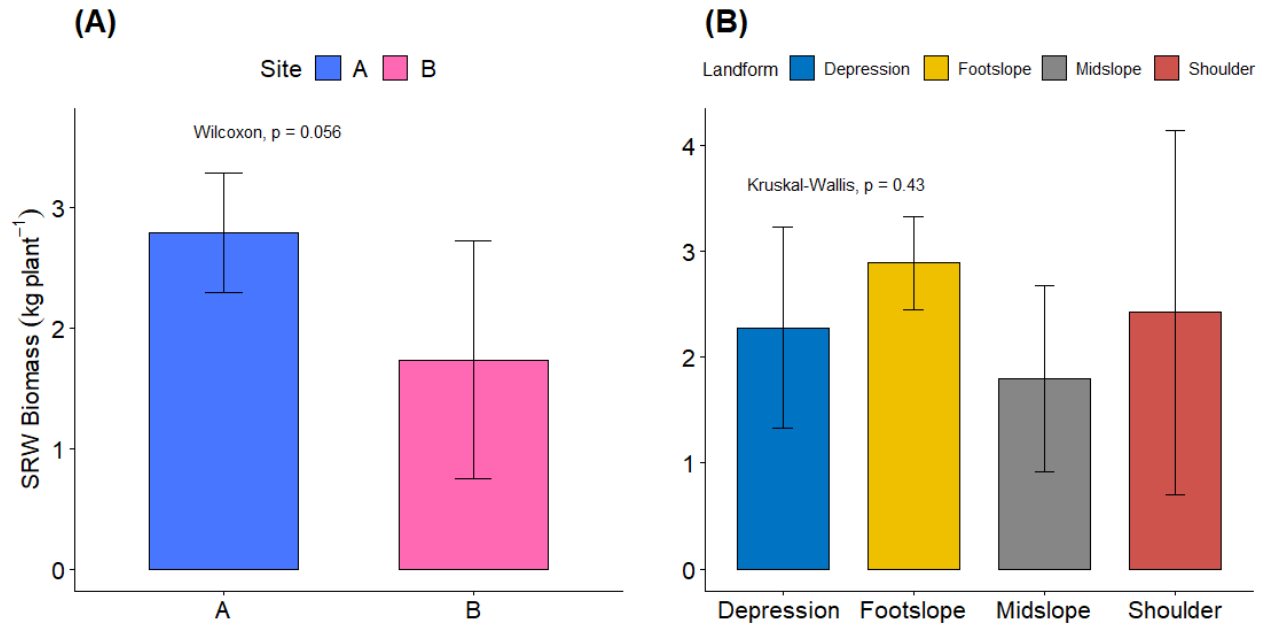


Figure A-S 6 Biomass of short rotation willow based on A) sites, and B) landforms.

^a *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; and rests are not significant ($p > 0.05$).

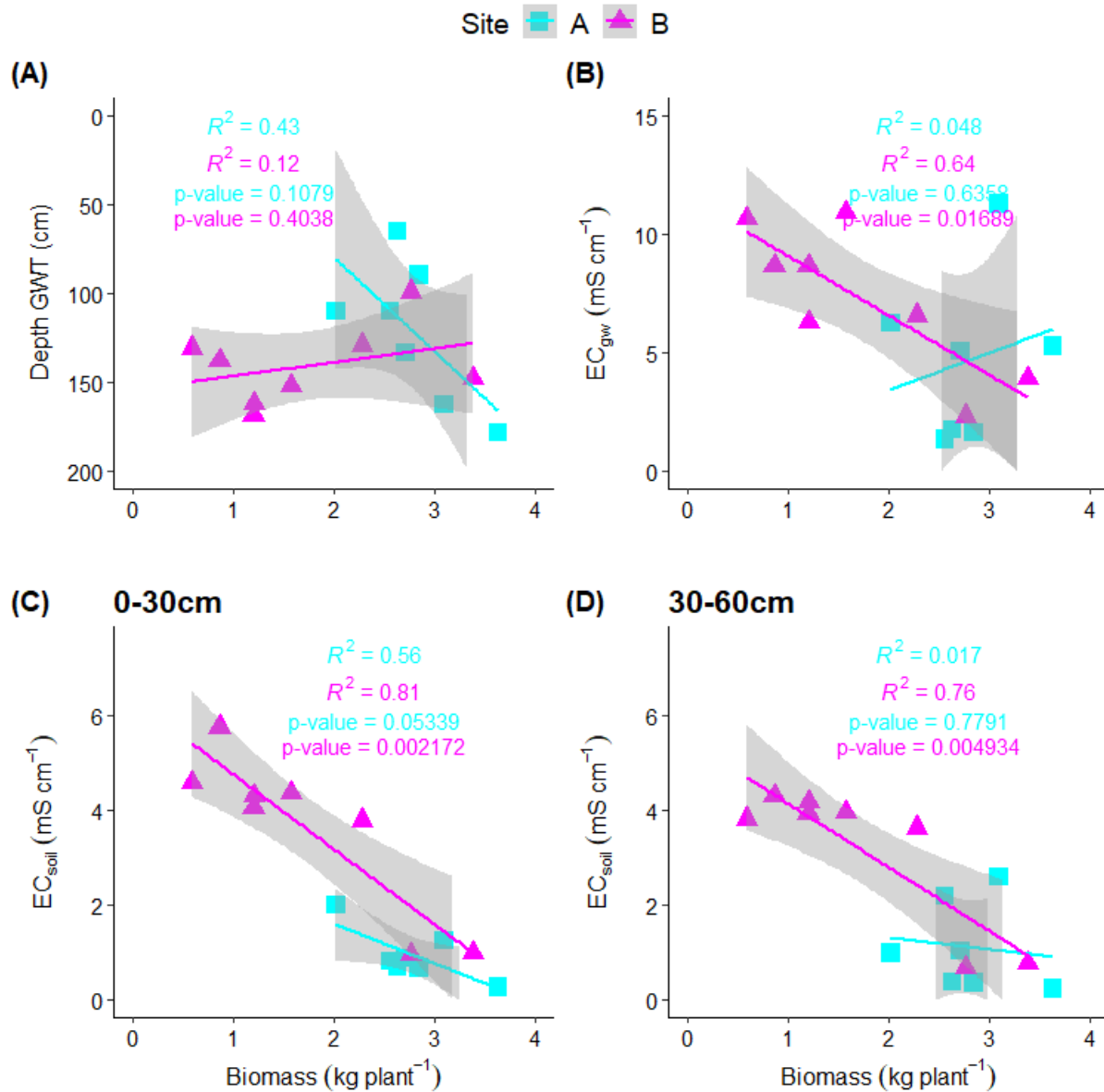


Figure A-S 7 Relationship between short rotation willow biomass with A) depth to groundwater table, B) groundwater electrical conductivity, C) soil electrical conductivity at 0-30 cm depth, and D) soil electrical conductivity at 30-60 cm from both sites.

^a The line shows linear regression, the shaded area shows 95% of the confidence interval, R² gives values for individual regression lines, and p values indicate the level of significance.

^b GWT = depth to groundwater table, EC_{gw} = groundwater electrical conductivity, EC_{soil} = soil electrical conductivity.

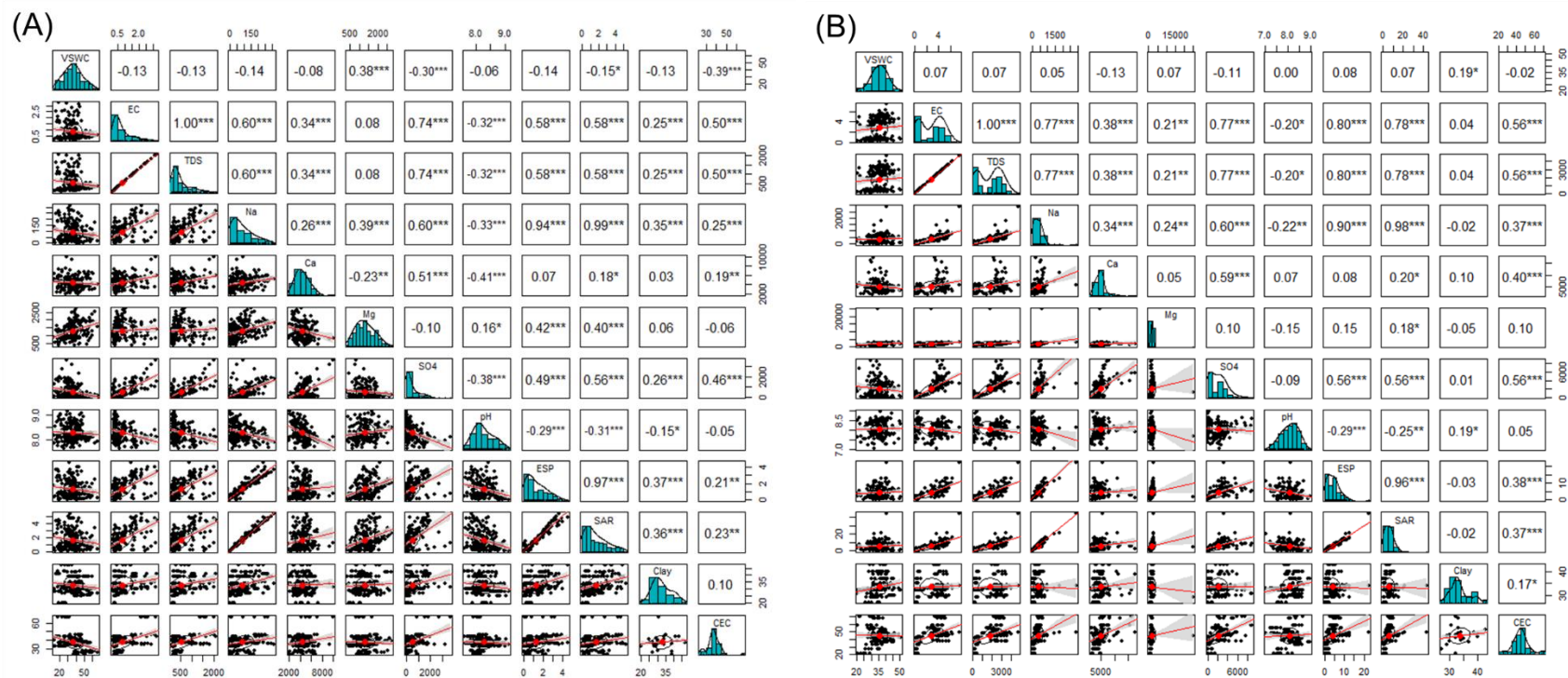


Figure A-S 8 Correlations (Pearson) among soil parameters measured during field experiment from A) site A, and B) site B.

^a The red line shows linear fit, the shaded area shows 95% of the confidence interval, values in the correspondent box indicates individual correlation coefficients (r), and p values indicate the level of significance.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of

^c VSWC = volumetric soil water content, EC = electrical conductivity, TDS = total dissolved salts, ESP = exchangeable sodium percentage, SAR = sodium adsorption ratio, CEC = cation exchange capacity.

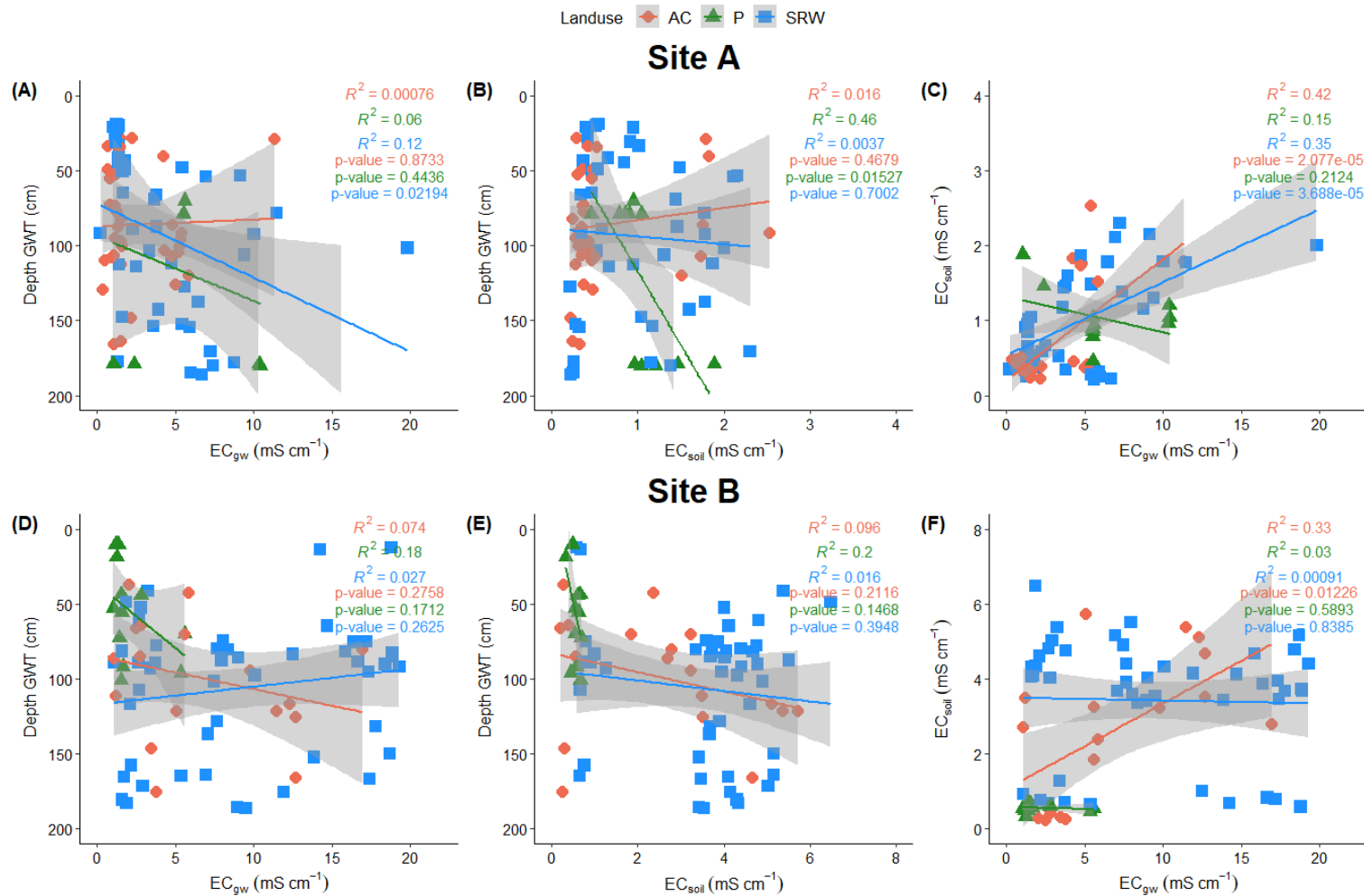


Figure A-S 9 Relationship between depth to GWT with groundwater EC and soil EC, and between groundwater EC with soil EC under three different land-use practices from A) site A, and B) site B.

^a The line shows linear regression, the shaded area shows 95% of the confidence interval, R^2 gives values for individual regression lines, and p values indicate the level of significance.

^b AC = annual crop, PA = pasture, SRW = short rotation willow, GWT = depth to groundwater table, EC_{gw} = groundwater electrical conductivity, EC_{soil} = soil electrical conductivity.

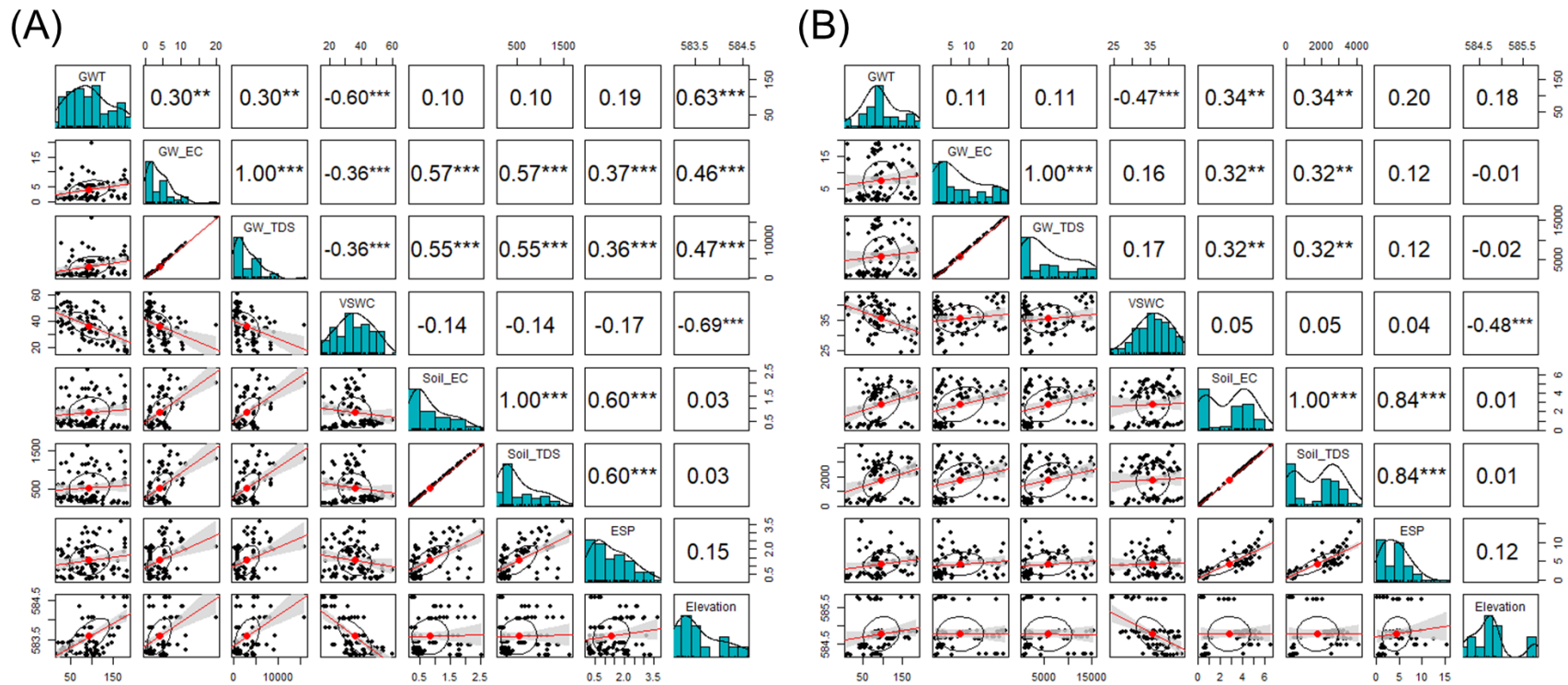


Figure A-S 10 Correlations (Pearson) among ground water and soil parameters measured during field experiment from A) site A, and B) site B.

