University of Alberta

Weeds promote greater arbuscular mycorrhizal fungi benefit in organically managed spring wheat (*Triticum aestivum* L.) cultivation system

By

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A thesis submitted to the Faculty of Graduate Studies and Research
in partial fulfillment of the requirements for the degree of
Master of Science
in
Plant Science

Department of Agricultural, Food and Nutritional Science

Fall 2012
Edmonton, Alberta

Abstract

Weeds appear to enhance soil microbial benefit in organic agricultuer. We examined effects of weed and microbial effects on spring wheat (*Triticum aestivum* L.) yield and quality under organic management systems. Canadian spring wheat cultivars were planted in weed-free and weedy treatments in 2010 and 2011, and soil microbial profiles were examined using phospholipid fatty acid (PLFA) analysis. The proportion of arbuscular mycorrhizal fungi (AMF) increased and was greater in the weedy plots than in the weed-free plots over the growing season. This greater proportion was associated with greater total plant biomass accumulation but lowered grain yield in the weedy plots. The high yielding semi-dwarf and later maturing cultivars appeared to be yield-stable in organic systems. It may be effective to choose late maturing semi-dwarf cultivars to achieve higher grain yields and benefit through soil microbial communities in organic systems.

Acknowledgment

My deepest appreciation goes to Dr. Dean Spaner for his patience and guidance throughout my Master of Science course work. I am privileged to have you as my supervisor.

I also would like to thank you for my thesis committee members, Dr. Sylvie Quideau and Dr. Miles Dyck for your advice and assistance during my thesis work.

I received generous supports and encouragements from Klaus Strenzke, Glen Hawkins, Lisa Raatz, Fabiana Dias, Alexander Pswarayi, Joseph Back, Ivan Adamyk, Henry Song, Atif Kamran, Graham Collier, Hua Chen, Muhammad Asif, Neshat Pazooki, Alison Nelson and Rachelle Rimmer in my wheat research group.

I would like to also thank Mathew Swallow, Mark Beasse, Pak Chow and Emily Lloret at Department of Renewable Resources in University of Alberta.

This research was supported by grants to the University of Alberta wheat breeding program from the Alberta Crop Industry Development Fund, Western Grains Research Foundation and an NSERC Discovery Grant to Dr. Dean Spaner.

Finally, I would like to thank my parents, family, girlfriend and friends. Without your support, this would not have been possible.

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List of Abbreviations

AMF: arbuscular mycorrhizal fungi

PLFA: phospholipid fatty acid

Symbiotic effects of weeds and soil microbial diversity on wheat under organically managed systems: a review of literature

1-0 Abstract

The presence of weeds and soil microbial community in organic management systems appear to be symbiotic in some manner although weeds generally inhibit crop growth especially in their early growth stage. This symbiotic relationship may compensate economical yield loss with the benefit of ecological sustainability. However, little is known about the effect of weeds on the soil microbial community. Since organic agriculture aims to promote sustainable agricultural management systems, it is important to reach a holistic understanding of the interactions among target crops, weeds and soil microbial communities. The following literature review provides a summary of organic agriculture, trends in global and Canadian Prairie Provinces organic wheat production, and of the interactions between microbial diversity, wheat and weed production.

1-1 Introduction

In recent years the global organic agricultural movement has been promoted as an alternative food production approach because of growing concerns for environmental degradation and worldwide natural resource shortages. Even though a number of studies have been conducted, many ecological charactristics of organic management systems remain unexplored.

Wheat is grown on the largest area of arable land among commercial crops, and is one of three most important cereal crops due to its adaptability and its versatile usage despite deficiency of some amino acids (Shewry, 2007). As the demand for organic products increased, research in aspects of organic wheat production has consequently increased.

Soil microbial communities are one of the key components in soil ecosystems. They have several beneficial roles for agro-ecosystems, such as decomposition of plant residues, carbon and soil nutrient cycling, suppression of soil borne pathogens, improving soil aggregation and production of many antibiotics and other bioactive compounds. Several microbial species have been reported to have symbiotic relationships with aboveground vegetation. These functions have attracted research attention in organic management systems. Accordingly, researchers started to explore specific microbial functions and more efficient crop production schemes by taking advantage of these microorganisms. Despite extended research focusing on the influence of soil microbial communities on target crops, little is known about the direct effect of weeds on microbial communities because weeds are considered a pest and removed from the system. As weeds play roles in nutrient cycling and are ubiquitous in agro-ecosystems, it is important to understand the effect of the presence of weeds on the soil microbial community.

The subject of this literature review is to summarize trends in global and Canadian organic agriculture and the significance of global and Canadian wheat production, review soil nutrient management and microbial diversity in organically managed systems, and examine the roles of weeds on soil microbial communities in wheat systems.

1-2 Wheat (*Triticum aestivum* L.)

1-2-1 General introduction and world wheat production

Wheat was one of the three most important cereal crops in the world in terms of cultivated area (220 million ha) and production (684 million t) in 2009 (Food and Agriculture organization of the United Nation [FAO], 2011b). Wheat makes up approximately 30% of the world's total cereal production and provides about 20% of total human calories (Carver et al., 2009; Braun et al., 2010). The ideal growth temperature of wheat is 25 ℃; it is able to grow between 3 to 32 ℃ (Heyne, 1987). Wheat requires 250 to 1750mm annual precipitation in the growing season (Heyne, 1987). Because of this versatility, wheat is grown in more than 120 countries, in a wide range of environments (from temperate to semi-tropical), even though it is recognized as a cool-season crop (FAO, 2011b). As the result of the implementation of modern techniques following the adaptation of what has been called Green Revolution technology (e.g. chemical fertilizer, large machineries, breeding programs and management improvement), world wheat production has grown steadily between the 1960's to the 90's, while its cultivated area has increased only slightly. This has meant that intensive agriculture has flourished (Heyne, 1987; Reynold & Borlaug, 2006).

The evolution of present polyploid wheat varieties started from natural and artificial selection in the Fertile Crescent. Wheat belongs to *Triticum* genus in the *Triticeae* tribe, and the most widely grown variety is common bread wheat (*Triticum aestivum*: BBAADD) which descends from bread tetraploid emmer wheat (*Triticum dicoccum*: BBAA*)* and wild diploid goat grass (*Aegilops tauschii*: DD) in the Fertile Crescent (Harlan, 1992; Hancock, 2004). Modern wheat cultivars are generally classified into two polyploid species: hexaploid common bread wheat and tetraploid hard or durum type wheat (*Triticum turgidum* subsp. *Durum*: BBAA). There are several other cultivated wheat groups in the world such as diploid: einkorn (*Triticum monococcum*), tetraploid: rivet (*Triticum turgidum* subsp. *turgidum*), Polish (*Triticum turgidum* subsp. *polonicum*), khorasan (*Triticum turgidum* subsp. turanicum) and Persian (*Triticum turgidum* subsp. *carthlicum*); and hexaploid: spelt (*Triticum asetivum* subsp. *spelta*), club (*Triticum aestivum* subsp. *compactum*), and Indian short (*Triticum aestivum* subsp. *sphaerococcum*) (Gustafson et al., 2009). These wheat groups are produced for biofuel or for human consumption in some areas.

Wheat is a self-pollinated plant; therefore, genetic diversity occurs in wild and primitive cultivated progenitors (Gustafson et al., 2009). Bread wheat breeding has been conducted using hybridized hexaploids to create cultivars, overcoming several biotic and abiotic stresses for yield (Acevedo et al., 2009). The unique property of hexaploid wheat is the presence of the gluten in endosperm, which is distinct from other cereals, and enables dough to rise for baking (Gustafson et al., 2009). This unique property made hexaploid bread wheat the predominant species among other wheat species the world over (Heyne, 1987; Hancock, 2004).

1-2-2 Canadian wheat production and trends

Canada produced 26.5 million tonnes of wheat; ranking 6th in the world for its production in 2009 (FAO, 2011a). Wheat consisted of approximately 35% of the total share in merchandise export and was valued at 6 billion Canadian dollars in 2010 (Statistics Canada, n.d.). There were approximately 12,000 wheat farmers in 2006 (Statistics Canada, 2008) and the wheat grain fractionation sector created more than 5,000 job opportunities nationwide (Invest in Canada Bureau, 2009). Therefore, wheat is an economically and politically important crop in Canada. Almost all Canadian wheat production occurs on the three prairie provinces of Alberta, Saskatchewan and Manitoba; on average 96% of Canadian wheat seeded area has been in these three provinces from 1908 to 2007 (McCallum et al., 2008). Three main wheat types are cultivated in these Prairie Provinces: spring and winter hexaploid bread wheat for baking, and durum wheat for pasta (United States Department of Agriculture Foreign Agricultural Service [USDA], n.d.). In Canada, wheat has 8 classes and named by growing region (western or eastern), hardness (hard or soft), colour (red or white) and growing season (spring or winter); and they are divided into subclasses (Canada Grain Commission, n.d.). Hexaploid spring wheat is the leading wheat type, and Canadian Western Red Spring (CWRS) has been the predominant class, grown in more than 60% of the wheat fields from 2008 to 2011 due to its excellent milling and baking quality (Grenier, 2008, 2009, 2010, 2011). Breeding of CWRS classes has a history of challenges to improve protein concentration, bread-making quality and rust resistance traits (McCallum et al., 2008). In recent years, Harvest and Lillian became two leading cultivars grown in the Prairie Provinces, accounting for 17.6% and 17.4% of total CWRS ha respectively (Grenier, 2011). Hexaploid winter wheat has been grown in the central to southern parts of the Prairies due to the severe winter climate, while the durum wheat cultivated area has increased steadily due to its commercial value and good adaptation to the dry semiarid climate in the brown and dark brown soil zones (McCallum et al., 2008).

1-3 Organic Agriculture

1-3-1 Introduction to Organic Agriculture

The origins of organic agriculture date back to the 19th century. The modern organic agricultural movement has been propelled by grassroots organizations, farmers and consumers; it has gradually grown since the late 1980's (Kristiansen et al., 2006). This movement has consequently spread from small local groups to a worldwide movement (Scialabba & Hattam, 2002), and has achieved political and legislative recognition (Kristiansen et al., 2006). Organic agriculture must take advantage of the natural nutrient cycling to achieve sustainable yields (International Federation of Organic Agriculture Movements [IFOAM], n.d.), while intensive conventional agriculture has sought to maximize yield through external inputs and industrialized monoculture (Main, 1999). Organic agriculture cannot be defined with a single and unified definition, and its standards differ in each country, however, the general understanding of organic agriculture derives mainly from three international groups. According to IFOAM, organic agriculture is defined as “a production system that sustains the health of soils, ecosystems and people. It relies on ecological processes, biodiversity and cycles adapted to local conditions, rather than the use of inputs with adverse effects” (IFOAM, n.d.). This definition is similar to the international guideline, The Codex Alimentarius Guidelines for Production, Processing, Labelling and Marketing of Organically Produced Foods. The Codex of Guidelines also highlights important ideas of organic products not by the products but rather by the certification process (Scialabba & Hattam, 2002; Bruinsma, 2003). The other international guideline, EU regulation 2092/91, describes the rules for imports of organic products into the EU, suggesting that there is no unified guideline for organic products (Willer & Menzler, 2008). These guidelines depict a wide range of different aspects among these three international standards. However, the fundamental idea of organic agriculture is globally understood as a comprehensive ecological crop production system designed to enhance agricultural ecosystems (Bruinsma, 2003; Kristiansen et al., 2006). This food production system is practiced with several techniques to maintain soil fertility with care and without mineral fertilizer, genetically modified crops, synthetic pesticides, herbicides or preservatives (Scialabba & Hattam, 2002; Bruinsma, 2003; Kristiansen et al., 2006).

