ENTOMOPATHOGENIC NEMATODE (EPN) PREVALENCE AND DIVERSITY IN ORGANIC AND CONVENTIONAL BEEF AND WHEAT PRODUCTION SYSTEMS AND ACROSS A STATE WIDE PRECIPITATION GRADIENT IN OKLAHOMA

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Bachelor of Science in Entomology

University of California at Riverside

Riverside, CA

2009

Submitted to the Faculty of the
Graduate College of the
Oklahoma State University
in partial fulfillment of
the requirements for
the Degree of
MASTER OF SCIENCE
December, 2012

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ACKNOWLEDGMENTS

I would first like to thank my family and friends for their continuing support over the last 2 ½ years. I wouldn't be graduating today without my major advisor Dr. Carmen Greenwood. Without her expertise and continual assistance, this project would not have been possible. I would also like to thank my committee members Dr. Kris Giles, Dr. Justin Talley, and Dr. Matt Bolek. I am grateful to everyone that helped me in the field: Xandra Robideau, Mackenzie Jochim, John Baggerly, Colby Gregg, Alli Giguere, Sunny Evans, Stephen Zhang, Ramesh Gunaratna, and Sharon Andreason. I appreciate the land owners and managers allowing me to sample with laboriously maintained fields. I would also like to thank Dr. DeSilva, and his lab tech Xiaoping Guo, and the Adams lab at BYU for their assistance with the molecular identification of my EPN. Finally I would like to thank Dr. Niblack and the Department of Entomology and Plant Pathology for funding my project.

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CHAPTER I

REVIEW OF LITERATURE

Biology and Life history

Entomopathogenic nematodes (EPN) are soil dwelling, obligate parasites of insects. Much like other parasitoids and predators, EPN have chemoreceptors and are motile. Like many pathogens utilized as biological control agents, they are highly virulent, easily cultured in vitro, have a broad host range, are arthropod specific, and have a high reproductive potential (H. K. Kaya & Gaugler, 1993). EPN are classified in two families: Heterorhabditidae and Steinernematidae. Each family is associated with a genus of gram-negative bacteria, *Photorhabdus spp*. and *Xenorhabdus spp*., respectively (H. K. Kaya & Gaugler, 1993). The bacteria are symbiotic and essential for EPN to complete their life cycle. EPN are ideal biological control agents for background suppression of soil-dwelling insect pests (Gaugler *et al.*, 1989). The Steinernematidae have two recognized genera and over forty species. The Heterorhabditidae contains only a single genus that currently has ten recognized species (Adams *et al.*, 2006a). The global distribution of these families is almost ubiquitous. Both families have been found on every continent except Antarctica, with North America having at least 20 confirmed species (Adams *et al.*, 2006a). Heterorhabditidae has been isolated primarily from sandy coastal soils, with some being found in porous and more calcareous soils inland. Steinernematidae are widely distributed in turf, weedy, and forest habitats (Adams *et al.*, 2006a).

The infective stage that actively moves through the soil is the Infective Juvenile (IJ) life stage. Once an IJ locates a host, it enters the hoemocoel of the insect using chemosensory cues. This can be accomplished through a natural opening such as the mouth, anus, spiracle, or, in some species, by entering directly through the cuticle. The IJ will then move into the hoemocoel by penetrating the midgut or the tracheal wall (H. K. Kaya & Gaugler, 1993). Some Heterorhabditidae IJs have a dorsal tooth that assist in direct penetration of the cuticle (H. K. Kaya & Gaugler, 1993). Once inside the insect host, the IJ regurgitates or defecates the symbiotic bacteria stored in its gut into the hoemocoel of its host. The bacteria then multiply rapidly, killing the host through septicemia within 48 hours. The EPN reproduce inside the insect cadaver for a few generations (2-3) until a new batch of IJ burst through the cuticle of the cadaver and begin to disperse back through the environment looking for a new host (H. K. Kaya & Gaugler, 1993).