^a Average value for both soil depths were used; only the measurements values from May, July and September used.

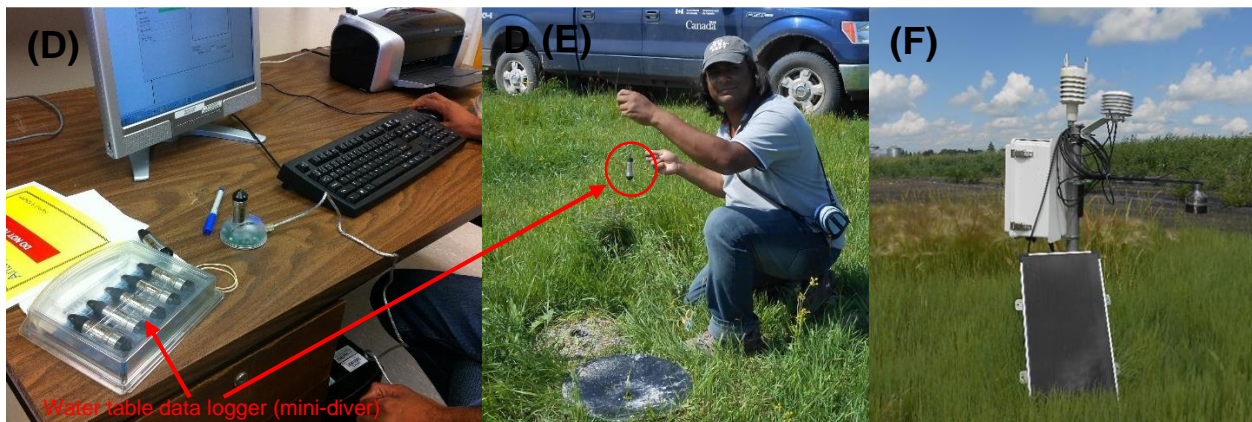
^b The red line shows linear fit, the shaded area shows 95% of the confidence interval, values in the correspondent box indicates individual correlation coefficients (r), and p values indicate the level of significance.

^c *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; and rests are not significant ($p > 0.05$).

^d GWT = depth to groundwater table, GW = groundwater, EC = electrical conductivity, VSWC = volumetric soil water content, TDS = total dissolved salts, ESP = exchangeable sodium percentage.

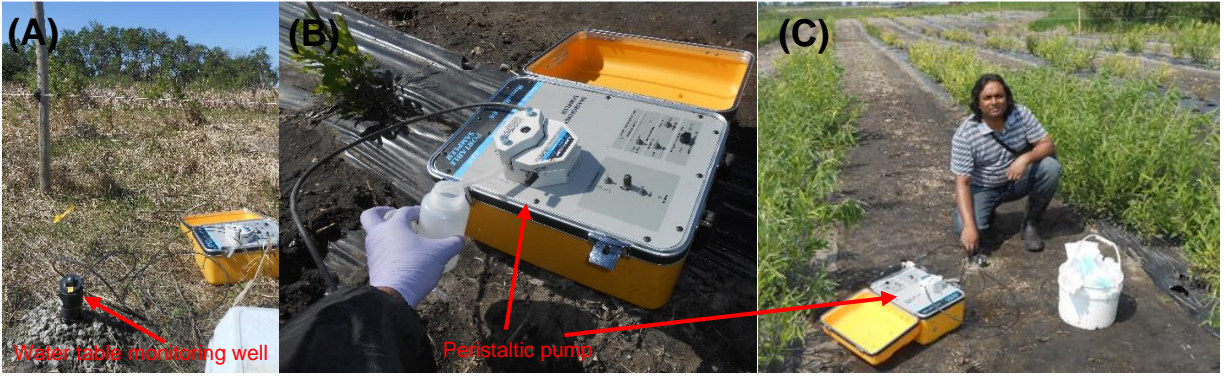


Installation of groundwater monitoring wells

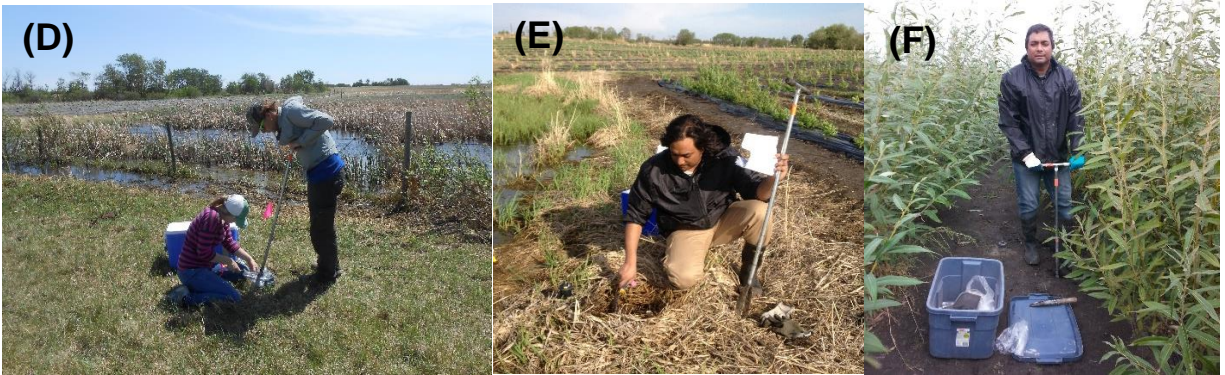


Installation water table data loggers (mini-diver) and mini weather station

Figure A-S 11 Photographs of the installation of shallow groundwater monitoring wells, water table data loggers (mini-diver), and weather station in the field experimental sites.



Groundwater sampling from the water table monitoring wells using peristaltic pump



Soil sampling



EM38 survey to measure apparent electrical conductivity of soil

Figure A-S 12 Photographs of the groundwater collection, soil sampling, and EM38 survey during the experimental period.

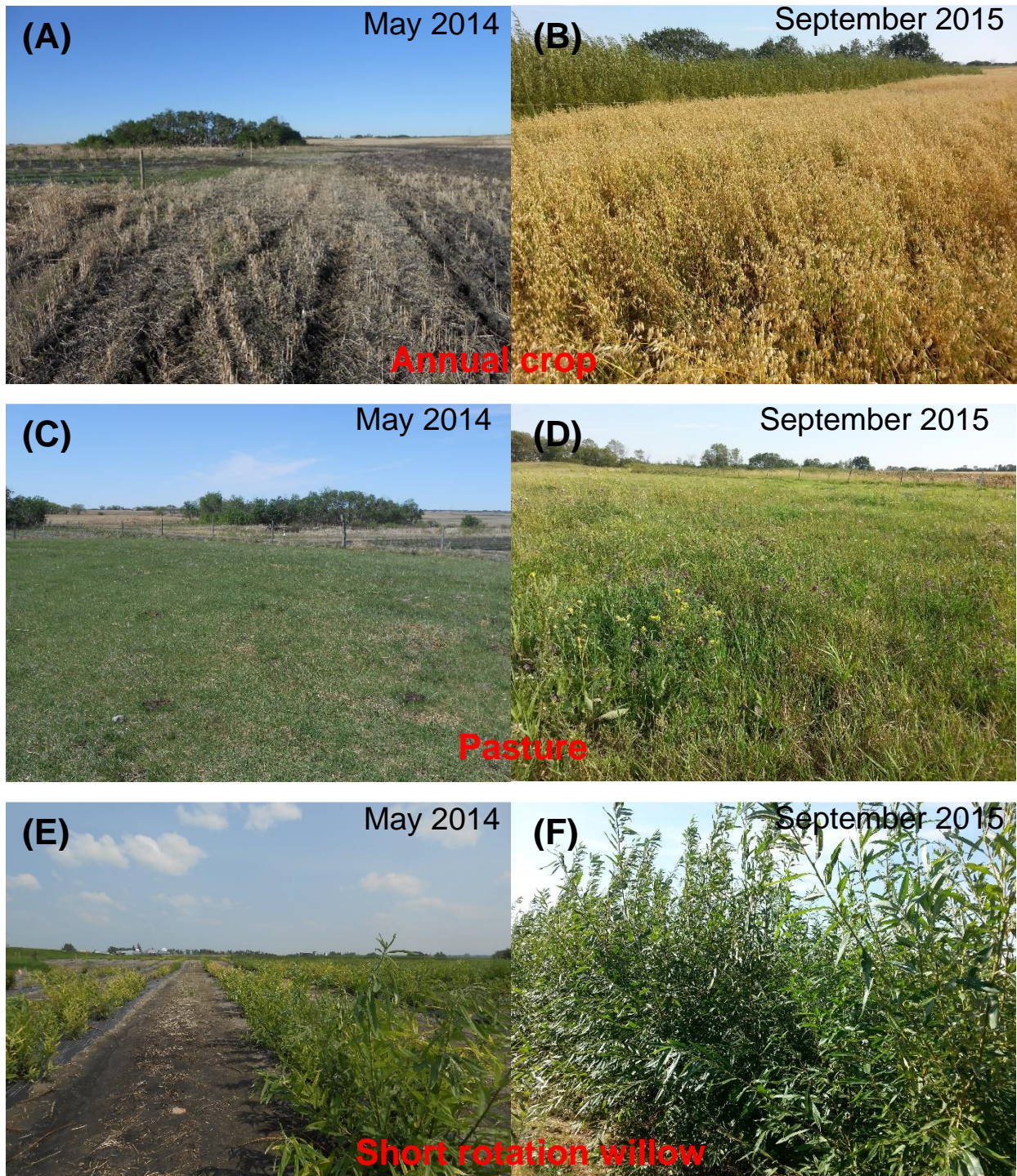


Figure A-S 13 Photographs of experimental field sites under annual crop (A and B), pasture (C and D), and short rotation willow (E and F) taken in May 2014 and September 2015.

APPENDIX B. SUPPLEMENTARY MATERIALS FOR CHAPTER 4

Table B-S 1 Mean (\pm SD) soil nutrients under different land-use practices from both sites.

Variable	Depth	Year	Site A			Site B		
			AC	PA	SRW	AC	PA	SRW
NH ₄ ⁺ -N (mg kg ⁻¹)	0-15cm	2014	3.93 \pm 1.96bB	3.82 \pm 0.30aB	3.99 \pm 1.99bB	7.44 \pm 5.05aB	5.49 \pm 0.83aB	3.56 \pm 1.76aB
		2015	6.37 \pm 2.58bA	8.89 \pm 2.77aA	6.27 \pm 2.62bA	7.02 \pm 2.51aA	10.25 \pm 3.68aA	7.42 \pm 2.83aA
	15-30cm	2014	3.08 \pm 1.16bB	2.78 \pm 0.23aB	3.19 \pm 0.74bB	4.17 \pm 1.46aB	3.59 \pm 0.99aB	2.94 \pm 0.81aB
		2015	5.24 \pm 1.39bA	6.17 \pm 1.56aA	5.51 \pm 2.88bA	6.59 \pm 1.57aA	6.76 \pm 1.71aA	7.77 \pm 3.84aA
	30-60cm	2014	2.71 \pm 0.64bB	2.48 \pm 0.27aB	2.99 \pm 0.60bB	3.54 \pm 0.47aB	3.93 \pm 1.15aB	3.10 \pm 0.99aB
		2015	4.54 \pm 1.85bA	5.77 \pm 1.22aA	5.47 \pm 1.65bA	6.34 \pm 0.98aA	4.57 \pm 1.49aA	7.29 \pm 3.03aA
NO ₃ ⁻ -N (mg kg ⁻¹)	0-15cm	2014	9.24 \pm 10.17aB	7.84 \pm 6.29aB	13.58 \pm 14.05aB	7.11 \pm 8.02bA	2.74 \pm 0.81bA	24.37 \pm 19.75aA
		2015	18.18 \pm 16.22aA	17.91 \pm 14.15aA	20.26 \pm 19.62aA	13.09 \pm 11.40bA	18.85 19.47bA	21.53 \pm 17.02aA
	15-30cm	2014	3.63 \pm 2.10aB	3.05 \pm 1.62aB	11.88 \pm 12.33aB	2.31 \pm 1.16bA	2.82 \pm 1.73bA	16.45 \pm 16.73aA
		2015	9.69 \pm 10.89aA	6.38 \pm 2.96aA	9.95 \pm 8.26aA	8.28 \pm 5.58bA	15.30 \pm 14.26bA	10.43 \pm 7.89aA
	30-60cm	2014	2.19 \pm 0.91aB	2.13 \pm 0.61aB	5.80 \pm 5.27aB	1.75 \pm 0.63bA	1.45 \pm 0.18bA	8.20 \pm 8.75aA
		2015	6.21 \pm 7.52aA	11.54 \pm 13.69aA	5.75 \pm 4.09aA	4.90 \pm 4.02bA	7.43 \pm 7.47bA	8.73 \pm 9.33aA
PO ₄ ³⁻ -P (mg kg ⁻¹)	0-15cm	2014	37.00 \pm 11.25bA	31.41 \pm 7.78bA	41.42 \pm 14.88aA	37.34 \pm 8.84bB	31.51 \pm 3.50bB	47.29 \pm 7.97aB
		2015	41.76 \pm 12.02bA	52.56 \pm 17.81bA	50.37 \pm 24.31aA	47.88 \pm 14.19bA	29.46 \pm 8.76bA	74.20 \pm 24.61aA
	15-30cm	2014	21.25 \pm 5.90bA	21.73 \pm 8.37bA	24.25 \pm 12.13aA	22.03 \pm 5.29bB	23.48 \pm 2.76bB	22.82 \pm 5.96aB
		2015	20.56 \pm 8.31bA	26.64 \pm 11.48bA	26.51 \pm 16.60aA	28.89 \pm 5.33bA	22.61 \pm 7.37bA	33.79 \pm 11.99aA
	30-60cm	2014	17.76 \pm 5.72bA	13.82 \pm 2.03bA	22.61 \pm 7.28aA	16.46 \pm 4.75bB	18.93 \pm 5.37bB	18.06 \pm 4.15aB
		2015	15.52 \pm 5.24bA	18.30 \pm 5.64bA	22.60 \pm 11.49aA	15.80 \pm 3.42bA	21.76 \pm 9.74bA	26.18 \pm 9.07aA
K ⁺ (mg kg ⁻¹)	0-15cm	2014	448.77 \pm 168.16aB	295.90 \pm 91.60bB	439.40 \pm 175.87bB	310.75 \pm 210.49aB	299.10 \pm 134.04aB	330.14 \pm 167.71aB
		2015	832.55 \pm 133.34aA	811.30 \pm 371.22bA	674.12 \pm 204.50bA	654.68 \pm 118.05aA	680.88 \pm 112.80aA	712.25 \pm 172.14aA
	15-30cm	2014	297.77 \pm 143.27aB	128.57 \pm 70.90bB	221.59 \pm 141.90bB	209.77 \pm 141.27aB	112.22 \pm 51.08aB	160.03 \pm 115.11aB
		2015	495.61 \pm 138.12aA	338.63 \pm 97.40bA	414.22 \pm 212.44bA	588.76 \pm 205.91aA	436.43 \pm 55.19aA	413.26 \pm 156.68aA
	30-60cm	2014	200.17 \pm 150.98aB	63.33 \pm 53.14bB	187.56 \pm 135.45bB	140.20 \pm 83.43aB	105.10 \pm 79.09aB	100.19 \pm 74.28aB
		2015	390.56 \pm 164.21aA	297.45 \pm 224.84bA	360.39 \pm 164.79bA	398.29 \pm 57.05aA	316.07 \pm 69.93aA	329.22 \pm 89.03aA

Table B-S 1 – continued

SO ₄ ²⁻ -S (mg kg ⁻¹)	0-15cm	2014	186.61± 327.64bB	13.49± 5.62bB	327.14± 527.89aB	1306.49± 1690.89bA	149.00± 91.55cA	2320.89± 1419.32aA
		2015	234.26± 376.91bA	54.25± 22.44bA	339.17± 361.71aA	2236.57± 1860.61bA	255.35± 160.69cA	2792.76± 1550.18aA
	15-30cm	2014	143.74± 265.77bB	131.57± 117.08bB	332.87± 537.16aB	1448.59± 1798.04bA	100.98± 95.99cA	3321.32± 3067.62aA
		2015	166.73± 255.98bA	154.22± 23.10bA	314.86± 338.90aA	1910.37± 1801.59bA	133.82± 69.07cA	2944.57± 1926.92aA
	30-60cm	2014	127.62± 251.83bB	354.58± 135.99bB	463.45± 733.38aB	1071.20± 1018.73bA	70.39± 62.48cA	3690.33± 3194.76aA
		2015	216.49± 413.95bA	909.22± 811.92bA	922.46± 1310.06aA	1544.98± 1381.74bA	99.25± 30.49cA	3164.21± 1945.99aA

^a Values represent mean ± standard deviations (±SD). Means were calculated from the values of soil nutrients measured from May and September every year.