1-3-2 Trends in global organic agriculture

Organic agriculture has become an alternative stream of the food production system worldwide, especially in developed countries, since the late 20th century (Bruinsma, 2003; Lynch, 2009). In recent decades, consumers' concerns for their health and food quality have accelerated this movement. In addition, global interest in environmental protection activities became another major driving force for the promotion of organic agriculture (Scialabba & Hattam, 2002; Holemes & Macey, 2009). Under the provisions of organic certification bodies, organic agriculture tries to conserve a sound agro-ecosystem and reduce external inputs through long-term management (Scialabba & Hattam, 2002; Bruinsma, 2003). Although still controversial, it is thought that organic agriculture is not only an environmentally friendly practice but also the less energy intensive system for both direct (i.e. fuel for machineries, application of fertilizer) and indirect (i.e. production and transportation of fertilizer) consumption (Dalgaard et al., 2001). Zentner et al. (2010) calculated the ratio of energy use efficiency comparing three agricultural management systems (i.e. conventional, conventional with reduced chemicals, and organic management) with three crop rotations and different crop diversity (i.e. a fallow-based low crop diversity, diversified annual crops, diversified annual cereals combined with perennial forage rotations). They concluded that the organic management system with a combination of grain and perennial forages in rotation was the most energy efficient system in terms of an energy output/ input ratio. As the result of these facts, organic agriculture is now practiced globally on approximately 37.2 million ha by more than 1.8 million farmers in 2009 (Willer & Kilcher, 2011). In terms of continents, Oceania has 12.2 million ha of organically managed land. Europe follows at 9.3 million ha and Latin America has 8.6 million ha (Willer & Kilcher, 2011). Although the organic sector is still a niche market, organically managed land has increased about 6% globally between 2008 and 2009 (Willer & Kilcher, 2011), and the range of organic products has increased. As demand of organic products increase, organic farmers face the challenge of expanding their production systems and yield to meet this demand.

1-3-3 Organic agriculture on the Canadian Prairie Provinces

By the middle of the 20th century, intensive monoculture, which applied mechanical deep tillage, chemical fertilizers, herbicides, pesticides, and frequent summer fallowing, became a common practice on the Canadian prairies; it also made Canada one of the largest cereal exporters in the world (Reganold et al., 1987; Zentner et al., 2001; Holemes & Macey, 2009). In recent years, several environmental problems including soil, water quality and biodiversity degradation, and large greenhouse gas emissions caused by these intensive agricultural practices became a concern (Entz et al., 2001; Badgley et al., 2007). Consequently, concerns for environmental pollution and anxiety for limited stocks of non-renewable energy sources have led some producers to shift toward less energy intensive food production systems (Zentner et al., 2010). Health concerns over pesticide residues on products and anxiety about genetically modified food, together with environmental issues, have helped develop the Canadian organic sector (Yiridoe et al., 2005; Cranfield et al., 2009; Holemes & Macey, 2009); Canada is a large exporter of raw and processed organic products (Holemes & Macey, 2009). As global demands for organic products increased, large-scale organic production systems started to be required (Nelson et al., 2004). Accordingly, Canadian organic farmers have been challenged to overcome organic production constraints of weed pressure, soil nutrient deficits, pest and disease damage, and crop/cultivar choices on a large scale (Degenhardt et al., 2005; Mason & Spaner, 2006). Weed and soil fertility management still remain as the two largest concerns for the farmers (Degenhardt et al., 2005).

Canadian organic farm land grew approximately 160% from 2002 to 2008 (Macey, 2003; Agriculture and Agri-Food Canada [AAFC], n.d.). There are 611,000 ha of certified organically managed land and 3,713 certified organic farmers in Canada (AAFC, n.d.). Although Canadian organic farmers are found in all provinces, the prairie provinces of Alberta, Saskatchewan and Manitoba account for 80% of Canadian organic land, operated by 40% of total organic farmers in 2008 (AAFC, n.d.). In these three provinces, hexaploid spring wheat is a main grain crop in both conventional and organic farms, and this organic wheat accounts for one of the most important organic export revenue streams in Canada (AAFC, n.d.).

1-3-4 Introduction to soil fertility in organic agriculture

The concept of soil fertility is complex and difficult to grasp; however, fertility generally refers to the amount of soil nutrients, the soil nutrient holding capacity, and availability of those nutrients for plants (Eash et al., 2008). Soil fertility is maintained by intertwined chemical, physical and biological properties; therefore, soil fertility varies according to soil properties, climate and other environmental factors, aboveground vegetation and field management systems, which potentially affect these three soil properties (Eash et al., 2008). While intensive agricultural systems aim for high yield by increasing soil fertility through chemical amendments (e.g. fertilizers, herbicides, pesticides and fungicides), organic farming systems try to maintain fertility without chemical amendments. Therefore, soil management sometimes differs between the two systems. The goal of soil fertility management in organic agriculture is to achieve sufficient yield by taking advantage of the natural soil nutrient cycle (IFOAM, n.d.; Nelson et al., 2004; Nelson & Janke, 2007; Birkhofer et al., 2008). Maintaining essential macronutrients for plants such as nitrogen, phosphorus, potassium and water depends upon inherent soil fertility and/or supplements from external inputs since these elements are taken away from the farm as end products. Water is usually renewable and replaceable by rain in areas where there is sufficient rainfall; soil amendments, such as manure and compost are commonly applied for maintaining other elements in organic systems (Emmens, 2003). However, these organic amendments are generally low in macronutrients and are slow to release them (Barker, 2011). Entz et al. (2001) reported that the average level of soil phosphate in organic farms in Manitoba, Saskatchewan and North Dakota was relatively lower than the level found in conventional farms in Manitoba. Furthermore, long-term cropping systems without phosphorus fertilization caused serious reduction of soil available phosphorus (Campbell et al., 1993). In recent decades, soil microorganisms have attracted attention as one of the solutions for the maintenance and optimal utilization of native soil fertility, especially in organic farming systems (Glassop et al., 2005; Gibson et al., 2007; Birkhofer et al., 2008; Cornejo et al., 2008; Smukler et al., 2008; Kahiluoto et al., 2009; Smith & Read, 2009; Barker, 2010; Verbruggen et al., 2010).

1-3-4-1 Nutritional inputs for soil fertility in organic agriculture

Organic agriculture strictly regulates the application of chemical fertilizers; therefore, natural amendments such as farmyard manures, green manure, crop residue compost and crop rotations with legumes are commonly applied in organically management systems (Scialabba & Hattam, 2002; Bruinsma, 2003; Emmens, 2003; Kristiansen et al., 2006; Dorais, 2007). An adequate amount of these amendments has multiple positive functions for maintaining soil fertility. (e.g. they are low amount but a great source of plant-available nutrients, they help retain sufficient soil moisture and nutrients, and maintain a favorable environment for soil microorganisms) (Bulluck et al., 2002). A number of studies reported positive effects of long-term applications of these organic amendments since these materials release nutrients slowly (Barker, 2011). Farmyard manure, green manure and compost increased soil microbial diversity (Mäder et al., 2002; Smukler et al., 2008), and consequently increased diversity of aboveground fauna (Birkhofer et al., 2008). Long-term farmyard manure application in organically managed fields maintained high levels of soil organic carbon and soil total nitrogen as a result of slow decomposition of organic matter and consequently promoted the mineralization of organically bound nitrogen by microorganisms (van Diepeningen et al., 2006; Birkhofer et al., 2008)

Management with organic amendments (i.e. composted cotton-gin mixed with composted poultry manure, uncomposted cattle manure or hay-cattle manure) had high levels of soil organic matter, cation exchange capacity and an increased density of specific beneficial fungi (e.g. *Trichoderma*) in a over 2-year application. Conventional management systems with chemical fertilizers tend to have high soil bulk density due to a lack of soil organic matter, which cause plant root growth restriction as well as poor movement of air and water (Bulluck et al., 2002). High bulk density also increased the density of pathogenic fungi (e.g. *pythium* and *Phytophthora*) (Bulluck et al., 2002). A 21-year organic agricultural study revealed increasing soil aggregate stability and a population of arthropods under different organic managements of bio-dynamic and bio-organic cultivation systems (Mäder et al., 2002). In another long-term field experiment, Arbuscular Mycorrhizal Fungi (AMF) external mycelia, which are known as nutrient transporters (Smith & Read, 2008), had higher densities in organically managed fields than in conventionally managed fields, suggesting that organic amendments increase general soil biological activity in long-term organic systems (Gryndler et al., 2006). Other studies observed that green manure and crop residues are adequate sources of organic matter, which maintain soil biotic and abiotic properties, thereby facilitating the entire nutrient cycle in fields (Reganold et al., 1987; Kristiansen et al., 2006).

1-3-4-2 Plant breeding in organic agriculture

Plant breeding is an effective strategy to overcome constraints in organic management systems (Wolfe et al., 2008; Reid et al., 2009 a,b). Organic wheat needs to be bred for improved nutrient utilization, competitive aboveground structure, early seedling vigour and root system development to outcompete weed pressure, disease resistance, and consistent quality of end products (Manson & Spaner, 2006; Wolfe et al., 2008; Reid et al., 2009 a,b). Specifically, nutrient utilization in organic systems may be achieved by well-developed rooting systems, developing a symbiotic relationship with soil microorganisms, and relocating absorbed nutrients from where they are stored to where the nutrients are most needed (Koide, 1991; Zou et al., 2002; Li et al., 2005, 2006, 2008; Löchenberger et al., 2008; Smith & Read, 2008). Some of the genetic information dealing with these traits has been reported, and breeders can use this information to breed suitable varieties under organic production systems (Zhu et al., 2001; Glassop et al., 2005; Jones et al., 2010). However, because of different growing conditions and stresses, certain agronomic traits in wheat cultivars bred in conventional systems do not necessarily perform in the same manner under organic management systems. Thus, wheat for organic cultivation systems should be bred and selected in organically managed systems (Reid et al., 2009 a,b). In addition, breeding should meet needs for overcoming various site-specific constraints. In the Canadian Prairie Provinces, wheat breeding programs try to overcome area-specific constraints through multilocation yield trials, and grain quality still remains the most important breeding objective (MaCallum & DePauw, 2008). Drought limitation has been an issue in southern Alberta and Saskatchewan, and early maturity is a critical trait for wheat growing in central and northern areas to avoid frost damage (McCaig & DePauw, 1995). Stem and leaf rust and other disease resistances are necessary for wheat growing in the Prairie Provinces, especially southern Manitoba and south western Saskatchewan where rust damage is critical (McCallum & DePauw, 2008). Mason et al. (2007) reported that spring wheat grown under organic management system in Alberta should have weed competitive ideotypes including ability for long-culmed fertile tillers with early maturity.

1-3-4-3 Microbial communities and their role in soil fertility in organic agriculture

Soil microorganisms are the organisms living in the soil. They are composed of unicellular or multicellular organisms and outnumber all other organisms above ground in terms of biomass and variety (Madigan et al., 2006; Lupwayi et al., 2010). They are classified into taxonomic categories as bacteria, algae, fungi, viruses, protists and nematodas (Buscot & Varma, 2005; Madigan et al., 2006); some of them take part in the function of decomposition of soil organic matter, nitrogen-fixation, denitrification and ammonia oxidization, solubilization of soil nutrients and biological control of soil born plant pathogens (Kaschuk et al., 2010; Lupwayi et al., 2010). In addition, soil microorganisms can store nutrients and facilitate many soil enzymatic activities, which are important for nutrient dynamics in the soil (Melero et al., 2008). Therefore, microbial diversity plays a significant role in aboveground plant growth, and the assessment of soil microbial diversity, and monitoring its community alteration are good indicators of soil health and fertility (Gunapala et al., 1998; Potthoff et al., 2006; Liu et al., 2007; Kaschuk et al., 2010; Lupwayi et al., 2010). Decomposers are an important component of microbial diversity, and these saprotrophic microbes decompose complex soil organic matter into simple forms, which other organisms are able to absorb (Kennedy, 1999). Hence, increasing the entire soil microbial diversity enhances the soil biological metabolic efficiency, which in turn, improves aboveground productivity. Greater mycorrhizal root colonization was observed in organic transitional fields (Smukler et al., 2008). This resulted in better plant absorption of nutrients through fungal mycelia, which led to efficient soil nutrient utilization. Some rhizosphere microbes promote plant root health and suppress corky root, soil-born fungal and bacterial diseases as well (Workneh & Bruggen, 1994; Bruggen & Semenov, 2000; Postma et al., 2008). Such functions indicate the importance of their relative diversity in organically managed fields. Due to their positive influence on entire agro-ecosystems, microbial diversity has attracted a great deal of research attention (Glassop et al., 2005; Gibson et al., 2007; Birkhofer et al., 2008; Cornejo et al., 2008; Smukler et al., 2008; Kahiluoto et al., 2009; Smith & Read, 2009; Barker, 2010;Verbruggen et al., 2010). Low-input and/or organically managed systems are reported to have richer microbial diversity than conventional systems (Workneh et al., 1994; Bulluck et al., 2002; Oehl et al., 2004; Birkhofer et al., 2008; Verbruggen et al., 2010), while some studies reported only a slight difference of microbial diversity among these managed fields (Kurle & Pfleger, 1996; Franke-Snyder et al., 2001). This disagreement might be caused by methods of analyzing and calculating microbial diversity, cultivation history and inherent soil properties (Frostegård et al., 2010).