EPN and their hosts are unable to develop a highly adapted host-parasite relationship due to rapid death of the host after EPN infection. This rapid host death allows EPN to exploit a wide host range that spans nearly all insect orders (H. K. Kaya & Gaugler, 1993). However, some host insects are not completely susceptible to EPN infection. Unrestrained *Popillia japonica* are capable of removing more than 60% of attacking nematodes from their cuticle by brushing with their legs or the abrasive raster on their abdomen (Gaugler *et al.*, 1994).

Many biotic and abiotic factors affect EPN in the soil. Abiotic factors that can affect EPN persistence in the environment include temperature, soil moisture, soil type, soil pH, and UV exposure (Kung *et al.*, 1990; Shang-Ping *et al.*, 1990; Grewal *et al.*, 2002; Karagoz *et al.*, 2009). Common biotic factors that can impact native EPN in the environment include predators, parasites, and pathogens of EPN (Stuart *et al.*, 2006).

While EPN can provide suppression of some agricultural pests in Oklahoma, they are susceptible to predation, antibiosis, and fungal parasitism in those same environments. EPN are preyed upon by many species of both mites and collembolans (C. Marie Greenwood *et al.*, 2011). Predators thrive in environments that promote EPN persistence as well. Antibiosis can occur when plants release volatiles from their roots into the soil, adversely impacting the IJ's ability to host seek in the immediate area (Harry K. Kaya & Koppenhöfer, 1996). This can cause a reduction of EPN persistence do to IJ lack of ability to successfully locate and infect a viable host. EPN can out-compete with most entomopathogenic fungi, with the exception of *Bacillus thuringiensis*. However, EPN can themselves be parasitized by nematophagous fungi. The most studied example of this is *Hirsutella rhossiliensis*, which has been shown to cause higher mortality in Steinernematid species than in Heterorhabditid species (Harry K. Kaya & Koppenhöfer, 1996). The combination of these biotic factors can impose heavy trophic pressure on native EPN populations in wheat and pasture environments.

Climate can have a major influence over the prevalence and community composition of EPN. *Steinernema weiseri* and *Steinernema feliae* are cold adapted and are most virulent between 10-15°C, whereas *Heterorhabditus bacteriophora* is warm-adapted and most virulent between 20-25°C (Karagoz *et al.*, 2009). Each species of EPN have their own thermal niche for infection, establishment, and reproduction that is independent and unaffected by their locality (Grewal *et al.*, 1994). Soil type plays a vital role in EPN persistence, due to the necessity of proper soil porosity for the dispersion of IJs. Sandy loam is the soil that IJs are most effective at dispersing though, followed by sand, clay loam, and finally clay (Kung *et al.*, 1990). These findings correlate with the porosity of the soils, with sandy loam being the most porous while still being able to maintain the moisture layer around particles essential for IJ dispersal. EPN persist well in a soil pH range of 4-8, and when approaching a pH of 10, survival drops dramatically (Shang-Ping *et al.*, 1990).

During periods of adverse conditions many EPN have the ability to molt to a long-lived, non-feeding, survival stage known as the Dauer Juvenile (DJ) (Dolan *et al.*, 2002). *Endotokia matricida* is a process through which the adult female EPN is consumed by its offspring. During embryo development, when the external conditions are unfavorable, *endotokia matricida* can occur within EPN, resulting in death of the adult reproductive and direct development of DJs instead of IJs (Johnigk & Ehlers, 1999).

The average LT_{90} (lethal time) of field collected strains of EPN vary from 6-16 weeks. *H. bacteriophora* has been shown to be capable of withstanding high temperatures up to 40ºC for 2 hours, exposure to 302nm ultra violet light (UV) for 5 minutes, 0% dissolved oxygen for 96 hours (hypoxia), and 25% glycerol (desiccation) for 72 hours. Results between populations isolated from different locations varied wildly. Overall longevity was most closely correlated with heat tolerance. These results show that patchy populations of EPN are highly fragmented suggesting a strong influence of meta-population dynamics (Grewal *et al.*, 2002).