^b Means within a row for land-use followed by the same small letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c Means within a column for year supported by the same capital letter are not significantly different ($p > 0.05$) using Tukey HSD.

^d SD = standard deviation, AC = annual crop, PA = pasture, SRW = short rotation willow.

Table B-S 2 Analysis of variance (ANOVA) with a linear mixed-effects model for measured nutrients in groundwater under three land-use practices from two sites during the growing season of two consecutive years.

Response variable	Sources of variation	Site A			Site B	
		df	F - value	p - value	F - value	p - value
NH ₄ ⁺ -N	Land-use	1	4.759	0.011 [*]	4.245	0.018 [*]
	Year	1	9.737	0.003 ^{**}	12.851	<0.001 ^{***}
	Month	2	1.732	0.183 ^{ns}	4.241	0.018 [*]
NO ₃ ⁻ -N	Land-use	1	3.284	0.042 [*]	3.636	0.031 [*]
	Year	1	3.197	0.078 ^{ns}	2.057	0.156 ^{ns}
	Month	2	3.393	0.038 [*]	0.596	0.554 ^{ns}
PO ₄ ³⁻ -P	Land-use	1	0.248	0.781 ^{ns}	0.972	0.383 ^{ns}
	Year	1	12.593	<0.001 ^{***}	102.595	<0.001 ^{***}
	Month	2	7.857	<0.001 ^{***}	5.386	0.007 ^{**}
K ⁺	Land-use	1	8.166	<0.001 ^{***}	19.882	<0.001 ^{***}
	Year	1	17.119	<0.001 ^{***}	44.927	<0.001 ^{***}
	Month	2	0.003	0.997 ^{ns}	0.706	0.497 ^{ns}
SO ₄ ²⁻ -S	Land-use	2	8.296	<0.001 ^{***}	8.132	<0.001 ^{***}
	Year	1	2.110	0.150 ^{ns}	1.092 ^{ns}	0.299 ^{ns}
	Month	2	1.110	0.335 ^{ns}	0.610 ^{ns}	0.546 ^{ns}

^a *, **, *** Indicate there is a statistically significant difference at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; ^{ns}, is not significantly different ($p > 0.05$).

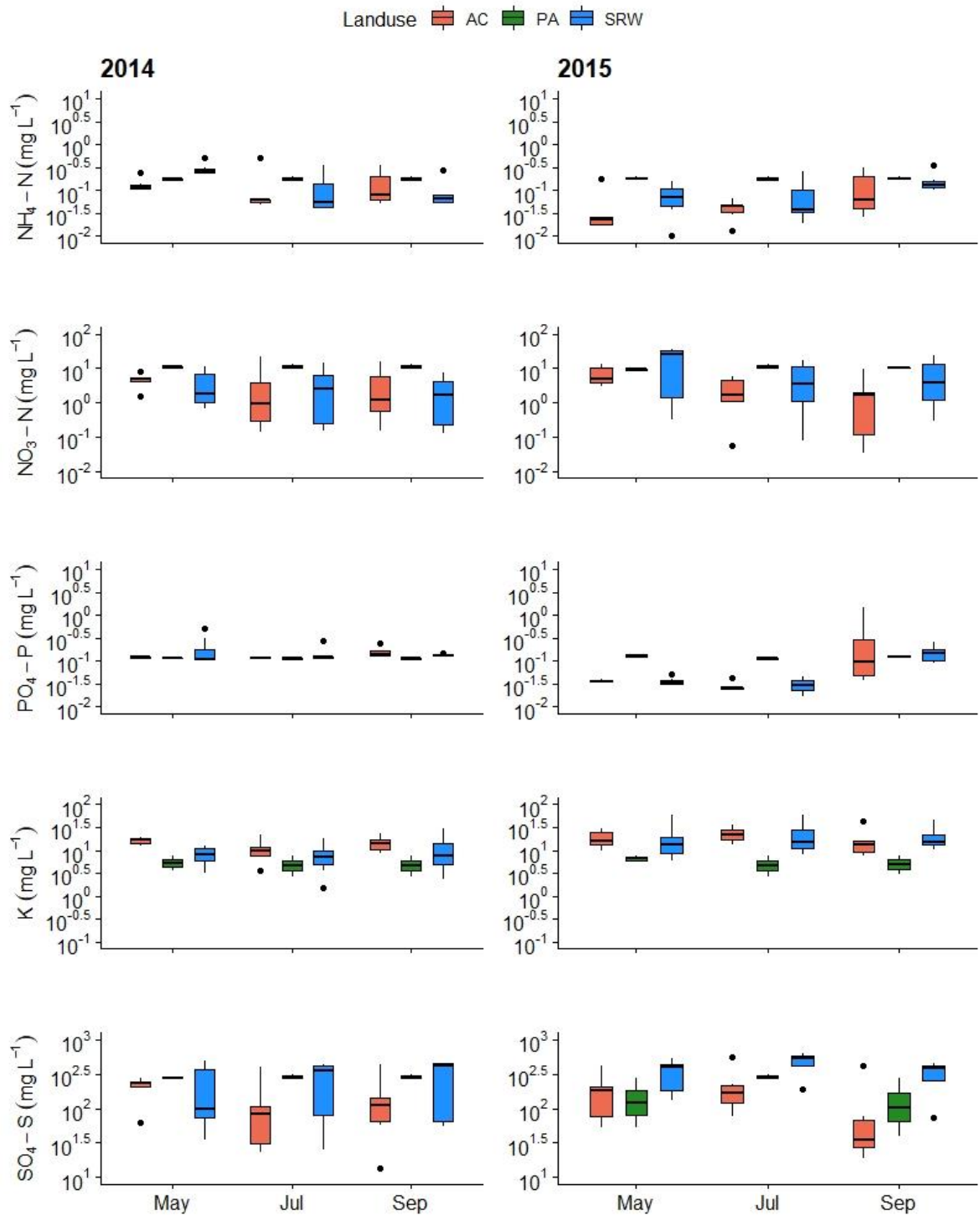


Figure B-S 1 Seasonal and annual variation of $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, K^+ , and $\text{SO}_4^{2-}\text{-S}$ concentration in groundwater from three different land-use practices during the growing seasons of 2014 and 2015 from site A.

^a Y-axis is transformed into \log_{10} .

^b AC = annual crop, PA = pasture, SRW = short rotation willow.

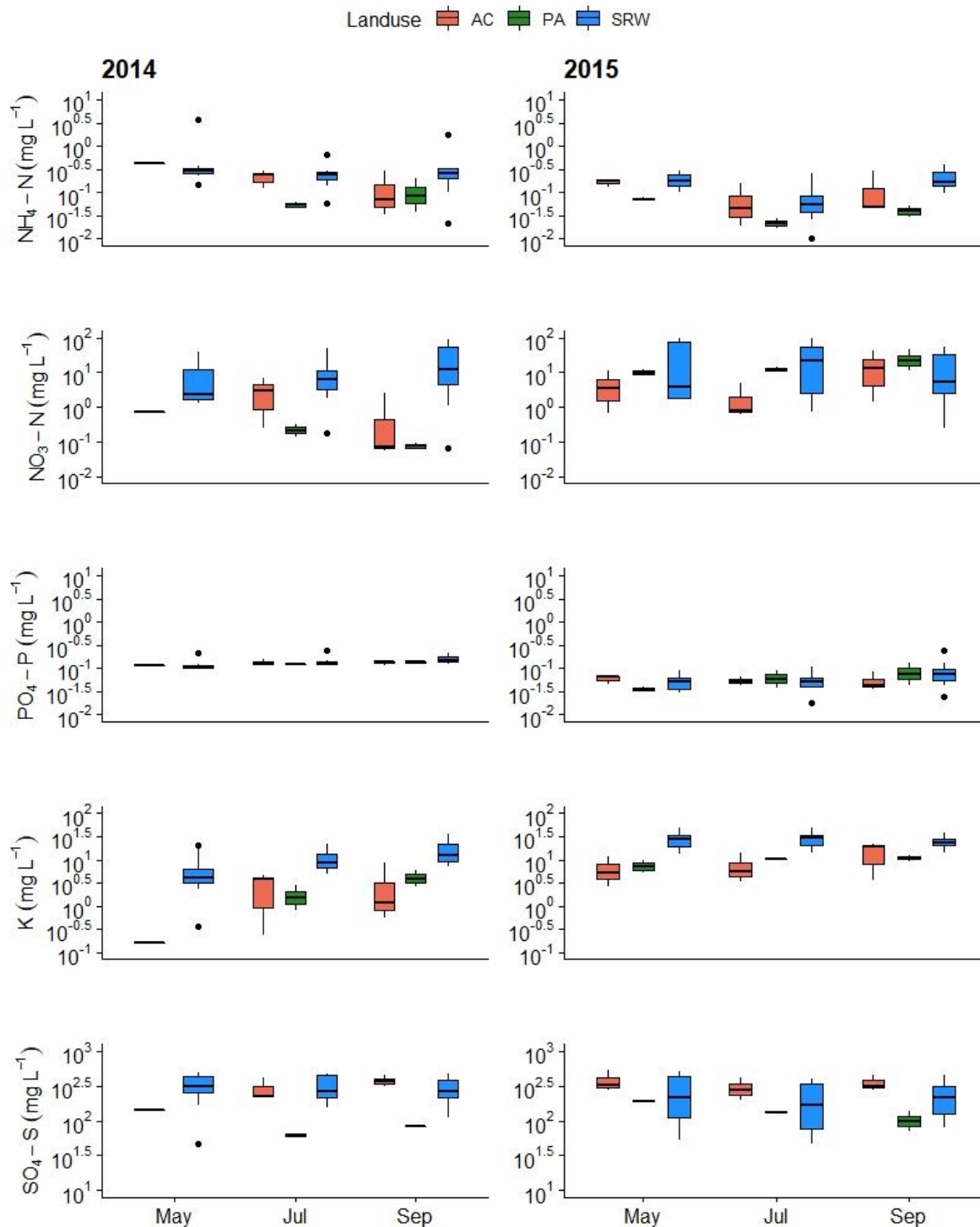


Figure B-S 2 Seasonal and annual variation of $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, K^+ , and $\text{SO}_4^{2-}\text{-S}$ concentration in groundwater from three different land-use practices during the growing seasons of 2014 and 2015 from site B.

^a Y-axis is transformed into \log_{10} .

^b AC = annual crop, PA = pasture, SRW = short rotation willow.

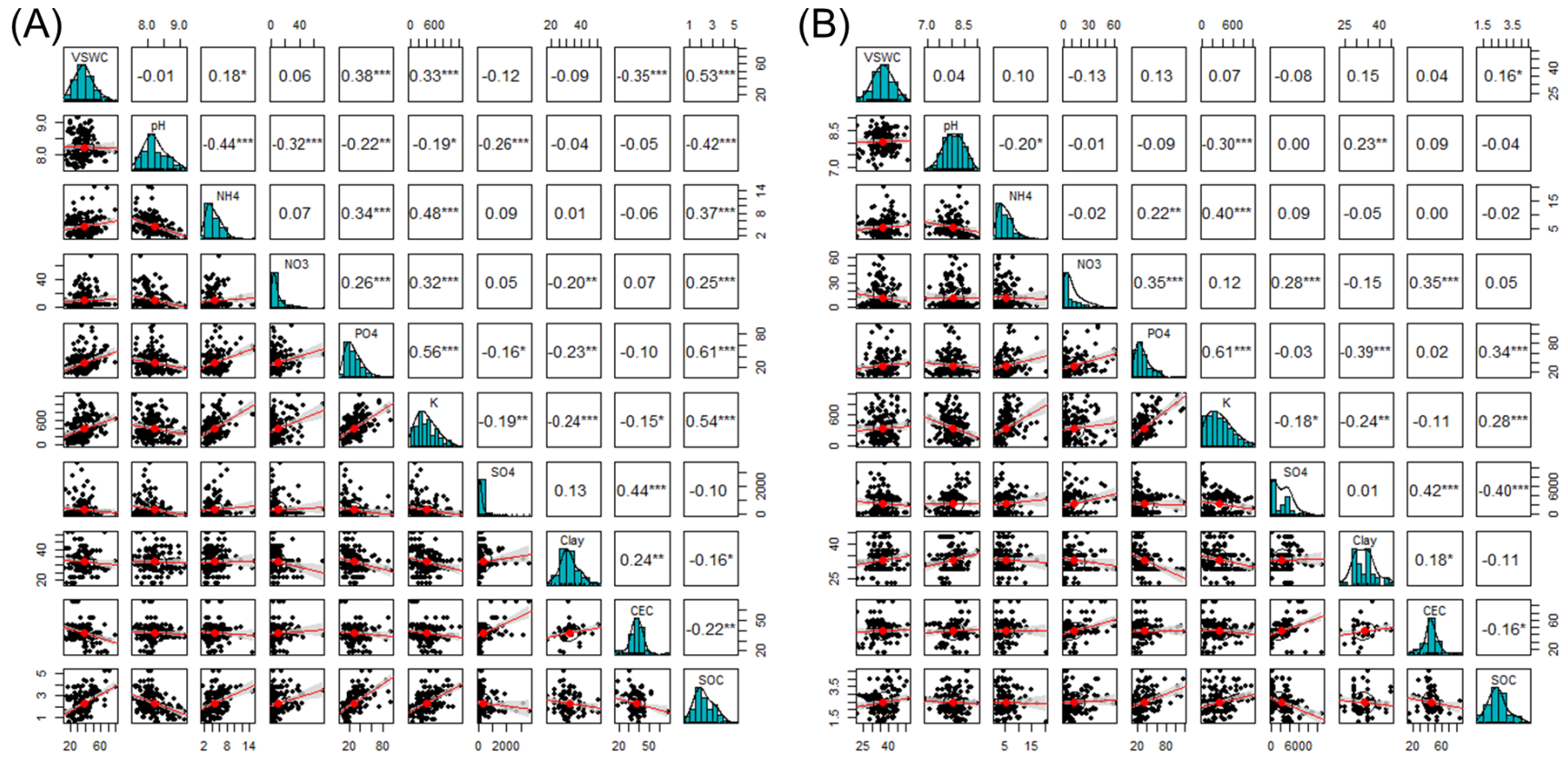


Figure B-S 3 Correlations (Pearson) among soil parameters measured during the field experiment from A) site A, and B) site B.

^a The red line shows linear fit, the shaded area shows 95% of the confidence interval, values in the correspondent box indicates individual correlation coefficients (r), and p values indicate the level of significance.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; and rests are not significant ($p > 0.05$).

^c VSWC = volumetric soil water content, CEC = cation exchange capacity, SOC = soil organic carbon.

