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Soil moisture dynamics in agriculturally-dominated landscapes after the introduction of native prairie vegetation

Jose Antonio Gutierrez Lopez
Iowa State University

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Soil moisture dynamics in agriculturally-dominated landscapes after the introduction of native prairie vegetation

By:

José Antonio Gutiérrez López

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Program of Study Committee:

Thomas Isenhart
Matthew Helmers
Heidi Asbjornsen
Alan D. Wanamaker

Iowa State University

Ames, Iowa

2012

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Dedication

A ti, Agustina
A mis padres
Gloria y René
A mis hermanos
Patricia y René
A todos mis amigos
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CHAPTER I

GENERAL INTRODUCTION

1.1 Contributions of this thesis to the overall mission of the Neal Smith National Wildlife Refuge

This research was developed to generate evidence of the benefits of the reintegration of perennial vegetation into agricultural dominated landscapes and also to provide key information about hydrological processes of native prairie ecosystems; particularly to provide information about water use and soil water content differences among prairie vegetation and cropland. This study also aims to help to understand how the location of prairie vegetation within a watershed can contribute to the hydrological balance of the entire watershed. This information is directly applicable to one of the Refuge’s priority goals: to reconstruct the original tallgrass prairie.

Additionally, information related to changes in depth of water uptake between prairie species and crops, and differences in soil water storage under prairie and crop vegetation can be incorporated into the Refuge’s prairie science class program, particularly the outdoor classroom activities that are currently being developed.

Finally, this research fits with the Refuge’s priorities to initiate a prairie/savanna land management and research demonstration program, which strives to advance problem solving via land-based research. Thus, the results from this research will contribute to the
Refuge’s goal, by providing information and tools for the Refuge’s priority of strengthening its prairie land management program.

1.2 Introduction

Soil moisture dynamics are the outcome of complex and highly interconnected processes. Some of the factors influencing these processes are vegetative cover, topographic position, soil properties, and precipitation among others. Soil moisture varies among ecosystems, across time, with depth and during the growing season (Isham et al., 2005; Zhang and Schilling, 2006; Tamea et al., 2009), and it is also highly affected by land cover primarily through water uptake (Fohrer et al., 2001).

Vegetative cover, in particular, plays an important role on the regional and local hydrologic balance (Cubera et al., 2004). Further, different changes in vegetative cover due to land use can have varying effects on the magnitude and direction of change in the water balance. Characteristics inherent to each species, such as rooting distribution and depth, photosynthetic pathway (e.g., C₃ vs. C₄), aerial biomass, phenology, etc., all dictate their influence on the hydrological balance.

Native prairie vegetation, because of its deeper root system, and longer annual life cycle, has shown to have a higher influence to the hydrological balance of the ecosystem when compared to short-rooted crop species. The phenology of native prairie species also greatly influences soil moisture dynamics and, in turn, ecosystem water budget. For example, certain C₃ prairie species, particularly cool season forbs, start their vegetative
cycle early in the growing season and remain active until late fall, thus providing soil cover when rowcrop species are not active.

In the Midwest United States, where native prairie vegetation has been dramatically replaced by row-crop species, the benefits that the native prairie vegetation provide to the hydrological balance of the ecosystem have been severely diminished, resulting in more frequent drought and flooding, higher fluctuations in streamflow, loss of nutrients and sediment and increased overland flow (Burkart and James, 1999; Schilling and Libra, 2000; Fohrer et al., 2001; Rabalais et al., 2002).

Increasing evidence suggests that the reincorporation of native prairie vegetation into agricultural dominated landscapes can mitigate the negative effects caused by the intensive agricultural row-crop production, by retaining soil, and controlling the hydrological balance through transpiration (Mersie and Seybold, 1997; Abu-Zreig et al., 2004; Blanco-Canqui et al., 2004; Gharabaghi et al., 2006). Currently, riparian buffers of perennial vegetation are being used to protect riverbanks from erosion and to reduce the discharge of agrochemicals into the stream (Schultz et al., 2004; Williard et al., 2005). Perennial vegetation is also being used to preserve soil on site through the Conservation Reserve (CRP) program, in which landowners of agricultural land can receive annual rental payments to establish long-term, resource conserving covers on eligible farmland.

However, some of the fundamental questions that the reintroduction of native vegetation raises, such as how much and where such vegetation should be planted on the landscape for maximizing benefits, have not been fully answered. This lack of information creates conflicts between production and conservation. Both productivity and
conservation are two fundamental elements of sustainable agriculture, which in the long term, define the stability and health of the ecosystem.

The question “Where on the landscape should plant perennial vegetation be planted?” refers to the relative position within a watershed. Prairie vegetation in the summit and side landscape position help to preserve soil in the upper parts of the watershed where large quantities of soil are eroded with rainfall, while placing it in the footslope position appears to contribute to reduce runoff and to regulate water discharge. Similarly, the question “How much should be planted?” is important to address to know the amount of vegetation that needs to be restored in order to bring back the benefits of native prairie vegetation without compromising production requirements. It is imperative then, to find the right balance to promote both productivity and conservation to assure farmland for future generations.

In this research, a total of 13 watersheds were used to provide evidence that can be used to answer these specific questions (the amount of native prairie vegetation that should be introduced and the location in which it should be planted). 5 treatments (100% row-crop, 10% prairie vegetation in the footslope, 10% prairie vegetation distributed in the watershed, 20% prairie vegetation distributed in the watershed, and 100% prairie vegetation) were assigned to these watersheds, which we monitored from 2007 to 2010 to assess the impacts of each treatment.
1.3 Thesis organization

This thesis is divided into four chapters; the first chapter is a general introduction that includes a literature review and general information related to this study, the overarching goal and specific objectives, and the hypotheses tested during the study. It also includes a general description of the data that were collected and a brief description of the methodology. The second chapter is a paper that explores soil moisture dynamics in detail, particularly how soil moisture patterns are influenced by different factors such as land cover, topographic position, soil depth, growing season, and precipitation. The third chapter is a paper in which individual plant species were studied, specifically dominant C₃ and C₄ prairie species and a C₄ crop (corn), to enhance understanding of the effects of their differences in water uptake depth on soil moisture dynamics. In this chapter we also address the importance of plant diversity in prairie restoration. The fourth and last chapter of this thesis is a general conclusion that will also discuss recommendations and suggestions for future research.

1.4 Literature review

1.4.1 History of native ecosystems in the Midwest

Historically, the area occupied by Iowa was covered in prairie; 162 million hectares covered an area known as the Great Plains that extended from Manitoba and Saskatchewan south through the eastern Dakotas, Nebraska, Kansas, Central Oklahoma and Texas to Mexico (Samson and Knopf, 1994; Brye and Moreno, 2006). The prairie
ecosystem was established thousands of years ago, driven by climatic, environmental factors and the deposition of large amounts of till by glaciers. Adapted to the environment, the prairie ecosystem was an integral part of more than 80% of the landscape (Smith, 1998).

After the European settlement in the 1800’s, a conversion started to turn areas occupied by native prairie vegetation into agricultural lands. This conversion, driven by the high fertility of the soils, led to an almost total replacement of native prairie vegetation by intensive rowcrop agriculture. From the estimated 162 million hectares of tallgrass prairie, only a fraction remains in reserves or isolated patches in a fragmented landscape. In Iowa alone, is it estimated that from the historic 12.5 million ha, only 12,140 remain. 99.9% of the original native vegetation was lost (Samson and Knopf, 1994) and with it, the ecosystem benefits that native prairie vegetation provides to society.

1.4.2 Soil moisture and its role in the ecosystem

Water plays an important role in the dynamics of terrestrial ecosystems (Tamea et al., 2009). It has a direct influence on ecosystem stability, and it is the major driver of plant productivity (Gholz et al., 1990). Soil water also constitutes a physical connection between soil, climate, and vegetation (Isham et al., 2005). This physically integrated system, where several processes take place interdependently, is analogous to links in a chain and is known as the soil-plant-atmosphere continuum (Philip, 1966).
The relationship between soil and plants is particularly important for the hydrological balance of the ecosystem. Through water uptake, plants have a strong influence on the local and regional hydrological balance through their role in cycling water between the soil and the atmosphere (Chahine, 1992; Mahmood and Hubbard, 2003). The presence of water facilitates the control of temperature variations in ecosystems. Wet, forested or vegetated areas show intermediate changes in temperature between day and night compared to hot and dry areas (Chahine, 1992). Soil formation and soil fertility are also greatly influenced by the presence of water. Several authors have described the importance of soil moisture in soil formation and development. They have attributed formation of soil structure and the production of tertiary minerals that define soil fertility to the presence and movement of water in the soil (Jenny, 1994; Hillel, 2004; Randall and Sharon, 2005).

In addition to the influence of soil water on these biological and chemical processes, physical processes, such as rainfall infiltration, percolation, runoff generation, capillarity, groundwater distribution, and pollutant transport into the soil matrix, are also directly controlled by soil water content (Gardner, 1936; Helmke et al., 2005; Tamea et al., 2009). Soil water represents a controlling factor in hydrological and geotechnical processes that are responsible for slope stability (Isham et al., 2005). At a larger scale, soil water is responsible for the formation of ore deposits, porosity occlusion, sediment cementation, petroleum migration, landslides and gas hydrate formation among other numerous features of geologic interest (Wood, 2002).
1.4.3 Factors influencing soil moisture

Despite being the major driver of plant productivity, soil moisture is subjected to the influence of factors that underline its dynamics within an ecosystem. Soil moisture dynamics are thus the outcome of complex and interconnected processes. Some of the factors influencing these processes are: vegetative cover, soil characteristics, topographic position and precipitation (Zhang and Schilling, 2006; Liu and Zhang, 2007; Wang et al., 2008; Kumagai et al., 2009; Qi and Helmers, 2010).

Understanding how these factors influence the hydrological balance of the ecosystem and the degree to which each impact this balance is imperative for the health of the ecosystems. More importantly, understanding the degree of influence of these factors under a given configuration of mixed annual-perennial vegetation, may allow researchers and scientist to make better decisions for conservation and productivity purposes.

1.4.3.1 Effects of land cover on soil moisture dynamics

Land cover has been recognized as a key factor controlling patterns of soil moisture by influencing infiltration rates, runoff, and evapotranspiration (Cubera et al., 2004). Different land covers have varying degrees of above and belowground biomass production. Aboveground biomass determines the amount of precipitation water that it is intercepted and thus the amount that reaches the ground (Brooks et al., 2003; Chang, 2006). Vegetation also deposits organic matter on the soil surface and belowground through root growth, which enhances infiltration rate and soil moisture holding capacity,
making surface runoff smaller, runoff timing longer, and water yield lower in vegetated areas than those in non-covered ones (Chang, 2006).

In the Midwest United States, two vegetative covers have historical significance; native prairie vegetation and agricultural crops (e.g. corn and soybean). These two land covers have contrasting effects on the hydrological balance due to their inherent differences in water use and uptake patterns and in plant phenology. After the European settlement (see 1.4.1), agricultural crops started to gain increasing influence on the hydrological balance of the entire region.

1.4.3.2 Effects of topographic position

Water moves in a watershed obeying laws of gravity, capillarity and suction primarily (Brooks et al., 2003; Hillel, 2004). Right after infiltration, as water penetrates into the soil profile and the length of the wetted part of the profile increases, suction gradient decreases, since the difference in pressure head divides itself over an ever-increasing distance. As this trend continues, the suction gradient of the upper part of the soil profile becomes negligible, leaving the gravitational head gradient as the only force to move the water downward (Hillel, 2004).

This gravitational gradient tends to move water from the upper parts of the watershed (i.e. recharge areas) towards lower parts (i.e. discharge areas). In these recharge areas, due to the effects of the gravitational gradient, there is often a rather deep unsaturated zone between the water table and the land surface. Conversely, the water table is found either close to or at the land surface in discharge areas (Fetter, 2001).
Water then moves naturally from the upper parts of the watershed and from shallower soil depths to deep horizons greatly influenced by gravity.

1.4.3.3 Effects of precipitation

Precipitation has a direct effect on soil moisture. Once net precipitation reaches the ground, it moves into the soil, forms puddles on the soil surface or flows over the soil surface, depending on preceding soil moisture content. The precipitation that enters the soil and is not retained by it, moves either downwards to groundwater or laterally to a stream channel (Brooks et al., 2003). As water moves into the soil profile, it influences directly soil moisture content before leaving the system through evaporation, plant water uptake or simply moves towards the lower parts of the watersheds due to the influence of gravity.

The rate at which net precipitation enters the soil surface depends on several soil properties as well as on soil surface conditions such as plant material or litter near the soil surface (Brooks et al., 2003). However, the intensity of a precipitation and the antecedent soil moisture are two important factors that control the effect of a precipitation event on the soil.

1.4.3.4 Water uptake by plants

Despite the laws of conservation, water uptake can in some ways be exceedingly wasteful (Hillel, 2004). Plants are often required to withdraw large quantities of water from the soil that is far beyond their essential metabolic needs. This water uptake has a
direct influence on the soil moisture content, which at a larger scale greatly influences the hydrological balance of the watershed.

However, water uptake varies among species, and is influenced by soil water availability. A study conducted in crops by (Araki and Iijima, 2005), found that water uptake was influenced by the extent of dryness in the topsoil. Specifically, they observed that the difference in water use among C₃ forbs and C₄ grasses varied according to water availability in the surface soil and that this difference was driven by recent precipitation history. When the water was available in the upper 30cm, the plants took water from shallower sources, but during dry periods, the C₃ species used proportionally more water from deep profiles than the C₄ species.

Studies have also found that there is a significant difference in plant water uptake depth under different rooting patterns (Nippert and Knapp, 2007). Previous studies have shown that on average corn (Zea mays) and big bluestem (Andropogon gerardii) (both C₄ species) take water only from the first 20-30 cm of soil (Asbjornsen et al., 2007; 2008). However, a greater seasonal variation has been observed in other studies with summer corn, that was highly influence by the development stage of the plant, showing that corn plants extract water from the upper 20 cm in the jointing and fully ripe stage, and from as deep as 50 cm during the flowering state (Wang et al., 2010).

1.4.4 Study of plant water uptake using stable isotopes

The study of plant water uptake requires the use of techniques that allow researchers to observe differences of water uptake not just by type of land cover (i.e.
crops, prairie), but also by species and vegetative plant types (e.g. C\textsubscript{3}, C\textsubscript{4}), and more importantly, to identify patterns and differences by vegetative type and species at different topographic positions, soil depths, and seasons.

In hydrological studies that aim to understand the effect of plant water uptake on soil moisture dynamics, several tracers have been used to track water flow in soils, such as chemical, fluorescent dye, radioactive, activable, biological, surface active, episodic, and isotopes (Emilian, 1987; Gaspar, 1987; Kendall and McDonnell, 1998; Signh and Kumar, 2005). Nevertheless, the stable isotopes of carbon (\textsuperscript{13}C), hydrogen (\textsuperscript{2}H) and oxygen (\textsuperscript{18}O) are by far the most widely used in hydrology processes, in part because they do not pose any threat to the health of humans or the environment, and they are naturally present in the hydrosphere. In nature, two stable isotopes of hydrogen can be found: protium \textsuperscript{1}H and deuterium \textsuperscript{2}H (or D) with an abundance of 99.985\% and 0.015\% respectively, as well as three stable oxygen isotopes, \textsuperscript{16}O, \textsuperscript{17}O, and \textsuperscript{18}O with average abundances of 99.756\%, 0.039\%, and 0.205\% respectively (Emilian, 1987; Brooks \textit{et al.}, 2003; Mook, 2006).

In soils were water movement is predominantly vertical, after a precipitation event the water in the topsoil becomes enriched in \textsuperscript{18}O and \textsuperscript{2}H, primarily through fractionation processes that take place during evaporation. Precipitation events drive this enriched water into the soil profile thereby creating a gradient in isotopic composition that can be measured. Because there is no isotopic fractionation during plant water uptake, the isotopic concentration of plant water extracted from non-photosynthetic tissue can be used to assess the approximate depth from which most water uptake occurs.
To assess depth of plant water uptake, soil samples at increasing depths are taken and the water contained in them is analyzed for $^{18}$O and $^2$H. At the same time non-photosynthetic plant tissue is collected and the water of this material is analyzed and compared with the signature of the soil. Similarities between $\delta^{18}$O and $^2$H in the water of the vegetative tissue and the soil are used to infer depth of plant water uptake, while mixing models are often used to derive more accurate estimates (Nippert and Knapp, 2007). Stable isotopes have been widely used to estimate water uptake depth of plants at the plot scale (Plamboeck et al., 1999; Asbjornsen et al., 2008; Rowland et al., 2008).

At the watershed scale, naturally occurring stable isotopes have been used to monitor water flowpaths according to well-defined methodologies (Emilian, 1987; Kendall and McDonnell, 1998; Salem et al., 2004a; Salem et al., 2004b). Stable isotope tracers offer unique virtues as water tracers in watersheds studies, as they are not subjected to chemical reactions during contact with the soil, they undergo evaporation and fractionation causing a gradient difference between meteoric and subsurficial water, changes in isotopic concentrations increase as they move through the unsaturated zone, and in theory isotopic concentrations in a given water will change only when it is mixed with other water resources having different concentrations (Buttle and McDonnell, 2005).

1.4.5 Importance of the study of soil moisture dynamics

As already mentioned, soil moisture is the outcome of complex and highly interconnected processes, these processes are at the same time highly influenced by biological and physical factors. Understanding how these factors influence soil moisture
and ultimately the hydrological balance of the ecosystem and the degree to which every of them impacts this balance it’s imperative for the hydrological balance an ecosystem.

More importantly, understanding the degree of influence of these factors, particularly how the use of prairie vegetation influences the hydrological balance of the ecosystem, may allow policy makers and land managers to make better decisions for soil conservation without compromising crop productivity benefits.

However, the study of soil moisture dynamics goes beyond just the hydrological balance of a watershed. At the ecosystem scale, the understanding of its role in the plant-soil-atmosphere continuum provides the elements necessary in crop management and it can also provide the basis for flood control management and sustainable agricultural practices.

1.5 Objectives

This research addresses three main objectives related to soil water dynamics under mixed annual-perennial (i.e. prairie and row-crop) vegetation. The literature review revealed a good range of studies that examined specific factors that influence soil moisture dynamics; however, very few of these studies were conducted under natural and applicable field conditions. Specifically, my research will contribute to filling these current knowledge gaps about the relationship of soil moisture to land cover type and topographical position by achieving the following three objectives:
1. Quantify the differences in water content under perennial and annual vegetation in a mixed agricultural watershed.

2. Estimate the depth of water uptake for dominant C\textsubscript{3} and C\textsubscript{4} species in a mixed agricultural watershed using stable oxygen and hydrogen isotope ratios.

3. Assess the effect of topographic position on soil moisture dynamics and depth of water uptake in perennial and annual row-crop vegetation.