EPN have a broad host range, are motile, have chemoreceptors for host seeking, are highly virulent and have high reproductive potential. These attributes make native EPN in agricultural systems valuable for low to no-cost addition background suppression of pests that have a soil dwelling life stage. However, the true pathogenicity of EPN is a result of the symbiotic bacteria stored in the IJs gut and released into the host's hoemocoel.

Bacteria EPN complex

Mutualistic and insecticidal relationships between nematodes and bacteria evolved on at least two separate occasions, leading to two lineages of Heterorhabditidae and Steinernematidae (Adams *et al.*, 2006b). The bacteria are symbiotic and essential for EPN to complete their life cycle. Each EPN species has a specific association with only one bacterial species, although some bacteria may be associated with more than one EPN species (H. K. Kaya & Gaugler, 1993). As the bacteria multiply, they release secondary metabolites that preserve the integrity of the cadaver's cuticle, repel or kill other bacteria and fungi to prevent breakdown of the cadaver, and turn the cadaver a different color specific to the bacterial species(H. K. Kaya & Gaugler, 1993). Studies show that the symbiotic bacteria of EPN in the genera *Xenorhabdus* and *Photorhabdus* compounds that deters a wide range of predators and scavengers (Zhou *et al.*, 2002). A particular compound has been identified for ant predators, called Ant Deterrent Factor (ADF). ADF protects nematodes from being consumed during reproduction within insect cadavers. The compound is produced by the symbiotic bacteria in culture and is found in the supernatants of the bacteria. It is filterable, heat stable, acid sensitive, and can pass through a 10-kDa-pore-size membrane. This leads researchers to believe that the compound is a small, extra cellular, non-proteinaceous compound (Zhou *et al.*, 2002).

The success of the compound depends upon the ant species, the strain, and age of the bacteria being tested.

Co-infection by two different species of EPN has been documented, but is uncommon. In most cases of co-infection one species of bacteria out-competes the other, resulting in only one species of EPN successfully reproducing and emerging from the cadaver. In one particular study, the frequency of *Steinernema feltiae* was unaffected by the presence of *Heterorhabditis marelatus*, however *H. marelatus* frequency dropped severely with the presence of *S. feltiae* (Gruner *et al.*, 2007). Two steinernematid species have been shown in a laboratory setting to be capable of parasitizing the same host. Members of one genus cannot feed on the symbiotic bacteria associated with a different genus, and commonly one species will be able to out compete the other (H. K. Kaya & Gaugler, 1993). Pathogenic symbiotic bacteria of EPN are essential for success of EPN to provide background suppression of pest insects. Besides the pathogenicity of symbiotic bacteria, another essential trait for EPN success is the host seeking ability of the IJ life stage. Many abiotic and biotic factors influence the success of host seeking by IJs. Land management practices in agricultural settings impact both the abiotic and biotic conditions of the soil environment.

Organic vs. conventional wheat and beef production in the southern great plains

80% of Oklahoma's land is used for agriculture. Beef and wheat production systems are some of the most prevalent, producing over 3 billion dollars a year in combined gross income (Shideler et al., 2011). Oklahoma is the 2nd largest beef cattle producing state in the nation with over 56,000 cattle operations and $2.54 billion in gross income a year. At the same time, Oklahoma is the 4th largest wheat producing state in the nation, with over $584 million in cash receipts. (Shideler et al. 2009).