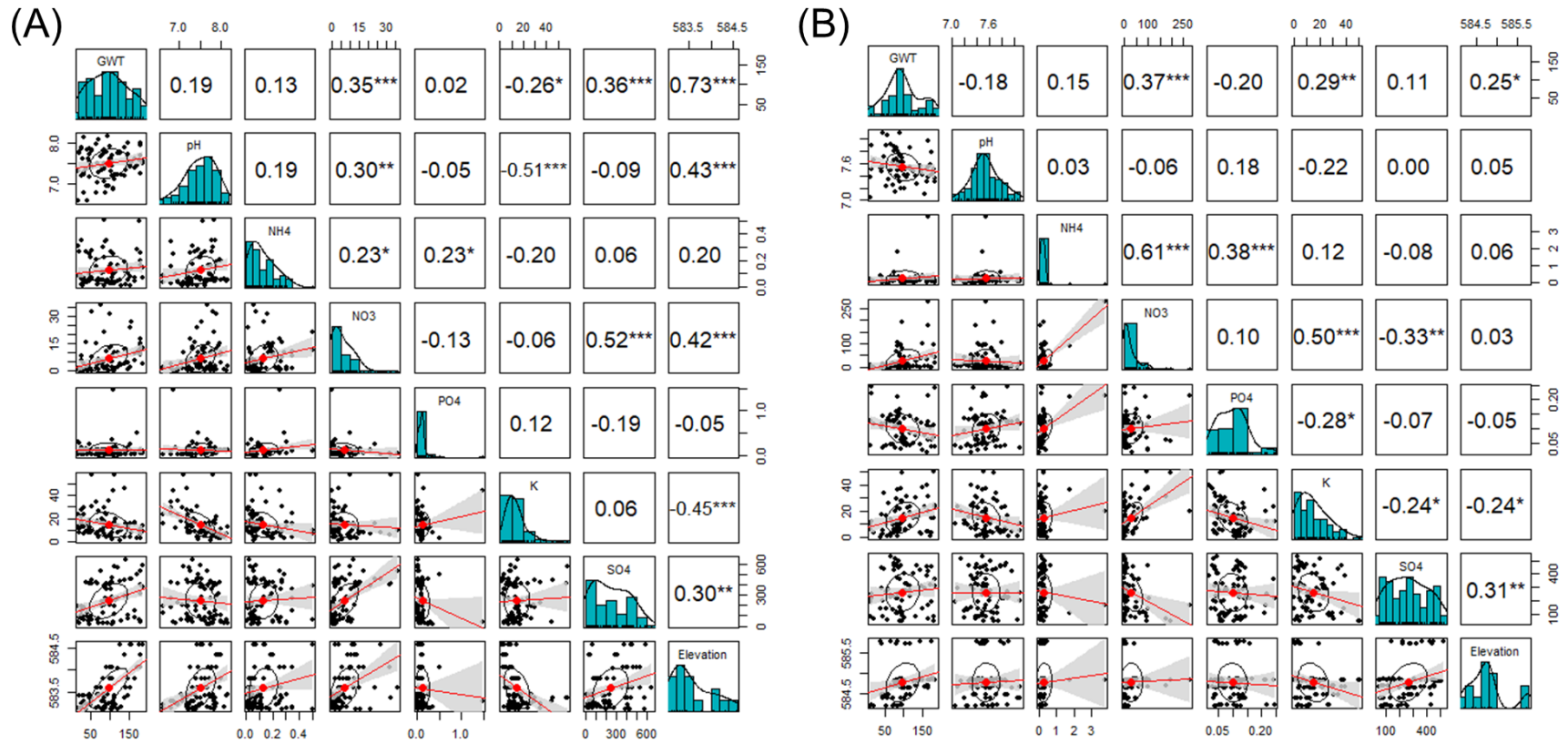


Figure B-S 4 Correlations (Pearson) among groundwater parameters measured during the field experiment from A) site A, and B) site B.

^a The red line shows linear fit, the shaded area shows 95% of the confidence interval, values in the correspondent box indicates individual correlation coefficients (r), and p values indicate the level of significance.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; and rests are not significant ($p > 0.05$).

^c GWT = depth to the groundwater table.

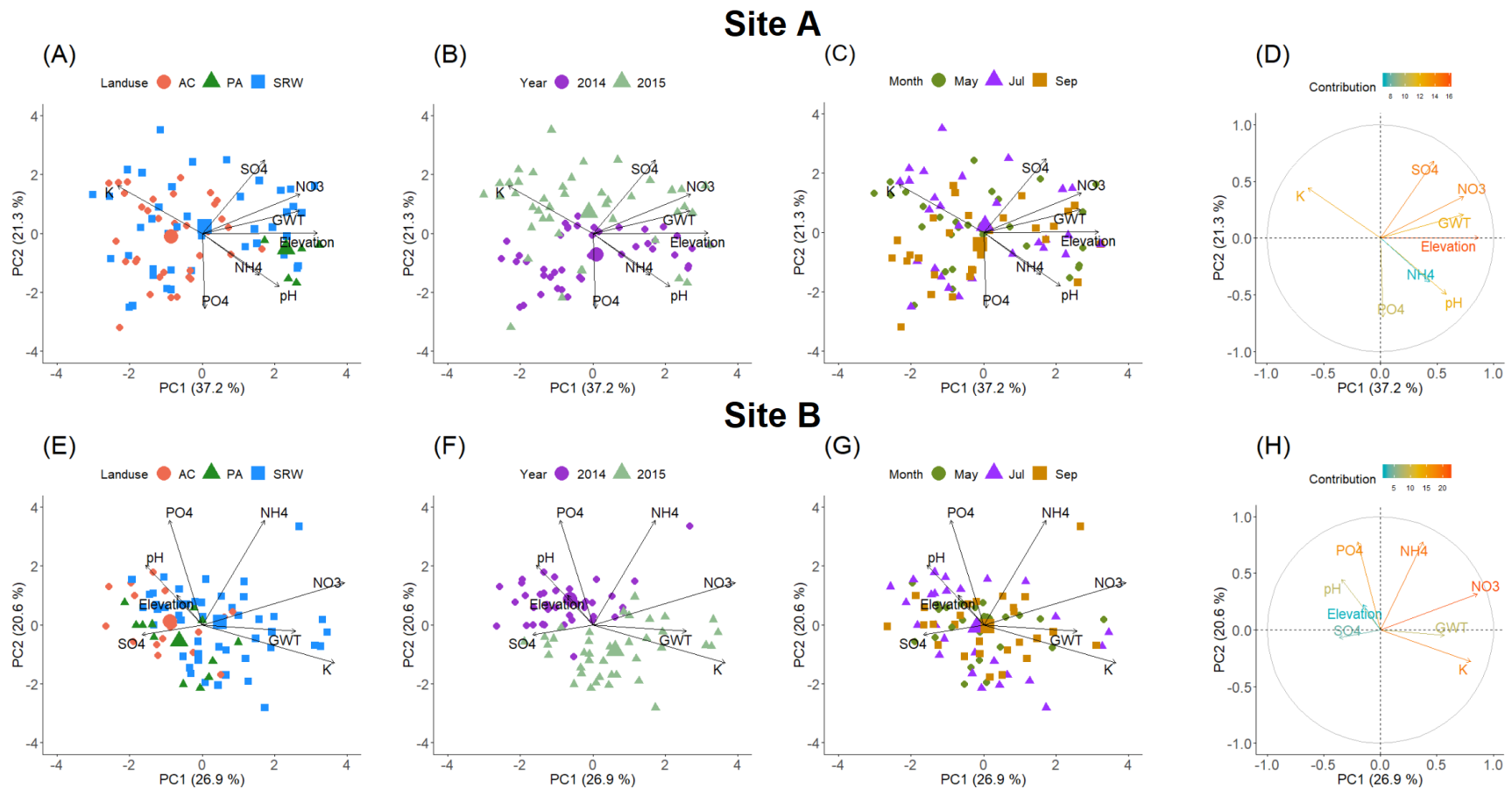


Figure B-S 5 Principal Component Analysis (PCA) of measured depth to GWT, pH, NH₄⁺-N, NO₃⁻-N, PO₄³⁻-P, K⁺, SO₄²⁻-S content, and elevation as observed variables based on land-use practices, years, and months of as factor variable, and contribution of variables from site A (A, B, C, D); and site B (D, E, F, G), respectively.

^a Data was transformed to square root.

^b Contribution (in percentage) of the variables to the principal components.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, GWT = depth to groundwater table.

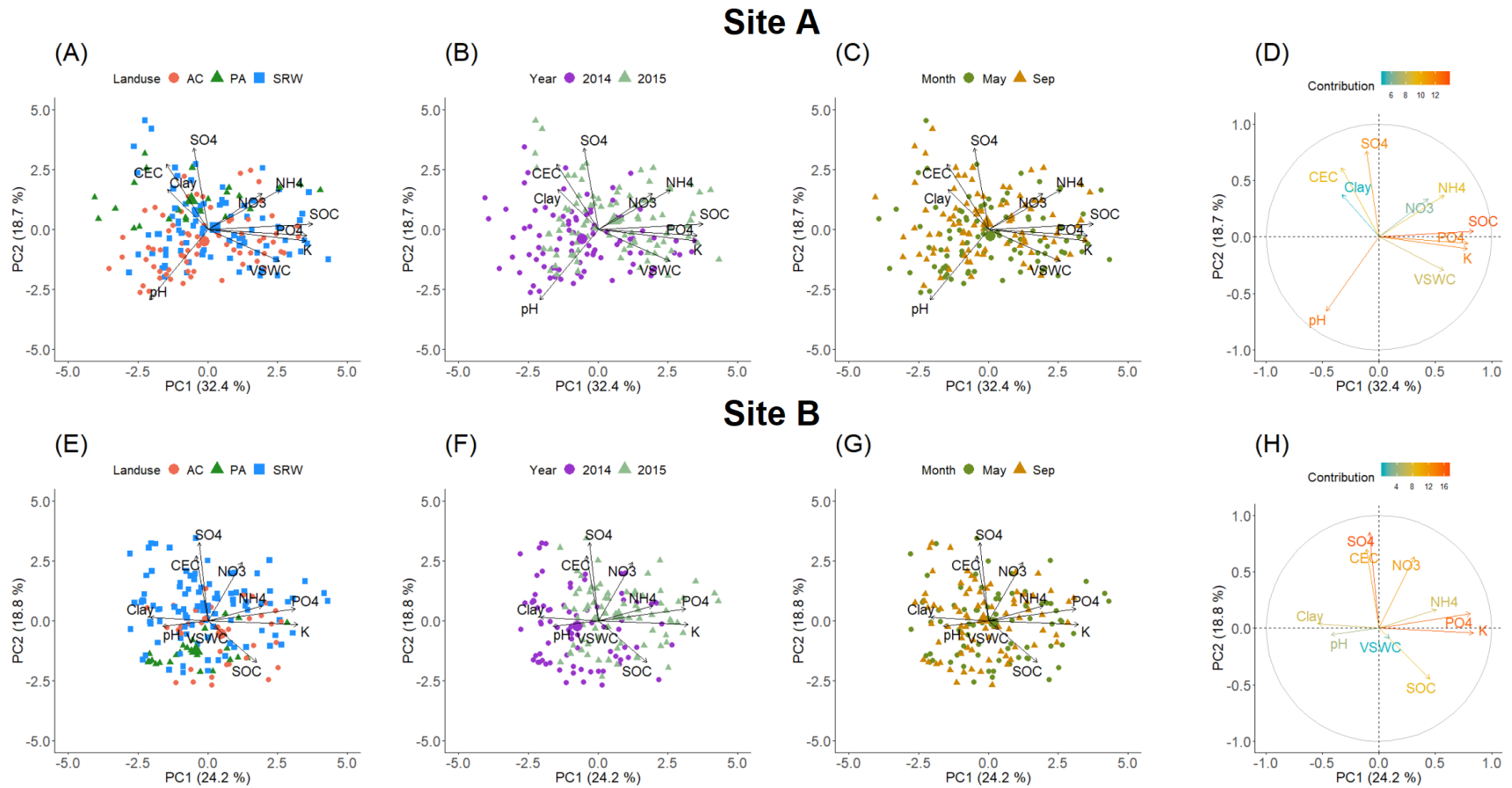


Figure B-S 6 Principal component analysis (PCA) of measured VSWC, pH, $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$, K^+ , $\text{SO}_4^{2-}\text{-S}$, SOC, CEC, and clay content as observed variables based on land-use practices, depths, years, and months as a factor variable, and contribution of variables from site A (A, B, C, D, E), and site B (F, G, H, I, J), respectively.

^a Data was transformed to square root.

^b Contribution (in percentage) of the variables to the principal components.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, VSWC = volumetric soil water content, CEC = cation exchange capacity, SOC = soil organic carbon, GWT = depth to groundwater table.

APPENDIX C. SUPPLEMENTARY MATERIALS FOR CHAPTER 5

Table C-S 1 Physical and chemical characteristics of soils from different land-use practices.

	Site A					Site B		
	Depth	Year	AC	PA	SRW	AC	PA	SRW
Bulk density (g cm ⁻³)	0-15cm	2013	1.4±0.10aA	1.3±0.01bA	1.2±0.14abA	1.3±0.08aB	1.2±0.05aB	1.2±0.07aB
	15-30cm		1.4±0.16aA	1.2±0.05bA	1.3±0.02abA	1.3±0.14aA	1.2±0.13aA	1.4±0.06aA
Clay (%)	0-15cm	2013	27.0±2.6bB	34.0±1.1aB	28.0±5.9bB	29.0± 5.2bB	32.0±3.3aB	30.0±1.8bB
	15-30cm		32.0±3.2bA	43.0±8.8aA	31.0±6.5bA	33.0±7.9bA	40.0±5.5aA	34.0±2.0bA
VSWC (%)	0-15cm	2013	34.0±5.8aB	33.3±6.7aB	38.7±6.9aB	35.0±8.07aB	40.3±3.48aB	35.2±4.80aB
		2014	45.0±11.0aA	31.0±4.0aA	43.0±15.0aA	43.0±1.60aA	41.0±6.20aA	39.0±6.30aA
		2015	40.0±9.9aB	25.0±10.0aB	30±10.7aB	32.0±1.25aB	36.0±0.15aB	32.0±3.62aB
EC (mS cm ⁻¹)	0-15cm	2013	0.75±0.44aA	0.88±0.19aA	1.15±0.91aA	2.30±1.80aA	0.78±0.14bA	2.54±0.92aA
		2014	0.87±0.76aA	0.35±0.07aA	0.82±0.53aA	2.4±2.40aA	0.8±0.08bA	3.7±1.78aA
		2015	0.83±0.60aA	0.34±0.18aA	0.99±0.57aA	2.9±2.26aA	1.2±0.02bA	3.5±1.70aA
pH	0-15cm	2013	8.0±0.33aA	8.1±0.51aA	7.9±0.43aA	7.8±0.36bA	8.2±0.45aA	8.0±0.20aA
		2014	8.1±0.33aA	7.7±0.12aA	8.1±0.14aA	7.6±0.22bA	8.3±0.47aA	8.0±0.20aA
		2015	8.0±0.32aA	7.7±0.10aA	7.9±0.16aA	7.4±0.26bA	8.1±0.37aA	8.0±0.17aA
TSC (Mg ha ⁻¹)	0-15cm	2013	71.0±24.0aA	72.0±20.0aA	79.0±32.0aA	58.0±23.2bA	83.0±23.1aA	61.2±17.0bA
		2014	89.0±28.9aA	87.0±4.3aA	92.0±32.8aA	69.0±8.5bA	95.0±17.9aA	68.0±17.8bA
		2015	76.0±18.4aA	83.0±2.8aA	83.0±18.7aA	58.0±8.8bA	88.0±12.1aA	62.0±11.5bA
TN (Mg ha ⁻¹)	0-15cm	2013	4.7±0.49aA	4.6±0.60aA	5.2±0.87aA	4.87±0.74bA	5.50±0.37aA	4.90±0.47bA
		2014	4.8±1.60aA	6.5±1.21aA	4.9±1.51aA	5.30±0.38bA	5.91±0.09aA	4.60±1.07bA
		2015	5.0±1.09aA	6.6±1.07aA	4.8±0.86aA	4.80±0.69bA	5.80±0.79aA	4.89±0.73bA
C/N ratio	0-15cm	2013	11.0±0.44aA	11.0±0.67aA	11.0±0.37aA	11.0±0.61aA	12.0±0.44aA	11.0±0.38aA
		2014	10.4±0.66aA	9.8±0.62aA	11.6±1.61aA	12.0±1.20aA	13.0±3.20aA	11.0±2.40aA
		2015	12.0±0.86aA	12.0±1.75aA	12.0±3.38aA	10.2±0.45aA	9.9±2.50aA	11.1±1.17aA
NH ₄ ⁺ -N (mg kg ⁻¹)	0-15cm	2013	12.0±11.20aA	14.0±1.60aA	26.0±26.00aA	7.3±4.60aA	7.5±1.70aA	6.6±6.80aA
		2014	2.9±0.68aB	3.9±0.24aB	3.0±0.84aB	3.7±0.80aB	5.3±0.74aB	2.9±0.43aB
		2015	4.7±1.83aB	6.5±0.67aB	5.5±0.50aB	5.8±2.19aA	7.6±2.99aA	5.9±0.83aA
NO ₃ ⁻ -N (mg kg ⁻¹)	0-15cm	2013	19.0±16.0aB	11.0±2.1aB	16.0±16.1aB	3.0±0.5aB	8.6±6.7aB	32.1±21.0aB
		2014	7.5±4.0aB	9.2±10.0aB	15.5±14.0aB	2.4±0.59aB	3.3±0.70aB	23.2±19.98aB

2015	33.0±8.8aA	24.0±21.1aA	29.0±21.1aA	23.0±4.4aA	36.0±3.1aA	20.0±9.8aA
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Table C-S 1 – continued

PO ₄ ³⁻ -P (mg kg ⁻¹)	0-15cm	2013	34.0±14.1aA	30.0±1.1aA	47.0±16.3aA	55.0±10.8aA	59.0±4.4aA	46.0±15.2aA
		2014	41.0±11.6aA	36.0±9.3aA	48.0±17.0aA	36.0±13.1aA	32.0±4.6aA	48.0±6.7aA
		2015	35.0±3.9aA	38.0±3.1aA	37.0±10.1aA	36.0±5.3aA	36.0±7.5aA	59.0±13.3aA
SO ₄ ²⁻ -S (mg kg ⁻¹)	0-15cm	2013	249.0±372.0aA	46.0±31.0bA	376.0±443.0aA	1260.0±1871.4aA	72.0±6.8bA	2265.0±1366.0aA
		2014	220.6±359.2aA	9.2±3.7bA	442.6±638.6aA	1123.0±1594.3aA	71.0±1.9bA	2144.0±1281.0aA
		2015	314.0±497.0aA	56.0±24.0bA	356.0±301.0aA	2659.0±2290.0aA	393.0±23.0bA	3064.0±1801.0aA
LFON (g fraction N g ⁻¹ soil N)	0-15cm	2014	0.021±0.018abA	0.036±0.010aA	0.007±0.003bA	0.004±0.002bA	0.015±0.003aA	0.008±0.005bA
		2015	0.019±0.022abA	0.041±0.004aA	0.008±0.002bA	0.009±0.004bA	0.016±0.003aA	0.011±0.005bA
TDN (mg N ha ⁻¹ soil)	0-15cm	2014	20.0±6.0aA	30.0±31.0aA	26.0±20.0aA	23.0±6.10aA	18.0±9.50aA	41.0±25.10aA
		2015	36.0±18.0aA	34.0±22.0aA	31.0±20.0aA	25.0±9.11aA	39.0±0.50aA	21.0±7.90aA

^a Values represent mean ± standard deviations (±SD).