The first and second objectives will be treated individually in chapter 2 and 3 respectively; the third objective will be part of both the 2\textsuperscript{nd} and the 3\textsuperscript{rd} chapters of this thesis. An overall conclusion will be provided in chapter 4.

1.6 Hypotheses

In this thesis I tested three main hypotheses, one concerning the effects of land cover on soil moisture dynamics, a second about the differences in water uptake among species, with particular interest in C\textsubscript{3} and C\textsubscript{4}, and a third about the effects of topographic position on soil moisture dynamics and plant water uptake. Each hypothesis and the supporting rationale is summarized below.
Hypothesis #1. Differences in soil water content

Soil water content will be lower (e.g., higher soil water storage capacity) under prairie vegetation compared to under corn crops early in the growing season (June-early July) when prairie plants are active but crops have not yet fully established, while the reverse pattern will occur during the peak growing season (late July-August) when crops have reached maximum growth rates.

Rationale. It is well known that different land covers have different effects on hydrological processes such as evapotranspiration. In a study conducted at the NSNWR comparing the effects of land cover on soil moisture, evapotranspiration and groundwater recharge, Zhang and Schilling (Zhang and Schilling, 2006) observed that grassland cover reduced soil moisture through evapotranspiration and that it was less susceptible to changes in groundwater changes as compared to bare ground. In another recent study of differences in water use between perennial plants big bluestem (*Andropogon gerardii*, a C$_4$ grass), coneflower (*Ratibida pinnata*, a C$_3$ forb) and corn (*Zea mays* a C$_4$ annual crop species), it was shown that at the ecosystem level, total evapotranspiration was similar under these two cover types, but on a per leaf area basis, transpiration for the two prairie species was significantly greater than for the corn (Mateos Remigio *et al.*, in review.). Understanding how differences in depth of water uptake among crop and prairie, and among C$_3$ and C$_4$ prairie species, contribute to different water use patterns is important for selecting appropriate species for maximizing ecohydrological functions of strategically located prairie strips in agricultural landscapes.
Hypothesis #2. Plant water uptake depth

During periods of adequate soil moisture availability (e.g., early in the growing season), crops and prairie species will obtain their water from relatively shallow depths in the soil profile. As soil moisture becomes more limiting (later in the growing season), prairie species (especially C₃ forbs) will shift their depth of water uptake to deeper depths in the soil profile, whereas corn and C₄ prairie species will have more limited capacity to obtain water from deeper depths. This variation of depth of water uptake (especially by C₃ forbs) can contribute to a better control of the hydrological balance in those watersheds with strips of perennial vegetation, due to the presence of roots at deeper profiles that allow plants to access water form deeper profiles.

Rationale. Previous studies have shown that on average corn (Zea mays) and big bluestem (Andropogon gerardii), both C₄ species, take water only from the first 20-30 cm of soil (Asbjornsen et al., 2007; 2008). However, a greater seasonal variation has been observed in other studies with summer corn, showing that corn plants extract water from as deep as 50 cm (Wang et al., 2010). Nippert an Knapp, (2007) in a study of C₃ and C₄ plants, observed that differences in water uptake depth between species were more variable when water was most limiting: C₄ plants shifted their depth of water uptake to deep soil profiles in months when water availability decreased, while C₃ forbs and shrubs appeared to avoid competition by taking water from even deeper profiles than C₄ plants. More research is need to better understand patterns of depth of water uptake between crop and prairie species and between C₃ forbs and C₄ grasses growing in reconstructed prairie communities.
Hypothesis #3. Effect of topographic position on soil moisture and plant water uptake

Prairie and crop species will use water from deeper depths in the soil profile in the summit position compared to the footslope position due to lower water availability during dry periods in the upper parts of the watershed.

Rationale. Topographic location of the prairie strips in the watershed will have impacts on the patterns of both depth of water uptake and soil water content. Specifically we expect to observe higher soil water content in the footslope position compared to the upslope position, and thus relatively more shallow depths of plant water uptake for both prairie and crop species in the toe position. In contrast, plants should take up water from deeper depths at the summit position where soil moisture is likely to be more limiting, while we expect C₃ species to have greater capacity to take up water from deeper depths than C₄ species. As previous research has shown, when water is available in shallow profiles, plants tend to use more of this shallower water (Nippert and Knapp, 2007).

1.7 Study site and description of the experimental setup

1.7.1 Study Site

The research was conducted in the Neal Smith National Wildlife Refuge, located in Prairie City, Iowa (NSNWR, 41°33´N, 93°16´W). The refuge was created in 1991 to convert over 3,400 ha of agriculturally dominated landscape to native perennial vegetation. To this day, the Refuge consists of a mosaic of reconstructions and
agricultural land uses with approximately 1,200 ha planted to tallgrass prairie through annually successive plantings. Most soils at the research sites are classified as Ladoga (Mollic Hapludalf) or Otley (Oxyaquic Argiudolls) soil series, which are highly erodible with slopes ranging from 5 to 14% (NRCS, 2010). The average precipitation is 910mm (mean from 1981-2010). A more detailed description of the sites is given in Chapter 2 and 3 of this thesis.

1.7.2 Experimental design

A total of 13 watersheds were used to test the hypotheses. Strips and buffers of prairie vegetation were planted at different topographic positions across agricultural fields, yielding a total of 5 treatments as shown in Figure A; 100% rowcrop, 10% prairie cover as buffer, 10% prairie vegetation distributed in strips and buffer, 20% prairie vegetation distributed in strips and buffer, and 100% prairie vegetation.

Strips and buffers of prairie vegetation were planted in the summer of 2007 in small watersheds under a corn-soybeans yearly rotation. The 100% prairie vegetation watershed is an 18-year-old restored prairie. Three fiberglass access tubes were installed per watershed at the summit, side and footslope positions to measure soil moisture and two groundwater wells in the summit and footslope.
The watersheds were distributed in three sites, Basswood, Orbweaver and Interim, containing 6, 3 and 4 watersheds respectively, as described in Table A.

Table A. Description of the 13 watersheds.

<table>
<thead>
<tr>
<th>Watershed</th>
<th>Site</th>
<th>Prairie cover (%)</th>
<th>Topographic location of prairie cover</th>
<th>Area (ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basswood 1</td>
<td>Basswood</td>
<td>10%</td>
<td>Footslope</td>
<td>0.53</td>
</tr>
<tr>
<td>Basswood 2</td>
<td>Basswood</td>
<td>10%</td>
<td>Footslope &amp; Summit</td>
<td>0.48</td>
</tr>
<tr>
<td>Basswood 3</td>
<td>Basswood</td>
<td>20%</td>
<td>Footslope &amp; Summit</td>
<td>0.47</td>
</tr>
<tr>
<td>Basswood 4</td>
<td>Basswood</td>
<td>20%</td>
<td>Footslope &amp; Summit</td>
<td>0.55</td>
</tr>
<tr>
<td>Basswood 5</td>
<td>Basswood</td>
<td>10%</td>
<td>Footslope &amp; Summit</td>
<td>1.24</td>
</tr>
<tr>
<td>Basswood 6</td>
<td>Basswood</td>
<td>100% rowcrop</td>
<td>None</td>
<td>0.84</td>
</tr>
<tr>
<td>Orbweaver 1</td>
<td>Orbweaver</td>
<td>10%</td>
<td>Footslope</td>
<td>1.18</td>
</tr>
<tr>
<td>Orbweaver 2</td>
<td>Orbweaver</td>
<td>20%</td>
<td>Footslope, Side &amp; Summit</td>
<td>2.40</td>
</tr>
<tr>
<td>Orbweaver 3</td>
<td>Orbweaver</td>
<td>100% rowcrop</td>
<td>None</td>
<td>1.24</td>
</tr>
<tr>
<td>Interim 1</td>
<td>Interim</td>
<td>10%</td>
<td>Footslope, Side &amp; Summit</td>
<td>3.00</td>
</tr>
<tr>
<td>Interim 2</td>
<td>Interim</td>
<td>10%</td>
<td>Footslope</td>
<td>3.19</td>
</tr>
<tr>
<td>Interim 3</td>
<td>Interim</td>
<td>100% rowcrop</td>
<td>None</td>
<td>0.73</td>
</tr>
<tr>
<td>Interim 4</td>
<td>Interim</td>
<td>100% prairie</td>
<td>None</td>
<td>0.60</td>
</tr>
</tbody>
</table>

To assess differences in depth of plant water uptake, three watersheds were selected at the Interim site, based on their relative close location and because of their
configuration regarding land cover. The watersheds we used were: Interim 1, Interim 3, and Interim 4. A total of 9 plots (three per watershed) were marked, two in the summit and one in the footslope positions as shown in Figure B.

![Figure B](image)

**Figure B.** Graphical representation of the watersheds used to assess depth of water uptake.

1.8 **Description of the overall methodology**

This is a general description of the methodology used in this research, a more specific description will be provided in Chapter 2, and Chapter 3 of this thesis.

Soil moisture readings were taken biweekly from previously installed access tubes under the perennial strips, annual crop, and reconstructed prairie from April to November during 2007, 2008, 2009 and 2010 using a Theta (ML2, Delta-T Devices, Cambridge, UK) and a PR2 Probe (PR2, Delta-T Devices, Cambridge, UK). The Theta Probe was used to measure soil moisture in the first 6 cm of soil and the PR2 probe to take readings
at 10, 20, 30, 40, 60 and 100 cm. Soil cores were collected from 0-15, 15-25, 25-35, 35-50, 50-70, and 70-10cm close to the access tube to calibrate Theta and PR2 readings.

Soil cores were taken to assess soil bulk density in the study site [perennial vegetation & annual crops]. The soil cores were collected in every watershed at three topographic positions; Summit, Side and Footslope at five depths 10, 20, 30, 60 and 100cm. The samples were weighed and dried in a drying oven at 105ºC for 24 hours. Soil bulk density was estimated dividing dry weight of every sample over its volume.

After the application of a Deuterium tracer in the study of depth of plant water uptake, soil and plant samples were collected in July 22-28 and August 29-30, 2010 to assess depth of water uptake. One set of six soil cores was collected per every plant sample, and two replicates were collected per plant. Soil cores were collected at increments from 0-10, 10-20, 20-30, 30-50, 50-70 and 70-100cm using a bucket auger at the Interim 1 and Interim 4 sites, and from 0-10, 10-20, 20-30, 30-50 and 50-70 in Interim 3 due to the shallower groundwater table.

Rainfall collectors were installed to sample water during rainfall events. These data were used to estimate the percentage of meteoric water entering the watersheds. To avoid evaporation a funnel was placed in the rain gauge, to allow the water to get into the gauge, but reducing the area of evaporation, other investigators have used a ping-pong balls, however, our collection flask was placed inside a wooden box to avoid evaporation and the samples were collected right after the precipitation event reducing fractionation in $\delta^{18}$O ‰ and $\delta$D ‰ values in rainfall water.
Groundwater was also collected to complement and compare concentrations of δ¹⁸O ‰ and δD ‰. The groundwater wells consisted of ¾ inch PVC piping capped at the bottom with a pointed tip. The wells were equipped with slits covering the bottom third of the piping to allow movement of groundwater into the well. Water table depths were determined biweekly over the course of the growing season with The Little Dipper (Heron Instruments, Inc, Burlington, Ontario).
### 1.9 References


CHAPTER II

ANALYSIS OF SOIL MOISTURE DYNAMICS UNDER MIXED ANNUAL-
PERENNIAL ECOSYSTEMS

A paper to be submitted to Geoderma
Jose Gutierrez Lopez, Heidi Asbjor森sen, Thomas Isenhart, Matthew Helmers

1 Introduction

Soil moisture dynamics reflect highly interconnected processes such as vegetative cover, soil characteristics, topographic position and precipitation. As a result, soil moisture varies among ecosystems under different land covers, across time, and by depth within the soil profile (Isham et al., 2005; Zhang and Schilling, 2006; Tamea et al., 2009). An understanding of soil moisture dynamics can provide land managers with critical information to increase productivity and better implement conservation practices.

Land cover has been recognized as an important factor controlling patterns of soil moisture by influencing infiltration rates, runoff, and evapotranspiration (Cubera et al., 2004). The variability of biomass under different land covers and its influence on rainfall interception is of particular importance (Brooks et al., 2003; Chang, 2006). Above and below-ground organic matter increases infiltration rate and soil moisture holding capacity, reducing surface runoff and decreasing water yield in vegetated areas (Chang, 2006).
In the Midwest United States, where rowcrops have replaced the majority of the native prairie vegetation, the impact of prairie on the hydrologic balance of the ecosystem has been severely diminished. In Iowa alone, it is estimated that of the historic 12.5 million ha of prairie, only 12,140 ha remain, representing a 99.9% reduction of the native vegetation (Samson and Knopf, 1994). Several researchers have indicated that the increases in the frequency of drought and flooding events, higher fluctuations in streamflow, and increased overland flow are the result of this landscape scale conversion of native prairie vegetation to rowcrop agriculture (Burkart and James, 1999; Schilling and Libra, 2000; Fohrer et al., 2001; Rabalais et al., 2002).

Over the past two decades, there has been growing emphasis on the potential hydrologic benefits of the incorporation of perennial vegetation in areas dominated by rowcrop agriculture (Tilman, 1999; Tilman et al., 2002; Boody et al., 2005). One of the earliest accounts, by Weaver and Flory (1934), suggested that increased drought resistance, the ability to take water from deeper soil depths, and greater plant diversity were advantages of native prairie vegetation over annual crops (e.g. corn, wheat, oats, rye, barley, and sorghums). Recent studies have shown that the incorporation of perennial covers have greater hydrologic benefits for the ecosystem than annual crops (Brye et al., 2000; Zhang and Schilling, 2006). In a smaller scale, Weaver (1941) observed higher water losses (ET) under prairie (Andropogon furcatus) cover than pasture (Bouteloua curtipendula). Additionally, he observed that the removal of aerial biomass lessened transpiration and increased evaporation from the soil surface (Weaver, 1941).
Prairie restoration or reconstruction was first suggested by Ada Hayden (1919), with the goal “to preserve a historic and banished ecosystem” and “to secure the present and the coming generations a heritage” (Smith, 1998). Recently, the restoration or reconstruction of native prairie vegetation has the goal not only to preserve a historic feature, but also to restore such functions as hydrologic regulation (Hernandez-Santana et al., In Press), nutrient regulation (Baer et al., 2002; Brye et al., 2003; Zhou et al., 2010), and water purification (Schilling, 2002), particularly in landscapes dominated by row crop agriculture. Much of the information about the use of perennial vegetation in agricultural areas comes from riparian buffer research, where native shrubs, grasses and trees are combined, primarily to mitigate sediment and nutrient loss (Schultz et al., 2004; Lowrance and Sheridan, 2005; Williard et al., 2005). While numerous studies have assessed the impact of buffers on sediment and nutrient loss and runoff (Schultz et al., 2004; Lowrance and Sheridan, 2005; Williard et al., 2005), very few have quantified the effects on soil moisture dynamics. For example, some work has examined the effect of perennial vegetative strips on soil hydrologic processes at the plot scale and under controlled rainfall conditions (Abu-Zreig et al., 2004; Blanco-Canqui et al., 2004; Gharabaghi et al., 2006), using tilted beds to simulate agricultural runoff in filter strips (Mersie and Seybold, 1997), and through modeling (Fox et al., 2009).

Overall, there is a lack of studies of the impacts of perennial vegetation on soil moisture dynamics, and few have studied the relevance of landscape position for hydrologic regulation (Hoover and Hursh, 1943; Ziadat et al., 2010). In particular, a robust understanding of soil-water dynamics within native prairie vegetation reintroduced
into rowcrop dominated landscapes requires a broader study under field-scale configurations and natural field conditions. Understanding the effects of the topographic location of the prairie strips within a watershed is also of great importance, as it will help in the management of runoff dynamics of zero order watersheds.

This study examines the effects of the reintroduction of native prairie vegetation into agricultural fields on the hydrological balance. The objectives of this study were [1] to assess differences in soil water storage at four intervals (0-30, 30-60, 0-60 and 1-100 cm) and volumetric water content at six depths (6, 10, 20, 30, 60 and 100 cm) under two land covers: native prairie and agricultural crops, and [2] to assess the effects of topographic position, season, soil depth and precipitation on soil moisture dynamics. It was hypothesized that soil water content will be lower (i.e. higher soil water storage capacity) under prairie vegetation compared to row-crops (corn, soybeans) early in the growing season (June, early-July) when prairie plants are active and crops are not fully established, while the reversed pattern will occur during the peak growing season (late July-August) when crops have reached maximum growth rates.

2 Materials and Methods

2.1 Study area

The research was conducted in the Neal Smith National Wildlife Refuge near Prairie City, Iowa (NSNWR, 41°33´N, 93°16´W). The refuge was created in 1990 with the central mission of converting over 3,400 ha of agriculturally dominated landscape to
native perennial vegetation. To date, the Refuge consists of a mosaic of reconstructions and agricultural land uses with approximately 2500 ha planted to tallgrass prairie through successive annual plantings.

Most soils at the research sites are classified as either Ladoga (Mollic Hapludalf) or Otley (Oxyaquic Argiudolls) soil series, which are highly erodible with slopes ranging from 5 to 14% (NRCS, 2010). Percentages of sand, silt and clay observed in each site are presented in Table II.1. The mean average precipitation registered over the last 30 years (1981-2010) was 910 mm, with the majority of the large storms occurring between May and August (NCDC, 2011). Precipitation data for this study were recorded at the MesoWest (ID = NSWI4) weather station located in the Neal Smith Wildlife Refuge in Jasper County, Iowa. The weather station registered data from March to November in 2007, 2008, 2009 and 2010.

2.2 Experimental design

Twelve zero-order watersheds (ephemeral in hydrologic flow regime) were used in this study under a balanced incomplete block design. The watersheds are distributed in four blocks, located in three sites: Basswood (two blocks), Orbweaver (one block), and Interim (one block), with six, three, and three watersheds per site, respectively (Table II.1). Watershed area ranged from 0.5 to 3.2 ha. One of four treatments was assigned to every watershed (three replicates per treatment). The treatments consisted of strips of native prairie vegetation (hereafter “NPV”) and corn and soybean row-crop vegetation (hereafter “ROWCROP”) planted in four configurations having different amounts and
topographical positions of NPV (100% rowcrop, 10% NPV in the footslope position, 10% NPV in the footslope, side and summit positions, 20% NPV distributed in the footslope, side and summit positions) (Figure II.1). NPV was planted in different topographic positions to assess optimal position to increase hydrological benefits. The NPV was planted in July 2007. The seed mixture used in the planting consisted of approximately 20 native prairie forbs and grasses with four primary species in the mix, including indiangrass (*Sorghastrum nutans* Nash), little bluestem (*Schizachyrium scoparium* Ness), big bluestem (*Andropogon gerardii* Vitman), and aster (*Aster spp.*). This mixture is similar to the one commonly used by the NSNWR staff in prairie reconstruction practices. The width of the NPV varied from 27 to 41 m at the footslope and from 5 to 10 m at the side and summit positions.