Arbuscular mycorrhizal fungi are among the most common and ubiquitous soil microorganisms existing in almost all soil systems (Smith & Read, 2008). They are classified in the phylum of Glomeromycota consisting of four orders (SchüBler et al., 2001; SchüBler, n.d.). They are able to form symbiotic relationships with a great number of plant species. Symbiotic relationships are necessary with their host plants. The relationship involves carbon additions through photosynthesis from host plants to AMF, and soil-derived nutrients from AMF to the plants (Smith & Read, 2008; Ryan & Tibbett, 2008). Due to this nutrient exchange function, AMF utilization in agricultural fields has been explored in organic agricultural systems as the alternative of choice to chemical fertilizers. Oehl et al. (2004) examined the levels of AMF diversity in the context of long-term field management effects and found higher AMF diversity in long-term organically managed fields than in conventional management systems. Diverse AMF assemblages, which were similar to those of semi-natural grasslands, were observed in organically managed systems, while fewer varieties of AMF were found in conventional systems in another study (Verbruggen et al., 2010). It appears in these studies that organic management systems may offer AMF a more favorable environment compared to conventional systems. Organically managed systems also tend to have higher diversity of other soil organisms (e.g. spiders, earthworms, gram-positive and gram-negative bacteria and actinomycetes) (Mäder et al., 2002; Smith & Read, 2008; Smukler et al., 2008; Nelson et al., 2011a). Ryan and Tibbett (2008) reviewed several studies and reported a tendency toward greater AMF diversity due to low levels of soluble phosphate fertilizer application and frequent diversified crop rotation, which are common characteristics of organic management systems.

There are many studies reporting the positive functions of AMF and other microbes on crops. For instance, mycorrhizal plants whose roots have mutualistic association with fungi (e.g. AMF) show improved nutrient absorption. Mycorrhizae: 1) explore new space and absorb phosphate by using their extraradical hyphae (Li et al., 2006, 2008; Smith & Read, 2008), 2) alter the rhizosphere pH as a result of root ion absorption which consequently enhances the availability of phosphate in the soil (Koide, 1991; Mohammad et al., 2005; Tawaraya et al., 2006), 3) alter other microbial communities which may enhance nutrient absorption (Marschner & Timonen, 2005). Li et al. (2006) reported that AMF hyphae are actually able to absorb phosphate that is otherwise inaccessible by wheat roots. They applied phosphorus isotope ^{32}P in compartment pots placed in mesh bags, which were inaccessible by the roots but only accessible by fungal hyphae. They observed ^{32}P only in arbuscular mycorrhizal inoculated wheat tissue, concluding that the mycorrhizal pathway of P absorption contributed to P acquisition, although plant shoot dry biomass seems to be low in arbuscular mycorrhizal inoculated wheat in this study. Mohammad et al. (2005) explained the function of the mycorrhizal pathway of P acquisition. In their study, lower pH was observed in mycorrhizal infected wheat rhizosphere due to alteration of root exudation, modification of microbial population and high rate of nitrification along with increasing carbon dioxide. This was thought to enhance absorption of immobile phosphate. According to Tommerup and Abbott (1981), arbuscular mycorrhizal infected vegetative hyphae survive at least six months, and Karasawa et al. (2001) reported that sunflower (a mycorrhizal host plant) positively affected the shoot dry weight and AM colonization of the succeeding maize crops. These studies imply the beneficial aspects of the long-term crop rotation including AMF host plants.

1-4 Microbial Diversity

1-4-1 Introduction to microbial diversity

Microbial diversity can be defined through species richness, abundance and species evenness or equitability (Wittebolle et al., 2009). A main research subject of soil microbial diversity in agriculture is to understand aspects of microbial diversity that are useful indicators of soil fertility, especially in organic farming systems (Gunapala et al., 1998; Potthoff et al., 2006; Liu et al., 2007; Kaschuk et al., 2010; Lupwayi et al., 2010). Soil microbiota is vulnerable and easy to alter through agricultural practices (Gunapala et al., 1998; Potthoff et al., 2006; Liu et al., 2007; Kaschuk et al., 2010; Lupwayi et al., 2010). Organic agriculture uses management systems which conserve more complex flora and fauna than conventionally managed fields (Hole et al., 2005), while conventional agriculture, which relies on intensive monoculture by chemical fertilizer, herbicide and pesticide, simplifies and reduces soil microbial diversity (Lynch, 2009). Smukler et al. (2008) reported that soil microbial biomass increased even during the transitional phase from conventionally to organically managed fields. Specifically, AMF community richness increased in a sequence of transitions to organic agriculture (Verbruggen et al., 2010). Observation of microbial variation and composition help us identify microbial benefits (Kristiansen et al., 2006). For instance, Mäder et al. (2002) compared economic benefits of organically managed systems in terms of microbial contributions. They found that mean crop yields in organic systems were only 20% lower than those in conventional management systems, whereas mean input of fertilizer (N, P, K) in organic management systems was 34 to 51% lower than in conventional systems over 21 years. They concluded that soil fertility can be improved by increasing faunal and microbial diversity (e.g. mycorrhizal infection, earthworms, arthropods) and greater number of weed species. Thus, in the long-term organically managed systems may exhibit relatively low rates of yield reductions in spite of an absence of chemical fertilizer. To characterize soil microbial diversity, phospholipid fatty acid (PLFA) analysis has been employed (Piotrowska-Seget & Mrozik, 2003). Phospholipids are one of the important types of lipids which exist in microorganism cell membranes and the lipids contain fatty acids bound to a glycerol backbone (Olsson, 1999). We can interpret soil microbial composition through the PLFA patterns (Piotrowska-Seget & Mrozik, 2003; Nelson et al., 2011a,b).

1-4-2 Microorganism interactions with wheat in organic agriculture

Agro-ecosystems consist of intricate connections between microorganisms and other organisms (Altieri, 1999). Consequently, the roles of microorganisms in organic management systems are diverse (Koide, 1991; Marschner & Timonen, 2005; Mohammad et al., 2005; Tawaraya et al., 2006; Li et al., 2006, 2008; Smith & Read, 2008;). There are many studies investigating the mechanisms of plant nutrient uptake enhancement by microorganisms (Mäder et al., 2002; Mohammad et al., 2005; Li et al., 2006, 2008; Smith & Read, 2008). Microbial communities highly depend on aboveground species as well as cultivation methods (Karasawa et al., 2001; Sanons et al., 2006; Birkhofer et al., 2008). Many aspects of modern agricultural practices such as tillage, agrochemicals and crop rotation alter soil properties and consequently limit microbial activities (Hetrick et al., 1995). Each plant species releases different types and amount of root exudates which directly affect the microbial communities in the rhizosphere where microbial symbiosis occurs (Lynch, 1994), and management practices highly influence microorganism composition (Kurle & Pfleger, 1995; Potthoff et al., 2006; Lynch, 2009). As previously mentioned, phosphorus deficiency has been reported for organic wheat producing farms (Entz et al., 2001; Manson et al., 2007; Martin et al., 2007). It is therefore important to explore the benefits of soil microbial interactions with wheat to improve soil phosphorus utilization and absorption under organic management systems. Phosphate acquisition of arbuscular mycorrhizal infected wheat may be attributed to the amount of phosphate in soil (Mohammad et al., 2005; Li et al., 2006, 2008; Ryan & Tibbett, 2008; Smith & Read, 2008), types and combinations of symbionts (Zaidi & Khan, 2005), timing of colonization (Hetrick & Bloom, 1984; Gryndler et al., 2006) and cultivars or genotypes of wheat (Hetrick et al., 1992, 1993, 1995; Xavier & Germida, 1998; Graham & Abbott, 2000; Zhu et al., 2001). Mohammad et al. (2005) observed a large amount of P and Zn absorption by AMF colonized wheat in a low soluble P soil while they reported no differences in P absorption between non-colonized and colonized wheat in a high P soil. Li et al. (1991) suggested that mycorrhizal plants produce more carbon dioxide, which enhanced nitrification. As a result, there is a progressive acidification of the rhizosphere, and phosphorus, which tends to be fixed due to excess calcium in alkalin soil and aluminium in acidic soil, becomes more mobile and available to the wheat root in soil. In another study, combinations of multiple microbial inoculations (with nitrogen-fixing: *azotobacter chroococcum,* phosphate solubilizing microorganisms: *Pseudomonas striata and Penicillium variable,* and AMF: *Glomus fasciculatum*) showed mostly positive but different effects on wheat yield and nutrient uptake (Zaidi & Khan, 2005). This result suggested that higher microbial diversity may have synergetic effects, resulting in better wheat nutrient absorption than simple microbial communities. This study also depicted intertwined relationship among soil microorganisms and the importance of higher soil microbial diversity.

Genomic affinity may control microbial symbiotic relationships. Hetrick et al. (1993, 1995) reported that mycorrhizal dependence of modern hexaploid wheat cultivars may be heritable and located on 5B and 7B chromosomes or on D genomes. The other factors may be related to morphological, physiological and phenological traits in wheat. Hetrick and his colleagues (1992, 1993 and 1995) concluded through a series of studies that mycorrhizal responsiveness was higher in standard height cultivars compared to semi-dwarf types. Zhu et al. (2001) reported that response to mycorrhizal colonization in old and modern wheat cultivars has no correlation to either agronomic phosphorus efficiency or soil phosphate uptake efficiency, however, there is a relationship between root biomass and mycorrhizal colonization. These studies suggest that the benefit of arbuscular mycorrhizal colonization may not be equal among wheat cultivars, depending on their physiological and genetic differences. Nelson et al. (2011a) reported no response differences among a small sample of Canadian cultivars. In another study, the Canadian wheat cultivar Columbus positively responded to arbuscular inoculum, resulting in greater yield and grain phosphorus use efficiency; however, another inoculated Canadian spring wheat cultivar (Katepwa) exhibited lower grain yield, shoot dry weight and grain phosphorus use efficiency (Xavier & Germida, 1998). Graham and Abbott (2000) reported that the benefits of mycorrhizal infection are dependent on the exchange balance between photosynthesized carbon from host plants and phosphate acquisition by AMF. Thus, the physiological performance relating to photosynthesis in wheat cultivars may affect their symbiotic balance. Xavier and Germida (1998) also suggested that no benefit of mutualism between AMF and wheat may be caused by the parasitic carbon requirement of the inoculants. These studies suggested that wheat physiological traits may be important for plants to have beneficial relationship with AMF.

1-4-3 Cereal micronutrients and soil microbial interactions in organic agriculture

The relative differences in nutritional quality between organically and conventionally grown cereals concerns both producers and consumers. There is a presumption that organic products are superior in their taste and nutritional value (Aertsens et al., 2009); however, no consistent studies about nutritional trends between organic and conventional products have been reported. For instance, Woese et al. (1997) reviewed relevant studies and maintained that there is no clear difference in grain minerals in cereals except a tendency of low protein in organically grown cereals. In addition, Mäder et al. (2007) reported no significant difference of plant macro and micro nutrients between organic and conventional managements systems. On the other hand, there are several reports that organic products contained higher iron (Worthington, 2001; Saha et al., 2007), magnesium (Worthington, 2001) and phosphorus (Worthington, 2001; Dangour et al., 2009). Some studies also reported higher protein content or no difference in organically grown wheat and rice (Manson et al., 2007b; Saha et al., 2007; Nelson et al., 2011a).