^b Means within a row for land-use followed by the same small letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c Means within a column for depth (for bulk density and clay) and years supported by the same capital letter are not significantly different ($p > 0.05$) using Tukey HSD.

^d AC = annual crop, PA = pasture, SRW = short rotation willow, VSWC = volumetric soil water content, EC = electrical conductivity, TSC = total soil carbon, TN = total nitrogen, LFON = light fraction organic nitrogen, TDN = total dissolved nitrogen.

Table C-S 2 Mean (\pm SD) relative abundance of FTIR absorbance intensities of the SOC chemical functional groups under different land-use practices and depths in both sites.

			1030 = Polysaccharides	1420 = Phenolic	1510 = Amides	1630 = Aromatic	1720 = Carboxylic
Site A	Land-use	AC	0.1647 \pm 0.0839a	0.0501 \pm 0.0526ab	0.0305 \pm 0.0265ab	0.0098 \pm 0.0070a	0.0009 \pm 0.0006a
		PA	0.0910 \pm 0.0219a	0.0025 \pm 0.0003b	0.0027 \pm 0.0003b	0.0067 \pm 0.0019a	0.0008 \pm 0.0003a
		SRW	0.1334 \pm 0.0547a	0.0632 \pm 0.0506a	0.0347 \pm 0.0225a	0.0068 \pm 0.0053a	0.0005 \pm 0.0004a
	Depth	0-15 cm	0.1252 \pm 0.0676a	0.0247 \pm 0.0230b	0.0178 \pm 0.0149b	0.0082 \pm 0.0059a	0.0007 \pm 0.0005a
		15-30 cm	0.1554 \pm 0.0678a	0.0750 \pm 0.0590a	0.0397 \pm 0.0278a	0.0079 \pm 0.0059a	0.0007 \pm 0.0005a
Site B	Land-use	AC	0.1656 \pm 0.0756a	0.0022 \pm 0.0009a	0.0022 \pm 0.0008a	0.0090 \pm 0.0042a	0.0010 \pm 0.0005a
		PA	0.1597 \pm 0.0244a	0.0485 \pm 0.0361a	0.0285 \pm 0.0153a	0.0090 \pm 0.0014a	0.0008 \pm 0.0004a
		SRW	0.1356 \pm 0.0570a	0.0379 \pm 0.0675a	0.0236 \pm 0.0318a	0.0073 \pm 0.0048a	0.0007 \pm 0.0006a
	Depth	0-15 cm	0.1256 \pm 0.0485b	0.0153 \pm 0.0175a	0.0125 \pm 0.0130a	0.0076 \pm 0.0031a	0.0007 \pm 0.0003a
		15-30 cm	0.1669 \pm 0.0609a	0.0474 \pm 0.0758a	0.0264 \pm 0.0353a	0.0083 \pm 0.0053a	0.0009 \pm 0.0007a

^a Values represent mean \pm standard deviations (\pm SD).

^b Means within a column for land-use and depth followed by the same letter are not significantly different ($p > 0.05$) using Tukey HSD.

^c FTIR = Fourier transform infrared, SOC = soil organic carbon, AC = annual crop, PA = pasture, SRW = short rotation willow.

^d Representative FTIR spectra absorbance band 1030 = polysaccharides (C-O), 1420 = phenolic (C-H), 1510 = amides (N-H), 1630 = aromatic (C=C), 1720 = carboxylic (C=O).

^e Obtained FTIR spectra are divided into five regions: 1) O-alkyl-C = 1030 = polysaccharides (C-O) (cellulose and hemicellulose, but also from proteins and side chains of lignin), 2) alkyl-C = 1420 = phenolic and aliphatic (C-H) (of lipids, fatty acids, and plant aliphatic polymers), 3) amide = 1510 = amides (N-H), 4) aryl-C = 1630 = aromatic (C=C) (deriving from lignin and/or protein), and 5) carbonyl-C = 1720 = carboxylic (C=O) (deriving from aliphatic esters, carboxyl groups, and amide carbonyls).

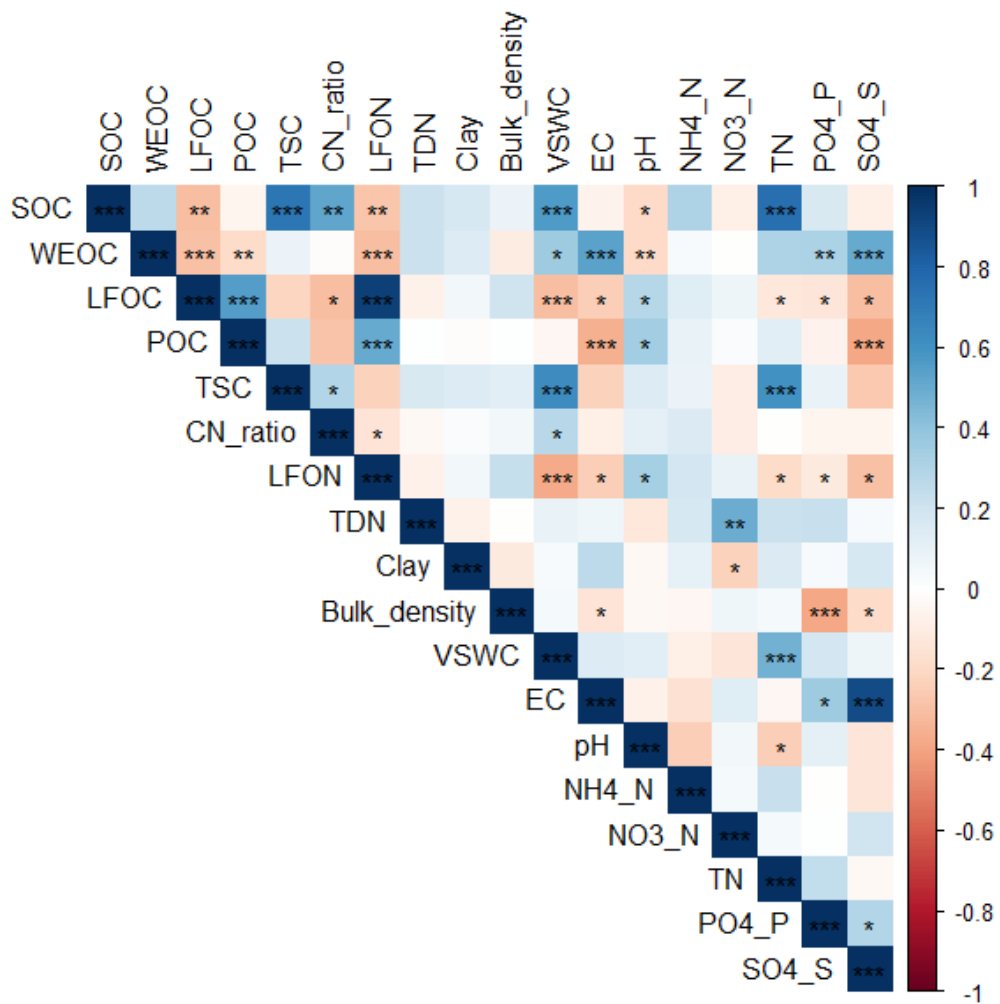


Figure C-S 1 Total soil organic and labile carbon fractions and their relationships with soil physical and chemical characteristics.

^a Negative correlations are depicted in red and positive correlations in blue. Increasing correlation strength is indicated by deeper color.

^b *, **, *** indicate there is a statistically significant relationship at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$ level of significance, respectively; and rests are not significant ($p > 0.05$).

^c SOC = soil organic carbon, WEOC = water extractable organic carbon, LFOC = light fraction organic carbon, POC = particulate organic carbon, TSC = total soil carbon, LFON = light fraction organic nitrogen, TDN = total dissolved nitrogen, VSWC = volumetric soil water content, EC = electrical conductivity.

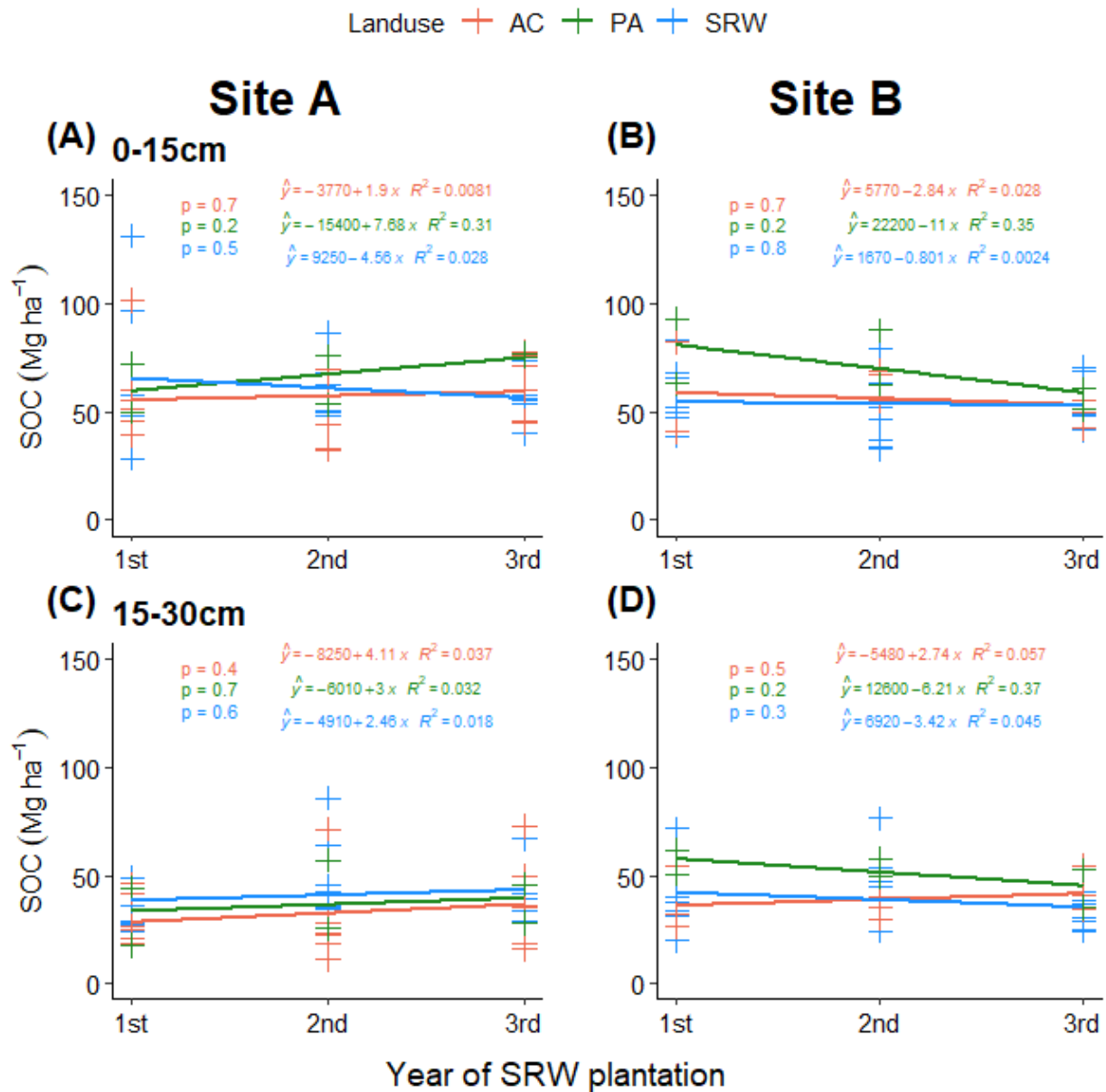


Figure C-S 2 Total soil organic carbon trends (in terms of the year of SRW plantation) from different land-use practices at 0-30cm depth from both sites.

^a The line shows linear regression of the change in SOC measured during the field study, R^2 gives values for individual regression lines, and p -values indicate the level of significance.

^b p -values ≤ 0.05 indicate a significant difference, whereas p -values > 0.05 are not significantly different.

^c AC = annual crop, PA = pasture, SRW = short rotation willow.