Fiberglass access tubes (Delta-T Devices), used to monitor soil moisture (Figure II.2) were installed in the summit, side and footslope positions of the watershed. These access tubes were installed inside the NPV or in the row-crop area, in the approximate center of the watersheds to evaluate the effects of land cover on soil moisture dynamics (Table II.2).

2.3 Data collection and processing

2.3.1 Conversion of dielectric constant to volumetric water content

Soil moisture was assessed by recording voltage output (mV) approximately every two weeks from April through November in 2007, 2008, 2009 and 2010 (Table II.2), with a HH2 Meter, using a Theta (ML2, Delta-T Devices, Cambridge, UK) and a
PR2 Probe (PR2, Delta-T Devices, Cambridge, UK). The Theta Probe was used to measure voltage outputs in the upper 5 cm of soil and the PR2 probe to take readings at depths of 10, 20, 30, 40, 60 and 100 cm (Figure II.2). Three readings were taken around each access tube using the Theta probe and three inside the access tubes at each depth using the PR2 probe, twisting the probe 120° between readings to get a more representative reading. Voltage outputs were averaged and then converted to the square root of permittivity as follows:

\[ \sqrt{\varepsilon} = 1.125 - 5.53V + 67.17V^2 - 234.42V^3 + 413.56V^4 - 356.68V^5 + 121.53V^6 \]  

(1)

Where \( \varepsilon \) is the permittivity, and \( V \) the voltage output (mV)

The square root of permittivity was then use to calibrate soil moisture readings via linear regression with the observed volumetric water content (see below for details).

2.3.2 Soil bulk density

Soil bulk density (\( \rho_b \)) was estimated by taking soilcores at 10, 20, 30, 60 and 100 cm at three topographic locations in every watershed (summit, side and footslope). Soil samples were taken using a soil corer fitted with aluminum rings of 7.5 cm in diameter by 7.5 cm in length. The samples were weighed and dried in a drying oven at 105°C for 48 h at the Porous Media Lab, Agricultural and Biosystems Engineering, Iowa State University. Soil bulk density (\( \rho_b \)) was estimated by dividing dry weight of every sample over the volume of the aluminum ring.
2.3.3 Calibration of soil moisture readings

Soil moisture readings were calibrated by assessing gravimetric water content ($\theta_g$) for one sensor of the Theta, and six sensors of the PR2 probe. Soil samples were collected at a distance of one meter around the access tubes during periods of high and low soil moisture contents (Figure II.2), at the same time the voltage output was recorded with the HH2 Meter. The number of samples collected varied per site, 2 to 4 sets of samples were collected each year, except in 2009 where no soil samples for calibration were collected. All the soil samples were dried for 48 h at 104°C in a drying oven and $\theta_g$ was estimated following the methodology indicated by Hillel (2004).

Soil parameters $a_0$ and $a_1$ (Equation 2) were estimated through a linear regression between observed $\theta_v$ and the root square of the permittivity ($\sqrt{\varepsilon}$), as estimated by Equation 1. The $\theta_v$ used in the calibration was estimated by multiplying the $\theta_g$ of a specific depth by the observed $\rho_b$ of that same depth. A total of 273 sets of parameters were estimated.

After the soil parameters ($a_0$ and $a_1$) were estimated, all voltage outputs were converted to $\theta_v$ using the following equation:

$$\theta_v = \frac{\sqrt{\varepsilon} - a_0}{a_1}$$  \hspace{1cm} (2)

Where $\theta_v$ is the volumetric water content (cm$^3$/cm$^3$). Default $a_0$ and $a_1$ parameters are provided by Delta-T Devices, for mineral (1.6, 8.4) and organic (1.3, 7.7) soils. However, for optimum accuracy we obtained soil parameters for our specific soil types.
\( \theta_v \) was converted to soil water storage (SWS) by multiplying the \( \theta_v \) by the area of influence of a given sensor in both the Theta and PR2 probes (Figure II.2). SWS was estimated at four intervals (0-30, 30-60, 0-60 and 0-100 cm), by summing the SWS from the corresponding depths of every interval. Depths 6, 10, 20 and 30 cm where used in the 0-30 interval, 40 and 60 cm for the 30-60 cm interval, depths 6, 10, 20, 30, 40 and 60 were used for the 0-60 cm interval, and all depths were used in the 0-100 cm interval.

2.4 Statistical analysis

Significant differences in \( \theta_v \) (\( \alpha = 0.05 \)) by individual depths (6, 10, 20, 30, 40, 60, and 100 cm) and by interval (0-30, 30-60, 0-60 and 0-100 cm) were determined using the Glimmix Procedure in SAS (SAS, Institute, 2001), with site, watershed, year, precipitation (the sum of the precipitation registered within the 7 days prior to the voltage output reading), landcover (NPV and ROWCROP), topographic position and season as fixed effects. Individual access tubes and observation dates were analyzed as random effects because of the nature of our analysis (repeated measurements), and to account for variability within seasons in the case of observation dates. The analysis of SWS was first run using all four years (hereafter, “four-year” analysis), to increase the power of our analysis for variables like landcover and topographic position, and then for each individual year from 2007 to 2010 (hereafter “annual” analysis), to detect specific differences within years in variables like season or landcover.

To assess the effects of the position of NPV within the watershed on soil water storage, the interaction between land cover and topographic position was included in the
model. The interactions of season and landcover and season with topographic position were also included to assess the effectiveness of different land covers on soil water storage throughout the growing season and how it is affected by topographic position. In the four-year analysis the year variable is included to account for the variability of the annual crop rotation (corn, soybeans).

3 Results

3.1 Precipitation

Precipitation from March to November was 900, 951, 866, and 1326 mm in 2007, 2008, 2009 and 2010 respectively (Figure II.3). The distribution of the precipitation varied in all four years with relatively wet and dry conditions observed in different months each year (Figure II.4). Relative to the 30-year average, annual precipitation was 1% lower, 4% higher, 5% lower, and 45% higher in 2007, 2008, 2009, and 2010, respectively.

3.2 Soil bulk density

The average $\rho_b$ of all samples (n=180) was 1.43 g·cm$^{-3}$, and ranged from 1.06 to 1.78 g·cm$^{-3}$ with a standard deviation of 0.12 g·cm$^{-3}$. For most of the sites sampled bulk density increased with depth (Figure II.5). Although visual differences were observed in Figure II.5, no statistical analysis was performed on bulk density samples.
3.3 Volumetric water content by depth

Marked differences in volumetric water content (VWC) were observed by sampling depth. Shallower depths (5, 10, 20, and 30 cm) showed greater variation than deeper depths (60 and 100 cm) in all four years. The variation of $\theta_v$ at shallower depths was closely associated with rainfall (Figure II.6 and Table II.3). There was no effect of year on mean annual $\theta_v$ estimated by depth (Table II.3).

The statistical four-year analysis indicates that VWC is highly influenced by site, watershed, precipitation, landcover, position and the interaction season*position (seasonal differences in VWC by topographic position) (Table II.3). Year, the interaction position*landcover (topographic differences in VWC by landcover), and season*landcover (seasonal differences in VWC by landcover) had less influence on the $\theta_v$ and no effect of year and season was observed for any depth (Table II.3).

3.4 Average soil water storage in the upper 60 cm

Average SWS in the upper 60 cm across all 12 study watersheds and for the entire four-year study period ranged from 15.3 to 38.9 cm, with a standard deviation of 2.7 over the four years of our study. Comparisons of SWS by year showed a slight increase in the SWS variability within the upper 60 cm from 2007 to 2010, with 2007 having the lowest SWS (15.3 cm) and 2009 the highest (38.9 cm) (Table II.4). Annual averages within this soil depth range were 25.4, 26, 25.4 and 26.5 cm for 2007, 2008, 2009 and 2010 respectively; these values closely mirrored the total precipitation observed by year.
Analysis of the annual average trend of SWS across all watersheds in the upper 60 cm showed that SWS increased early in the growing season around the months June-July, and decreased in July, August and September (Figure II.7). An increase of SWS was observed after September in the four years of our study. The low inputs of water from precipitation in the months of October and November in 2010 resulted in an overall decrease in the observed SWS in the upper 60 cm during this period.

3.4.1 Soil water storage by depth increments

The four-year statistical analysis by soil depth increment showed that watershed, precipitation, landcover, position and the interaction season*position (seasonal differences in SWS by topographic position) had a strong influence on the SWS observed in our study (Table II.5). Site, the interaction position*landcover (topographic differences in SWS by landcover) and season*landcover (seasonal differences in SWS by landcover) showed less influence on the SWS of the entire soil profile, while year and season had no effect at any depth increment.

The annual statistical analysis showed effects of land cover from 0-30 cm in 2007 (Table II.6), and no effect of land cover in any of the other years for any of the four increments analyzed (Tables, II.6, II.7, II.8 and II.9). Our results showed a significant effect of the interaction season*landcover and season*position for nearly all soil depths in 2007 (Table II.6), and for the interaction season*landcover on two soil depth depths in 2009 (Table II.8). In 2008, only the interaction season*position showed statistical differences in the 0-100 cm interval, the other factors appear to have no effect on SWS.
3.5 Effects of land cover on soil water storage

As shown on Table II.5, in the four-year analysis, landcover had a significant effect on SWS at all the four increments analyzed (0-30, 30-60, 0-60 and 0-100 cm). For the soil depth increment 0-60 cm, we observed differences in soil water storage among land covers that peaked around mid-growing season and decreased towards the end of the growing season, with the largest differences found in early August in 2007, mid October in 2008, mid August in 2009 and late June in 2010 (Figure II.8; Table II.12). The annual statistical analysis showed no significant effects of land cover at this increment (increment 0-60 in Table II.6, II.7, II.8, and II.9).

When comparing mean annual SWS in the upper 60 cm, SWS was lowest under ROWCROP in 2007, and under NPV in 2008 and 2010, while there were no significant differences in 2009 (Table II.10; see Tables II.6, II.7, II.8, and II.9 for statistical analyses). Trends of average SWS by land cover shifted seasonally and by observation date (Figure II.8). In 2007, SWS was slightly lower under NPV than ROWCROP from May until the end of June, but the trend shifted from July to mid-October with a difference of as much as 12% in early August. The annual analysis of SWS for this soil
increment (0-60 cm) showed significant (p=0.001) seasonal differences between land covers (interaction season*position; Table II.6). In 2008 SWS was lower under NPV for most of the observation dates, with as much as 5% lower SWS under NPV than ROWCROP. The only exceptions were early-September and mid-October, when SWS under ROWCROP was 5 to 6% lower than NPV. The annual statistical analysis showed no influence of any variable at this depth (Table II.7). In 2009 SWS was lower under NPV from May through late-July, with a maximum difference of 4% in late June. This trend shifted from August to mid-October in 2009 when SWS was lower under ROWCROP, with the greatest difference of 5% observed in late-August. The annual analysis showed significant (p=0.0029) seasonal differences among land covers (interaction season*landcover; Table II.8). In 2010, SWS was lower under NPV on 15 of the 16 observation dates. Maximum differences of 5 and 4% were observed in late-June and mid-September, respectively. Differences in SWS under NPV and ROWCROP were less pronounced in 2010, due to the influence of precipitation, which had a significant (p=<0.0001) influence on SWS for both land covers (Table II.9).

When comparisons of SWS between land covers are limited to the top 0-30 cm, additional differences were observed by season and observation date (Figure II.9). On an annual basis, SWS within this increment was higher under NPV than ROWCROP in 2007, and lower under NPV than ROWCROP in 2008, 2009, and 2010 as the prairie strips were becoming better established. Annual totals are shown in Table II.4. The annual statistical analysis of this soil increment (0-30 cm) indicates significant (p=0.0056) differences in SWS by land cover in 2007 (Table II.6), but no significant
differences in consecutive years (Table II.5). The four-year analysis showed no effect of season on SWS in any of the soil depths analyzed for this soil increment (0-30 cm) (Table II.6, II.7, II.8 and II.9). Conversely, the analysis of seasonal SWS variations of land cover by season (interaction season*landcover), significant differences were observed in the upper three soil intervals in 2007 (Table II.6), 2008 showed no differences (Table II.7), 30-60 and 0-60 cm were significantly different in 2009 (Table II.8) and no significant differences were observed in 2010 (Table II.9 and II.12). SWS was lower under NPV early in the growing season and at the end of the growing season in 2008, 2009, and 2010 by 0.4 cm, 1 cm, and 0.9 cm, respectively.

3.6 Effects of topographic position on soil water storage

Topographic position (summit, side and footslope) strongly affected SWS, as shown in the four-year statistical analysis for all increments (0-30, 30-60, 0-60 and 0-100 cm) (Table II.5 and II.11). Mean annual SWS in the upper 60 cm was consistently higher in footslope than in the summit position in each of the four monitored years. This pattern was not consistent when comparing differences between footslope and sides positions. Annual average SWS in the side position was lower than summit in 2007 and 2009 (Figure II.10).

When comparing results by observation date (Figure II.11), SWS in the upper 60 cm was higher in the footslope position than the summit position for the great majority of the observed dates (Figure II.11). This trend was consistent under conditions of both low and high total SWS. SWS in the summit positions exhibited a greater response to
precipitation than footslope positions. Side positions exhibited the greatest variation in SWS among all the measured dates, with values as low as 22.7 cm in August 2007, and as high as 29.91 cm in June 2010. In the analysis by year, no statistical differences were found for any soil increment in any year (Tables II.6, II.7, II.8 and II.9). However, in the four-year analysis, a single depth interval (0-100) showed significant (p=0.0032) differences of SWS by topographic position among land covers (interaction position*landcover; Table II.5). The annual statistical analysis showed no statistical differences in SWS by topographic position in any depth analyzed in any year (interaction position*landcover in Tables II.6, II.7, II.8, and II.9). The four-year analysis found significant seasonal differences of SWS by topographic position for all intervals (Table II.5 and II.12). However, the annual analysis showed seasonal differences in SWS by topographic position for all the soil intervals analyzed in 2007 (interaction season*position in Table II.6), significant differences in one interval only in 2008 (Table II.7), no significant differences in 2009 (Table II.8), and no differences in 2010 (Table II.9). The annual averages observed by are presented in Table II.12.

3.7 Other factors influencing soil moisture

In three sites, Basswood 4 and 5 in the sides position and Interim 3 in the summit position, excessive water at the soil surface created a wet soil environment that resulted in consistently higher readings of VWC at 5 and 10 cm at these sites with an average for the entire observation period (2007 to 2010) of 0.49 \( \text{cm}^3\cdot\text{cm}^{-3} \) at 5 cm and 0.50 \( \text{cm}^3\cdot\text{cm}^{-3} \) at 10 cm in Basswood 4 and 0.50 \( \text{cm}^3\cdot\text{cm}^{-3} \) at 5 and 10 cm in Basswood 5. The average of
the gravimetric samples collected throughout the study at Basswood 4 position side from 0-15 cm was 0.33g/g (standard deviation = 0.05), and the average of the gravimetric samples in Basswood 5 side was 0.31g/g (standard deviation 0.05), indicating the little variation in soil water content in these sites.

Conversely, in the watersheds Basswood 6 and Interim 3, low VWC values were recorded throughout our study in the side position, with little variation over time. In Basswood 6 side position, VWC values varied very little at 20 cm (mean = 0.34 cm$^3$ cm$^{-3}$, SD = 0.014), 30 cm (mean = 0.34 cm$^3$ cm$^{-3}$, SD = 0.000) and 100 cm (mean = 0.34 cm$^3$ cm$^{-3}$, SD = 0.020) during the four years of our study. The mean of 6 gravimetric samples collected in Basswood 3 in the sides position, over the four years of this study showed an average of 0.23 g/g (SD = 0.02) at 20 cm, 0.22 g/g (SD = 0.02) at 30cm and 0.18 g/g (SD = 0.01) at 100 cm, showing the natural low variation in gravimetric water content at these depths.

Similarly, in Interim 3, position side, low VWC values were observed at all three depths (20 cm: mean = 0.29 cm$^3$ cm$^{-3}$; SD = 0.01; 40cm: mean = 0.25 cm$^3$ cm$^{-3}$; SD = 0.02; 60 cm: mean = 0.27 cm$^3$ cm$^{-3}$; SD = 0.01; 100 cm: mean = 0.25 cm$^3$ cm$^{-3}$; SD = 0.00). The mean of the gravimetric samples collected for these points were: 0.22 g/g at 20cm (SD = 0.03), 0.18 g/g at 40 cm (SD = 0.02), 0.18g/g at 60cm (SD = 0.02) and 0.16 g/g at 100cm (SD = 0.01), showing also natural low variation of gravimetric water content at these depths.
4 Discussion

Previous research has shown that the reintroduction of NPV can reduce total SWS via evapotranspiration, or increase infiltration through the modification of soil properties such as porosity (Weaver, 1927; Weaver and Flory, 1934; Weaver, 1954; Ehrenreich and Aikman, 1963). By reducing the total amount of water in the soil via evapotranspiration, native prairie vegetation allows for the retention of higher amounts of precipitation or runoff (Hernandez-Santana et al., In Press), which would otherwise contribute to surface runoff and transport of nutrients (Schilling, 2002; Fox et al., 2009) and sediments (Gharabaghi et al., 2006) to receiving waters.

In this study we analyzed the effects of the reintroduction of strips of NPV into agricultural lands on $\theta_v$ and SWS in twelve small watersheds. The influence of precipitation, season, soil depth and topographic position on SWS characteristics was analyzed. Our results indicate that by the second year after establishment, the reintroduction of NPV can contribute to the water balance of the watershed and to the ecosystem, by reducing soil moisture content via evapotranspiration, thereby increasing soil moisture storage capacity and controlling surficial water flow (Hernandez-Santana et al., In Press).

4.1 Variations under land cover

In our study, the four-year analysis revealed that land cover significantly affected both VWC and SWS (Table II.3 and II.5). Zhang and Schilling (2006), conducted a study in the same research area to study the effects of land cover (grassland and bare ground)
on soil moisture, evapotranspiration and groundwater recharge, and found similarly to our results that land cover directly influences soil moisture dynamics. In that study, in the absence of land cover (bare ground), soil moisture remained higher due to a lower ET, which in turn resulted in higher fluctuations and higher recharge of groundwater in the bare ground compared to the grass-covered areas. Qi et al. (2011) monitored SWS from 2006 to 2008 and observed similar results in a comparison of six land covers including perennial forage and conventional corn and soybean crops. Their results support our findings that perennial vegetation helps to reduce SWS by increasing ET. In our analysis of VWC by depth, two shallow depths (5 and 10 cm) and a deep one (60 cm), showed no effect of land cover. Precipitation in the upper two depths significantly influenced VWC (p=>0.0001). Conversely, the analysis of SWS showed a significant influence of land cover at the four increments analyzed. This result could be due to the significant influence of factors such as precipitation that might overcome the influence of land cover in shallow depths (5 and 10 cm) when analyzed individually, as shown in Table II.3.