Because several agricultural management factors (e.g. fertilization, tillage, crop rotation, organic agricultural practices, soil moisture management and breeding) are intertwined and all affect crop nutritional quality in addition to geographical and environmental variations (Wang et al., 2008), knowledge about microorganism function for plant nutrient uptake may help to understand the effect of organic management on grain nutrition. Correlations between abundant microbial communities and high levels of grain micronutrients have been reported (Nayyar et al., 2009; Nelson et al., 2011a). Particularly, many studies concerning the relationship of AMF and plant micronutrients have been reported. Cornejo et al. (2008) suggested interactive effects of different types of fertilization and microbial symbiosis on grain micronutrients in acidic soils. They reported that N sources (ammonium and nitrate) affected AMF mycelium development differently, which indirectly altered plant shoot micronutrient contents, resulting in higher absorption rate of Zn and P in mycorrhizal colonized wheat. They concluded that a greater effect of mycorrhizal colonization on particular micronutrients (P and Zn) resulted from NO_3^- fertilization due to the maintenance of root electroneutrality. Ryne et al. (2004) reported that organically produced wheat grain contains slightly higher levels of Zn and Cu, but lower Mn and P than conventionally cultivated grains. They explained this by suggesting that soluble P fertilizers increased P uptake but decreased mycorrhizal infection. Consequently, Zn absorption decreased. These results agreed with many other studies that mycorrhizal plants increase P and Zn absorption under low-P conditions; however, plants absorbed P by themselves in highly soluble P conditions, which reduced mycorrhizal infection. Nelson et al. (2011b) reported a positive correlation between Cu and fungal PLFAs in an organically managed system in Alberta. Summarizing these studies, the interaction between crop nutrients and soil microbial community (particularly AMF) may vary depending on soil properties. The mutual nutritional exchange may be affected only when demands of both plant and microorganisms are complimentary because plant symbiotic relationships with AMF are balanced by mutual nutrient exchange (AMF to plants: soil relatively immobile nutrients, plant to AMF: carbohydrates)(Smith & Read, 2008). Differences in micronutrient concentrations in AMF colonized wheat reflect nutrition efficiency in crops and soil fertility in organic systems. Clark and Zeto (2000) reported by reviewing relevant articles that positive effects of micronutrient uptake (e.g. P, N, Zn, Cu, K, Ca, Mg) on mycorrhizal infected plants depended on the range of soil pH. All these reports may be summarized by stating that grain micronutrient concentration depends on the type of microorganisms, nutrient efficiency in cultivars, soil type and soil nutrient levels, and the management system (Wang et al., 2008).

1-5 Weeds in organic agricultural systems

1-5-1 Weeds in organic agriculture

Weeds are one of the biggest constraints in crop production. Weeds cause enormous economic losses (e.g. $984 million in 58 commonly planted crops and $300 million in wheat obtained by the sum of each provincial reports during 1985 to 1989) in Canada (Swanton et al., 1993). Conventionally managed monoculture fields are generally well maintained through tillage, crop rotation and agrochemical application to avoid yield loss by weeds. Achieving maximum yield is the main target in conventional agriculture; therefore, monoculture and simplification of the workload in agricultural systems has been practiced. Thus, conventional fields are likely to lose well balanced biodiversity. On the other hand, more plant species were observed in organically managed fields (Gabriel & Tscharntke, 2007; Gibson et al., 2007). Thus, while the biggest constraint in fields, weeds may also be considered as one of the important components of field biodiversity (Chen et al., 2004). For instance, only twelve cereal species are planted globally on most agricultural lands (Fowler & Mooney, 1990). On the other hand, several studies have emphasized that species diversity (including weeds) in organic agricultural fields are relatively high and this diversity supports the important functions of ecological services, especially in terms of soil nutrient cycling (Altieri, 1999; Main, 1999; Jordan et al., 2000). In addition, increasing concerns about the environmental impact of agrochemicals have made some producers hesitate to use excessive herbicides to control weeds, leading them to seek other ways to deal with them (Swanton et al., 1993).

1-5-2 Weed management in organic agriculture

Weed management is one of the most time and energy consuming tasks for organic farmers in Canada (Entz et al., 2001; Degenhardt et al., 2005). Spatial and nutritional competition is greater under organic management systems due to coexistence of many species. In organic systems, it was reported that weeds caused about 40% grain yield reduction in spring wheat cultivars growing in Alberta (Manson et al., 2007). Organic farmers have to deal with greater numbers and diversified weed species without chemical herbicides. Organic weed management strategies include differential seeding rates, use of clean seed, altering seeding dates, cultivar choices and crop rotations (Samuel & Guest, 1990; Emmens, 2003; Manson et al., 2007; May et al., 2009; Anderson, 2010). Seeding double the recommended seed rate suppressed weeds and maintain spring wheat yield in Alberta (Manson et al., 2007). The authors concluded that this strategy was more efficient to suppress weed pressure than cultivar choice. Derksen et al. (2002) also reported that increased seeding rate lessened the weed seed bank for the subsequent growing season. Crop rotation could also prevent or/minimize weed seed bank build-up (Kristiansen et al., 2006; Anderson, 2010). Arranging seeding date is a good strategy to avoid weed damage as well. O'Donovan et al. (1985) reported that emergence of weed seedlings prior to crop seedlings causes great yield loss. Therefore, delaying seeding date following tillage to remove weed seedlings in barley was an efficient weed control method (Mckenzie et al., 2005). However, this practice shortens the growing period, causing grain quality problems and potential yield reduction.

Plant physiological traits (e.g. height, early maturity and tillering capacity) have been studied as important weed competitive traits in spring wheat (Mason et al., 2007; Reid et al., 2009b). Tall genotypes, great tillering capacity and canopy structuring, maximizing light interception, early biomass accumulation, flag leaf length, ground cover and timing of spike emergence are all considered as important plant structural factors to compete against weeds (Mason et al., 2007). For organically managed fields, higher seeding rates and the choice of proper cultivars to meet needs for their site-specific competitors are important considerations for weed management. Mechanical tillage is an efficient weed management tool; however, frequent tillage compresses and alters soil surface structure and reduces some microorganisms, causing a critical loss for the entire agro-ecosystem (Mazzoncini et al., 2009).

1-5-3 Weed and soil microorganism relationships in organic agriculture

Despite improved weed management techniques, little is known about the effects of weed communities on entire agro-ecosystems in organic management systems. Soil microbial diversity is supported by soil physiological, chemical and biological properties as well as interactions with aboveground vegetation, including weed communities (Chen et al., 2004). Several indirect benefits of weeds on soil microorganism function in organic systems have been reported (Feldmann & Boyle, 1999; Jordan et al., 2000; Suárez et al., 2001; Loranger-Merciris et al., 2006), while there are also reports on the negative impacts of weed biomass and density on some crop yields (Milberg & Hallgren, 2004; Degenhardt et al., 2005; Eslami et al., 2006; Pollnac et al., 2009). Pollnac et al. (2009) investigated the effect of neighbor plants (including several weed species) on wheat yield. They reported that neighboring weed densities, especially early in the season, altered wheat physiological performance and consequentially reduced grain yields. However, there appears to be a small negative impact of increased weed diversity on wheat yield. Suárez et al. (2001) suggested that weed variety richness prevented soil degradation, which indirectly protects soil micro biota. Loranger-Merciris et al. (2006) also reported greater microbial activities and catabolic diversity in grasslands with larger vegetative diversity compared to fields with simple vegetation. Positive correlations between weed species and microbial diversity have been reported in other studies (Feldmann & Boyle, 1998; Chen et al., 2004). Some studies report that modern wheat cultivars and ancestral wheat cultivars have different responsiveness to mycorrhizal colonization (Hetrick et al., 1992, 1993, 1995). As previously mentioned, some of these differences in response to mycorrhizal colonization may be attributed to the differences in nutritional acquisition ability in modern bred compared to wild wheat cultivars, and a similar trend has been reported among weed species. One of the problematic weeds in Alberta, wild oats (*Avena fatua* L.), exhibited differential responsiveness to mycorrhizal infection from cultivated oats (Koide et al., 1988). Wild oat has inherent physical plasticity and a vigorous root system to grow well in less fertile conditions, while cultivated oats have been bred for improved productivity under high input conditions. The inherent tolerance of wild oats to low soil fertility may be the result of differential responses of mycorrhizal infection (Koide et al., 1988). Haynes et al. (1991) grew wild oats and cultivated oats in a growth chamber. Wild oats developed higher dry root matter than modern bred oats in both low and high P conditions, suggesting wild oats physiologically more competitive nutrient absorption through well-developed root systems. This allowed wild oats to acquire more phosphate than modern bred oats (Haynes et al., 1991).

There are several studies reporting the relationship between weed communities and soil organisms (e.g. beneficial insect, mycorrhizal fungi) in arable lands (Feldman & Boyle, 1999; Kytöviita et al., 2011). A recent study reported that suppression of soil fungal communities by fungicide application increased performance of mycorrhizal non-host weed spices in corn fields (Jordan & Huerd, 2008). This result indicates that soil fungal communities affect weed community compositions, and reduce competitiveness of mycorrhizal host weeds against non AMF host weeds due to loss of the symbiotic benefit through AMF. This may cause a shift in weed diversity, affecting crop yield. In addition, elimination of AMF host weed species possibly causes loss of AMF beneficial effect on crop growth and yields (Feldmann & Boyle, 1999; Kabir & Koide, 2000). In another study, Kabir and Koide (2002) compared fallow management systems with cover crops. Cover crops of fall rye (*Secale cereal* L.) and/or oats (*Avena sativa* L.) increased mycorrhizal colonization and density of mycorrhizal hypha in subsequent sweet corn (*Zea mays* L.) crops. This study also reported that planting cover crops resulted in higher corn yield and a higher shoot phosphate concentrations in the corn plants. They maintained two reasons for the possible role of these cover crops on high yield and high phosphate concentration in the subsequent crop. Firstly, they hypothesized that cover crops facilitated better conditions for a diverse microorganism community, which helps corn absorb phosphorus from decomposed cover crops. Secondly, cover crops may physically protect topsoil from erosion. As a result, the field exhibited greater aggregated soil where crop root extension and microbial activity were enhanced. Burrows and Pfleger (2002) also examined the impact of aboveground plant species diversity on arbuscular mycorrhizal sporulation and species composition. They found a positive correlation between the number of aboveground plant species and AMF sporulation. These results may indicate that increasing aboveground biomass and/or species richness, regardless of species, increased the diversity of microbial communities. As a result, the entire agro-ecosystem is maintained. Several studies reported positive correlations between organic management systems and soil microbial communities (Mäder et al., 2002; Birkhofer et al., 2008; Verbruggen et al., 2010; Nelson et al., 2011a); however, there are few actual studies which investigated weed-microbial community interactions under organically managed fields. Nelson et al. (2011a) reported that weed seed yield and soil microbial diversity were positively correlated suggesting that high aboveground plant biomass and diversity created enhanced levels of soil microbial diversity. It is known that microbial diversity is altered by several agricultural practices such as agrochemicals (Mohammad et al., 2005; Santos et al., 2006; Ryan & Tibbett, 2008; Smith & Read, 2008), tillage (Menéndez et al., 2001), aboveground vegetation and crop rotations (karasawa et al., 2001; Kabir & Koide, 2002; Loranger-Merciris et al., 2006).

1-6 Conclusion

Organic farmers rely mainly on natural materials and techniques to maintain adequate soil fertility which is derived from integrated biological activities. Weed stress is one of the biggest constraints for organic farmers; however, there are few studies which examine the symbiotic relationship between weeds and microbial diversity on crop yield. In organic agriculture, maximizing soil nutrient utilization with limited materials and techniques is critical to achieve sufficient yield, and soil microorganisms have been recognized as a key factor to enhance nutrient absorption. The microbial community is highly influenced by soil management systems as well as aboveground vegetation. Research is required to elucidate the relationship between weeds and microbial diversity to uncover more efficient and sustainable agricultural management systems.