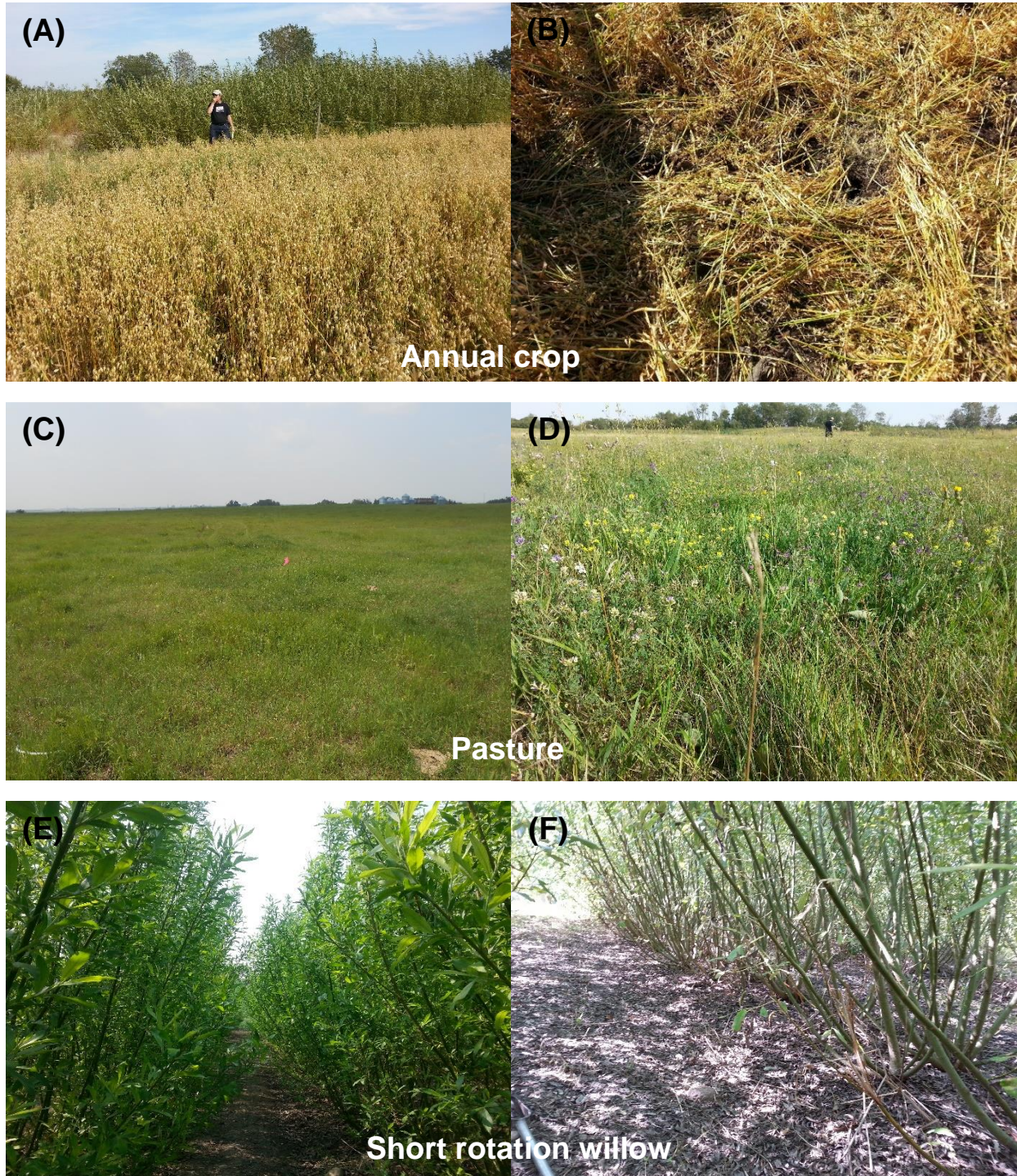


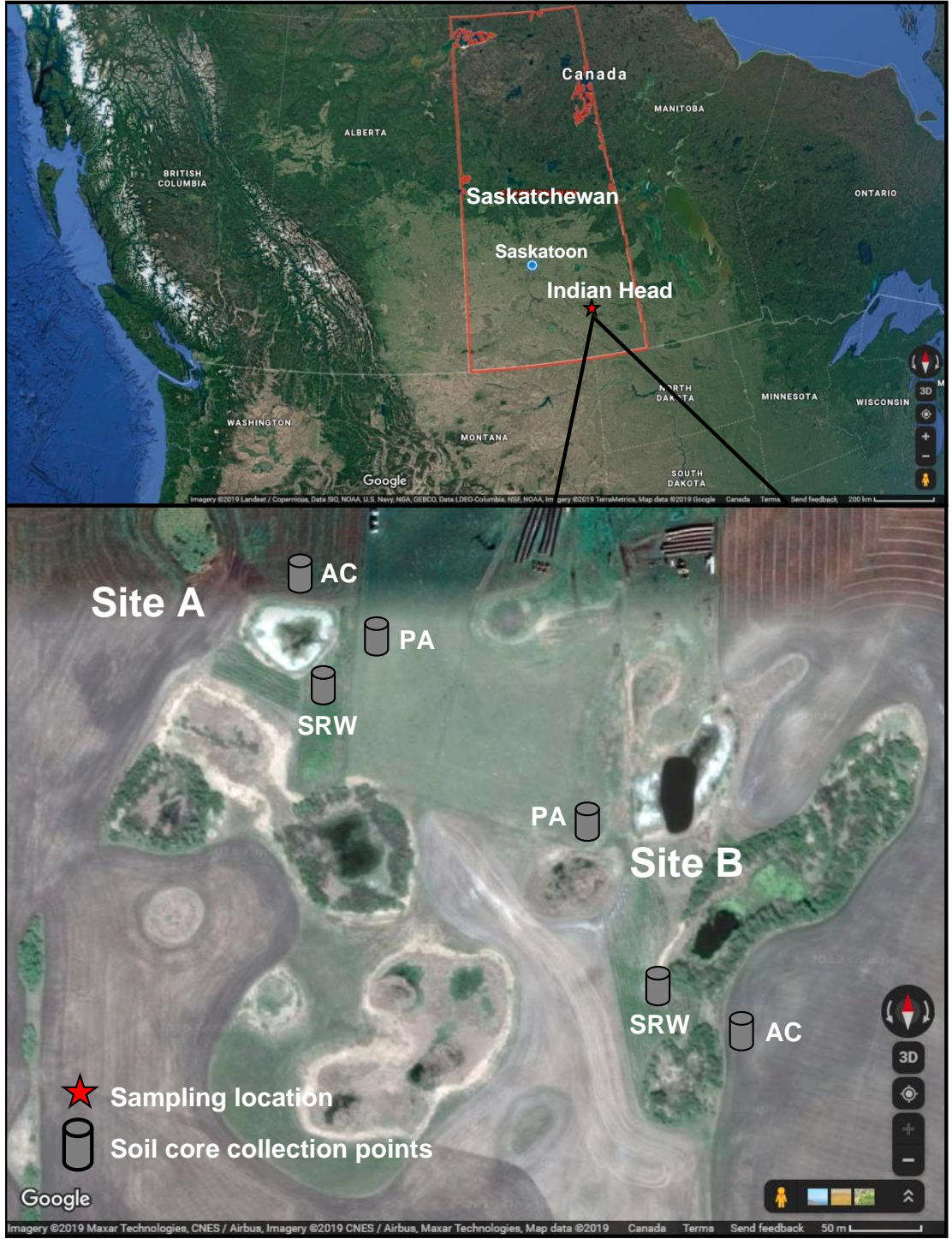
Figure C-S 3 Photographs were taken from different land-use practices during the field experiment in the Fall of 2015. Photographs of the left side represent the land-use practices (A, C, E), and the right side represents the residue status of the representative land-use practices (B, D, F).

APPENDIX D. SUPPLEMENTARY MATERIALS FOR CHAPTER 6

Table D-S 1 Physiochemical characteristics of soil used for microcosm experiment.

Site	Land-use practice	pH	EC (mS cm ⁻¹)	Soil texture	Clay (%)	Bulk density (g cm ⁻³)	CEC (cmol _c kg ⁻¹)	TSC (%)	SOC (%)	WEOC (mg C kg ⁻¹)	TN (%)	NH ₄ ⁺ -N (mg kg ⁻¹)	NO ₃ ⁻ -N (mg kg ⁻¹)	WEON (mg N kg ⁻¹)	C/N ratio	PO ₄ ³⁻ -P (mg kg ⁻¹)	SO ₄ ²⁻ -S (mg kg ⁻¹)
Site A	AC	8.4	1.4	CL	33.0	1.4	36.7	4.1	2.2	3.5	0.2	4.3	25.1	8.1	11.3	24.9	82.1
	PA	7.8	0.6	CL	34.0	1.3	43.9	3.4	2.9	5.2	0.3	6.7	7.0	3.7	11.6	28.5	98.2
	SRW	8.0	1.9	SCL	27.0	1.3	37.4	3.7	2.2	5.2	0.2	6.0	12.5	5.1	10.8	21.3	641.5
Site B	AC	7.8	1.0	CL	33.0	1.3	48.7	3.1	2.5	5.9	0.2	6.0	17.0	4.5	9.8	30.0	2353.2
	PA	8.0	2.6	CL	32.0	1.2	42.5	4.9	2.7	4.6	0.3	5.2	33.4	8.1	10.8	31.2	274.4
	SRW	7.8	2.8	CL	30.0	1.4	45.9	2.8	2.4	6.2	0.3	6.3	13.9	4.3	9.4	37.8	3496.2

^a AC = annual crop, PA = pasture, SRW = short rotation willow, EC = electrical conductivity, CL = clay loam, SCL = sandy clay loam, CEC = cation exchange capacity, TSC = total soil carbon, SOC = soil organic carbon, WEOC = water extractable organic carbon, TN = total nitrogen, WEON = water extractable organic nitrogen, C/N ratio = carbon and nitrogen ratio.



Source: Google Imagery (2019)

Figure D-S 1 Location map of study sites at Indian Head, SK from where intact soil cores collected.

^a AC = annual crop, PA = pasture, SRW = short rotation willow.

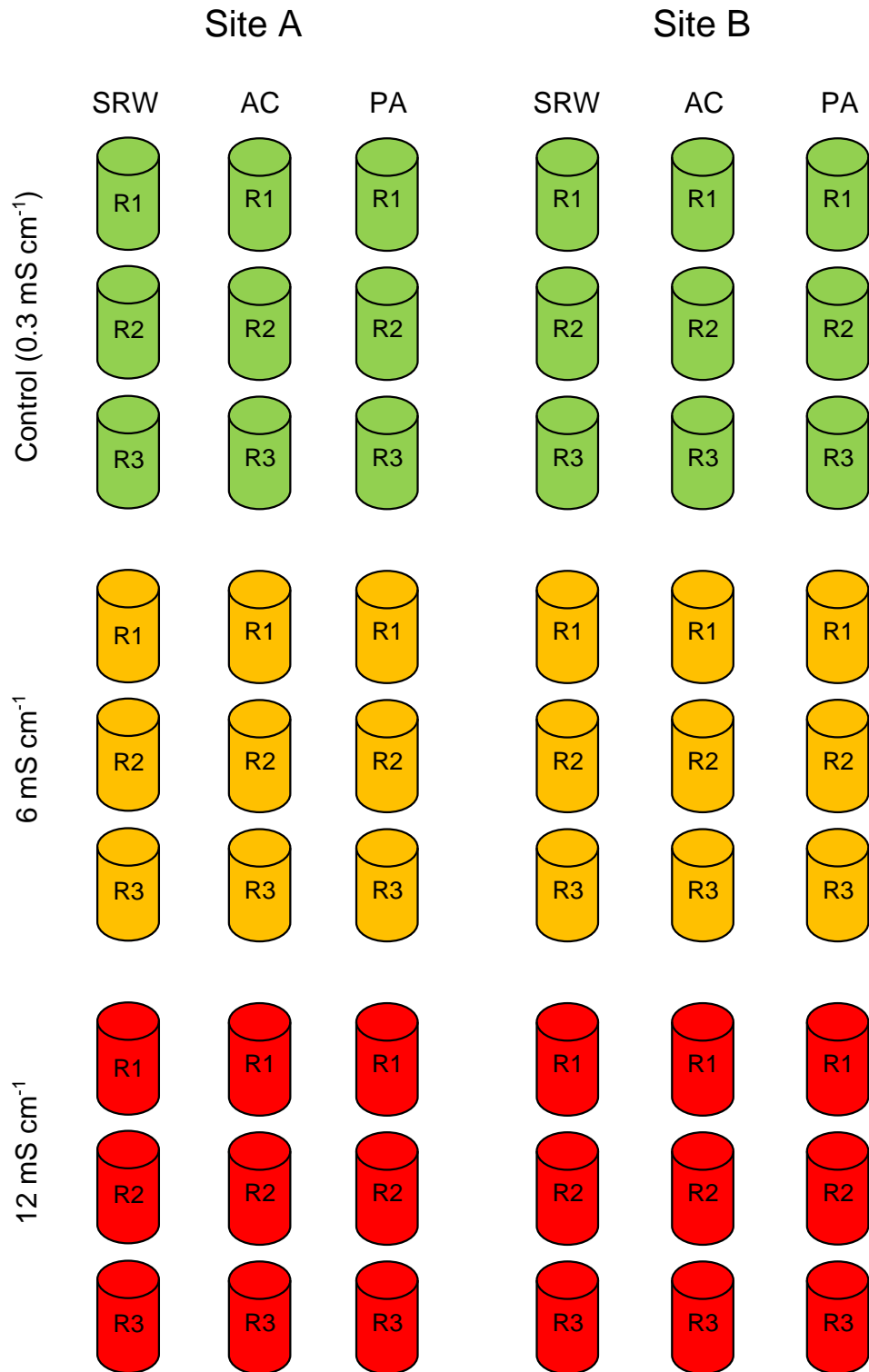


Figure D-S 2 Layout for microcosm incubation experiment in the greenhouse using intact soil cores (Note: diagram is not to scale).

^a SRW = short rotation willow, AC = annual crop, PA = pasture, R = replication.

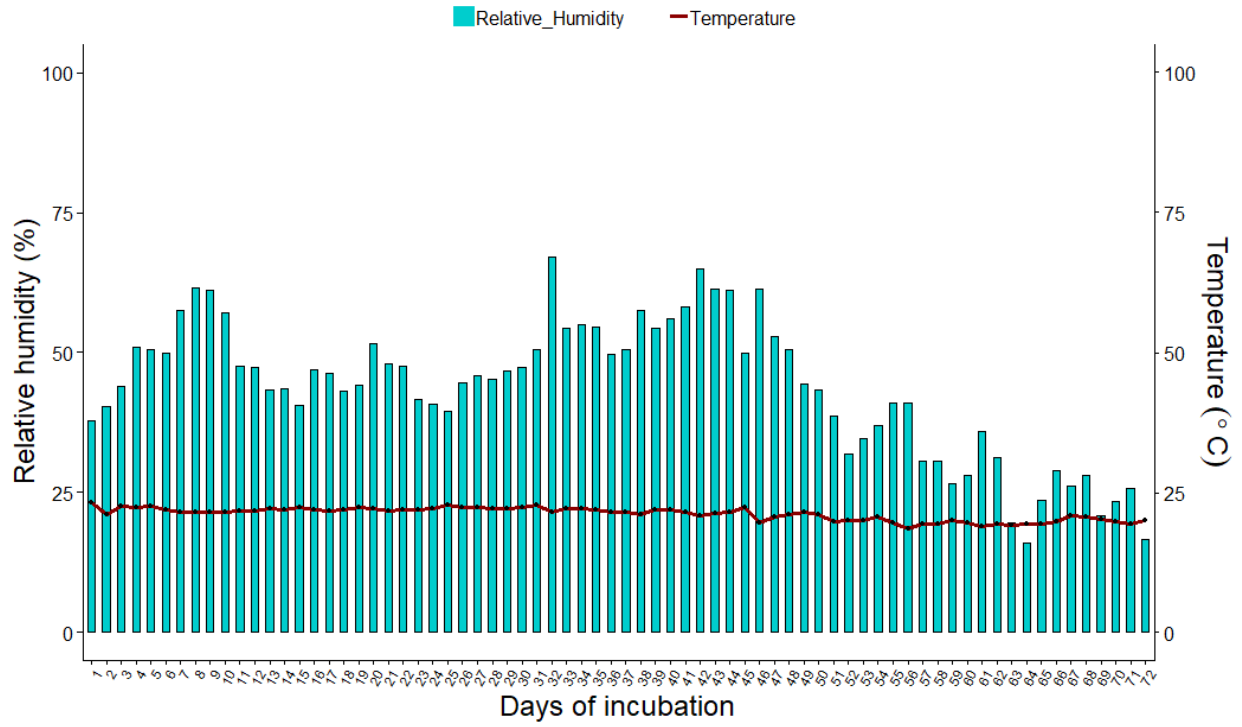


Figure D-S 3 Temperature and relative humidity of the greenhouse chamber used for microcosm incubation study.

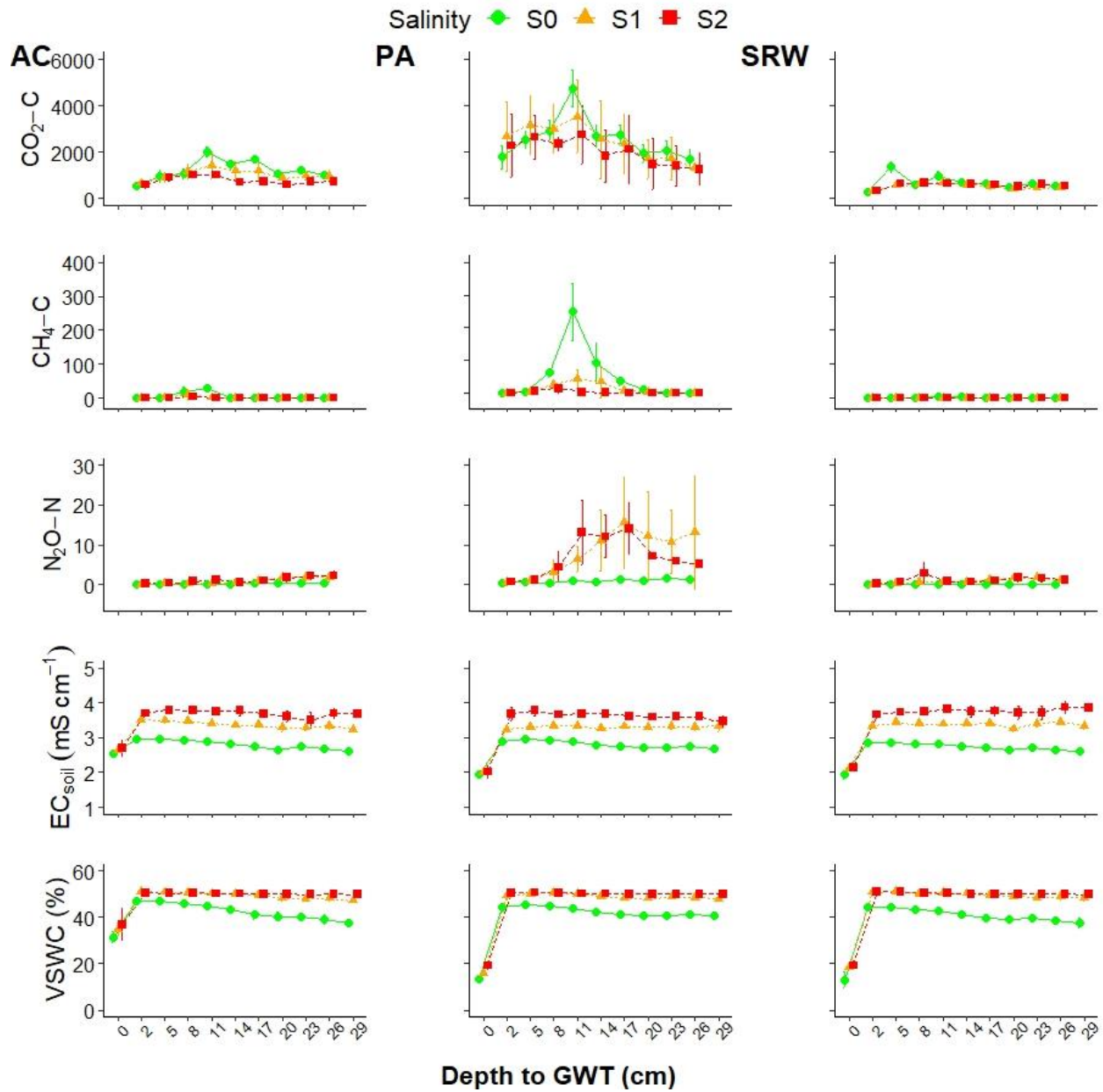


Figure D-S 4 Time series (mean \pm SD) of GHG emissions ($\text{mg m}^{-2} \text{d}^{-1}$), soil EC, and VSWC measured weekly with their equivalent depth to groundwater table and salinity treatments from core soils collected from different land-use practices from site A.

^a Error bar stands for standard deviations (\pm SD).

^b S0 = control, S1 = 6 mS cm^{-1} , S2 = 12 mS cm^{-1} , AC = annual crop, PA = pasture, SRW = short rotation willow, EC_{soil} = soil electrical conductivity, VSWC = volumetric soil water content, GWT = depth to groundwater table.