Oklahoma is unique in that many producers in the central "winter wheat belt" portion of the state use the early stages of wheat growth as a cattle forage (Horn, 2006). For most of the state, highest wheat yields are achieved by planting in the first two weeks in October.. If the goal is to utilize the wheat pasture only for cattle forage then planting can be done any time after August 20th (Cuperus, 2000). Wheat has many pests that spend part of their life cycle in the soil, making them susceptible to EPN infection, including armyworm (Lepidoptera), wireworm (Coleoptera), and cutworms (Lepidoptera) (Cuperus, 2000)

Many species of EPN, including *Steinernema masoodi*, *S. seemae*, *S. carpocapsae*, *S. glaseri*, *S. thermophilum*, , *S. weiseri*, and *Heterorhabditis bacteriophora* have been tested in laboratory settings against multiple pest species of Lepidoptera, which include gram pod borer (*Helicoverpa armigera*), greater wax moth (*Galleria mellonella*), rice moth (*Corcyra cephalonica*), acorn moth (*Cydia splendana*), and Oriental leafworm moth (*Spodoptera litura*) (Ali *et al.*, 2008; Karagoz *et al.*, 2009; Seth *et al.*, 2009). All of the EPN species tested successfully infected and reproduced within the Lepidopteran pests that they were tested against. *S. masoodi*, *S. seemae*, and *S. carpocapsae* were found to be the most pathogenic, killing the rice moth within 24hrs. The Greater wax mothwere the most susceptible and best for rearing large quantities of IJs for the majority of EPN tested (Ali *et al.*, 2008).

Introducing EPN into the wheat field environment can also have positive indirect effects for a producer, such as reducing the number of plant parasitic nematodes in the soil. This is mainly attributed to spatial and environmental displacement of plant parasitic nematodes by EPN (Jagdale *et al.*, 2002). Considering this effect in addition to EPN direct effects, conservation or augmentation of naturally-occurring EPN could provide one component of a comprehensive IPM program designed to promote sustainable wheat production (Koppenhöfer *et al.*, 2000).

Cattle have multiple pests with the potential to be susceptible to EPN control as well, including hornflies (*Haematobia irritans*), cattle grubs (Hypoderma), stable flies (*Stomoxys calcitrans*), black flies (Simuliidae), horse and deer flies (Tabanidae), sand flies (Psychodidae), house flies (*Musca domestica*), and hard ticks (Renn, 1998; de Carvalho *et al.*, 2010).The most economically important pests in Oklahoma beef cattle systems include horn flies, stable flies, house flies and hard ticks; these will be the main focus of the following studies. EPN have been shown to successfully control prominent pests of several systems, including members of Coleoptera, Diptera, and Lepidoptera (Grewal et al., 2001). *S. carpocapsae* alone has been shown to infect more than 250 insect species in over 75 families in 11 different orders (Grewal et al., 2001). EPN have been shown to successfully infect ticks, even engorged females (de Carvalho *et al.*, 2010). To reduce the complexity of so many confounding variables, however, the following studies will focus on wheat production systems not grazed by cattle and pastureland systems used specifically for beef production.

Effects of organic vs. conventional wheat production on EPN

EPN depend upon suitable biotic and abiotic soil conditions in order to infect insects and reproduce successfully. The conditions of the soil between organic and conventional agricultural practices may vary dramatically due to the wide range of land management strategies, which may include application of pesticides, herbicides, fertilizer, and mechanical disturbances, such as tillage and compaction. Common abiotic factors that influence EPN success include soil moisture level, soil type (pore size), temperature, and soil pH. Common biotic factors that can impact native EPN in the environment include predators, parasites, and pathogens of EPN (Stuart *et al.*, 2006).

When comparing organic and conventional practices, the amount of tillage in the systems can be a major factor on the success of nematodes in the local food web, most likely due to the drying of the soil (Briar *et al.*, 2007). It is common practice in conventional agricultural practices in Oklahoma to add anhydrous ammonia to soil in order to increase nitrate levels. This has been shown to inhibit microbiological and biochemical activity in the soil communities (Deng et al., 2005). In contrast, the addition of manure to otherwise clay heavy soils, which is the common practice in organic agriculture, has many benefits for the soil biologic communities (Obi et al., 1994). These benefits include a decrease in bulk density and an increase in structural aggregation and porosity, which allows for better movement and dispersal for the EPN through the environment. Clay soils have very small pores between particles, making dispersal harder for EPN, whereas moist sandy soils, or soil with manure additions, have large pores between particles and are more ideal for EPN dispersal into the environment (H. K. Kaya & Gaugler, 1993). The increase in nematode activity can also be attributed to the increase in food resources associated with the enrichment of the soil by readily degradable compounds (Mahran *et al.*, 2009)**.**