1-7 Statement of Purpose

Knowledge of Canadian organic farming systems has been developed with increasing demand for organic products. Despite numerous studies claiming substantial positive effects on wheat at an experimental level, practical knowledge of soil microbial diversity under organic conditions is relatively small. Understanding weed functions on microbial communities under organic management would give direction for weed management techniques in organic agriculture. Organic producers may find merit in maintaining adequate levels of weeds to maintain a holistic agro-ecosystem.

Conventional agricultural farmers also would have a chance to reduce pesticide use, keeping a balance between yields and weed pressure if weeds have some positive influence on microbial diversity, which promotes plant nutrient uptake.

The objectives of this research are,

1, To examine potential differences in soil microbial communities between weed-free and weedy environments under organic management systems over wheat growing seasons.

2, To examine the relationship of wheat cultivar performance in weed-free and weedy environments under organic systems.

3, To examine the relationship between wheat grain nutrition and microbial diversity in weedy and weed-free environments under organic management systems.

The underlying hypotheses are,

1, There are differences in soil microbial structure between weed-free and weedy environments under organic management systems over wheat growing seasons. It is higher in weedy than in weed-free environments in both early and late seasons.

2, Modern Canadian cultivars exhibit higher grain yields than other wheat cultivars in both environments.

3, There are differences in grain micronutrient contents between the two systems due to different microbial composition among treatments and cultivars.

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Weeds promote greater arbuscular mycorrhizal fungi benefit in organically managed spring wheat (*Triticum aestivum* L.) cultivation system

2-1 Introduction

Global interest in environmental protection and consumers’ health concerns have become major driving forces for the increase in organically managed fields (Scialabba & Hattam, 2002; Holemes & Macey, 2009; Willer & Kilcher, 2011). As demand of organic products increases, organic farmers face the challenge of expanding their production systems or increasing yield to meet demand. Canada is a large exporter of raw and processed organic products (Holemes & Macey, 2009). The prairie provinces of Alberta, Saskatchewan and Manitoba account for 80% of Canadian organic land, operated by 40% of total organic farmers in 2008 (Agriculture and Agri-Food Canada [AAFC], 2010). In these three provinces, hexaploid spring wheat is a main grain crop in both conventional and organic farms, and this organic wheat accounts for one of the most important organic export revenue streams (AAFC, 2010). Thus, large-scale organic production systems are the norm for organically produced grain in Canada (Nelson et al., 2004). Accordingly,

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A version of this chapter has been submitted for publication. Hiroshi Kubota, Sylvie A. Quideau, Pierre J. Hucl. and Dean M. Spaner. Weeds promote greater arbuscular mycorrhizal fungi benefit in organically managed spring wheat (Triticum aestivum L.) cultivation system. Agronomy Journal.

Canadian organic farmers have been challenged to overcome organic production constraints of weed pressure, soil nutrient deficits, pest and disease damages and crop/cultivar choiceses on a large scale (Degenhardt et al., 2005; Mason & Spaner, 2006). Weed and soil fertility managements still remain large concerns for organic farmers (Degenhardt et al., 2005).

In recent decades, soil microorganisms have attracted attention as one of many potential methods for the maintenance and optimal utilization of native soil fertility, especially in organic farming systems (Glassop et al., 2005; Gibson et al., 2007; Birkhofer et al., 2008; Cornejo et al., 2008; Smukler et al., 2008; Kahiluoto et al., 2009; Smith & Read, 2009; Barker, 2010; Verbruggen et al., 2010). Arbuscular mycorrhizal fungi (AMF) are among the most common and ubiquitous soil microorganisms existing in almost all soil systems (Smith & Read, 2008). They are able to form symbiotic relationships with a great number of plant species. Their relationship involves carbon cycling through photosynthesis from host plants to AMF, and soil-derived nutrients from AMF to the plants (Smith & Read, 2008; Ryan & Tibbett, 2008). Due to this nutrient exchange function, AMF utilization in agricultural fields has been explored in organic agricultural systems as an alternative of choice to chemical fertilizers (Oehl et al., 2004; Mohammad et al., 2005; Smith & Read, 2008; Smukler et al., 2008; Nelson et al., 2011).

Weeds are one of the biggest constraints in agriculture and cause enormous economic losses (e.g. $300 million loss in wheat during 1985 to 1989) in Canada (Swanton et al., 1993). Despite economic loss through weed pressure, several studies have emphasized the importance of aboveground species diversity (including weeds) in organic agricultural fields. Aboveground species diversity supports functions of ecological services, especially in terms of nutrient cycling by the soil microbial community (Altieri, 1999; Main, 1999; Jordan et al., 2000; Chen et al., 2004). The elimination of weed species that are AMF hosts possibly causes the loss of AMF beneficial effects on crop growth and yield (Feldmann & Boyle, 1999; Kabir & Koide, 2000). This indicates that increasing aboveground biomass and/or species richness, regardless of species, may increase the diversity of soil microbial communities. Positive correlations between organic management systems and soil microbial communities has been reported (Mäder et al., 2002; Birkhofer et al., 2008; Verbruggen et al., 2010; Nelson et al., 2011); however, there are few studies investigating weed-microbial community interactions in organically managed fields. Nelson et al. (2011) reported that weed seed yield and soil microbial diversity were positively correlated, suggesting that high aboveground plant biomass and diversity created enhanced levels of soil microbial diversity.

There is a presumption that organic products are superior in their taste and nutritional value (Aertsens et al., 2009); however, studies pertaining to nutritional trends between organic and conventional products have been inconsistent. Because several agricultural management factors (e.g. fertilization, tillage, crop rotation, organic agricultural practices, soil moisture management and breeding) are intertwined and all affect crop nutritional quality, in addition to geographical and environmental variations (Wang et al., 2008). Knowledge about beneficial microorganism for plant nutrient uptake may help to understand the effect of organic management on grain nutrition.

A better understanding of weed effects on soil microbial communities under organic management systems would give us direction for weed management strategies. Organic producers may find merit in keeping adequate levels of weeds to maintain an agro-ecosystem with greater biodiversity. Conventional agricultural farmers also would have a chance to reduce pesticide use, keeping a better balance between yields and weed pressure if weeds have some positive effects on soil microbial diversity, which may promote plant nutrient uptake. Therefore, the objectives of the present study are to: 1) examine potential differences in soil microbial communities between weed-free and weedy environments under organic management systems over wheat growing seasons, 2) examine the relationship of wheat cultivar performance in weed-free and weedy environments under organic systems and 3) examine the relationship between wheat grain nutrition and microbial diversity in weedy and weed-free environment under organic management systems.

2-2 Materials and Methods

2-2-1 Experimental Site Description

This experiment was carried out during two consecutive years (2010 and 2011) at the University of Alberta research centre (55^{o} 34’ N, 113^{o} 31’ W). The soil at the site is classified as an Orthic Black Chernozem (Alberta Agriculture, Food and Rural Development [AAFRD], 2011). Soil samples were collected by randomized sampling, and initial soil fertility levels were determined by a modified Kelowna extracting solution for nitrogen (N), phosphorus (P) and potassium (K). The fertility levels were adequate for wheat cultivation in both years, with the exception of N levels (Table 2.1). The field has been organically managed since 1999, and compost (consisted of dairy manure, sawdust, wood chips and straw) was applied in the experimental field for five consecutive years starting in 1999 at the rate of 50 to 62 t ha^{-1}. No compost was applied in the field after 2005. The field has been rotationally managed with the sequence of wheat (*Triticum aestivum* L.) - fall rye (*Secale sereale* L.) - triticale (*X Triticosecale* Wittmack) and field pea (*Pisum sativum* L.) mixture until 2009, and the sequence of fall rye - barley silage (*Hordeum vulgare* L.) - wheat from 2010. Plant residues from prior years were ploughed into the field several weeks before seeding. A single tillage operation was performed in the spring to control initial weed emergence prior to seeding and no herbicide was used during the growing seasons (Table 2.2).

2-2-2 Experimental Design

The experiment was seeded as a split-plot design with four replications. Weed treatment (weed-free and weedy) was the main effect and cultivar (thirteen cultivars) was the subplot in the field. Each plot was 4 m by 1.4 m with six rows (23 cm row spacing) in 2010 and five rows wide (23 cm row spacing) in 2011. The plots were seeded by a self-propelled, no-till double-disk plot drill (Fabro Enterprises Ltd., Swift Current, SK, Canada). Thirteen western Canadian wheat cultivars were chosen according to different registered year and physical traits (Table 2.3). Weed-free plots were maintained by hand-weeding at weekly intervals throughout the growing seasons to avoid soil surface disruption, and the hand-weeding was applied until the end of stem extension stage.

2-2-3 Data Collection

Physiological maturity was recorded when 75% of wheat in a plot has lost green colour in the peduncle. Plant height measurement and single tiller counting were conducted when wheat completed stem elongation. Weed samples were collected from 25 cm x 25 cm quadrat in each plot when wheat vegetative growth was completed, and the weed samples were weighed after drying in a dryer. Wheat grain was harvested by a Wintersteiger plot combine in late September (Table 2.2). Harvested grain was stored in a dryer at 40 °C for approximately 48 h before measurement. Yield components (grain yield, test weight, thousand kernel weight) were recorded after cleaning the grain using 2-mm mesh sieves and a grain blower.

Grain nutrition data (Zn, Mn, Cu, K) were determined in the Crop Development Centre at University of Saskatchwan, Saskatoon, SK. A subsample of 80 g of dry grain was taken from each plot. The subsamples were ground and a standard HNO_3 H_2O_2 method was used to obtain grain nutrient concentration (Thavarajah et al., 2007, 2009). CDC Redberry lentil and organically grown hard red spring wheat were used as reference, and micronutrient concentrations were measured using flame atomic absorption spectrometry (AJ ANOVA 300, Lab Synergy, Goshen, NY, USA). Grain protein content (%) was determined using Near-infrared Reflectance (NIR) spectroscopy by SpectraStar RTW system (version 3.8.0, Unity Scientific, Connecticut, USA).

Soil samples for microbial community analysis were taken twice during each growing season. The first soil samples were taken within one week after seeding, and the second sampling was carried out within one week before harvest (Table 2.2). Soil cores were taken from 10 to 20 cm depth using a core sampler from two randomly chosen spots in each plot and mixed in a sterilized plastic bag. Samples were stored at -80 ^{o}C until subsampling. Subsamples were sieved to 4 mm, and approximately three grams of the subsamples were freeze-dried. Soil microbial community structure was characterized by phospholipid fatty acid (PLFA) analysis as described in Hannam at al. (2006). Briefly, fatty acids were extracted from freeze-dried subsamples using methanol, chloroform and citric buffers. Extracts were separated on solid phase extraction columns (Agilent Technologies, Wilmington, DE) to isolate polar lipids. These were methylated using mild alkaline methanolysis to form fatty acid methyl esters and analyzed on a Hewlett Packard 5890 II Plus gas chromatograph with the MIDI peak identification software (MIDI, Inc., Newark, DE).

To express the composition of the soil microbial community, PLFA concentrations were calculated as nmol g^{-1} PLFA per gram of dry soil, using straight-chain saturated lipid 19:0 as the recovery standard. Indicator PLFAs were used to calculate the relative contribution (as % of total microbial biomass) of several soil microbial groups, including gram-positive bacteria, gram-negative bacteria, actinomycetes, AMF, and other fungal groups (Table 2.4). Shannon’s diversity index (Krebs, 1989) was used to describe microbial diversity. Evenness was calculated using Shannon’s diversity index. Species richness was described as the number of unique PLFA s in each sample. Formulas for Shannon’s diversity index and evenness are described in the following equations.

Shannon’s diversity index

$$H' = \sum_{i=1}^{s} - (P_i * \ln P_i)$$

Evenness

$$E = H' / \ln S$$

where H’ is the Shannon’s diversity index, P_i is the fraction of each individual PLFA, E is evenness, and S is the number of PLFAs in a sample. A greater value for H’ indicates a more diverse community. E is representative of equitability of species in a sample. Richness indicates how many different PLFAs are in a sample.