Studies that have compared the influences of land cover on soil moisture dynamics, have found more significant differences among land covers when these are analyzed comparing soil intervals (Weaver, 1941; Brye et al., 2000; Cubera et al., 2004; Enloe et al., 2004; Qi and Helmers, 2010; Qi et al., 2011), rather than specific depths. Further, in locations having high moisture content soils during spring and fall (i.e. central Iowa), the effects of land cover on soil moisture dynamics can be better studied at shorter soil intervals (from 10 to 30 cm). In this study, we found different statistical differences
when SWS was analyzed for the entire soil profile than when it was analyzed by smaller soil intervals.

The observed SWS by land cover (Figure II.8) closely followed the development of the strips of native prairie vegetation over time. Researchers have found that the development stage of the vegetation greatly influences soil moisture dynamics (Weaver, 1941; Brye et al., 2000; Cubera et al., 2004; Qi and Helmers, 2010; Qi et al., 2011). In a controlled experiment designed to compare water loss between prairie and pasture using phytometers, Weaver observed that the water loss profile was dictated by the amount of functioning vegetation demanding water (Weaver, 1941). In this study, the initially higher SWS observed in 2007 under prairie could have been a response of soil moisture accumulation in the soil due to the lack of actively transpiring vegetation together with the relatively shallow rooting depth of young plants, which resulted in a statistically significant difference among land covers, particularly in the first 30 cm of soil (Table II.6).

While the NPV was planted in the strips in July 2007, little vegetation was observed throughout the growing season. The NPV developed more aerial biomass in 2008, principally weeds and a few targeted prairie species, which were mowed for the first time from the 19 to the 21 of June 2008 and a second time in late August 2008. In this year, SWS in the upper 60 cm was lower under NPV, with exception of some sampling dates (May 16, September 2, and October 16, the later was included in the analysis but is not shown in the figures). The first observation seemed to be a continuation of the tendency observed at the end of 2007. Shi et al (2007), and Chen et al
(2007), have identified antecedent or pre-existing soil moisture conditions as a factor influencing the soil moisture differences observed at a given time under a given land cover. Values observed on September 2, 2008 could be a result of the removal and thus the modification of the aerial biomass conducted in late August, an effect on soil hydrology previously documented for forested (Hamilton et al., 1983), open woodlands with scattered oak trees (Cubera et al., 2004), crop lands (Nejadhashemi et al., 2011; Qi et al., 2011; Bagley et al., 2012), and native grasslands (Ehrenreich and Aikman, 1963). The low SWS under ROWCROP observed on October 16, 2008 appear to be an effect of the number of values averaged for that date and their specific values. For this date only the observations for the Interim site were included, due to an equipment malfunction. As discussed earlier, low values of VWC and thus SWS were observed in the side position of Interim 3 (100% rowcrop treatment), which highly influenced mean SWS under rowcrop.

Removal of the aerial biomass through diverse processes (i.e. grazing, mowing, burning, cutting) has been shown to influence soil moisture and run off dynamics (Brooks et al., 2003; Hillel, 2004). In forested and grassland areas, the removal of the land cover has shown to increase downstream water yields (Hamilton et al., 1983; Bruijnzeel, 1990; FAO, 2008). Light or selective removal of land cover appears to have little impact on the total water yield, however it has been shown that the effects in water yield increases as the removal of the land cover increases (Bruijnzeel, 1990).

In 2009, the NPV was mowed on June 25, which could have led to the high SWS observed under NPV in the upper 60 cm. As the land cover is removed, soil water loss decreases due to a decrease in ET, and evaporation increases when insolation due to the
presence of soil cover increases (Weaver, 1941; Hillel, 2004). SWS in the upper 60 cm under prairie was higher after August, with this difference decreasing by the end of the year. However, the observed SWS from 0-30 cm (Figure II.9) under NPV was similar to SWS under ROWCROP in August and then progressively decreased under NPV as the year progressed, showing lower SWS under NPV starting mid-September. Land cover thus appeared to have a higher effect in the upper 30 cm of soil. Contrary to these results, in their comparison of the water budget of prairie and maize land covers, Brye et al (2000), found consistently higher values of volumetric water content under prairie, compared to other two land covers consisting of corn, however, the comparisons in this study were done in large soil intervals (0-70, and 80-140 cm). We have indicated previously that among small plant species, it appears that differences in soil moisture are easier to identify when compared in shorter intervals (from 10 to 30 cm).

In 2010, no management was imparted to the NPV until the end of the year. In this year, after the crops were harvested, the biomass in the NPV was cut and baled on October 30, 2010, with an average of 12.7 ton·ha⁻¹ harvested, as part of the management of the NPV. Despite being the year that received the highest total precipitation (1326 mm from Match to November), SWS was lower under NPV during most of the year in the 0-60 cm (Figure II.8) increment and during the entire year from 0-30 cm (Figure II.9). The study conducted by Hernandez-Santa et al, (In Press) showed that runoff was lower in watershed covered with NPV. This result may be attributed to the higher soil water use under native prairie cover and its more advanced stage of establishment and maturity. Early studies of the prairie ecosystem by Weaver (1927; 1934; 1941) found that the water
demands of the ecosystem are highly controlled by the vegetative stage and development of the plants. In a recent study, Mateos-Remigio et al. (in review.) used the heat balance method to measure plant water use at the NSNWR, and showed that water use was strongly governed by plant phenology and season, and that cumulative water use on a leaf area basis was greater for native prairie C$_3$ and C$_4$ plants compared to the annual C$_4$ crop, corn.

4.2 Variations under topographic position

It has been noted by different authors that slope or topographic position are key factors that contribute to soil moisture dynamics (Chang, 2006; Shi et al., 2007; Ziadat et al., 2010). In fact, Roessel (1950) advised caution when comparing watersheds based on their land cover only, since topographical factors may override the effects of land cover. In the four-year analysis of SWS and VWC by topographic position, we observed significant differences among topographic positions on the observed VWC in the upper six soil depth intervals and SWS in all four intervals analyzed (Table II.3 and II.5). In contrast, the analysis of SWS by year, showed no effect of topographic position in any of the years, which could be due to the variability within each year, or due to a loss in estimation power of the statistical analysis when it is split by year.

The effect of topographic position has been previously studied, with varying results. Ziadat et al (2010), studied soil moisture content differences in four topographic positions (summit, shoulder, backslope and toeslope) in five different transects of different slope characteristics and found no marked differences among topographic
positions in the five studied transects. Conversely, the results reported by Fu et al. (2003), showed a strong influence of topographic position on soil moisture content in each of the five land cover studied, with a steady increase in soil moisture content from summit to footslope. Averaged on an annual basis (Figure II.10), our results indicate that the upper parts of the watershed (summit) tend to have lower SWS than the lower parts (foot). Particularly in the first 30 cm of soil profile. The middle part of the watersheds (side) showed varying results each year, however it was never higher than the foot. These results follow the water movement laws and principles dictated by homogeneous porous medium: gravity, capillarity and suction primarily (Brooks et al., 2003; Hillel, 2004). Hillel indicates that after infiltration, the gravitational head gradient is the only force that moves water into the soil (Hillel, 2004), and this gravitational gradient tends to move water from the upper parts of the watershed towards the lower parts (Fetter, 2001), which can explain the annual average values found in this study.

We evaluated the effects of the strategic placement of strips of perennial vegetation into three different topographic positions within a watershed. Independent of the effects of topographic position on soil moisture dynamics, one of our objectives was to determine to what extent soil moisture is affected by the topographical position of the NPV placement within a watershed. Our four-year statistical analysis showed that SWS does not vary significantly by topographic position under each vegetative cover studied (interaction position*landcover; Table II.5), as the relative position of the vegetative cover only had a significant effect on VWC at a single interval (0-100 cm). Roessel (1950), mention that the effects of topographic position can in some cases override
effects of land cover on soil moisture dynamics, which we observed in our four-year analysis of VWC (Table II.3) and SWS (Table II.5), which showed strong influence of topographic position when analyzed as independent variable. However, the four-year analysis of the VWC differences by topographic positions under each land cover (interaction position*landcover) showed only one depth increment with significant differences (0-100 cm; Table II.4). The annual analysis showed no significant differences at any depth increment for year (Tables II.6, II.7, II.8 and II.9) which could be due to: (1) differences are not constant enough to be identified as significantly different by our analysis, (2) differences by watershed are greater than the differences among land covers in one watershed, as shown in Table II.5, or (3) that differences in SWS tend to be more distinguishable on a seasonal basis.

VWC and SWS were no significantly different when analyzed by season as an independent factor in the four-year analysis (Table II.5). When analyzed as the interaction season*landcover (seasonal changes in soil moisture under a given land cover), we found little influence on VWC (Table II.3) and statistically different SWS in two soil depths (Table II.5). The analysis by year showed significant seasonal differences of SWS under each land cover in three soil depth intervals in 2007 (Table II.6), no differences in 2008 (Table II.7), two depths with significant seasonal differences in 2009 (Table II.8) and no difference sin 2010 (Table II.9). Our data indicates strong seasonal variations in SWS in the upper soil depth increments (0-30, 30-60 and 0-60 cm) of soil, that are likely the result of a combination of precipitation patterns and seasonal changes in water use by the different vegetation. On average, June-July had the highest values of
SWS in the upper 60 cm (26.5 cm), August September had the lowest (25.1 cm). Table II.12 shows the seasonal averages found by year under each land cover.

Based on our results, it also appears that the benefits of the reintroduction of native prairie vegetation into agricultural watersheds, present a threshold effect at high and low soil moisture contents. In 2009, when we had the lowest values of SWS, no significant differences were detected among the two land covers studied, particularly in the upper 30 cm of soil (Figure II.9). In 2010, when we had the highest precipitation among the four years of our study, the differences in SWS between land covers was reduced, as compared to other years (Figure II.8).

4.3 Limitations to this study

Several factors were thought to influence the observed VWC apart from the experimental treatments, and may have influenced the results obtained in this study. First, the four-year analysis showed significant differences in both VWC by depth and SWS by depth increments as shown in Table II.3 and II.5. High VWC values observed in the side positions in Basswood 4 and 5 and at the summit position of Interim 3 are possibly a result of: (a) a broken subsurface drainage tile that releases water at these positions, (b) the presence of a soil layer with low hydraulic conductivity that impedes water from moving into the soil matrix, or (c) topographic and geological conditions that cause groundwater to flow out of the soil surface to create return flow.

Low VWC values observed in the side position of Basswood 6 and Interim 3 could have resulted from: (a) little or no contact area between the soil and the fiber glass
access tubes caused by a incorrect installation, soil contraction or physical deterioration of the contact between access tube and soil over time, yielding a low voltage output, (b) physical characteristics of the soil (e.g. high porosity, high organic matter content) or of a structure (e.g. roots) within the soil at that given point, or (c) presence of openings in the soil (burrows) created by animals or roots from pre-existing trees.

Although we acknowledge the intrinsic differences among some watersheds, it is important to denote that our statistical analysis accounts for variations within access tubes or observation points (specific position at a specific watershed within a specific site), by including these as random effects in our analytical model. Similarly, the results of our analysis include all the watersheds, combining data for VWC or the SWS across of all the watersheds and topographical positions at a given depth or increment in the case of SWS. Natural variation in SWS and VWC was thus accounted for in our statistical analysis, as one of our original goals is to study soil moisture dynamics under natural field conditions.

5 Conclusions

In this study, we monitored twelve small zero-order watersheds to assess how the reintroduction of native prairie vegetation into agriculturally dominated landscapes affected soil moisture dynamics, specifically volumetric water content (VWC), at seven soil depths (5, 10, 20, 30, 40, 60, 100), and soil water storage (SWS) within four
increments (0-30, 30-60, 0-60 and 0-100). We analyzed the effects of precipitation, land cover, topographic position, season and year on each of these variables.

Lower seasonal SWS was observed under ROWCROP than under NPV in 2007, when the prairie strips were planted. This trend shifted in 2008, 2009 and 2010, which had the greatest amount of total precipitation. Although SWS was not consistently lower under NPV in 2008, 2009 and 2010, significant differences were observed in our statistical analysis by year. The timing of the establishment of NPV played a critical role in explaining observed differences, with SWS under this vegetation type decreasing with time since prairie establishment, except in 2010 where our statistical analysis indicates that precipitation had the highest influence than the other variables in explaining differences in SWS. The SWS differences among land covers were manifested to a greater extent within the upper 0-30 cm relative to 0-60 cm of soil. Topographic position had a direct influence on SWS, with the upper parts of the watershed exhibiting less water storage on average, but higher responses to precipitation. In contrast, high SWS and low response to precipitation was observed in the lower landscape positions.

The lack of significant differences in 2008 and 2010, an anomalously low and high rainfall year, respectively, led us to propose that SWS variations between NPV and ROWCROPS may represent a threshold effect, since SWS differences were not statistically different under these land covers when precipitation and thus soil moisture in the upper soil depths was either lower or higher than average.

Our results have important implications for land managers and scientists, if similar studies are conducted in other agricultural watersheds, NPV can help regulate the
hydrological balance of the watershed. However, special care must be taken since effects on the hydrological balance are not observed the year NPV is planted. During the first year, SWS is likely to be higher under NPV due to the lack of water-demanding vegetation, which eventually may lead to increased runoff and potentially to lower groundwater recharge. Further research is needed to understand the implications for runoff and water yield.
6 References


Hyden, A., 1919. Iowa Parks: Conservation of Iowa Historic, Scenic and Scientific Areas. State Board of Conservation, Des Moines, IA.


Weaver, J.E., 1927. Some ecological aspects of agriculture in the prairie. Ecology 8, 1-17.


Figure II-1. Graphical representation of the four treatments applied to the watersheds. Proportions are not at real scale.
Figure II-2. Diagram of the Theta and PR2 probes, the area of influence used for every sensor and the depths of gravimetric samples used in the calibration.
Figure II-3. Cumulative precipitation by year

Figure II-4. Precipitation registered by day (2007-2010)
Figure II-5. Bulk density by watershed and topographic position
Figure II-6. Average volumetric water by depth

Lines in Figure 6 represent the average of all topographic positions and watersheds for that specific depth. The average includes both land covers (CROP and NPV).
Figure II-7. Average soil water storage in the upper 60 cm for the entire site of study

Average soil water storage was estimated using all topographic positions and watersheds for this interval. The average includes both land covers (CROP and NPV). Gray vertical lines represent the standard error of the mean.
Figure II-8. Average soil water storage in the upper 60 cm by land cover

Soil water storage was averaged using all the topographic positions and watersheds corresponding to a given land cover. Gray vertical lines represent the standard error of the mean. P=Prairie vegetation planted July 7, 2007; M=Prairie vegetation mowed; B=Prairie vegetation baled. Significant statistical differences using Glimmix in SAS are indicated with “*”. See Appendix A and B for specific details. “n” = or < 4 are not shown in this graph.
Soil water storage was averaged using all the topographic positions and watersheds corresponding to a given land cover. Gray vertical lines represent the standard error of the mean. P=Prairie vegetation planted July 7, 2007; M=Prairie vegetation mowed; B=Prairie vegetation baled. Significant statistical differences using Glimmix in SAS are indicated with "**". See Appendix C and D for specific details. “n” = or < 4 are not shown in this graph.
Figure II-10. Average soil water storage in the upper 60 cm in three topographic positions

Annual average soil water storage estimated using values from both land covers of a given topographic position (Summit, Side and Foot). The average is estimated using all readings observed in each of the monitored years (2007, 2008, 2009 and 2010)
Figure II-11. Average soil water storage in the upper 60 cm in three topographic positions

Lines in Figure 11 represent the average soil water storage estimated using values from both land covers of a given topographic position (Summit, Side and Foot)
### Table II-1. Characteristics of the twelve watersheds

<table>
<thead>
<tr>
<th>Watershed</th>
<th>Site</th>
<th>Total prairie cover (%)</th>
<th>Topographic position of prairie cover</th>
<th>Area (ha)</th>
<th>Sand (%)</th>
<th>Silt (%)</th>
<th>Clay (%)</th>
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<tbody>
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<td>Basswood 1</td>
<td>Basswood</td>
<td>10%</td>
<td>Footslope</td>
<td>0.53</td>
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<td>68.88</td>
<td>28.58</td>
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<td>16.81</td>
<td>57.43</td>
<td>25.76</td>
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<td>Footslope &amp; Summit</td>
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<td>28.58</td>
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<td>16.81</td>
<td>57.43</td>
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<td>Footslope &amp; Summit</td>
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<td>57.43</td>
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<tr>
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<td>Basswood</td>
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<td>16.81</td>
<td>57.43</td>
<td>25.76</td>
</tr>
<tr>
<td>Orbweaver 1</td>
<td>Orbweaver</td>
<td>10%</td>
<td>Footslope</td>
<td>1.18</td>
<td>2.26</td>
<td>66.89</td>
<td>30.85</td>
</tr>
<tr>
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<td></td>
<td>12.99</td>
<td>61.22</td>
<td>25.79</td>
</tr>
<tr>
<td>Orbweaver 2</td>
<td>Orbweaver</td>
<td>20%</td>
<td>Footslope, Side &amp; Summit</td>
<td>2.40</td>
<td>2.26</td>
<td>66.89</td>
<td>30.85</td>
</tr>
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<td>12.99</td>
<td>61.22</td>
<td>25.79</td>
</tr>
<tr>
<td>Orbweaver 3</td>
<td>Orbweaver</td>
<td>100% rowcrop</td>
<td>None</td>
<td>1.24</td>
<td>2.26</td>
<td>66.89</td>
<td>30.85</td>
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<td></td>
<td></td>
<td></td>
<td>12.99</td>
<td>61.22</td>
<td>25.79</td>
</tr>
<tr>
<td>Interim 1</td>
<td>Interim</td>
<td>10%</td>
<td>Footslope, Side &amp; Summit</td>
<td>3.00</td>
<td>3.75</td>
<td>69.89</td>
<td>36.38</td>
</tr>
<tr>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>10.52</td>
<td>66.01</td>
<td>23.47</td>
</tr>
<tr>
<td>Interim 2</td>
<td>Interim</td>
<td>10%</td>
<td>Footslope</td>
<td>3.19</td>
<td>3.75</td>
<td>69.89</td>
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<td>Interim</td>
<td>100% rowcrop</td>
<td>None</td>
<td>0.73</td>
<td>3.75</td>
<td>69.89</td>
<td>36.38</td>
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<td>10.52</td>
<td>66.01</td>
<td>23.47</td>
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↑ = Summit  
↓ = Foot  
Soil percentages correspond to the upper 30 cm of soil
<table>
<thead>
<tr>
<th>Watershed</th>
<th>Seasons monitored per year</th>
<th>Position of the access tubes and its land cover</th>
</tr>
</thead>
</table>
| Basswood 1 | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Prairie buffer |
| Basswood 2 | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Prairie buffer |
| Basswood 3 | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Prairie strip  
Side: Rowcrop  
Footslope: Prairie buffer |
Side: Rowcrop  
Footslope: Prairie buffer |
| Basswood 5 | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Prairie buffer |
| Basswood 6 | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Rowcrop |
| Orbweaver 1| 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Prairie buffer |
| Orbweaver 2| 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Prairie strip  
Side: Rowcrop  
Footslope: Rowcrop |
| Orbweaver 3| 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Rowcrop |
| Interim 1  | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Prairie strip  
Side: Prairie strip  
Footslope: Prairie buffer |
| Interim 2  | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Prairie buffer |
| Interim 3  | 4 [Apr-May, Jun-Jul, Aug-Sep, Oct-Nov] | Summit: Rowcrop  
Side: Rowcrop  
Footslope: Rowcrop |
Table II-3. Glimmix Procedure by depth 2007-2010. Dependent variable: volumetric water content (%)