Soil food web indices based on nematode assemblages are a reliable method of predicting trophic composition of functional characteristics of soil mite assemblages. Bacteriophagous and predatory nematodes, together with predatory mites, are more abundant in organic-no till treatments than in conventional- standard till treatments. Conventional-standard tillage treatments have high abundances of fungiphagous and plant-parasitic nematodes and algivorous mites (Sánchez-Moreno *et al.*, 2009). With the promotion of predatory mites and nematodes, organic-no till treatments can increase the predation on EPN, while maintaining abiotic conditions that promote EPN success. conventional-standard tillage does not promote predators or competitors of EPN, thereby reducing those pressures on the local EPN community. However, conventional-standard tillage promotes abiotic factors that are not conducive to EPN persistence.

Many soil-borne insect pests are managed by the application of soil insecticides. Under current federal re-evaluation many soil insecticides will no longer be available for use in many crops. In order to achieve reduced synthetic inputs and sustainable agriculture, growers will need to manage soil organisms to promote nutrient cycling and to suppress pests, and biological and cultural pest management alternatives will be necessary. Naturally occurring EPN have the potential to be an effective force in controlling agricultural soil-dwelling pest species. It will be especially important to understand how production systems and practices affect beneficial and pest organisms. This information can be used to devise ways to exploit soil properties and beneficial soil organisms, such as predators and pathogens of soil-dwelling insect pests, and to enhance agricultural sustainability (Millar & Barbercheck, 2002).

Effect of organic vs. conventional pastureland management on EPN

It is common practice, in conventional pastureland management, to use pesticides as the main source of parasite control for cattle (Svensson *et al.*, 2000). The practice of using Ivermectin as a prophylactic can have a large non-target effect on the local arthropod communities, killing off beetle grubs and fly larvae due to excretion of the pesticide in the feces (Römbke *et al.*, 2010). Fresh manure is breeding substrate for horn flies, whereas older manure serves as breeding substrate for stable flies, both of which are major pests of cattle. The amount of pesticide excreted has been shown not to have a detrimental effect on local soil dwelling nematode populations (Yeates *et al.*, 2007). The treatments may still affect abundance and diversity of EPN due to the lethal effects on their hosts and added stress on the local EPN. Stable habitats, light soils, and an abundance of hosts creates an environment for successful EPN persistence and insect suppression (Mrácek *et al.*, 2005). Due to restrictions against the use of pesticides in organic practices, parasite control is mainly achieved through grazing management, nutritional supplements, and forage. The grazing is managed in such a way as to avoid pastures that were grazed by any herd animals the previous season, this greatly reduces the number of parasites in the grazing environment encountered by cattle (Svensson *et al.*, 2000).

Pesticides and fertilizers can have positive, neutral, or even negative effects on the local EPN populations and must be considered when looking at EPN for successful biological control of a pest (Shapiro-Ilan *et al.*, 2006). Swine effluent (SE) is particularly acidic and high in salinity, which can have a detrimental effect on the soil nematode community. The sodium adsorption ratio of soils under SE treatment at the Goodwell field site increased following five cumulative, annual manure additions, a potential long-term problem for this production system (Turner *et al.*, 2010). Thus, application of SE may be contributing to salinity stress on soil dwelling invertebrates, and a negative trend for microarthropod abundance in treated soils will most likely result if no action is taken to correct this. It is necessary to take land management practices into account when looking to conserve native EPN (Campos-Herrera *et al.*, 2008).