2-2-4 Statistical Analysis

All collected data were analyzed using the PROC MIXED procedure of SAS software (version 9.2, SAS® Institute 2008). The data were analyzed as a split-plot with weed treatment as the main plot and cultivar as the subplot. Weed treatment, cultivars and weed treatment × cultivars were considered as fixed effects, while year and year × weed treatment were considered as random effects. Preliminary analysis of variance (ANOVA) was conducted to detect significant differences of weed treatment, cultivar, and those interactions for agronomic traits, grain nutrients. For PLFA analyses, sampling time was considered as a fixed effect, and ANOVA was computed to detect significant differences of weed treatments and sampling times. Pearson's correlation coefficients were computed to examine correlations among agronomic traits, grain nutrients, weed dry biomass and soil microbial communities.

2-3 Results

2-3-1 Agronomic properties, yield and grain nutrients

In 2010, grain yield averaged 5.12 t ha^{-1} in weed-free plots and 4.29 t ha^{-1} in weedy plots. In 2011, grain yield averaged 4.53 t ha^{-1} in weed-free plots and 3.58 t ha^{-1} in weedy plots (data not shown). The weed-free treatment exhibited higher ($P<0.01$) grain yield and the number of tillers m^{-2} with much lower weed biomass. Total biomass production (grain yield + weed dry biomass) was greater in weedy plots than in weed-free plots ($P<0.01$). Cultivars differed

($P<0.01$) for all wheat agronomic traits (Table 2.5), and weed treatment × cultivar was not significant for all measured traits (Table 2.5). The modern cultivars AC Andrew, AC Superb, AC Harvest, and CDC Go yielded the most grain in both weed-free and weedy treatments, and the historical and older cultivars Garnet, Katepwa and Marquis yielded the least grain in both treatments (Table 2.5).

Weed-free plots exhibited lower Mn ($P<0.05$) in the grain but weed treatments did not alter grain concentration of Zn, Cu, K and grain protein (Table 2.6). Cultivars differed ($P<0.01$) for Zn, Cu, K and grain protein in weedy plots, and differed for Zn and protein ($P<0.01$) and K ($P<0.05$) in weed-free plots. Weed treatment × cultivar interaction was not significant for all measured grain quality traits. The high yielding soft white wheat cultivar AC Andrew had the lowest levels of Zn, Mn, Cu, and grain protein and the highest levels of K in both treatments (Table 2.6).

In the weedy treatment, days to maturity and grain yield were positively correlated; however, plant height and grain yield, and the number of tillers and weed dry biomass were negatively correlated one another. Grain yield and grain protein content were negatively correlated in weed-free treatment; however, there was no correlation between them in the weedy treatment (Table 2.7). Weed dry biomass and grain protein content were positively correlated in weedy and weed-free treatments, but stronger correlations occurred in the weedy treatment. Protein concentration was positively correlated with Mn and Cu and negatively correlated with K in both treatments (Table 2.8). Zn was negatively correlated with grain yields and thousand kernel weight in both treatments; however, Zn was positively correlated with protein only in the weed-free treatment (Table 2.8).

2-3-2 Soil microbial community structure

The weedy treatment exhibited a greater proportion of AMF ($P<0.01$) than the weed-free treatment while other soil microbial groups, species richness, diversity index and evenness were not altered by weed presence (Table 2.9). The proportion of AMF (16:1ω5 and 20:1ω9c), actinomycetes and overall species diversity increased over the growing season, while the proportions of gram-negative and other fungal biomarker PLFAs (18:1ω9c and 18:3ω6c) decreased (Table 2.9). Effects of soil sampling time were significant in gram-negative bacteria, actinomycetes, AMF, other fungi, species diversity and evenness at $P<0.01$ or $P<0.05$ although each microbial group was affected differently.

The treatment × soil sampling time interaction was significant ($P<0.01$) for AMF and species diversity (Table 2.9). When treatments were analyzed separately, Gram-negative, AMF and other fungal groups showed significant differences in sampling time ($P<0.01$) in both treatments (Table 2.9). Microbial diversity was significantly different by soil sampling time in weedy treatments but not in weed-free plots. The diversity increased over the wheat growing season (Table 2.9).

Protein content was positively correlated with gram-negative bacteria and fungal group communities and negatively correlated with gram-positive bacteria in both treatments at two sampling times, while there were positive correlations between grain protein content and AMF at two sampling times in only the weedy treatment (Table 2.10). When data was analyzed without the weed treatments effect, AMF and other fungal groups were positively correlated with weed dry biomass and total biomass production at two sampling times (Table 2.11).

2-4 Discussion

In this study we uncovered four major findings: 1) the proportion of AMF increased over the growing season and increased significantly in weedy plots, 2) this greater proportion was associated with greater total biomass accumulation but lowered grain yield in weedy plots, 3) individual wheat cultivars did not alter soil biological properties in either weed-free or weedy treatments, and 4) the high yielding semi-dwarf and later maturing cultivars were appeared to be yield stable in organic systems.

2-4-1 Response of agronomic trait and grain yield with respect to weed presence

Grain yield and the number of tillers were greater in weed-free treatments, while weeds did not alter days to maturity or plant height. Several studies have suggested that modern wheat cultivars selected in high-input conventional cultivation systems exhibit high yield potential in optimal environments and lower potential when grown under stressed conditions (Ceccarelli, 1996; Guarda et al., 2004). Most of modern wheat cultivars grown in the present study were high yielding in both weeded and weed-free treatments, and the least four yielding cultivars (Red Bobs, Marquis, Katepwa and Garnet) were all historical or older cultivars. CDC Go and AC Andrew were the highest yielding cultivars in both treatments, and these two cultivars were the two shortest cultivars (semi-dwarf) in this study. Under the natural weed competition, weed stress was not equal, and wheat competitive traits altered according to extent and variability of weed stress (Mason et al., 2008). In fact, yield and plant height were negatively correlated in weedy plots (r=-0.51). Many studies reported that taller plant height was a greater weed competitive ability resulting higher yield than semi-dwarf cultivars in spring and winter wheat (Hucl, 1998; Korres & Froud-Williams, 2002; Yenish & Young, 2004; Zerner, et al., 2008). Such studies may not be comparable to the present study because they used cultivated oats (*Avena sativa* L.) and jointed grass (*Aegilops cylindrical*) to evaluate wheat competitive ability against weeds. They were unnatural and single competitors in those studies, while indigenous weed groups (i.e. Canadian thistle, Shepherd purse, Wild buckwheat, Field penny-cress and Lambsquarter) were competitors for wheat in our study. It is conceivable that competitive traits in wheat may be different according to wheat sultivars and/or weed species, which differ in spatial and nutritional requirements. Semi-dwarf cultivars yielded relatively greater than taller cultivars in both treatments in the present study. This may suggest that some factors other than plant height (e.g. flag leaf area index, length of each growth stages) along with the influence of microbes affected wheat competitive ability. In addition, both semi-dwarf cultivars had relatively longer days to maturity and greater thousand kernel weight. Knott and Talukdar (1971) suggested that thousand kernel weight was an important yield component rather than the number of kernels to determine grain yield; however, grain weight and grain number were always negatively correlated each other (Reynolds et al., 1996). This means if one yield component increased, some other components decreased. Early maturity is also an important trait to overcome weed stress and achieve more grain yield in northern organic wheat cropping systems (Manson et al., 2007). However, in the present study, days to maturity showed a positive correlation to grain yield and thousand kernel weight in weedy treatment. This reflects the genetic potential of the higher yielding modern cultivars with late-maturing and semidwarf traits. It may also be due to photosynthesis and nutritional repletion over longer growing periods and adequate levels of soil nutrients. It has been documented that wheat yield is sink-limited in post- anthesis period especially by the number of grains rather than grain weight (Acreche et al., 2008).

2-4-2 Response of microbial communities to aboveground vegetation

Our original hypothesis was that the weedy treatment facilitated greater microbial diversity than the weed-free treatment. Zornoza et al. (2009) reported that abandoned agricultural lands had a high rate of AMF. In addition, organically managed soil was likely to be more similar to perennial grasslands soil than conventionally managed soil in terms of AMF diversity (Verbruggen et al., 2010). Organically managed soils have greater aboveground biodiversity which results a greater proportion of AMF in soils, thereby mitigating against agricultural management and climatic stresses (Wu et al., 2007). In the present study, weeds apparently offered a similar condition as abandoned agricultural environment, which possibly facilitated a greater propostion of AMF in weedy plots than in weed-free plots.

Several studies have examined relationships between aboveground plant diversity and soil microbial communities, and results are not consistent (Porazinska et al., 2003; Carney & Matson, 2005; Hamel et al., 2006; Habekost et al., 2008; Liu et al., 2010). It has been reported that microbial PLFAs and overall microbial biomass are positively correlated to aboveground plant biomass (Hamel et al., 2006; Liu et al., 2010) and plant species diversity (Carney & Matson, 2005). Soil microbial groups have important functions in agro-ecosystems; affecting and altering both abiotic and biotic soil properties as well as soil nutrient cycling. These soil functions change with aboveground growth stages as these soil microbial groups partly rely on root exudates as their energy sources (Habekost et al., 2008). Habekost et al. (2008), suggested that functional weed species such as legumes exert dominant control on nitrogen input, affecting microbial activity and soil organic matter. In their study, mixtures of four plant species, including legumes, resulted in greater proportions of particular soil PLFAs than mixtures of 8 and 16 species with legumes. This may emphasize an importance of soil carbon and nitrogen balances for soil microbial community composition. In the present study, AMF in the weedy treatment exhibited a greater proportion compared to those in the weed-free treatment. We observed 5 main weed species in our experimental plots (Canadian thistle (*Cirsium arvense* L.), Shepherd purse (*Capsella bursa-pastoris* L.), Wild buckwheat (*Polygonum convolvulus* L.), Field penny-cress (*Thlaspi* arvense L.) and Lambsquarter (*Chenopodium berlandieri*) (data not shown), and no legume species was observed. However, these weed species evidently altered both wheat agronomic traits and the proportion of microbial communities by modifying the surrounding system.

To a large extent, a micro-environmental alteration (i.e. soil chemical and physical properties, light reception, moisture and temperature), which is governed by above ground vegetation, may explain changes of microbial communities in a short term (Wardle & Parkinson, 1990). The weedy treatemnt exhibited greater overall plant biomass accumulation, which may have released a greater amount of root exudates into soil (Grayston et al., 2001; Yang & Crowley, 2000; Jones et al., 2004). These exudates may also differ in quality and quantity according to aboveground plant species and their growing stages (Lynch & Whipps, 1990; Jones, 1998; Pinton et al., 2001; Hartmann et al., 2009), and that may attract specialized microbial species and/or groups. Root exudates are dominated by low-molecular weight carbohydrates, amino acids and organic acids, and plants release about 3 to 10% of their photosynthesized carbon into soil according to their species and growth stages (Jones, 1998; Pinton, et al., 2001; Farrar et al., 2003; Hartmann et al., 2009). AMF are obligated to have symbiotic relationships with almost all plants species, and rely on their host plants for most of their carbon (Smith & Read, 2008). Plant root exudates are likely to change composition of AMF as root carbohydrate metabolism changes (Bago et al., 2003). In the present study, the proportion of AMF was greater in the late growing season in both treatments, and the proportion of AMF in the weedy treatment was greater than that in the weed-free treatment. Photosynthetic capacity varies between species; however, plants gradually increase this capacity along with their growth (Parry et al., 2011). As a consequence, root exudates increase, thus likely to attract more AMF. In addition, wheat cultivars with long days to maturity may have an advantage to nurture more AMF than cultivars with short days to maturity by releasing root exudates in a long term.

According to a review by Jones (1998), plants increase their carbon efflux (i.e. root exudation) along with their growth stages in order to alleviate abiotic and biotic stress (e.g. nutrient deficiency, drought, pathogen, soil CO^2 concentration). It is possible that a higher weed level in the weedy treatment stressed all aboveground vegetation to the point where plant roots stimulated to release more exudates in the present study. This may explain the elevated AMF level, resulting in the promotion of the development of colonized AMF (Vierheilig et al., 1998).