<table>
<thead>
<tr>
<th>Depth</th>
<th>Site</th>
<th>Watershed</th>
<th>Year</th>
<th>Precipitation</th>
<th>Land cover</th>
<th>Position</th>
<th>Season</th>
<th>F (Position)</th>
<th>p (Position)</th>
<th>F (Land cover)</th>
<th>p (Land cover)</th>
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<tbody>
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<td>5</td>
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<td>F=3.37</td>
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<td>F= 8.82</td>
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<td>F= 0.32</td>
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<td>F=8.80</td>
<td>F= 0.76</td>
<td>F= 23.88</td>
<td>F= 20.88</td>
<td>F= 13.50</td>
<td>F= 0.92</td>
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<td>F= 2.87</td>
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<tr>
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<td>F= 7.49</td>
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<td>F= 0.74</td>
<td>F= 2.29</td>
<td>F= 3.43</td>
<td>F= 16.70</td>
<td>F= 1.53</td>
<td>F= 2.59</td>
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<td>p&lt;0.0008</td>
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<td>F= 0.74</td>
<td>F= 2.29</td>
<td>F= 3.43</td>
<td>F= 16.70</td>
<td>F= 1.53</td>
<td>F= 2.59</td>
<td>p=0.0030</td>
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<td>p&lt;0.0008</td>
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<tr>
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<td>F= 9.60</td>
<td>F= 3.02</td>
<td>F= 1.22</td>
<td>F= 5.98</td>
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<td>F= 2.29</td>
<td>F= 3.43</td>
<td>F= 16.70</td>
<td>F= 1.53</td>
<td>F= 2.59</td>
<td>p=0.0030</td>
<td>p&lt;0.0001</td>
<td>p&lt;0.0008</td>
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<td>F= 0.24</td>
<td>F= 6.79</td>
<td>F= 9.60</td>
<td>F= 3.02</td>
<td>F= 1.22</td>
<td>F= 5.98</td>
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<td>F= 16.70</td>
<td>F= 1.53</td>
<td>F= 2.59</td>
<td>p=0.0030</td>
<td>p&lt;0.0001</td>
<td>p&lt;0.0008</td>
</tr>
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<td>F=8.88</td>
<td>F= 0.24</td>
<td>F= 6.79</td>
<td>F= 9.60</td>
<td>F= 3.02</td>
<td>F= 1.22</td>
<td>F= 5.98</td>
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<td>p&lt;0.0001</td>
</tr>
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<td>F= 3.43</td>
<td>F= 16.70</td>
<td>F= 1.53</td>
<td>F= 2.59</td>
<td>p=0.0030</td>
<td>p&lt;0.0001</td>
<td>p&lt;0.0008</td>
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</tbody>
</table>

Depth: in cm
Site: Basswood, Orbweaver and Interim
Watershed: 12 individual watersheds
Precipitation: sum of the precipitation registered in the previous 7 days
Land cover: native prairie vegetation vs. cropland (corn, soybean)
Position: Summit, Side, and footslope
Season: Four seasons, two months each (April-May, Aug-Sept, Jun-Jul, Oct-Nov)
Table II-4. Annual values of soil water storage in the upper 30 and 60 cm

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<td>2009</td>
<td>2010</td>
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<td>1.7</td>
<td>1.7</td>
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<td>23.5</td>
<td>23.8</td>
<td>23.5</td>
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<td>455</td>
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<td>0-60</td>
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<td>Site</td>
<td>Watershed</td>
<td>Year</td>
<td>Precipitation</td>
<td>Land cover</td>
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<td>p=0.7806</td>
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Depth: increments expressed in cm
Site: Basswood, Orbweaver and Interim
Watershed: 12 individual watersheds
Precipitation: sum of the precipitation registered in the previous 7 days
Land cover: prairie vegetation vs. cropland (corn, soybean)
Position: Summit, Side, and footslope
Season: Four seasons, two months each (April-May, Aug-Sept, Jun-Jul, Oct-Nov)
Table II-6. Analysis of SWS by increment. Year: 2007

<table>
<thead>
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<th>Depth</th>
<th>Site</th>
<th>Watershed</th>
<th>Precipitation</th>
<th>Land cover</th>
<th>Position</th>
<th>Season</th>
<th>Season</th>
<th>Season</th>
</tr>
</thead>
<tbody>
<tr>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
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<td>F=1.87</td>
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</tbody>
</table>

Depth: increments expressed in cm
Site: Basswood, Orbweaver and Interim
Watershed: 12 individual watersheds
Precipitation: sum of the precipitation registered in the previous 7 days
Land cover: prairie vegetation vs. cropland (corn, soybean)
Position: Summit, Side, and footslope
Season: Four seasons, two months each (April-May, Aug-Sept, Jun-Jul, Oct-Nov)
Table II-7. Analysis of SWS by increment. Year: 2008

<table>
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<th>Site</th>
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<th>Land cover</th>
<th>Position</th>
<th>Season</th>
<th>Position *Land cover</th>
<th>Season *Land cover</th>
<th>Season *Position</th>
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<td>p=0.2960</td>
<td>p=0.3136</td>
<td>p=0.0109</td>
</tr>
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Depth: increments expressed in cm
Site: Basswood, Orbweaver and Interim
Watershed: 12 individual watersheds
Precipitation: sum of the precipitation registered in the previous 7 days
Land cover: prairie vegetation vs. cropland (corn, soybean)
Position: Summit, Side, and footslope
Season: Four seasons, two months each (April-May, Aug-Sept, Jun-Jul, Oct-Nov)
### Table II-8. Analysis of SWS by increment. Year: 2009

<table>
<thead>
<tr>
<th>Depth</th>
<th>Site</th>
<th>Watershed</th>
<th>Precipitation</th>
<th>Land cover</th>
<th>Position</th>
<th>Season</th>
<th>Position *Land cover</th>
<th>Season *Land cover</th>
<th>Season *Position</th>
</tr>
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<tbody>
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<td>p= 0.1435</td>
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<td>p= 0.4632</td>
<td>p= 0.1342</td>
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Depth: increments expressed in cm  
Site: Basswood, Orbweaver and Interim  
Watershed: 12 individual watersheds  
Precipitation: sum of the precipitation registered in the previous 7 days  
Land cover: prairie vegetation vs. cropland (corn, soybean)  
Position: Summit, Side, and footslope  
Season: Four seasons, two months each (April-May, Aug-Sept, Jun-Jul, Oct-Nov)
Table II-9. Analysis of SWS by increment. Year: 2010

<table>
<thead>
<tr>
<th>Depth</th>
<th>Site</th>
<th>Watershed</th>
<th>Precipitation</th>
<th>Land cover</th>
<th>Position</th>
<th>Season</th>
<th>Position *Land cover</th>
<th>Season *Land cover</th>
<th>Season *Position</th>
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<td>p=0.6249</td>
<td>p=0.3281</td>
</tr>
<tr>
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<td>F=13.42</td>
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<td>F=0.15</td>
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<td>p=0.0905</td>
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<tr>
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<td>F=2.23</td>
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</table>

Depth: increments expressed in cm  
Site: Basswood, Orbweaver and Interim  
Watershed: 12 individual watersheds  
Precipitation: sum of the precipitation registered in the previous 7 days  
Land cover: prairie vegetation vs. cropland (corn, soybean)  
Position: Summit, Side, and footslope  
Season: Four seasons, two months each (April-May, Aug-Sept, Jun-Jul, Oct-Nov)
Table II-10. Annual average soil water storage in the upper 60 cm by land cover

<table>
<thead>
<tr>
<th>Land cover</th>
<th>Estimate</th>
<th>2007</th>
<th>2008</th>
<th>2009</th>
<th>2010</th>
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</thead>
<tbody>
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<td>Rowcrop</td>
<td>Mean</td>
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<td>26.2</td>
<td>25.4</td>
<td>26.7</td>
</tr>
<tr>
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<td>Standard Deviation</td>
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<td>2.8</td>
<td>3.1</td>
<td>3.4</td>
</tr>
<tr>
<td></td>
<td>Min</td>
<td>15.3</td>
<td>17.4</td>
<td>16.2</td>
<td>17.5</td>
</tr>
<tr>
<td></td>
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<td>38.9</td>
<td>38.1</td>
</tr>
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<td>303</td>
<td>281</td>
<td>341</td>
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<tr>
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<td>Mean</td>
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<td>25.8</td>
<td>25.5</td>
<td>26.2</td>
</tr>
<tr>
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<td>Standard Deviation</td>
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<td>1.8</td>
<td>1.7</td>
<td>2.1</td>
</tr>
<tr>
<td></td>
<td>Min</td>
<td>22.8</td>
<td>19.1</td>
<td>19.7</td>
<td>19.2</td>
</tr>
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<td>Max</td>
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<td>29.6</td>
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Table II-11. Average annual soil water storage in the upper 60 cm and standard deviations in three topographic positions (Summit, Side, Foot) under two land covers (Rowcrop, Native prairie)

<table>
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<tr>
<th>Position</th>
<th>Estimate</th>
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<th>2008</th>
<th>2009</th>
<th>2010</th>
</tr>
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<tr>
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</tr>
<tr>
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<td>Max</td>
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<td>29.2</td>
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<tr>
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<td>38.9</td>
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<td>26.6</td>
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<tr>
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<td>2.0</td>
<td>2.1</td>
<td>2.5</td>
</tr>
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### Table II-12. Average annual soil water storage in the upper 60 cm and standard deviations by season under two land covers

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<td>19.0</td>
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<td>Jun-Jul</td>
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<td>24.6</td>
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<td>17.4</td>
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<td>69</td>
<td>36</td>
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</table>

RC = rowcrop
NP = native prairie)
CHAPTER III

ASSESSING WATERFLOW AND UPTAKE DEPTH PATTERNS UNDER MIXED ANNUAL-PERENNIAL ECOSYSTEMS USING STABLE OXYGEN (\textsuperscript{18}O) AND HYDROGEN (\textsuperscript{2}H) ISOTOPES

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Jose Gutierrez Lopez, Heidi Asbjornsen, Thomas Isenhart, Matthew Helmers, Alan Wanamaker

1 Introduction

Understanding the complexity of ecohydrological processes in agroecosystems, such as depth of plant water uptake and the effects of vegetation on soil hydrology requires research approaches that assess the interactions between multiple important and relevant factors that influence water fluxes at a specific site. Given that water is the major factor determining plant productivity (Gholz et al., 1990) and that vegetation directly affects water balance and streamflow (Fohrer et al., 2001; Schilling, 2002), understanding the mechanisms and processes that determine patterns of plant water uptake from soil is crucial for managing agroecosystems for sustained productivity and other ecosystems services.

Variation in water use patterns among plant species has been studied using different methods that assess changes in soil moisture as a measure of ET: direct methods include the use of metal recipients (phytometers) containing transplanted sods of different plant species (Weaver, 1941) and weighing lysimeters (Young et al., 1996; Evett et al.,
2009; Bryla et al., 2010), while indirect methods include neutron scattering probes (Yoder et al., 1998) and time-domain reflectometry (TDR) probes (Qi and Helmers, 2010a; Qi and Helmers, 2010b). However, although these methods provide good information about soil moisture differences among plant communities or soil covers and water use differences at specific depths in the case of the neutron and TDR probes, they lack the capacity to provide information about from where in the soil profile individual plants or species are obtaining water. Such plant or species-specific information on plant water uptake patterns may be particularly important when selecting species for specific management practices (e.g. hydrological services, landscaping) in highly diverse mixed agroecosystems or native prairie communities or when establishing strips of native prairie vegetation (SNPV) for ecosystem restoration purposes.

In agricultural landscapes in temperate-northern regions, most of the water use by crops takes place during the growing season and in the months with the greatest evaporative demand. In the Midwestern U.S., studies have shown that annual crops take most of their water from the upper 30 cm of soil which is where most of the root system is concentrated in crop species like corn and soybeans (Asbjornsen et al., 2007; Nippert and Knapp, 2007; Asbjornsen et al., 2008). Conversely, native prairie vegetation is characterized by a fine and extremely branched root system, that extends to depths greater than 1.5 m (Weaver, 1931; Weaver et al., 1934), allowing it to access water from deeper soil profiles than rowcrop species. These contrasting rooting patterns can result in differences in depth of plant water uptake (DWU) among different plant species and vegetative cover types (Zhang and Schilling, 2006).
Recent studies are starting to investigate differences in DWU using stable isotopes, a method that allows researchers to infer from which depths within the soil profile co-existing plant species are acquiring water (Araki and Iijima, 2005; Asbjornsen et al., 2007; Nippert et al., 2010; Wang et al., 2010). These studies have provided good insights about DWU in both annual and perennial vegetation. For example, Nippert an Knapp, (2007) in their study of C₃ and C₄ plants growing in a native prairie in Kansas, observed interspecific differences in DWU in response to changes in water availability: when water was available in the upper 30 cm, all plant species took water from shallower sources, but during dry periods, C₃ species used proportionally more water from deeper depths than C₄ species. In a study comparing water uptake patterns by corn and native prairie species in central Iowa, Asbjornsen et al. (2007; 2008) found that early in the season when water was abundant, C₃ and C₄ plants extracted water from the upper 20 cm of soil, but as water became progressively more limiting, C₃ shrubs and trees in the savanna and woodland ecosystem (Quercus alba, Symphoricarpos orbiculatus and Carex sp.), shifted their water uptake to deeper horizons, and C₄ species (Andropogon gerardii and Zea mays) used water from shallower soil depths. Seasonal variation in water uptake has also been documented for annual crops. For example, Wang et al. (2010), found that summer corn extracted water from the upper 20 cm in the jointing and fully ripe stage, and from as deep as 50 cm in the flowering state. Under controlled conditions of water availability and soil compaction, Araki and Iijima (2005) found that variations in depth of water uptake by rice (Oryza sativa L.) was also influenced by the availability of water in the top soil, when water was restricted in the upper layers of soil, plants took water from
deeper soil layers.

Despite the substantial research conducted on DWU in agricultural crops and prairie vegetation (above), and the use of perennial vegetation in waterways or buffer strips for conservation purposes (Schultz et al., 2004; Williard et al., 2005; Gharabaghi et al., 2006; Fox et al., 2009; Zhou et al., 2010), we are unaware of previous studies that have examined DWU uptake patterns between native prairie vegetation and annual crops established as mixed agroecosystems. Under these conditions, competition over water resources may develop between SNPV and crops that may compromise the health of the entire ecosystem. More research is needed to enhance understanding of patterns of DWU between annual crops and reconstructed prairie vegetation to allow land managers and scientists to make more informed decisions regarding the incorporation of NPVS to enhance regulation of the hydrological balance in rowcrop dominated landscapes. In particular, because C₃ forbs and C₄ grasses may vary widely in water use patterns, knowledge about these differences can be critical for determining the most effective combination of plant species when designing NPVS for specific objectives.

The use of stable isotopes to assess DWU by plants relies on: (a) the presence of a natural gradient in $\delta^{18}$O and $\delta^2$H in the soil profile ($< 3 \%e$ and $< 30 \%e$ in $\delta^{18}$O and $\delta^2$H respectively preferentially), and (b) the ability to obtain the right isotopic signature from a given soil depth. However, natural gradients are not always present and there are significant limitations to relying solely on naturally occurring stable isotopes, specially without the presence of clear isotopic gradients, especially in humid environments where frequent rainfall together with mixing of water having different source isotopic
concentrations can create ‘noisy’ vertical concentrations in the soil profile rather than a clear and continuous gradient. In other words, when assessing DWU the isotopic concentration of the plant tissue may match the isotopic value of more than one layer of soil (Moreira et al., 2000). Numerous studies have faced this problem, which has limited the interpretation of their data and ability to infer DWU (Moreira et al., 2000; Asbjornsen et al., 2007; Asbjornsen et al., 2008). As an alternative approach, researchers have used manipulative irrigation experiments whereby water enriched in the stable isotope is applied to the study area as a means of artificially establishing the isotopic gradient in the soil profile (Yoder et al., 1998; Araki and Iijima, 2005; Rowland et al., 2008).

In clay rich soils, water extraction for isotopic analysis in DWU studies presents a mayor challenge, since strong intramolecular forces (e.g. van der Waals) tend to retain hydrogen and oxygen molecules (Hillel, 2004), thus increasing the water extraction time needed to get a unfractionated sample. Araguás-Araguás et al (1995), extracted water from clay rich soils (50 to 80 % clay content) for up to 7 hours to get unfractionated samples, and suggested that calibration is required for specific soil types. In a recent study, West et al (2006) proposed a minimum extraction time of 40 minutes for clay soils, but no clay content is provided to make direct comparisons.

This research comprises part of a long-term study involving the integration of native perennial vegetation strips (NPVS) into rowcrop agricultural fields to assess their effectiveness in restoring the natural hydrological balance of agroecosystems (Zhou et al., 2010; Hernandez-Santana et al., In Press). The goals of the present study were to compare DWU by dominant plant species within an annual rowcrop system and prairie
vegetation in central Iowa, at two different topographical positions (upslope and footslope) and in three watersheds having different configurations of rowcrop and prairie vegetation, to propose a calibration method for water extraction techniques for clay rich soils, and to test the applicability of artificially created isotopic gradients in mixed agricultural ecosystems in DWU studies. Our specific objectives were to: (a) assess the average DWU of dominant annual crop and native prairie species within each watershed during the growing season using two methods: natural variability in stable isotope concentrations and a stable isotope tracer δD, and (b) assess the effects of landscape position and soil water content on depth of plant water uptake under each of these cover types.

We hypothesized that (1) during periods of adequate soil moisture availability (e.g., early in the growing season), corn and prairie species will obtain their water from relatively shallow depths in the soil profile. As soil moisture becomes more limiting (later in the growing season), prairie species (especially C_3 forbs) will shift their depth of water uptake to deeper depths in the soil profile, whereas corn and C_4 prairie species will have more limited capacity to obtain water from deeper depths, and (2) prairie and crop species will use water from deeper depths in the soil profile in the summit position compared to the footslope position due to lower water availability during dry periods in the upper parts of the watershed.
2 Study design and methods

2.1 Study area

This study was conducted at the Neal Smith National Wildlife Refuge (NSNWR; 41°33´N, 93°16´W) located in Prairie City, Jasper County, Iowa. The NSNWR, which comprises 3500 Ha administrated by the National Fish and Wildlife Service, and was created by an act of Congress in 1990 with the mission to reconstruct presettlement vegetation on the landscape, particularly native tallgrass prairie. To date, the NSNWR has converted approximate 2250 Ha of previous agricultural land into reconstructed native prairie vegetation, while areas that are still awaiting reconstruction are currently maintained under pasture or corn-soybean rotation.