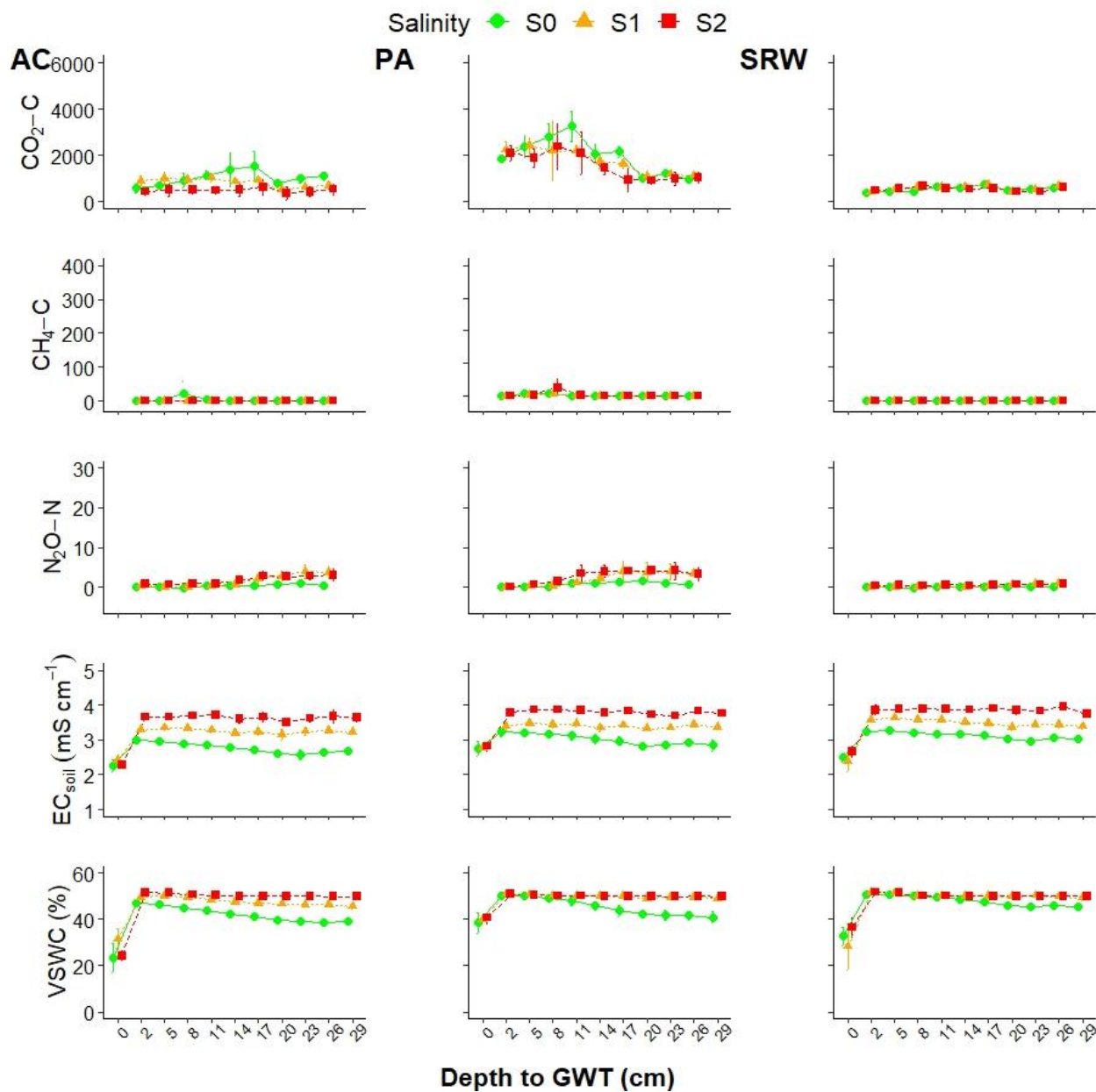
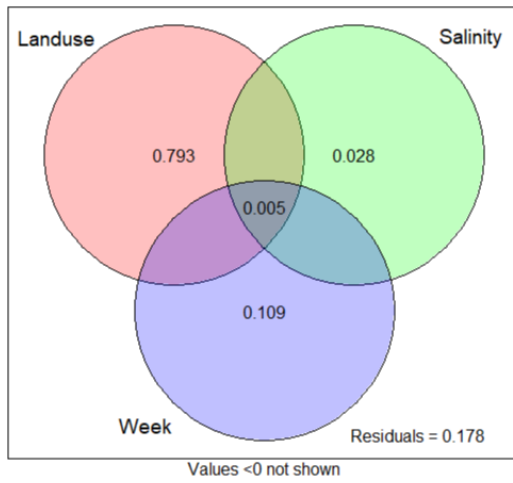


Figure D-S 5 Time series (mean \pm SD) of GHG emissions ($\text{mg m}^{-2} \text{d}^{-1}$), soil EC, and VSWC measured weekly with their equivalent depth to groundwater table and salinity treatments from core soils collected from different land-use practices from site B.

^a Error bar stands for standard deviations (\pm SD).

^b S0 = control, S1 = 6 mS cm^{-1} , S2 = 12 mS cm^{-1} , AC = annual crop, PA = pasture, SRW = short rotation willow, EC_{soil} = soil electrical conductivity, VSWC = volumetric soil water content, GWT = depth to groundwater table.

(A)



(B)

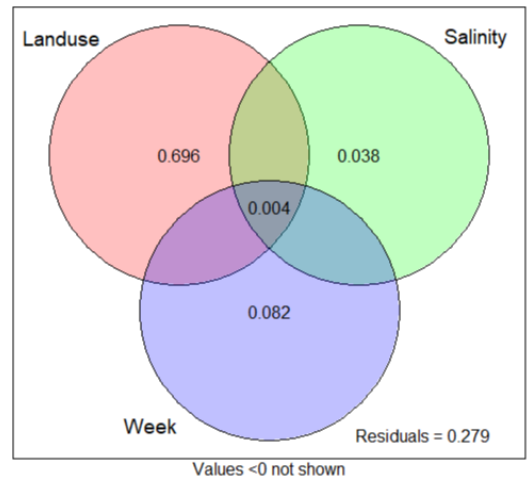


Figure D-S 6 Venn's diagram showing variation partitioning analysis (VPA) illustrates the contribution of land-use practices, salinity, and groundwater table depth to variation in GHG from A) site A, and B) site B.

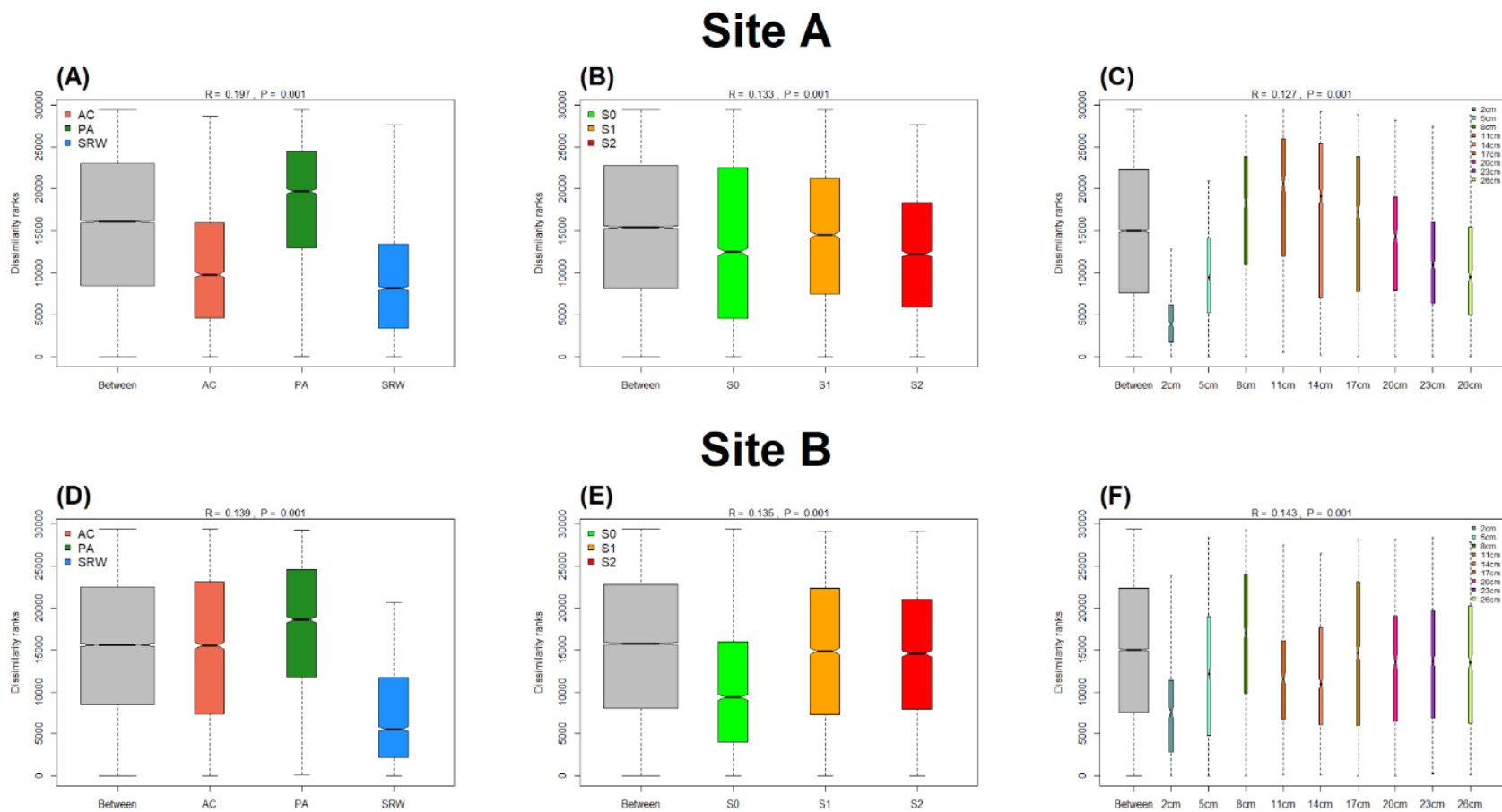


Figure D-S 7 Analysis of similarities (ANOSIM) with dissimilarity ranks between and within soil GHG (CO_2 , CH_4 , N_2O) measured weekly with their equivalent groundwater table depths during the incubation period in the core soils collected from three different land-use practices, under groundwater salinity treatments and from two sites.

^a R-value (i.e., the strength of the factors on the samples) varies between 0 and 1 (a value close to 1 indicates the high separation between levels of a factor, while R-value close to 0 indicates no separation between levels of a factor).

^b AC = annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm^{-1} , S2 = 12 mS cm^{-1} , GHG = greenhouse gas.

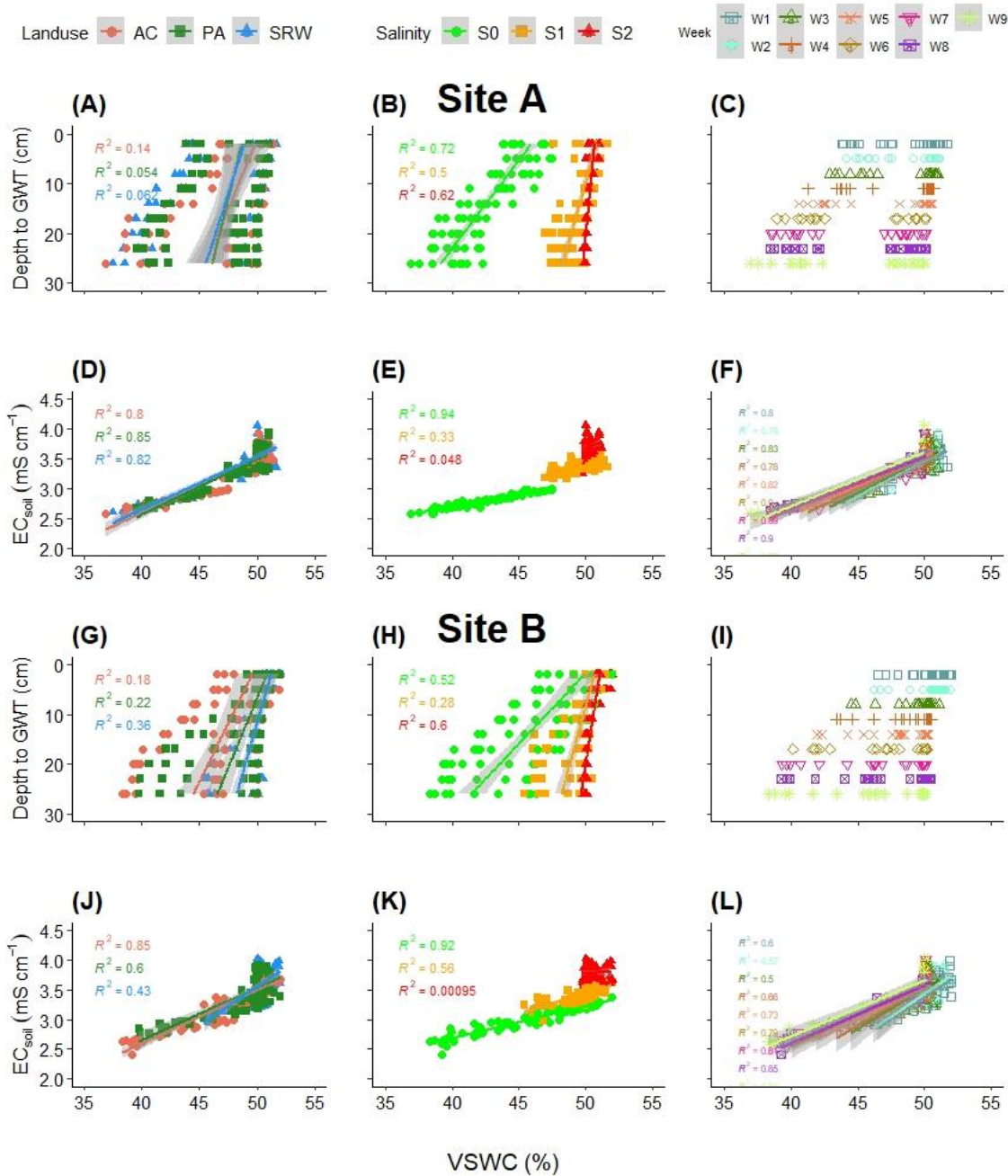


Figure D-S 8 Relationship between declining depths to groundwater table, volumetric soil water content, and soil electrical conductivity in soil cores from three land-use practices, salinity, and groundwater table depths treatments two sites.

^a The line shows linear regression of the change in VSWC and soil EC measured during the experiment against groundwater and salinity treatments; shaded area shows 95% of the confidence interval, R^2 gives values for individual regression lines, and p -values indicates the level of significance.

^b p -values ≤ 0.05 indicates a significant difference, whereas p -values > 0.05 are not significantly different.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm⁻¹, S2 = 12 mS cm⁻¹, W = week of measurement, GWT = depth to groundwater table. EC_{soil} = soil electrical conductivity. VSWC = volumetric soil water content.



Figure D-S 9 Photographs of intact soil core samples collected from the experimental field: A) from the annual crop, B) from pasture, C) from short rotation willow, D) empty soil corer ready to mount with punch truck, E) soil corer unmounted from the punch truck after soil samples collection, F) soil core samples capped both sides and ready to transfer into the greenhouse, and G) an individual intact soil core sample.



Figure D-S 10 Photographs of some activities from the microcosm incubation experiment: A) intact soil cores collected from the field, B) an individual experimental unit with and without the headspace, C) bucket filled with gravels, D) groundwater salinity measurements, E) volumetric soil water content and soil electrical conductivity measurements in the cores, F) gas samples collected from the headspaces, G) transfer of gas samples into the vials, and H) arrangement of the incubation experiment in the greenhouse.