Hamel et al. (2006) reported that soil sampling time (i.e. temporal variation) had stronger influence on soil microbial composition than soil amendments. Other studies have also reported similar trends of seasonal variations on soil microbial composition (Bossio et al., 1998; Bardgett et al., 1999; Schutter et al., 2001; Petersen et al., 2002; Spedding et al., 2004; Habekost et al., 2008). A temporal change in microbial communities during a plant growing season may indicate that transitions of active microbial groups occur along with aboveground vegetative growth (Bardgett et al., 1999; Habekost et al., 2008). In the present study, all microbial groups except the gram-positive bacteria underwent significant seasonal changes. Increases in the relative proportions of actinomycetes and AMF, and species diversity and evenness were observed in the late growing season. Some studies have reported that an early plant growing season had a relatively low amount of PLFAs, microbial biomass carbon and basal respiration compared to an end of the growing season (Bardgett et al., 1999; Habekost et al., 2008); however, specific microbial groups were not investigated in these studies. Spedding et al. (2004) and Habekost et al. (2008) observed higher fungal abundance in fall. This trend was explained by Kabir et al. (1997), who stated that the seasonal establishment of symbiotic microbes was largely affected by their host plant growth since many of these microbes rely on their host plants for their energy. However, reports of seasonal variations in soil microbial dynamic are somewhat inconsistent among previously published studies. Some studies reported that a summer season had higher total microbial activity (Griffiths et al., 2003; Hamel et al., 2006) and total microbial biomass (Bardgett et al., 1999; Griffiths et al., 2003). Other studies reported high active microbial abundance in spring (Bardgett et al., 1999; Grayston et al., 2001) or in fall (Spedding et al., 2004). Jangid et al. (2008) reported less impact of seasonal influence on microbial communities than that of fertilizer application. This inconsistency in temporal influence on microbial communities may be caused by variations of soil temperature and moisture, aboveground species, available soil nutrition, soil texture, pore-size distribution, cultivation methods and histories as well as different methods in soil microbial analysis.

No difference in response of soil microbial communities to wheat cultivars in the presence study suggested that there was little or no relationship between specific soil microorganisms and wheat cultivars. Previous studies suggested that the gradient of microbial affinity partly depended on wheat genetic variation, which have been selected through the history of wheat breeding (Hetrick et al., 1992, 1993, 1995); evidently the cultivars used in this study did not exhibit a great deal of variation.

2-4-3 Influence of weed presence on wheat grain quality

For Canadian wheat farmers, high protein concentration in wheat grains is critical due to its direct impact on price. Wheat grain protein concentration is affected by both genotype and environmental conditions (Graybosch et al., 1995; Mason & Madin, 1996; Georget et al., 2008; Hristov et al., 2010). In the present study, AMF were positively correlated with grain protein contents, but only in the weedy plots. The correlation (*r*=0.46 and *r*=0.27 for the first and second soil sampling times, respectively) between AMF and grain protein concentration in weedy plots may indicate some functions of AMF in wheat protein accumulation relating to weeds presence. A few studies have reported AMF effects resulting in increased protein content in soybean (Bethlefalvay et al., 1997) and wheat (Roesti et al., 2006). This mechanism may be explained by mediating functions of AMF on soil carbon and nitrogen cycles. For wheat, post-anthesis nitrogen supplementation results in increased grain protein concentration in winter wheat (Woolfolk et al., 2002; Burhan, 2010). In addition, AMF have a positive role in supplying nitrogen sources to their host plants (He et al., 2003; Smith & Read, 2008). In the rhizosphere, activity of AMF increased turnover of soil organic matter, and thus mineralized immobile nitrogen (Hamel, 2004). Consequently, wheat grains accumulate a greater amount of nitrogen for protein synthesis. In addition, Chen et al. (2004) reported that microbial biomass carbon and nitrogen increased as the number of weed species increased in early growing season, resulting in increased plant aboveground biomass and plant available nitrogen later in the season. In our present study, aboveground total biomass production may have been partly supported by AMF that provided a supplemental source of nitrogen to wheat spikes through AMF channels (Smith & Read, 2008). This microbial benefit could explain the mechanisms through which the increased proportion of AMF communities potentially increased grain protein contents in the weedy plots. Seed protein concentration had a strong positive correlation with Zn in weed-free plots while there was no correlation between them in weedy plots. This may be because the concentration of micronutrients in grain depends on the amount of absorbed nutrients through roots and the amount of relocation from a plant body (Pahlavan-Rad & Pessarakli, 2009). N is easier to be translocated than Zn. In addition, in the present study, greater interspecific competition for Zn between wheat and weed may preponderate over the function of wheat grain as the sink for Zn in weedy plots (Cakmak et al., 2004).

2-5 Conclusions

Understanding the structure of soil microbial communities in organic management systems is important to develop more efficient and sustainable agricultural management systems. In the present study, the proportion of AMF increased over the growing season, and this increase was significantly greater in the weedy plots than in the weed-free plots. This greater proportion was associated with greater total plant biomass accumulation but lowered grain yield in weedy plots. Individual wheat cultivars did not alter soil microbial properties in either weed-free or weedy treatments. The high yielding semi-dwarf and later maturing cultivars were appeared to be yield stable in organic systems.

Choosing late maturing semi-dwarf cultivars may yield better accompanied with optimal benefit through microbial communities under organically managed systems. Further investigation on effects of both temporal fluctuation and aboveground vegetation including weed species on microbial community is needed to improve organic wheat yield and grain quality.

2-6 Tables

Table 2.1. Soil properties for 0-20 cm depth soil samples at the organic sites in 2010 and 2011.

		Organic	
	Recommendation†	**2010**	**2011**
NO_3-N (kg/ha)	190	76	25
P (kg/ha)	67	134	64
K (kg/ha)	146	1244	539
SO_4-S (kg/ha)	22	46	16
Zn (ppm)	0.34	9.5	9.7
Mn (ppm)	0.56	11.2	10.2
Cu (ppm)	0.22	1	1
Fe (ppm)	0.78	222	222
B (ppm)	0.09	1.8	1.9
pH	6.0-7.0	6.7	6.4
EC (dS/m)	< 1.5	0.7	0.35
OM (%)	>3-4	10.8	12.6

†Recommendation rates were based on Government of Alberta (2003).

Table2.2. Seeding, soil sampling and harvesting dates in 2010 and 2011.

	Treatment	**Seeding**	**Soil Sampling**†		**Harvesting**
			1st	**2nd**	
2010	**Weed-free**	18-May	25-May	22-Sep	24-Sep
	Weedy	18-May	25-May	22-Sep	24-Sep
2011	**Weed-free**	10-Jun	13-Jun	26-Sep	28-Sep
	Weedy	10-Jun	13-Jun	26-Sep	28-Sep

† Soil samples were taken twice each year (1st collection: within one week before seeding, 2nd collection: within one week prior to harvest).

Table 2.3. Description of thirteen Canadian spring wheat (*Triticum aestivum* L.) cultivars used in the study in 2010 and 2011 in Edmonton, AB¶.

Caltivar	Class†	Registered year	Country of origin	Height	Matulity (+/- AC Barrie)
Marquis	historical	1910	Agriculture Canada, Ottawa, ON	113	+2
Garnet	CWRS	1925	Agriculture Canada, Ottawa, ON	104	-6
Red Bobs	historical	1926	University of Alberta	105	-3
Park	CWRS	1963	Agriculture Canada, Lacombe, AB	99	-3
Sinton	historical	1975	Agriculture Canada, Regina and Swift Current, SK	102	-2
Katepwa	CWRS	1981	Agriculture Canada, Winnipeg, MB	97	-3
CDC Teal	CWRS	1991	Crop Development Centre, Saskatoon, SK	96	-2
AC Barrie	CWRS	1994	Agriculture Canada, Swift Current, SK	91	101
AC Superb	CWRS	2000	Agriculture Canada, Winnipeg, MB	85	+2
AC Harvest	CWRS	2003	Agriculture, Canada, Winnipeg, MB	83	-1
CDC Go	CWRS	2004	University of Saskatchewan	83	-
CDC Kernen	CWRS	2009	University of Saskatchewan	92	+1
AC Andrew	CWSWS	2004‡	Agriculture Canada, Lethbridge, AB	92	+11

† CWRS: Canadian Western Hard Red Spring, CWSWS: Canadian Western Soft White Spring (Canadian Grain Commission, 2011).

‡ full registration.

¶ Information on this Table was taken from Mason et al. (2007), Government of Alberta (2012) and Government of Alberta, Agriculture and Rural Development (2012).

Table 2.4. Biomarker fatty acids of selected microbial groups.

Table 5. Biomarker fatty acids of selected microbial groups

group		Biomarker
Bacteria	Gram-positive†	i14:0, i15:0, a15:0, i16:0, i17:0, a17:0
	Gram-negative‡	16:1ω7c, 16:1ω9c, 18:1ω7c, cy17:0, cy19:0
	Actinomycete§	10Me16:0, 10Me18:0, 10Me19:0
Fungi	AMF¶	16:1ω5c, 20:1ω9c
	Other fungal groups#	18:1ω9c, 18:3ω6c

†Hedrick et al. (2005).
‡ modified from Macdonald et al. (2004).
§ Zelles, (1999).
¶ Olsson, (1999) and Sakamoto et al. (2004).
# Frostegård & Bååth (1996) and Hamman et al. (2007).

Table 2.5. Least square means and statistical analyses of agronomic traits for thirteen Canadian spring wheat cultivars grown in organically managed weed-free and weedy conditions in Edmonton, AB in 2010 and 2011.

	Grain yield	**Number of tillers/m^2**	**Days to maturity**	**Height**	**Weed dry biomass**	**Total biomass production**
	t ha^{-1}	(tillers)	d	cm	(dw)kg ha^{-1}	t ha^{-1}
Treatment (T)						
weed-free	4.79	665	102	101	7	4.8
weedy	3.94	533	103	107	2013	5.86
F-test	**	*	ns	ns	**	**
SE$_{treatment}$‡	0.32	11.07	2.68	10.61	0.11	0.35
F-test$_{cultivar}$	**	**	**	**	ns	**
SE$_{cultivar}$	0.37	23.77	2.5	10.41	0.27	0.46
F-test$_{TxC}$	ns†	ns	ns	ns	ns	ns
			Weed-free			
Marquis	4.01	666	108	118	7	4.01
Garnet	3.87	652	88	106	0	3.87
Red Bobs	4.48	596	97	109	8	4.49
Park	4.52	713	95	101	4	4.52
Sinton	4.52	611	108	111	12	4.53
Katepwa	3.95	662	100	104	5	3.96
CDC Teal	5.07	670	101	100	4	5.08
AC Barrie	4.69	755	102	103	0	4.79
AC Superb	5.43	751	109	94	2	5.43
AC Harvest	5.25	750	92	91	6	5.26
CDC Go	5.24	631	106	86	28	5.27
CDC Kernen	5.03	623	108	107	21	5.06
AC Andrew	5.98	565	114	91	0	5.98
F-test	**	**	**	**	ns	**
SE	0.25	10.6	3.64	12.98	4.4	0.25
			Weedy			
Marquis	3.15	507	104	123	2079	4.89
Garnet	3.03	547	89	112	1847	4.87
Red Bobs	3.19	463	97	115	2200	5.15
Park	3.36	525	95	106	1955	5.32
Sinton	3.99	469	111	115	2103	6.10
Katepwa	3.05	529	100	111	2893	5.59
CDC Teal	3.98	544	103	111	3506	7.49
AC Barrie	4.16	584	100	105	1600	5.76
AC Superb	4.61	598	111	97	1619	6.23
AC Harvest	4.35	586	96	98	1635	5.98
CDC Go	4.76	567	106	91	1538	6.30
CDC Kernen	4.61	544	109	110	1050	5.45
AC Andrew	4.99	470	120	97	2137	7.13
F-test	**	*	**	**	ns	**
SE	0.36	11.5	1.07	7.53	149.6	0.41

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

† Non significant at the 0.1 probability level

‡ Standard Error.

Table 2.6. Least square means and statistical analyses of nutritional quantity for thirteen Canadian spring wheat cultivars grown in organically managed weed-free and weedy conditions in Edmonton, AB in 2010 and 2011.