The NSNWR includes part of the southern Iowa drift plain, characterized by the presence of steep rolling hills of Wisconsin-age loess on pre-Illinoian till (Prior, 1991). Walnut Creek is a third order stream that drains into the Des Moines River at the upper end of the Red Rock Reservoir. Most soils at the research sites are classified as Ladoga (Mollic Hapludalf) or Otley (Oxyaquic Argiudolls) soil series, which are highly erodible with slopes ranging from 5 to 14%. Texture of Ladoga soils is silt loam and silty clay loam for Otley soils, with clay contents from 15 to 42% and 20 to 42% respectively (NRCS, 2010). The mean average precipitation registered over the last 30 years (1981-2010) is 910 mm, with the majority of the large storms occurring between May and August (NCDC, 2011). Precipitation data for this study were recorded at the MesoWest (ID = NSWI4) weather station located in the Neal Smith Wildlife Refuge in Jasper County, Iowa, from March to November 2010.
2.2 Experimental design

Three experimental zero-order (intermittent in hydrological outflow) watersheds were used in this study, each subjected to a different treatment: Interim 1, with 10% prairie vegetation distributed as contour strips of native prairie vegetation (hereafter referred to as “SNPV”) within a crop matrix, Interim 3 with 100% row-crop (hereafter “CROP”) and Interim 4 with 100% reconstructed prairie vegetation (hereafter “PRAIRIE”; Figure III.1). SNPV were planted in July 2007 and PRAIRIE in 1994. The seed mixture used in the plantings consisted of 20 native prairie forbs and grasses with four primary species in the mix, including indiangrass (*Sorghastrum nutans* Nash), little bluestem (*Schizachyrium scoparium* Ness), big bluestem (*Andropogon gerardii* Vitman), and aster (*Aster* spp. L.). This mixture is similar to the one commonly used by the NSNWR staff in prairie reconstruction practices. Fire is also used as a management tool in the prairies under reconstruction at the NSNWR; the PRAIRIE watershed was burned in the spring of 2010.

In each study watershed, two 2 m² plots were marked in the summit and one in the footslope position, yielding a total of 9 plots (3 per watershed). A deuterated tracer (i.e. highly enriched δD values) was applied in two plots per watersheds, one in the summit and one in the footslope; the second summit plot was left as a control (i.e. no tracer was applied and naturally occurring isotopic concentrations were assessed; see details below).

In the CROP watershed, corn (*Zea mays*), an annual C₄ crop, was analyzed in all the plots. In the SNPV watershed, we selected 3 dominant species for assessment of
depth of water uptake: coneflower (*Ratibida pinnata*), a C$_3$ forb, brome grass (*Bromus ciliatus*), a C$_3$ grass, and wild rye (*Elymus canadensis*), a C$_3$ grass. In the PRAIRIE watershed, two species were selected: big bluestem (*Andropogon gerardii*), a C$_4$ grass, and coneflower (Table III.1). Coneflower was thus sampled in two different watersheds, SNPV and PRAIRIE. These species were selected based on dominancy at the watershed level and their presence in all three plots within each watershed. Other species (e.g. *Aster* spp. L.) were also dominant but not present in all plots in a given watershed.

2.3 Soil moisture monitoring

Fiberglass access tubes (Delta-T Devices) were used to monitor soil moisture using a Theta Probe (ML2, Delta-T Devices, Cambridge, UK) and a PR2 Probe (PR2, Delta-T Devices, Cambridge, UK). The Theta Probe was used to measure voltage outputs in the upper 5 cm of soil and the PR2 probe to take readings at soil depths of 10, 20, 30, 40, 60 and 100 cm. Data conversion and calibration details can be found in Chapter II of this thesis. Soil moisture readings were taken to coincide with the timing of the deuterated water tracer application and the collection the soil and plant sample collection.

2.4 Isotopic tracer application

A solution of 500 mL of D$_2$O [deuterium oxide 99.9%] diluted in 12 L of regular tap water of known $\delta^{18}$O ‰ (VSMOW) and $\delta$D ‰ (VSMOW) was applied on DOY 184, 2010 in 6 plots (two per watershed) using a backpack water pump. The deuterated tracer was applied prior to a forecasted rainfall event the next day (DOY 185) of 24 mm to
ensure rapid vertical movement of the tracer into the soil profile. Two L of labeled water were applied per plot, equivalent to 1 mm of precipitation. The application was conducted in the late afternoon and early evening in order to minimize fractionation of the stable isotope due to evaporation. The tracer was applied covering as evenly as possible the soil surface. Special care was taken to apply the δD tracer slowly and precisely within each plot to avoid immediate runoff as well as minimize contact with the vegetation, as this would reduce the amount of tracer applied to the soil.

Previous to the application of labeled water, one set of soil samples was collected at six intervals (0-10, 10-20, 20-30, 30-50, 50-70 and 70-100 cm) from each plot using a bucket auger from the soil surface at a depth of 100 cm. Each sample was placed in vials and stored in a freezer at -4ºC for future 18O and δD analysis to provide baseline ratios prior to the irrigation application of the deuterated water.

2.5 Collection of soil and plant samples

Soil and plant samples were collected in two periods during the 2010 growing season: July (DOY 203 and 206) and August (DOY 240, 241 and 242; hereafter July and August sample). Coneflower plants collected in the PRAIRIE watershed were not fully developed due to a prescribed burning treatment applied to this watershed in the spring. Big bluestem, bromegrass, coneflower and corn were collected on DOY 203 and wildrye was collected on DOY 206 in the July sample. Bromegrass and corn were sampled on DOY 240, coneflower and big bluestem on DOY 241 and wildrye on DOY 242 in the August sample. One set of six soil cores was collected adjacent to each plant sample using a bucket auger. Soil cores were collected at increments from 0-10, 10-20, 20-30,
30-50, 50-70 and 70-100 cm in SNPV and PRAIRIE, and from 0-10, 10-20, 20-30, 30-50 and 50-70 in CROP due to the shallower groundwater table (Figure III.3). A total of 282 soil and plant samples were collected during the July and August sampling periods. For plants, stems and leaf were sampled and analyzed, however only stems were used to assess DWU (see below).

For sampling of plant tissue, we collected non-photosynthetic stem tissue from the base of each of the study species, based on the principle of no fractionation upon water uptake (White et al., 1985; Roden and Ehleringer, 1999). For plants with small stems, tissue from several stems was pooled into one sample. In corn plants vertical segments of two stems were combined into one sample. Soil and plant samples were collected in vials and immediately placed in a cooler with ice to avoid evaporation and promote stomatal closure. Samples were transported to the Stable Isotope Lab at Iowa State University and kept frozen (-4°C) until analysis.

2.6 Rainfall and groundwater collection

Rainwater samples were collected from June to September, 2010 after a precipitation event in two watersheds, SNPV and CROP, using custom-design rainfall collectors consisting of a funnel connected through a house to a collector bottle, which was inside a wooden box to avoid fractionation due to evaporation. All the samples were collected within two hours after the precipitation events that occur during the daytime, and early in the morning for nighttime precipitation events. These data were used to estimate the percentage of meteoric water entering the watersheds. Groundwater was collected to complement and compare concentrations of δ^{18}O ‰ and δD ‰ in the soil.
One well was installed at the summit and footslope positions of the SNPV and CROP watersheds to a depth of approximately 6 m. The groundwater wells consisted of $\frac{3}{4}$ inch PVC piping capped at the bottom with a pointed tip. The wells were equipped with slits covering the bottom third of the piping to allow movement of groundwater into the well.

2.7 Water extraction and isotopic analysis

Water from plant and soil samples was extracted using a custom-design vacuum cryogenic distillation apparatus (Figure III.3). The water extraction apparatus consisted of five extraction arms attached via an 18/9 ball joint to a 2.54 cm o.d. vacuum line powered by a vacuum pump C Plus Maxima model M4C. A Millitorr Vacuum Gauge was attached to the vacuum line to measure pressure. A Chem-Vac high vacuum valve (CG-962-01) was used to isolate each extraction arm from the main vacuum line. Each extraction arm was attached to an extraction tube on one side and to a collection tube on the other. Extraction and collection tubes were 2.54 cm o.d. each, attached to their respective extraction arm with a stainless steel Ultra-Torr vacuum fitting (SS-16-UT-6). A 25 Watt incandescent light bulb was used as a heat source and a thin cardboard circle covered in aluminum foil was used to keep the light bulb in place and the heat insulated inside the “heat lamp dewar”.

Prior to extraction, plant and soil samples were removed from the freezer and allowed to thaw at room temperature. In the case of soils, the sample was removed from the vial, homogenized with a spatula and roots were removed to obtain the isotopic signature of the soil alone and discard the value of the water being transported in these roots from an unknown depth. The sample was then placed in the extraction tube and a
custom-made filter consisting of a plastic ring of about 1 cm in length covered with filter paper was placed inside the extraction tube to avoid soil particles from moving into the vacuum line, extraction arm or collection tube. Another filter was placed in the collection tube for better protection. Both the extraction and collection tubes were attached to the extraction arm via Ultra-Torr fittings. A Dewar with liquid nitrogen was placed under the extraction tube and the sample submerged in the liquid nitrogen until the sample was completely frozen (about 5 min). Once the sample was frozen, the isolation valve was opened and the extraction arm and the tubes pumped down to at least 50 mTorr. Once the desired vacuum was reached, the isolation valve was closed and the Dewar with liquid nitrogen was replaced with the heat lamp Dewar. The Dewar containing liquid nitrogen was refilled and placed under the collection tube. The extraction time was 60 min for soil samples and 30 to 60 min for plant tissue (see 2.8 for further details). Once the extraction time was completed, collection and extraction tubes were removed from the extraction arm and the collection tube was sealed with Parafilm and allowed to thaw at room temperature. The extracted water was then transferred to a 10 mL vial and stored in a cooler at 4°C for isotopic analysis.

All plant and soil water extraction samples were measured for $\delta^D$ and $\delta^{18}O$ on a Picarro L1102-i Isotopic Liquid Water Analyzer attached to an autosampler and using ChemCorrect software, at the Department of Geological and Atmospheric Sciences at Iowa State University. Each sample was measured a total of six times, and to account for memory effects (Barbour, 2007), only the last four injections were used to calculate mean isotopic values. Reference standards (OH-1, OH-2, OH-3, OH-4) were used for isotopic
corrections, and to assign the data to the appropriate isotopic scale. At least one reference standard was used for every five samples. The combined uncertainty (analytical uncertainty and average correction factor) for $\delta^{18}O$ was $\pm 0.09\%e$ (VSMOW) and $\delta D$ was $\pm 0.45\%e$ (VSMOW).

As indicated by West et al, infrared spectroscopy is highly influenced by the presence of organic compounds in water samples (West et al., 2010), to overcome this problem, all the $\delta^{18}O$ samples flagged as contaminated (with presence of organic compounds) by the ChemCorrect software, and 22 (8%) of the non-flagged samples (for precision comparison purposes) were measured on a Finnigan MAT Delta Plus XL mass spectrometer in continuous flow mode connected to a Gas Bench with a CombiPAL autosampler at Iowa State University (Department of Geological and Atmospheric Sciences) using reference standards [OH1, OH2, OH3, ISU Tap (lab internal std)] for isotopic corrections, and to assign the data to the appropriate isotopic scale. At least one reference standard was used for every eight samples. The combined uncertainty (analytical uncertainty and average correction factor) for $\delta^{18}O$ was $\pm 0.16\%e$ (VSMOW). Further, only corrected and calibrated $\delta^{18}O$ (aided by $\delta D$ when necessary) values were used in the results section.

2.8 Precision and reliability of the water extraction apparatus

To assess the precision of the water extraction apparatus, a series of soil samples were collected in all study watersheds at different topographic positions and depths. All soil samples were mixed together and homogenized. Plant roots and other plant materials
and rocks were removed and the remaining soil was dried in the oven drier for 24 h at 104°C. The dried soil was then dampened with tap water of known isotopic composition (internal lab standard) to a ratio of 400 mL of water per every Kg of soil. The soil was homogenized one more time to assure even moisture and subsamples were run through the extraction process.

To estimate the optimum time for extraction, samples were run from 10 to 109 minutes (See Appendix E). The $\delta^{18}$O ‰ and $\delta$D ‰ values of the water extracted from these soils were compared with the known values of the tap water that was applied to the soils. The differences in isotopic concentrations were estimated and the mean of the differences was regarded as the average extraction systematic error, or extraction error. We observe differences of less than 0.5 ‰ (observed – standard) starting at 39 min, similar to the results observed by West et al, (2006), however consistent differences were observed after 50 min. Average error estimated for extraction times greater than 30 min was $\pm 0.51$ ‰ and $\pm 7.69$ ‰ for $\delta^{18}$O and $\delta$D, with a standard error of 0.06 ‰ and 0.22 ‰, respectively. The extraction error for extraction times greater than 59 min was $\pm 0.44$ ‰ and $\pm 7.42$ ‰ for $\delta^{18}$O and $\delta$D, with a standard error of 0.07 ‰ and 0.29 ‰, respectively. Since the extraction time for our samples varied from 30 to 60 minutes, we used the mean extraction systematic error for extraction times greater than 30 min to adjust our isotopic data prior to assessment of DWU. The average extraction error for extraction times greater than 30 min was chosen, to include all our extraction times, as previously mentioned the extraction times ranged from 30 to 60 min including plant and soil samples. Adjusted isotopic values of $\delta^{18}$O and $\delta$D of soil water from non-flagged samples
(plot 1, 4, and 7) were plotted to verify the linearity of the fractionation form our extraction apparatus (See Appendix F for further details).

2.9 Estimation of depth of water uptake

Once the $\delta^{18}O$ values were adjusted with the extraction error, depth of water uptake was estimated using the direct inference method (White et al., 1985; Brunel et al., 1995), in which the isotopic values of the water extracted from the soils at different depths are compared with the isotopic values of the plant. Similarly, in this study we compared the $\delta^{18}O$ values of water extracted from soils at different depth intervals (Figure III.2) with the $\delta^{18}O$ values of the water extracted from a plant (only stems were used). Each of the 36 plant samples was compared to its own set of soil samples. The isotopic value of one of the 6 corresponding soil samples with the highest similarity to the value of the plant samples was regarded as the probable DWU.

$\delta^{18}O$ values were used as the main tracer of DWU, since no $\delta D$ values were determined in the mass spectrometer for samples flagged by the ChemCorrect Software. However, in cases where the $\delta^{18}O$ value of the plant matched the $\delta^{18}O$ value of more than one depth (e.g. 0-10 and 50-70), the artificial gradient created with the deuterated tracer, which had a higher positive signature, was used to eliminate ambiguous DWU suggested by $\delta^{18}O$. The analytical error of the measuring instruments was accounted for at the time of assessing probable DWU. From the 36 soil sets used to determine DWU, 18 of them had clear gradients (i.e. clear gradient with no overlapping values), 9 had similar or
overlapping isotopic values in only two intervals, and 9 had overlapping values in 3 or more depths.

2.10 Statistical analysis

Significant differences between mean DWU per species observed for the two sampling periods (July, August; Table III.2) were determined using the GLM procedure in the statistical program SAS. Vegetative type (C3 and C4) and the five different species (big bluestem, bromegrass, coneflower, corn and wild rye), were the independent variables, with DWU as the dependent variable. We analyzed the effects of plot, watershed, sampling time, volumetric water content in the topsoil and topographic position on the variation observed in DWU.

3. Results

3.1 Precipitation

The total precipitation registered during the study period was 1326 mm (Figure III.4), 45\% greater than the average precipitation of 910 mm recorded from 1981 to 2010 (NCDC, 2011), with precipitation events greater than 10 mm observed from early-April to mid-November.

Analysis of the isotopic signature of precipitation samples revealed variations in $\delta^{18}O$ from -12.6 to -1.1 ‰, and in $\delta D$ from -85.3 to -3.3 ‰ (Figures III.5 and III.6). The variations in the isotopic concentrations appeared to be a response of the frequency and
intensity of the precipitation. $\delta^{18}O$ and $\delta D$ values became more positive as the frequency and the intensity of the precipitation increased, and more negative as they decreased. Additionally, the graph showing $\delta^{18}O$ and $\delta D$ precipitation values versus the global meteoric water line (GMWL) (Craig, 1961), and the local meteoric water line (MWL) (Simpkins, 1995), demonstrates that the collection protocol did not influence the isotopic composition of precipitation (i.e. evaporation effects) (Figure III.7).

3.2 Groundwater

Unlike the isotopic values of rainfall water, groundwater showed little variation during the monitored period (Figure III.7). $\delta^{18}O$ showed an average of -7.30 ‰ and $\delta D$ - 45.60 ‰ with standard deviations of 0.31 ‰ and 2.32 ‰, respectively. Plotting $\delta D$ versus $\delta^{18}O$ values, we observed linear fractionation of both isotopes ($R^2=0.94$) with a light enrichment of $\delta D$. The $\delta^{18}O$ and $\delta D$ values were plotted against the global (Craig, 1961) and the local meteoric water line (Simpkins, 1995) to denote the $\delta D$ enrichment, which remained constant in three of the four sites where samples were collected (Figure III.8, III.9). Differences in isotopic composition were detected by topographic position. Plotted by watershed and observation date, $\delta D$ values of groundwater samples showed enriched values in summit position of the SNPV (Interim 1) watershed, and lower $\delta D$ values in the Foot position of the SNPV (Interim I) and CROP (Interim 3). Only the Summit position of the CROP (Interim 3) watershed showed significant changes in isotopic concentration with time (Figure III.9).
3.3 Soil moisture content

$\theta_v$ declined in the upper layers of the soil profile during the study period (Figure III.10). Deeper depths remained fairly constant and were not as dynamic as shallower depths. Mean $\theta_v$ in the upper 5 to 10 cm of soil at the time the tracer was applied was 0.36 and 0.38 respectively. Lower parts of the watershed showed higher $\theta_v$, compared with the upper parts, particularly in the SNPV and PRAIRIE watersheds (Figure III.11). In contrast, the CROP watershed maintained relatively higher $\theta_v$ compared to the other two watersheds, particularly in the upper layers of the summit position (Figure III.11).