APPENDIX E. SUPPLEMENTARY MATERIALS FOR CHAPTER 7

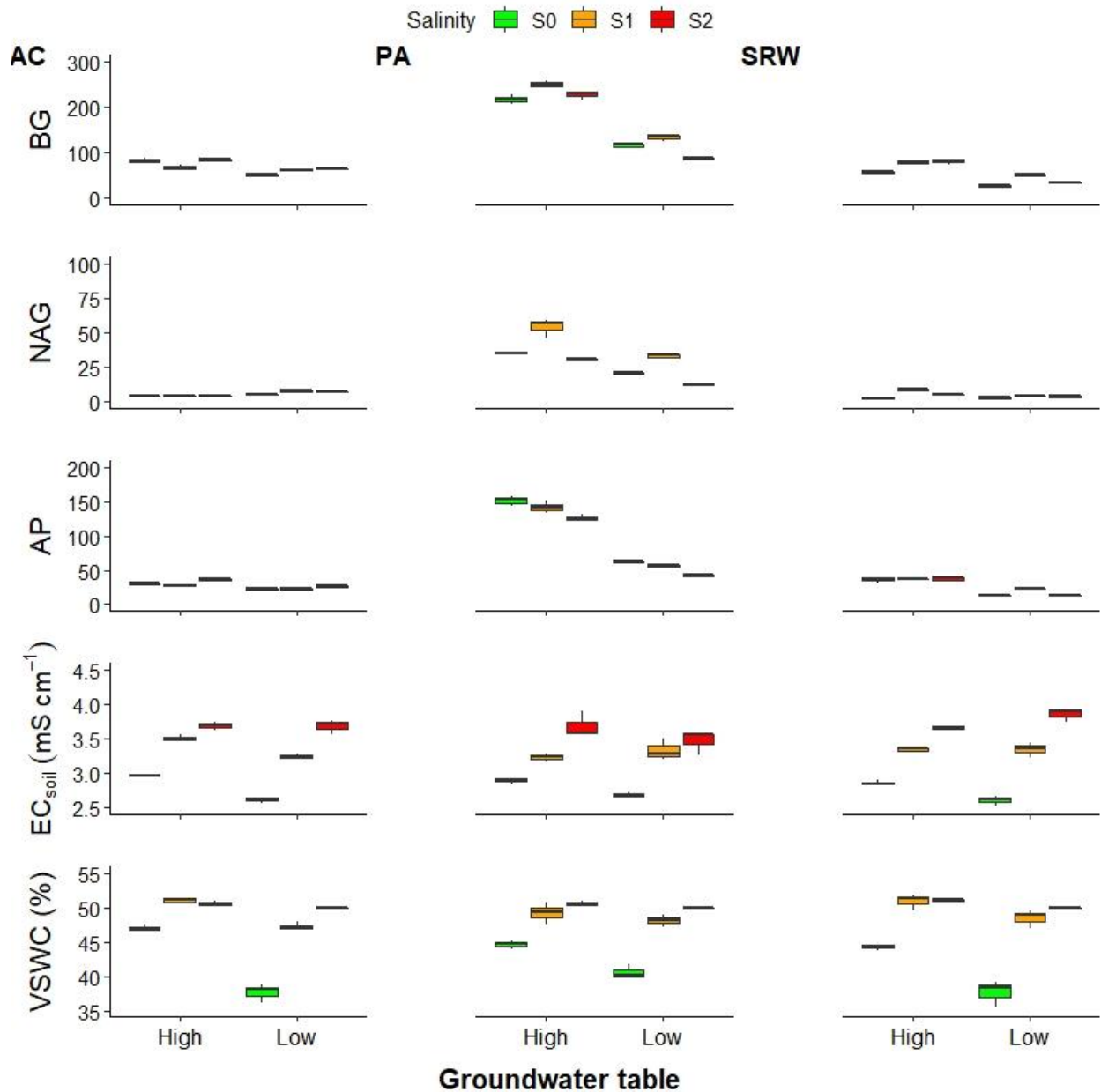


Figure E-S 1 Soil EEAs ($\text{nmol activity g}^{-1} \text{ C h}^{-1}$), EC_{soil} , and VSWC measured under different groundwater salinity treatments and groundwater table levels in the core soils collected from different land-use practices from site A.

^a Error bar represents standard deviations (\pm SD).

^b AC= annual crop, PA = pasture, SRW = short rotation willow, BG = β -glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase, EC_{soil} = soil electrical conductivity, VSWC = volumetric soil water content, EEAs = extracellular enzyme activities.

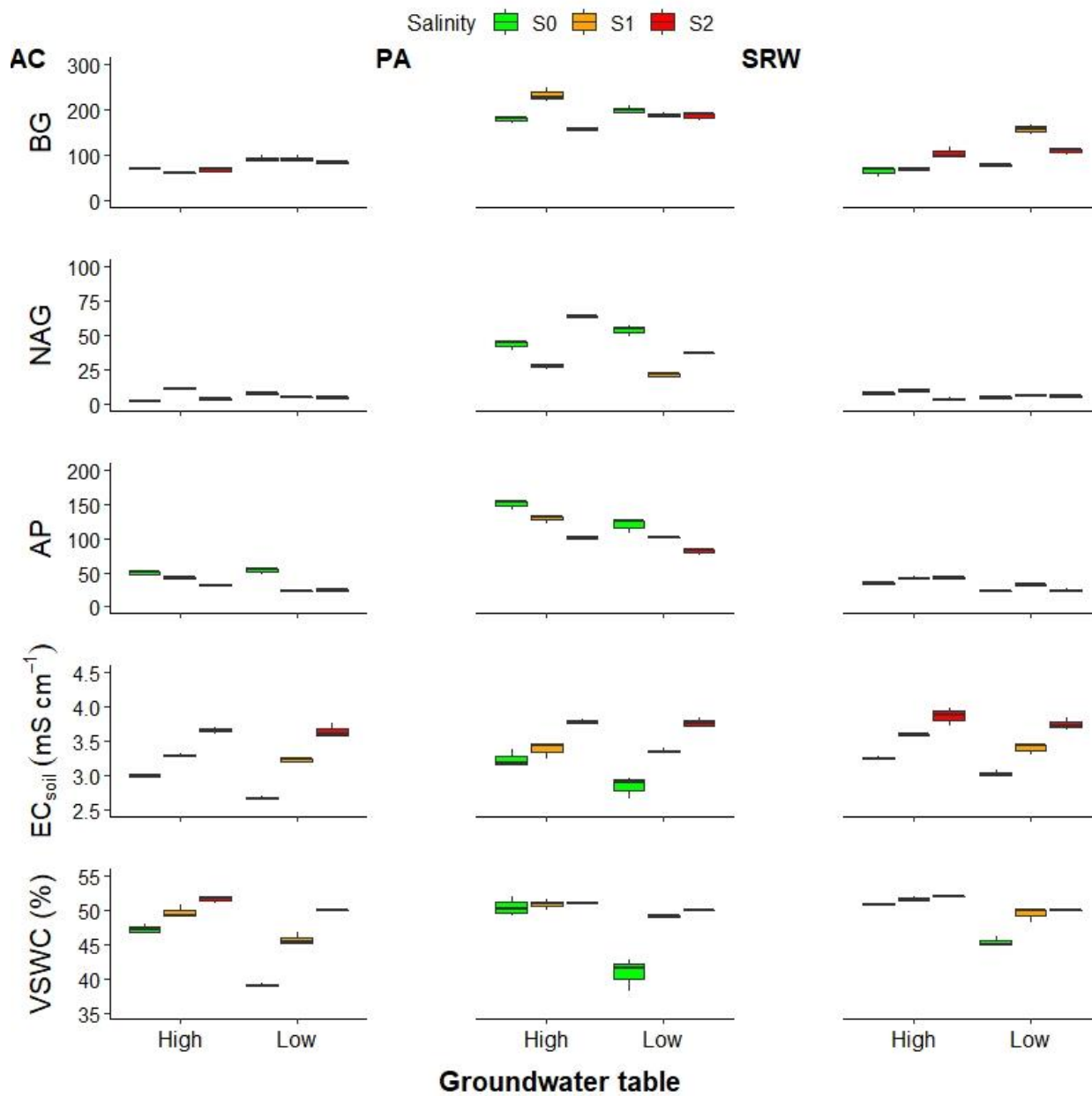
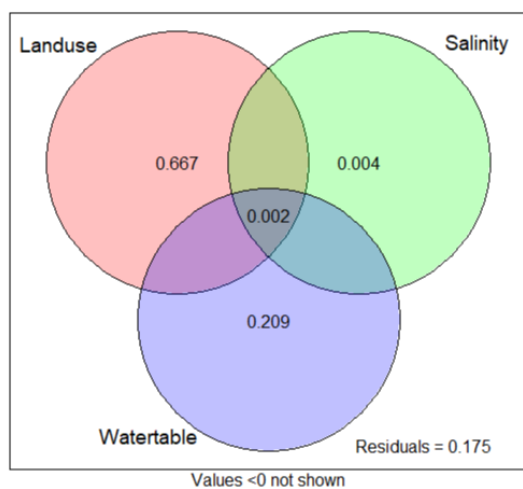


Figure E-S 2 Soil EEAs (nmol activity g⁻¹ C h⁻¹), EC_{soil}, and VSWC measured under different groundwater salinity treatments and groundwater table levels in the core soils collected from different land-use practices from site B.

^a Error bar represents standard deviations (± SD).

^b AC= annual crop, PA = pasture, SRW = short rotation willow, BG = β-glucosidase, NAG = N-acetyl glucosaminidase, AP = alkaline phosphatase, EC_{soil} = soil electrical conductivity, VSWC = volumetric soil water content, EEAs = extracellular enzyme activities.

(A)



(B)

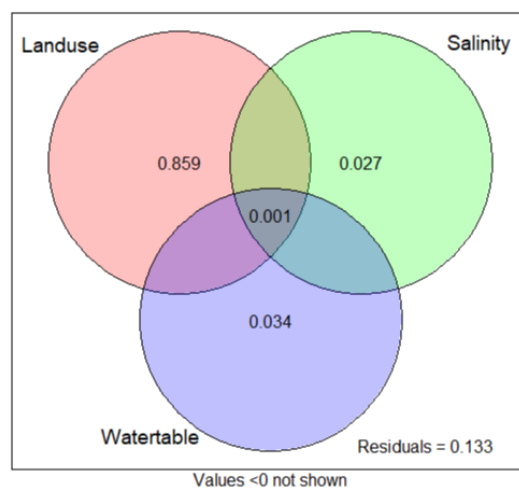
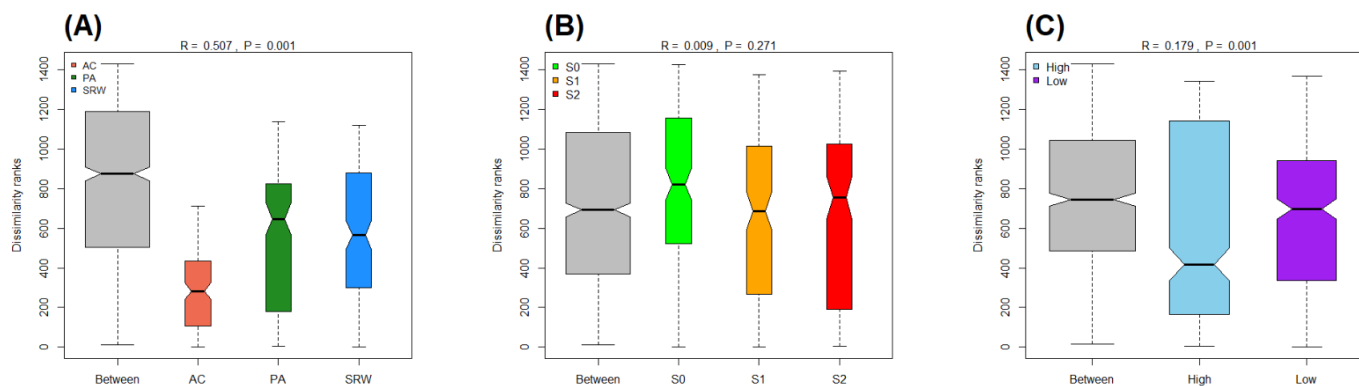


Figure E-S 3 Venn's diagram showing variation partitioning analysis (VPA) illustrates the contribution of land-use practices, salinity, and groundwater table depth to variation in EEAs from A) site A, and B) site B.

^a EEAs = extracellular enzyme activities.

Site A



Site B

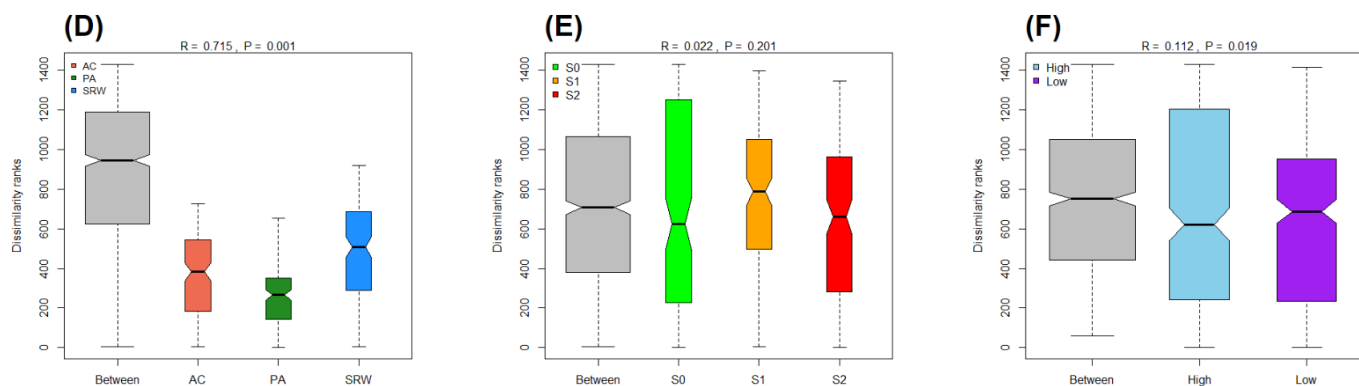


Figure E-S 4 Analysis of similarities (ANOSIM) with dissimilarity ranks between and within soil EEAs measured at week- 1 and 10 of incubation period in the core soils collected from different land-use practices from two sites treated under grounder salinity and water table depths.

^a An R-value close to "1.0" suggests dissimilarity between groups; R-value close to "0" suggests an even distribution of high and low ranks within and between groups; R-value below "0" suggests that dissimilarities are greater within groups than between groups.

^b AC= annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm⁻¹, S2 = 12 mS cm⁻¹, EEAs = extracellular enzyme activities.

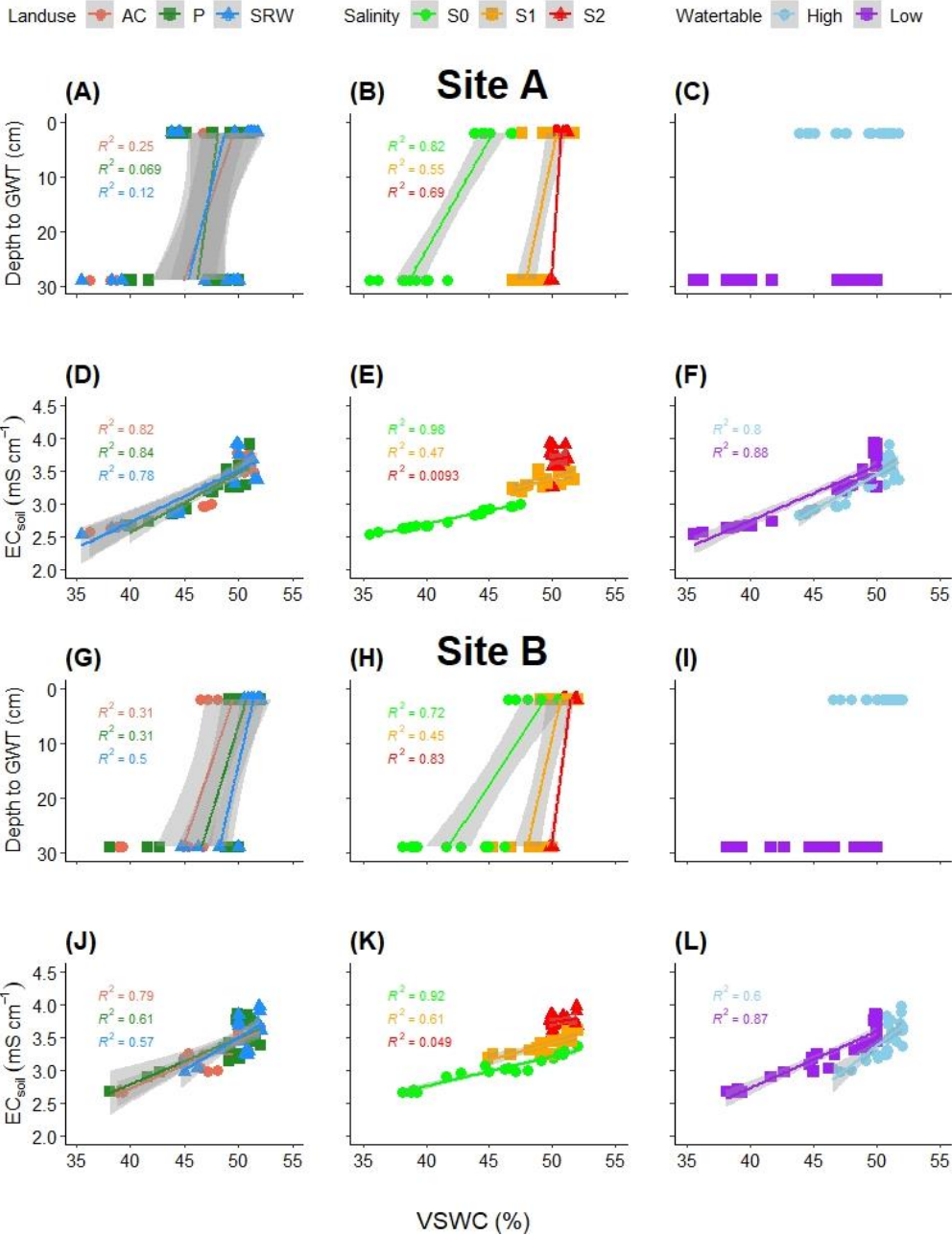


Figure E-S 5 Relationship between declining groundwater table depths (GWT), volumetric soil water content (VSWC), and soil electrical conductivity (EC) in soil cores from three land-use practices, salinity, and groundwater table depth treatments from two sites.

^a The line shows linear regression of the change in VSWC and soil EC measured during the experiment against groundwater table and salinity treatments; shaded area shows 95% of confidence interval, R^2 gives values for individual regression lines, and p -values indicates the level of significance.

^b p -values ≤ 0.05 indicates a significant difference, whereas p -values > 0.05 are not significantly different.

^c AC = annual crop, PA = pasture, SRW = short rotation willow, S0 = control, S1 = 6 mS cm⁻¹, S2 = 12 mS cm⁻¹, GWT = depth to groundwater table. EC_{soil} = soil electrical conductivity, VSWC = volumetric soil water content.