	Zn	**Mn**	**Cu**	**K**	**Protein**
	ppm	ppm	ppm	ppm	%
Treatment (T)					
weed-free	40.7	31.9	4.7	3629.1	12.2
weedy	36.8	34.1	5.1	3671.3	11.5
F-test	ns[†]	*	ns	ns	ns
$\mathrm{SE}_{\mathrm{treatment}}$[‡]	2.62	3.73	0.12	119.6	0.85
$\text{F-test}_{\mathrm{cultivar}}$	**	ns	**	**	**
$\mathrm{SE}_{\mathrm{cultivar}}$	2.36	4.13	0.39	118.3	0.87
$\text{F-test}_{\mathrm{TxC}}$	ns	ns	ns	ns	ns
			Weed-free		
Marquis	45.7	31.4	4.2	3416.6	13.3
Garnet	48.2	33.0	5.6	3475.0	13.2
Red Bobs	40.7	31.2	4.6	3549.3	12.1
Park	38.5	31.7	4.2	3472.5	12.8
Sinton	39.8	31.6	4.2	3501.0	11.9
Katepwa	43.7	33.8	4.7	3501.0	12.4
CDC Teal	39.5	33.1	4.1	3666.5	12.7
AC Barrie	44.6	35.4	6.2	3584.9	12.3
AC Superb	38.6	37.3	4.1	3844.0	11.6
AC Harvest	42.1	29.8	5.0	3764.9	11.9
CDC Go	36.5	30.2	5.9	3782.4	12.0
CDC Kernen	40.7	31.7	5.1	3601.0	12.4
AC Andrew	31.1	25.1	3.4	4019.4	9.5
F-test	**	ns	ns	*	**
SE	3.02	3.29	0.17	58	0.92
			Weedy		
Marquis	39.8	35.2	5.8	3576.1	11.9
Garnet	39.3	33.3	5.7	3736.4	12.3
Red Bobs	38.7	30.8	4.6	3579.6	11.4
Park	36.3	34.4	5.2	3559.3	11.8
Sinton	35.0	36.0	4.8	3595.9	11.3
Katepwa	40.4	37.1	5.0	3518.5	11.7
CDC Teal	35.7	38.0	5.1	3646.1	11.9
AC Barrie	39.6	34.1	5.8	3309.6	11.8
AC Superb	34.8	37.2	5.0	3951.0	10.8
AC Harvest	38.9	33.2	5.1	3759.6	11.4
CDC Go	31.5	32.0	5.0	3771.9	11.5
CDC Kernen	36.6	32.8	6.4	3658.0	12.0
AC Andrew	31.5	29.3	3.1	4064.9	9.3
F-test	**	ns	**	**	**
SE	2.15	4.11	0.139	159.11	0.79

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

† Non significant at the 0.1 probability level.

‡ Standard Error.

Table 2.7. Pearson correlation coefficients between plant agronomic traits, weed biomass and grain protein content for thirteen Canadian spring wheat cultivars grown in Edmonton, AB in 2010 and 2011. Only parameters with significant correlations at P<0.05 are presented.

		Weedy								
		Days to maturity	**Height**	**Number of tillers**	**Kernel weight**	**Grain yield**	**Test weight**	**Weed dry biomass**	**Total biomass production**	**protein**
Weed-free	**Days to maturity**				0.51**	0.51**	-0.40**		0.47**	
	Height	0.24*			-0.44**	-0.51**			-0.42**	
	Number of tillers						0.38**	-0.26*		
	Kernel weight	0.32**	-0.42**			0.77**			0.48**	-0.25*
	Grain yield		-0.71**		0.74**				0.60**	
	Test weight	-0.51**		0.32**					-0.25*	
	Weed dry biomass	-0.24*	-0.40**						0.74**	0.35**
	Total biomass production		-0.72**		0.74**	0.99**				
	Protein	-0.38**				-0.29**		0.24*	-0.29*	

**r* values significant at the 0.1 probability level, ***r* values significant at the 0.05 probability level.

Table 2.8. Pearson correlation coefficients between plant agronomic traits, weed biomass and grain nutrient measurements for thirteen Canadian spring wheat cultivars grown in Edmonton, AB in 2010 and 2011. Parameters with significant correlations at P<0.05 are presented.

	Zn		**Mn**		**Cu**		**K**	
	Weed-free	**Weedy**	**Weed-free**	**Weedy**	**Weed-free**	**Weedy**	**Weed-free**	**Weedy**
Days to maturity	-0.25*	-0.43**			-0.26*			
Height		0.41**			-0.23*		-0.50**	
Number of tillers		0.34**		-0.24*				
Kernel weight	-0.38**	-0.46**					0.34**	
Grain yield	-0.48**	-0.46**					0.48**	
Test weight				-0.25*	0.23*			
Weed dry biomass								
Total biomass production	-0.48**	-0.43**					0.48**	
Protein	0.55**		0.52**	0.27*	0.32**	0.24*	-0.44**	-0.69**

*_r_ values significant at the 0.1 probability level, **_r_ values significant at the 0.05 probability level.

Table 2.9. Least square means and statistical analyses for PLFA in organically managed weed-free and weedy environment in Edmonton, AB in 2010 and 2011.

	Gram positive	Gram negative	Actinomycetes	AMF	Other fungal groups	Diversity	Evenness
	%	%	%	%	%	-	-
Treatment (T)							
Weed-free	25.7	12.3	12.2	3.0	7.7	3.08	0.83
weedy	25.5	12.6	12.0	3.4	7.9	3.08	0.83
F-test$_{\text{treatment}}$	ns[†]	ns	ns	**	ns	ns	ns
SE$_{\text{treatment}}$[‡]	2	0.9	0.39	0.38	0.9	0.0117	0.0036
Sampling time (ST)							
1st	25.6	12.9	11.9	2.9	8.0	3.07	0.82
2nd	25.6	11.9	12.2	3.5	7.5	3.09	0.83
F-test$_{\text{sampling time}}$	ns	**	**	**	**	**	*
F-test$_{\text{cultivar}}$	ns	ns	ns	ns	ns	ns	ns
F-test$_{\text{TxC}}$	ns	ns	ns	ns	ns	ns	ns
F-test$_{\text{TxST}}$	ns	ns	ns	**	ns	**	ns
				Weed-free			
F-test$_{\text{cultivar}}$	ns	ns	ns	ns	ns	ns	ns
SE$_{\text{cultivar}}$	2.2	0.8	0.2	0.4	0.9	0.015	0.004
ST1	25.6	12.8	12.0	2.8	7.9	3.08	0.82
ST2	25.8	11.9	12.3	3.1	7.5	3.09	0.83
F-test$_{\text{sampling time}}$	ns	**	ns	**	**	ns	ns
SE$_{\text{sampling time}}$	2.2	0.8	0.3	0.4	0.9	0.012	0.001
F-test$_{\text{CxST}}$	ns	ns	ns	ns	ns	ns	ns
				Weedy			
F-test$_{\text{cultivar}}$	ns	ns	ns	ns	ns	ns	ns
SE$_{\text{cultivar}}$	1.9	1.0	0.6	0.4	1.0	0.015	0.006
ST1	25.6	13.0	11.8	2.9	8.2	3.07	0.82
ST2	25.5	12.0	12.2	3.9	7.6	3.09	0.83
F-test$_{\text{sampling time}}$	ns	**	ns	**	**	**	ns
SE$_{\text{sampling time}}$	1.8	1.0	0.5	0.3	0.9	0.011	0.005
F-test$_{\text{CxST}}$	ns	ns	ns	ns	ns	ns	ns

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

[†] Non significant at the 0.1 probability level.

[‡] Standard Error.

Table 2.10. Pearson correlation coefficients between plant agronomic traits, yield, weed dry biomass and PLFA measurements by treatments for thirteen Canadian spring wheat cultivars grown in Edmonton, AB in 2010 and 2011.Parameters with significant correlations at P<0.05 are presented.

	Zn		**Mn**		**Cu**		**K**	
	Weed-free	**Weedy**	**Weed-free**	**Weedy**	**Weed-free**	**Weedy**	**Weed-free**	**Weedy**
Days to maturity	-0.25*	-0.43**			-0.26*			
Height		0.41**			-0.23*		-0.50**	
Number of tillers		0.34**		-0.24*				
Kernel weight	-0.38**	-0.46**					0.34**	
Grain yield	-0.48**	-0.46**					0.48**	
Test weight				-0.25*	0.23*			
Weed dry biomass								
Total biomass production	-0.48**	-0.43**					0.48**	
Protein	0.55**		0.52**	0.27*	0.32**	0.24*	-0.44**	-0.69**

**r* values significant at the 0.1 probability level, ***r* values significant at the 0.05 probability level.

Table 2.11. Pearson correlation coefficients between plant agronomic traits, yield, weed dry biomass and PLFA measurements for thirteen Canadian spring wheat cultivars grown in Edmonton, AB in 2010 and 2011 Parameters with significant correlations at P<0.05 are presented.

	Sampling time	Grain yield	Weed dry biomass	Total biomass production	Protein
Gram positive	1st	-0.27**	-0.17*	-0.34**	-0.41**
	2nd	-0.25**	-0.22**	-0.35**	-0.46**
Gram negative	1st		0.26**	0.23**	0.41**
	2nd	0.23**	0.18*	0.29**	0.39**
Actinomycetes	1st		-0.26**		-0.29**
	2nd				
AMF	1st		0.21**	0.21**	0.30**
	2nd		0.43**	0.31**	
Other fungal groups	1st	0.17*	0.25**	0.29**	0.40**
	2nd		0.21**	0.26**	0.34**
Diversity	1st		-0.19*		-0.18*
	2nd	-0.21**			-0.21**

*r values significant at the 0.1 probability level, **r values significant at the 0.05 probability level.

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3-0 General Discussion and Conclusions

Organic farmers need to have different methods to overcome weed pressure, soil nutrient deficits, pest and disease damages and crop/cultivar choices from conventional methods. Weed pressure and soil nutrient deficits are two major problems; however, they seem to be related in the function of soil microorganisms. Soil microbes play a significant role in soil nutritional cycling providing nutrients to aboveground plants.

Diverse aboveground vegetation facilitates higher compositions of beneficial soil microbes, which may provide more nutrients to the plants.

Understanding the relationship between weeds and soil microbial communities for target crops may help us find new approaches for weed management techniques in organic agriculture.

The objectives of our study are:

1, To examine potential differences in soil microbial communities between weed-free and weedy environments under organic management systems over wheat growing seasons.

2, To examine the relationship of wheat cultivar performance in weed-free and weedy environments under organic systems.

3, To examine the relationship between wheat grain nutrition and microbial community in weedy and weed-free environment under organic management systems.

The following are summaries of our study explaining these objectives:

- Soil microbial community compositions were affected by both temporal variability and weed presence. Specifically, the proportion of AMF increased greater in the weedy treatment than in the weed-free treatment over the wheat growing seasons.
- Late maturing semi-dwarf wheat cultivars were stable in yield in both weedy and weed-free organically managed systems; however, this result may be different according to weed species and wheat cultivars.
- There was positive correlation between grain protein concentration and AMF in only the weedy treatment; however, the rate of protein concentration in wheat grains was higher in weed-free treatment.

3-1 Recommendation for Future Research

Microbial community varied in both weedy and weed-free environments through the wheat growing seasons, and weedy plots appeared to have stronger influence on AMF communities. Weed presence may directly or indirectly affect the beneficial functions of AMF on aboveground vegetation. We drew broad conclusions of the possible benefit of weed presence in organic management systems associated with soil microbial functions. Although weeds are considered as one of the main constraints, it may be beneficial to explore a new approach for the maximum outcome by coexistence of weed managements and soil microbial functions under organic management systems. In addition, choice of late maturing semi-dwarf wheat cultivars may give us greater advantages of soil microbial benefit than other wheat cultivars.

This study may provide a base on which future studies are able to draw practical conclusions regarding how we should manage weeds and draw maximum benefit from soil microbial communities under organic management systems. Further investigation on effects of temporal fluctuation and aboveground vegetation including weed species on microbial community is needed to improve organic wheat yield and grain quality.