3.4 Isotopic signature of the soil water by depth

Fifty percent of the samples analyzed showed clear natural isotopic gradients, and 25% of them had similar or overlapping isotopic values in only two depths, which facilitated the estimation of depth of water uptake. $\delta^{18}O$ values ranged from -1.48 to -9.06 ‰ with a mean average difference (gradient) of the 36 soil sets of 3.24 ‰ between the uppermost and deepest soil depths. The gradients observed per watershed were: 3.25 ‰, 2.39 ‰ and 3.66 ‰ in the SNPV, CROP and PRAIRIE, respectively. Despite the expectation of noisy gradients due to the precipitation registered in 2010 (45% above the 30 year average), only 9% of the soil sets showed undefined gradients. Analysis of $\delta^{18}O$ showed well-defined gradients in most of the plots in the SNPV and PRAIRIE watersheds (Figure III.12). The majority of the observations with noisy gradients were collected from the plots 4, 5 and 6, which were located in the CROP watershed.
The δD analysis showed that the application of the deuterated tracer significantly altered the isotopic gradient in the soil. Values of δD ranged from -63.02 to 1552.89‰, with the highest δD values observed during the August sample 20 days following application of the tracer. The highest δD value observed in the July sample was 282.18‰. The artificial gradient created with the deuterated tracer varied by plot and watershed (Fig. 13), with the SNPV and PRAIRIE watersheds having the most pronounced isotopic gradients. The strength of the gradients also decreased significantly by the second observation period in all watersheds (Figure III.14).

3.5 Plant water uptake

The analysis of depth of DWU indicates that on average the upper 70 cm of soil were the main source of water for all species. Despite the overall high water content observed during our study in the three watersheds (See 3.1), all species shifted their depth of water uptake between the two sampling periods. In most cases plants shifted to deeper soil water sources in August, as compared to their DWU in July (Table III.2). The statistical analysis of DWU indicated that collection date (July, August) and variations in θ, among observation dates had a significant (p=0.0401 and p=0.0092, respectively) influence on DWU for all the species (Table III.3). Further, our results suggest that big bluestem obtained water from 10-50 cm in July and shifted to a depth of 20-50 cm in August (Fig. 12). Bromegrass showed a DWU of 0-20 cm in July and 10-50 cm in August. Coneflower, which was sampled in two different watersheds (SNPV and PRAIRIE), showed different patterns of DWU in each watershed. In SNPV, coneflower shifted from
10-70 cm in July to a shallower depth, 0-30 cm, in August. In PRAIRIE, the inverse pattern was observed, as coneflower shifted from a shallower depth (0-10 cm) in July to a deeper depth, 20-70 cm, in August (see Discussion section for further details). Corn acquired water from 10-50 cm in July and from 20-70 cm in August. Wild rye shifted from 0-30 cm in July to a shallower depth, 0-20 cm, in August.

3.6 Effects of topographic position and soil water content on DWU

Our statistical analysis showed no significant relationship between topographic position on DWU for all species (Table III.3). Changes in $\theta_v$ had a greater influence than topographic position for all species. In the toe position during the July sampling period, big bluestem, bromegrass, coneflower and wild rye showed shallower DWU for at least during one observation date, while corn exhibited the deepest DWU of 30 cm (Table III.2). In the summit position, the deepest DWU observed across all species in July corresponded to coneflower from the PRAIRIE watershed (50-70 cm). In August, most species shifted to deeper depths, independent of topographic position (Table III.2).

The watersheds SNPV and PRAIRIE showed lower $\theta_v$ in the upper parts of the watershed (0.32 and 0.33), compared to their $\theta_v$ in the lower parts (0.36 and 0.38) particularly in the shallower depths (5-10 cm). However, only PRAIRIE showed a consistently lower $\theta_v$ in the summit position at most depths, compared with the Toe position (Figure III.8). The CROP watershed showed a higher $\theta_v$ (0.49) in the summit compared to the toe position (0.36). The differences in $\theta_v$ appeared to influence patterns of DWU, affecting primarily C$_3$ species according to our statistical analysis.
3.7 Water uptake by functional group: C₃ and C₄ species

Our data showed no difference between functional groups in the DWU patterns observed (Table III.3). The statistical analysis by functional groups however, showed that C₃ plants were more influenced by changes in θᵥ than C₄ plants (Table III.3). The C₃ forb, coneflower, from watershed PRAIRIE exhibited the deepest DWU across all study species (30-70 cm in August). The same species from watershed SNPV showed a range of DWU from 0-30 cm in August (Figure III.15). The C₃ grass, wild rye, showed the shallowest range in DWU, 0-30 cm in July to 0-20 cm in August. Both C₄ species (big bluestem and corn) showed similar patterns of DWU in July and shifted to a similar depth in August but their ranges were not different from the ones observed for C₃ species.

An inverse pattern in DWU was observed for both the C₃ forbs coneflower and wild rye collected from the SNPV watershed (deeper DWU in July than August). When these two species were collected in July, their DWU was deeper compared to in August, 10-70 cm in July to 0-30 cm in August for coneflower and 0-30 cm in July to 0-20 cm in August for wild rye.

4. Discussion

4.1 Using δ¹⁸O and δD as indicators of depth of water uptake

We used δ¹⁸O and δD stable isotopes to determine the effects of variations in θᵥ, topographic position and season on patterns of depth of plant water uptake in four
dominant prairie species (big bluestem, bromegrass, coneflower, wildrye) and one crop (corn) in three small watersheds. Several studies have used this approach to assess DWU in a variety of land covers such as rice (Araki and Iijima, 2005), grassland (Nippert and Knapp, 2007), crops (Wang *et al.*, 2010), shrubs (Nippert *et al.*, 2010), woody plants (Midwood *et al.*, 1998) and mixed agricultural-perennial ecosystems (Asbjornsen *et al.*, 2007; Asbjornsen *et al.*, 2008). In restoration efforts, water dynamics and particularly water use patterns are of great importance for the entire ecosystem of interest. There is a need of research techniques to study these dynamics, such as the use of stable isotopes as an important tool for researchers and land managers. Most of these techniques rely on natural occurring isotopic gradients in the soil to assess. In saturated soils or where the preferential infiltration is not vertical, the isotopic concentration does not always present the necessary gradient for the assessment of DWU.

In order to overcome the difficulties posed by the usually persistent wet soils present at our study site and hence the expected “noisy” isotopic gradients due to excessive rainfall, we applied a deuterated tracer to help determine DWU. Several studies of DWU have been conducted using δD tracers in different ecosystems and land covers (Yoder *et al.*, 1998; Plamboeck *et al.*, 1999; Moreira *et al.*, 2000; Rowland *et al.*, 2008), which facilitates the assessment of DWU. In this study the use of the deuterated tracer helped significantly the assessment of DWU in the plots where neither δ18O nor δD natural gradients were clear, by eliminating ambiguous DWU suggested by these isotopes, and introducing a more positive δD value. Using soils from our research sites, the water extraction apparatus was calibrated and minimum times of extraction to get
consistent differences was estimated. The average extraction systematic error was accounted for when DWU was estimated.

4.2 Isotopic signature of rainfall water

Fractionation of rainfall water is a well-known process that is highly influenced by different parameters such as altitude, latitude and distance from the coast, the fraction precipitated from a vapor mass (Clark and Fritz, 1997; Mook, 2006), and also due to the effects of a Rayleigh-type distillation, in which condensation *distills* the heavy isotopes ($\delta^{18}O$ and $\delta D$) from an air mass, depleting the air mass as rainout occurs (Clark and Fritz, 1997). This variation and changes in isotopic concentration in precipitation water was observed in our data (Figure III.5 and III.6), where consecutive rainfall events showed slightly similar isotopic values, compared to isolated events. Previous studies have found similar results in Central Iowa (Simpkins, 1995), Kansas (Nippert and Knapp, 2007), Hebei, China (Li *et al.*, 2007), Shanxi, China (Wang *et al.*, 2010). The global meteoric water line (GMWL) developed in 1961, by Harmon Craig, which describes the linear fractionation of $\delta^{18}O$ and $\delta D$ in meteoric waters, and the regional meteoric water line developed by Simpkins in 1995 (Craig, 1961; Simpkins, 1995), closely follow the linear regression of our precipitation data ($R^2=0.9635$), showing no statistical significant differences ($P=\text{<0.001}$) for both Craig and Simpkins lines.
4.3 Isotopic signature in soils and groundwater

Approximately 50% of the soil profiles examined in our plots had clear gradients, 25% had similar values (or within the estimation error) at two depths, and 25% had similar values at more than two depths. The majority of the soil profiles that exhibited noisy gradients were located in the CROP watershed (Figure III.12), which has naturally occurring lateral seepage in the upper parts (summit)\(^1\). This seepage is present beginning early spring and depending on the rainfall conditions can remain active until late fall, creating conditions of increased \(\theta_v\) content in the upper parts of the watershed. Of the three study watersheds, it is the only one that presents higher volumetric water content in the upper parts and also the one with the least variation in \(\theta_v\) observed during our study (Figure III.10).

Several authors have studied the influence of soil moisture dynamics on the development of isotopic gradients in the soil (Barnes and Allison, 1988; Gat, 1998; Leibundgut et al., 2009). This isotopic gradient, represents a balance between the upward convective flux and the downward diffusion of the evaporative signature (Barnes and Allison, 1988), and it is caused by hydrodynamic dispersion within the soil (Leibundgut et al., 2009). Then, the amount of water that moves into the soil directly affects the development of the isotopic gradient in the soil (Dalton, 1989; Gat, 1998; Leibundgut et al., 2009). This gradient was observed for most of our samples, particularly in the SNPV and PRAIRIE watersheds; however, it is possible that the gradient observed in plots 4, 5

\(^1\) See Chapter II of this thesis for further details.
and 6 in the CROP watershed in July, was a result of an uncontrolled movement of surface water into the soil profile in this watershed. Another possible explanation is that water originated from the lateral seepage modified or alter the attenuation of the isotopic signature of water into the soil, as discussed by Leibundgut et al, (2009). The attenuation of the isotopic gradient in the soil profile they mention was observed in these plots (4, 5, 6). The depleted values in the light isotopes observed in July at the intervals 10-20, 20-30 and 30-50 cm that created these irregular gradients, appeared to get enriched in the heavy isotopes ($\delta^{18}O$ and $\delta D$) in August and thus creating a clearer gradient (Figure III.12).

Despite the care that was taken to apply equal amounts of tracer in each plot, the concentration observed varied by plot and by watershed. Judging by the amount of tracer retained in every plot, we observed clear differences by watershed, with PRAIRIE retaining the highest and CROP retaining the least amount of tracer (Figure III.13). The movement of the tracer into the soil profile depends largely on the infiltration properties of the soil (Leibundgut et al., 2009). The uppermost layer of the soil plays a critical role in the infiltration process, vegetative cover, organic matter deposited on the ground and biomass aboveground control the amount of precipitation that it is intercepted and the amount that reaches the ground (Brooks et al., 2003; Chang, 2006). Plant cover also plays a critical role in the reduction of surface runoff, affecting with this the isotopic composition of recharge flux (Gat, 1998). In this study, values of the deuterated tracer observed in the first sampling period (Figure III.13, July period) may be the result of the different land cover properties among these sites. The thick soil cover in the PRAIRIE watershed, second only by the soil cover in the SNPV watershed, had greater potential to
retain higher amounts of tracer closer to the mineral soil surface than the bare ground in the CROP watershed, and thus promoting a higher infiltration rate of the tracer into the soil in the SNPV and PRAIRIE watersheds, than the amount of tracer that entered the soil surface in the CROP watershed, as observed in Figure III.13.

Without taking into account the summit position in the CROP watershed, the values for groundwater observed in this study remained constant during the observation period in each of the remaining sampling sites (Figure III.9). These results match the underlying principle of the combination of water having different isotopic values, in which if a small amount of water (i.e. infiltration) having a different isotopic value is combined with a large body of water (i.e. groundwater), its effects on the isotopic value of the receiving water body will be minimal. Similarly, as indicated by Leibundgut et al, the temporal variability of stable isotopes in groundwater is influenced by the variations in the isotopic values of meteoric waters. However, due to the attenuation of the isotopic values with depth, the effects observed on groundwater are minimal, and other approaches based on mass balance can be more appropriate to study these variations (Leibundgut et al., 2009). In the position summit in the CROP watershed, it is possible the a higher infiltration, or a high interaction of groundwater with surficial water (due to shallow groundwater) caused water enriched in the light isotopes to change the isotopic composition of groundwater. As shown in Figure III.9, the isotopic composition of groundwater in this particular position gradually changed from δD -47.4 ‰ June to δD -41.6 ‰ August.
4.4 Variations in plant water uptake

Characteristics specific to annual crops and native prairie vegetation, such as root depth, water use patterns, phenology and adaptation to variations in soil moisture, gives each of these land covers the ability to access water from different soil profiles when they are subjected to conditions of limited water availability. In this study we observed differences in DWU in bluestem, bromegrass, coneflower (collected in SNPV) and corn. In the July sampling period when the average $\theta_v$ for all the sites in the upper 20 cm was 0.39, these plants used proportionally more water from shallower sources (15 cm on average) and shifted to a deeper depth (33 cm on average) in August when the mean $\theta_v$ was 0.35. Not including the CROP watershed, which had a great influence on the $\theta_v$ average of all the sites due to the presence of lateral seepage, the average $\theta_v$ was 0.38 in July and decreased to 0.33 in August. The statistical analysis indicates that $\theta_v$ had a significant ($p=0.0092$) influence on the DWU observed in both July and August when the analysis was run for all species (Table III.3).

Changes in DWU have been observed in other studies conducted on C$_3$ shrubs and C$_4$ crops and grasses (Asbjornsen et al., 2007; Nippert and Knapp, 2007; Asbjornsen et al., 2008). However our results differ in the range of PWU observed by previous research. For example, Asbjornsen et al (2007; 2008), observed that corn and big bluestem obtained water from 0 to 30 cm of soil during the growing season of 2008, results that were similar to a previous study where in 2007 similar results were observed for the range of water uptake for corn and bigbluestem (from 5 to 20 cm) In this study, the range of DWU observed for corn (including July and August) was 10-70 cm, and for
big bluestem it was 10-50 cm. Given the lower (515 mm) precipitation registered during 2007, a larger range of water uptake could be expected, however it is possible that the relative slope of our sites, which ranged from 6.6 to 10.3%, caused corn plant to take water from deeper soil profiles in the upper parts of the watershed where water was more limiting in the upper layers, and thus creating a larger range in DWU (See 4.3). Although Asbjornsen *et al.* (2007; 2008) provided information about the precipitation registered during each of the studies, it is not possible to assess to what extent differences in the range of DWU were due to dryness of the topsoil. The $\theta_v$ in the CORN watershed, primarily in the summit position, remained fairly stable (*approx.* 0.40) due previously described high soil moisture content, nonetheless our findings indicate that there was a change in DWU between sampling dates among species. In the watershed PRAIRIE, where big bluestem was sampled, $\theta_v$ in the upper 20 cm changed from 0.38 in July to 0.32 in August. The range of DWU we found for corn is similar to a study conducted in summer corn by Wang *et al.*, (2010), which reported that summer corn extracted water from 20-50 cm in the flowering state. According to their data, soil water potential decreased to almost -50 KPa during the flowering state, indicating a $\theta_v$ of approximately to 15%, which could have explained the wider range of DWU observed.

Plant phenology among the studied species also seems to influence the patterns of DWU. Weaver found that water use requirements are highly influenced by the functioning vegetation demanding water (Weaver, 1941). As mentioned earlier, the NSNWR conducts prescribed burning on the restored areas like the interim 4 site, which was burned in the spring of 2010. Our field records indicate that at the time of collection
in July sampling period, coneflower and wildrye were in the flowering state, by the second sampling period in August, coneflower had initiated leaf senescence. Growing at natural field conditions, C₃ species tend to grow earlier in the season when temperatures are more favorable for photosynthesis (Pearcy and Ehleringer, 1984) and due to a temporal displacement of C₃ and C₄ as a function of the differential temperature responses to photosynthesis (Kemp and Williams, 1980). Coneflower in the PRAIRIE watershed was in an early vegetative stage (due to prescribed burning) and by the time the second sample was taken in late August, coneflower was already in flowering state.

The study of summer corn and cotton conducted by Wang et al., (2010) found that DWU was highly linked to the vegetative state of the plant, rather than the time of the year. A study conducted by Mateos-Remigio et al., (In Review) of sap flow in a C₄ grass (Andropogon gerardii), a C₃ shrub (Ratibida pinnata) and a C₄ crop (Zea mays), found that water use by plants was influenced by water requirements caused by phenological and physiological differences among functional groups. Given that the phenology of the plant is related to its development, this is indicative that plants shift their depth of plant water uptake in response to a variety of factors such water availability in the topsoil, precipitation, vegetative type and also metabolic needs during different development stages.

The relative topographic position of a plant within a watershed also influences its biomass production. Lower parts of the watershed tend to have higher soil moisture contents, relative to the upslope (Brooks et al., 2003; Hillel, 2004), and since water and soil moisture regulate plant productivity (Gholz et al., 1990), higher amounts of biomass
may expected in the lower parts of the watershed (provided that soil moisture does not exceed plant limits), compared to the upslope, and thus higher water use due to the higher amounts of vegetation demanding water (Weaver, 1941). It would also be expected to see higher fluctuations in the DWU in plants located in upslope positions as compared to plants in the downslope, however our statistical analysis showed no effect of topographic position in changes of DWU in the analysis for all the plants (p=0.5549) and in the analysis by vegetative type [C₃ (p=0.8501) and C₄ (p=0.5549)]. These results could have been a response of the high precipitation and overall high $\theta_v$ observed during this study, but this explanation contradicts our findings about the influence on $\theta_v$ (See Table III.3). Another possible explanation can be the influence of the inverse patterns of DWU observed for coneflower and wildrye (both from the SNPV watershed) on the statistical analysis.

Analyzed by photosynthetic pathway functional groups, soil moisture in the top 20 cm of soil had a higher influence on C₃ (p=0.0136) than C₄ (p=0.4051) plants. C₄ plants are known for a lower leaf conductance and therefore transpiration than C₃ plants due to a lower operational intercellular CO₂ (Pearcy and Ehleringer, 1984). This provides C₄ plants with higher water-use efficiencies (Pearcy and Ehleringer, 1984) and thus lower water requirements, compared with C₃ plants. Our results may then reflect the adaptation of C₄ plants to hot and dry environments, and the lower water-use efficiency of C₃ plants. Since C₄ plants are more adapted to lower water contents, the need to access water from a deeper soil profiles may not be as crucial as for C₃ plants with lower water-efficiency.
Other studies have found that on average C₃ plant species take water from deeper soil profiles than C₄ species when water becomes more limiting than C₄ species (Nippert and Knapp, 2007; Asbjornsen et al., 2008), supporting the water-use efficiency factor in DWU. However, taking into consideration the change in DWU observed for the C₃-forb coneflower collected in the PRAIRIE watershed, which was in a much younger state when first sampled, it is also possible that inverse DWU observed in coneflower samples collected in SNPV were a response of the phenology of the specie, which were at the flowering stage at the time of the first collection (July), compared with the “sprout” state of the samples of coneflower collected in PRAIRIE in July. This suggests that although photosynthetic pathways are essential in water-use patterns (and thus DWU), other factors such life form, phenology, changes in soil moisture in the top soil or the relative position of a plant within a topographic gradient, are also important factors to consider in the analysis of DWU.

5. Conclusions

The study of plant water uptake and its role within the hydrologic cycle requires the use of research approaches that assess the interactions between multiple important and relevant factors that influence water fluxes at a specific site. Understanding of the effects of species composition and diversity on soil moisture dynamics in restoration efforts is of major importance to assess hydrological impacts at the long term. We used natural gradients of δ¹⁸O and δD and artificially created gradients of δD to determine depth of plant water uptake. The δD tracer significantly helped to in the assessment of
DWU when natural $\delta^{18}O$ gradient was not observed. A calibration process was developed for our specific soil type and our water extraction apparatus was modified to reduce extraction time. The estimated extraction error for extraction times greater than 30 min was $\pm 0.51$ ‰ and $\pm 7.69$ ‰ for $\delta^{18}O$ and $\delta D$, with a standard error of $0.06$ ‰ and $0.22$ ‰, respectively, which were taken into account when assessing DWU.

Our results support that C$_3$ and C$_4$ plant communities shift their depth of plant water uptake when soil water becomes more limiting. We found deeper ranges of water uptake in cone flower and corn than previously observed, that could have been influenced by the topographic positions and the relative volumetric water content differences documented among them. The development stage of the plants appeared to influence shifts in DWU, plants that were sampled in their early development states in July and were fully developed in August shifted to deeper soil depths in the second sampling period. Plants sampled in their full development state in July and had started to senesce in August shifted to shallower depths. This provides supporting evidence for the water use dynamics of native prairie ecosystems and their role in the control of the water balance of the ecosystem, as well as the importance of C$_3$ and C$_4$ plant diversity in prairie restoration efforts.
6 References


Kemp, P.R., Williams, G.J., 1980. A Physiological-Basis for Niche Separation between Agropyron-Smithii (C-3) and Bouteloua-Gracilis (C-4). Ecology 61, 846-858.


7 Figures

Figure III-1. Design of the three experimental watersheds used in this study, each black rectangle represents a plot.
Figure III-2. Scheme of the soil and plant samples collected
Figure not at real scale
Figure III-3. Scheme of the vacuum cryogenic distillation apparatus used in this study
Figure III-4. Precipitation registered from May to October 2010

Figure III-5. $\delta^{18}$O values of the precipitation registered during this study
Figure III-6. $\delta$D values of the precipitation registered during this study

Figure III-7. Precipitation water plotted against the Global Meteoric Water Line (Craig, 1961), and the Local Meteoric Water Line (Simpkins, 1995)
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Figure III-12. $\delta^{18}O$ values of the soil samples averaged by plot. Horizontal gray lines represent the SE of the mean.
Figure III-13. δD values of the soil samples averaged by plot. Only plots where the tracer was applied are shown.
Figure III-14. δD tracer differences among observation periods. Averaged by watershed
Figure III-15. Depth of plant water uptake by species within observation periods. Black circles represent the mean of the water uptake range and the gray vertical lines the standard deviation.
## Table III-1 Description of the 3 watersheds and species sampled per watershed

<table>
<thead>
<tr>
<th>Watershed</th>
<th>Total prairie cover (%)</th>
<th>Topographic position of prairie cover</th>
<th>Species sampled</th>
<th>Vegetative type</th>
<th>Watershed area (ha)</th>
<th>Sand (%)</th>
<th>Silt (%)</th>
<th>Clay (%)</th>
</tr>
</thead>
</table>
| Interim 1 [SNPV] | 10%                     | Summit Side Footslope                 | Cone flower *Ratibida pinnata*  
Brome grass *Bromus ciliatus*  
Wild rye *Elymus canadensis*      | $C_3$ forb             | 3.00                 | $\uparrow$3 | 70       | 27       |
|             |                         |                                       |                                                                                 | $C_3$ grass    | $\downarrow$12    | 65       | 23       |
| Interim 3 [CROP] | 100% rowcrop            | None                                  | Com *Zea mays*                                                                 | $C_4$ crop     | 0.73                | $\uparrow$4 | 70       | 26       |
|             |                         |                                       |                                                                                 | $C_3$ grass    | $\downarrow$12    | 63       | 25       |
| Interim 4 [PRAIRIE] | 100% Rec. prairie       | Summit Side Footslope                 | Big bluestem *Andropogon gerardii*  
Cone flower *Ratibida pinnata*       | $C_4$ grass             | 0.60                 | $\uparrow$4 | 70       | 26       |
|             |                         |                                       |                                                                                 | $C_3$ forb     | $\downarrow$12    | 63       | 25       |

$\uparrow$ = Summit  
$\downarrow$ = Foot  
Percentages of sand, silt and clay correspond to the upper 30 cm of soil
Table III-2 Depth of water uptake by topographic position in July and August

<table>
<thead>
<tr>
<th>Depth</th>
<th>Big bluestem ($C_4$)</th>
<th>Bromegrass ($C_3$)</th>
<th>Coneflower ($C_3$)</th>
<th>Corn ($C_4$)</th>
<th>Wildrye ($C_3$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-10</td>
<td></td>
<td></td>
<td>Foot</td>
<td>Sum</td>
<td>Foot*</td>
</tr>
<tr>
<td>10-20</td>
<td>Foot</td>
<td>Sum</td>
<td>Sum</td>
<td>Sum</td>
<td>Foot</td>
</tr>
<tr>
<td>20-30</td>
<td>Foot</td>
<td></td>
<td>Sum</td>
<td></td>
<td>Sum*</td>
</tr>
<tr>
<td>30-50</td>
<td>Sum</td>
<td>Sum</td>
<td></td>
<td></td>
<td>Foot</td>
</tr>
<tr>
<td>50-70</td>
<td></td>
<td></td>
<td>Sum</td>
<td></td>
<td>Foot*</td>
</tr>
<tr>
<td>70-100</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Summit=Sum
*Corresponds to coneflower samples collected in the Interim 4 site

The topographic position of a given species is placed on the estimated depth of water uptake. For any given date and plant two plant samples were collected in the summit and one in the foot, hence three topographic positions (2 summit and 1 foot) are used in each sampling period (July and August)
Table III-3 GLM procedure by species and vegetative type (C₃ and C₄)

<table>
<thead>
<tr>
<th></th>
<th>C₃</th>
<th>C₄</th>
<th>Average DPWU* of all species</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Topographic position</strong></td>
<td>F=0.04</td>
<td>F=0.91</td>
<td>F=0.36</td>
</tr>
<tr>
<td></td>
<td>p=0.8501</td>
<td>p=0.3633</td>
<td>p=0.5549</td>
</tr>
<tr>
<td><strong>Month</strong></td>
<td>F=3.23</td>
<td>F=3.37</td>
<td>F=4.55</td>
</tr>
<tr>
<td></td>
<td>p=0.1390</td>
<td>p=0.0963</td>
<td>p=0.0401</td>
</tr>
<tr>
<td><strong>Volumetric water content</strong></td>
<td>F=5.31</td>
<td>F=1.40</td>
<td>F=4.09</td>
</tr>
<tr>
<td></td>
<td>p=0.0136</td>
<td>p=0.4051</td>
<td>p=0.0092</td>
</tr>
<tr>
<td><strong>Specie</strong></td>
<td>F=1.29</td>
<td>F=0.37</td>
<td>F=1.58</td>
</tr>
<tr>
<td></td>
<td>p=0.2959</td>
<td>p=0.5580</td>
<td>p=0.2052</td>
</tr>
<tr>
<td><strong>Functional group</strong></td>
<td>NA</td>
<td>NA</td>
<td>F=3.08</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>p=0.882</td>
</tr>
</tbody>
</table>

*Corresponds to the mid-point of the observed range of depth of plant water uptake
CHAPTER IV

GENERAL CONCLUSIONS

We studied the effects of the establishment of native prairie vegetation on soil water dynamics within landscapes dominated by row-crop agriculture. This research is a component of the larger Science-based Trials of Rowcrops Integrated with Prairies (STRIPS) project that has the objective to quantify the influence of different proportions and landscape configurations of annual (e.g., corn and soybean) and perennial (e.g., prairie, savanna, agroforestry) plant communities on the storage, cycling, and output of nutrients, water, and carbon at the field and catchment scale. The research described herein specifically compared the depth of plant water uptake by annual and perennial vegetation and the resulting effect on soil water storage. Results were discussed in the context of regulation of larger-scale hydrologic balance, impacts on watershed management, and implications for sustainable agricultural practices. A calibration process was developed for vacuum cryogenic distillation extractions, which can be applicable to DWU studies in clay rich soils.
Major findings

Lower soil water storage was observed under native prairie vegetation than annual rowcrops within one year after establishment of prairie vegetation. This pattern contrasted with results in 2007 prior to prairie establishment when soil water storage was lower under rowcrop, and illustrates the potential critical role of perennial vegetation in regulating soil water balance. The pattern of lower soil water storage under prairie vegetation was less significant in 2010, when annual precipitation was 45% above the long-term annual average, which seems to indicate the presence of a threshold effect in the benefits the prairie ecosystem, as far as its benefits on soil water storage. Observed differences in soil water storage were more pronounced within the upper 30 cm of soil than upper first 60 cm.

Topographic position had a significant influence on soil water storage, with lower average storage in the summit slope positions compared to footslopes. Greater fluctuations in soil water storage were also observed in the upper slope positions. Precipitation, which was analyzed as the total precipitation during the seven days previous to the monitoring date, significantly influenced the variability in soil water storage, particularly in 2010 when it seemed to override other factors.

In our study of depth of water uptake we found that the first 70 cm were the main source of water for all species for both observation periods, with variations within this range among species and observation dates. The deuterated tracer artificially applied significantly helped to determine depth of water uptake in plots where the isotopic gradient in the soil was not clear, by eliminating ambiguous depths suggested by the
naturally occurring oxygen isotopes. It was also observed that available water in the topsoil, and observation date influenced all species. When analyzed by functional group, C₃ plants appeared to be more responsible to changes in soil water in the topsoil than C₄ plants.

Phenological and physiological differences among the plants also influenced the depth of water uptake observed. Plants sampled in their full development state in July that had started to senesce in August shifted to shallower depths. While plants that were sampled in their early development stages in July, shifted to deeper depths in August. This provides supporting evidence for the water use dynamics of native prairie ecosystems and their role in the control of the water balance of the ecosystem.

**Implications for watershed management**

This study highlights the importance of understanding the effects of vegetation on soil water dynamics. Such information is useful in predicting the impact at the catchment-scale of the re-incorporation of native prairie vegetation into agricultural landscapes. Our 4-year study indicates that the re-incorporation of native prairie vegetation can lower average annual soil water contents within one-year after the establishment of prairie vegetation. Soils with low water storage (i.e. larger storage capacity) are more likely allow precipitation to infiltrate, thereby reducing runoff and sediment and nutrient flux to receiving waters. In areas with tile drainage, the presence of strips of native prairie vegetation can help to regulate water yield.
Increasing evidence indicates that watersheds with strategically established native prairie or other perennial vegetation have lower surface runoff than those dominated with annual rowcrops [e.g. corn, soybean]. In places like Iowa, where the native prairie vegetation has been replaced with annual crops, watershed hydrology has been dramatically altered, resulting in increased loss of nutrients and sediments and an increased risk of flooding. Understanding the need for agricultural goods, and the importance of sustainable agricultural practices, the use of small amounts of prairie vegetation to regulate the water balance of an ecosystem appears to be a viable and sustainable alternative, that requires only to assign a small percentage of cropland surface to native prairie vegetation, which can in turn return multiple benefits to land owners and managers.

**Challenges faced in this research**

In this study, volumetric water content was measured using a standing wave measurement to determine the impedance of a sensing rod array using a combination of a Theta and PR2 Probes (Delta-T Devices, Cambridge, UK). The acquisition of accurate data from such sensors requires precise calibration. Since we decided to calibrate every sensor of the PR2 and one of the Theta probes used in this study, the number of gravimetric samples required for the calibration increased exponentially. In order to achieve a reliable calibration, it is important to collect gravimetric samples over a range of water contents, especially at lower water contents. During our study there were few
periods of time when the soil water content was low enough to collect calibration samples for this range. The few opportunities to collect gravimetric samples at lower water contents, combined with the large number of access tubes and depths to calibrate, made the calibration process a challenging task.

For our study of depth of plant water uptake we decided to collect a set of soil samples for every plant sampled. Including stems, leaves and the soil samples, we extracted water from 282 samples (including stems, leaves and soils), and although we did not use the leaf samples in our interpretation, the time required to process and to extract water from all the samples made this process a time consuming task. We strongly recommend future studies of depth of plant water uptake, to determine isotopic variability in the soil, prior to decide the number of samples needed per plot, and take a decision about the number of samples needed to get a representative and accurate sample.

**Recommendations for future research**

**Study of the effects of management practices of the prairie strips**

It is important to get a better understanding of the effects of the removal of the aboveground biomass in the prairie strips on soil water dynamics. The effects of mowing and burning are not well understood under these configurations of mixed annual-perennial vegetation. This study would provide important information relevant for the future management of the prairie strips. If it were decided to harvest the prairie strips for
biomass, biofuels, etc., it would be important to have a prior understanding of the impacts on soil water dynamics.

The watersheds Interim 1, Interim 4, Orbweaver 2 and Basswood 3 would be four potential sites for such a study. It would be important to cover as much of the area of the prairie strip as possible. In Interim 1 and 4, there are already three access tubes per topographic positions, which appear to be enough to detect changes in soil water content. More access tubes would be needed in Orbweaver 2 and Basswood 3. If possible, this should be tested during several growing seasons, and the removal times of the aboveground biomass should be arranged at specific times to compare soil water storage differences with the rowcrop area.

**Water infiltration using stable isotope tracers**

Understanding the effectiveness of strips of native prairie to increase infiltration requires the use of techniques that are not limited by the capability of the equipment used to estimate infiltration. Traditional techniques are limited by the area covered by the infiltration equipment, the number of replicates that can be conducted per day, and are limited in their ability to assess seasonal changes in infiltration. Advantages of using stable isotopes as tracers for estimating infiltration include a larger area of measurement, a small sample volume that can be later processed for isotopic analysis. Samples can be collected periodically at several soil depths and the movement of the tracer into the soil profile can be followed during an entire growing season. Such a study would require
significant laboratory work processing soil samples, but with the extraction apparatus and the methodology already developed, this should be a straightforward task.

Intensive study of depth of plant water uptake

Our study of depth of plant water uptake provides supporting evidence of the ability of prairie species to shift their depth of water uptake in response to environmental conditions. Without the use of tracers, this would be difficult to assess. We included several dominant prairie species in our study; however there are others equally important that were not included. An intensive study over more than one growing season that included more species would provide additional insight into the depth of plant water uptake patterns in mixes annual-perennial watersheds. Differences in depth of plant water uptake can be used as a tool for selecting species in prairie re-establishment based on characteristics, which may regulate the hydrologic balance these ecosystems.

Site-specific characteristics often present trade-offs for experimental design. For example, in our study the watershed Interim 1 has a higher slope allowing the assessment of the effects of topographic position in depth of plant water uptake, while the prairie strip in the watershed Orbweaver 2 is wider which allows the establishment of larger plots. Also, Orbweaver 2 is a lot less disturbed than Interim 1. Thus, the watersheds Interim 1, Orbweaver 2, Interim 3, Orbweaver 3 and the 100% reconstructed prairie Interim 4 would be good sites for the suggested study. In a future study, we strongly recommend even numbers of plots per topographic positions.
Design of a water sampler for isotopic analysis

In isotopic analysis, the time required to process samples, particularly water extraction, limits the number of samples that can be analyzed. Thus, the design of a soil water sampler for use in isotopic studies would open up new possibilities for studies requiring large sample numbers. In addition to its use for studies of depth of plant water uptake, such equipment could potentially be used for studies assessing water residence times within soils under different vegetation. While this principle has been used before, it could be improved through the use of nests of micro-lysimeters installed at different depths. These micro-lysimeters should be specially designed to reduce evaporation within the lysimeter. They should also be small in diameter to reduce the opening in the soil, and to better seal the walls of the opening and avoid surficial water from contaminating the sample. These nests of micro-lysimeters could be placed at different topographic positions under several land covers to address questions about subsurface water movement.

Improved PR2 Probes calibration

During this study we were able to collect enough gravimetric samples at low water contents, which dramatically improved the precision of our instruments. However, due to the high clay content and a shallow depth to groundwater in some areas of our study sites, the gravimetric water content in a few depths in some access tubes was not low enough for precise calibration, resulting in a low $R^2$ of the regression. This was a
greater challenge in the deeper depths, where the groundwater level prevented the collection of gravimetric samples at different water contents in several access tubes.

Laboratory calibration could help to improve the calibration in areas with saturated soils. A calibration apparatus can be developed using soil from the saturated areas, filling PVC cylinders at the same density as indicated in the records by fitting a given mass of homogenized soil into a given volume of the PVC cylinders. The PVC cylinders do not need to be more than 30 cm in height for the 100 cm sensor of the PR2. Larger [110 cm] calibration apparatus can be built if an entire profile needs to be calibrated. However, this might be difficult because the cylinders need to be weighted constantly.
## APPENDIX A. AVERAGE SWS IN THE UPPER 30 CM BY OBSERVATION

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APPENDIX C. AVERAGE SWS IN THE UPPER 60 CM BY OBSERVATION

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# APPENDIX D. AVERAGE SWS IN THE UPPER 60 CM BY OBSERVATION

## DATE AND LAND COVER [2009 AND 2010]

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APPENDIX E. DIFFERENCES BETWEEN δ\textsuperscript{18}O OBSERVED AND THE STANDARD USED TO DETERMINE WATER EXTRACTION TIMES

Soil samples were run at different extraction times to determine the optimum point between extraction time and precision. A total of 55 samples were run and two of them were lost. In this graph we show 53, including the samples where we observed loss of pressure during the extraction process.
This graph shows the linear fractionation observed between $\delta^{18}\text{O}$ and $\delta\text{D}$ in each of the watersheds studied where no tracer was applied. n=15 in Interim 1, 8 in Interim 3, and 24 in Interim 4. Where 1 observation = [$\delta^{18}\text{O}$ and $\delta\text{D}$ pair].
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To my advisors

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The U.S. Forest Service Northern Research Station
Iowa State University College of Agriculture and Life Sciences
USDA-NCR-SARE
USDA-NIFA-AFRI-Managed Ecosystems

To the Porous Media laboratory research group, department of Agricultural and Biosystems Engineering
To the Stable Isotope Paleo Environments Research Group, department of Geological and Atmospheric Sciences
To the Ecohydrology laboratory staff, department of Natural Resource Ecology and Management
To Jonathan Hobbs and Dennis Lock from the Statistics Department
Soil moisture dynamics in agriculturally-dominated landscapes after the introduction of native prairie vegetation

José Antonio Gutiérrez López. 2012