EPN have been shown to successfully infect many Dipteran pests including: midges (Cecidomyiidae), fruit flies (Tephritidae), sand flies (Psychodidae), and house flies (Muscidae) (Secundino *et al.*, 2002; Georgis *et al.*, 2006; Corlay *et al.*, 2007; Malan & Manrakhan, 2009). Not all of these species are of great concern for cattle farmers; however it is easy to assume that if these can all be successfully controlled by EPN then the main pests of cattle, almost all of which are in the family Muscidae, may be as well. Conservation or augmentation of naturally-occurring EPN may serve as one component of a comprehensive IPM plan aimed at reducing populations of blood-feeding flies in livestock production.

EPN abundance and diversity across a geographical precipitation gradient

EPN require a water film over soil particles for successful active dispersion, making soil moisture levels and type important factors in the efficacy of EPN as pest control (H. K. Kaya & Gaugler, 1993). Oklahoma exhibits a distinct precipitation gradient, with west to east rainfall levels ranging from 40cm to 150cm, respectively. Soil moisture has been shown to be one of the most important abiotic factors affecting EPN community composition. EPN are most effective at moderate soil moisture levels (-10 to -100 kPa) and become less successful at infecting hosts in high moisture content soils (> -1kPa) and lower moisture content soils (< -1000 kPa) (Koppenho¨fer & Fuzy 2007).

Oklahoma has a wide range of soil types as well. Clay soils have very small pores between particles, making dispersal harder for EPN; whereas moist sandy soils with large pores between particles are more ideal for EPN dispersal into the environment. (H. K. Kaya & Gaugler, 1993). A state wide survey to sample native EPN diversity must take these factors into consideration.

Seasonal variation must be considered when looking to conserve naturally occurring EPN. Seasonal dynamics of EPN are characterized by an initial explosion of individuals, followed by a steady decrease, and an eventual stabilization (Puza & Mrácek, 2005). EPN abundance has a strong negative correlation with host populations in the enviornment. Intraspecific and interspecific competition for limited resources (hosts) is a major component to EPN abundance in the field (Puza & Mrácek, 2005). When host abundance is low, there is feirce competition between EPN, infection of a host by too many EPN causes complete loss of the host, without complete utilization of the available resources by any of the EPN. When competition is high, EPN populations crash, allowing the host populations to increase with less trophic pressure.

Conservation of naturally occurring EPN, through the use of land use practices that are conducive to EPN proliferation is one way producers can enhance background pest suppression (Carmen M. Greenwood & Rebek, 2009). With the high virulence, broad host range, and massive reproductive potential of EPN, this may be potent pest control option that producers cannot afford to overlook.

EPN communities can be heavily influenced by environmental factors, both biotic and abiotic. Land management practices and precipitation levels can influence environmental factors that are crucial for EPN success and persistence in the environment. The first objective of the following study is to analyze the impact of organic and conventional management regimes on the EPN communities in wheat and cattle pasture agricultural systems in Oklahoma. The second objective of this study is to analyze how a varying precipitation gradient across Oklahoma impacts the EPN communities in conventional cattle pastures across the state. This information will be helpful in understanding and implementing EPN conservation in agricultural systems across Oklahoma.

CHAPTER II

COMPARISON OF EPN PREVALENCE AND DIVERSITY IN ORGANIC AND CONVENTIONAL BEEF AND WHEAT PRODUCTION SYSTEMS IN OKLAHOMA

Introduction

Entomopathogenic Nematodes (EPN) are small soil-dwelling round worms that are obligate pathogens of arthropods. There are two families of EPN, the Steinernematidae and Heterorhabditidae; both of which have a bacterial symbiont (*Xenorhabdus spp.* and *Photorhabdus spp.* respectively) that are found only in the guts of these nematodes (Harry K. Kaya *et al.*, 2006). EPN have one free living life stage called the infective juvenile (IJ) stage. IJs are motile and use chemosensory cues to track down potential hosts in the environment (Gaugler *et al.*, 1989). Once an IJ finds a host, it must find an opening where is can pass into the hoemocoel of the host. This is commonly done by entering the digestive tract through the mouth or anus and then penetrates the midgut wall. IJs are also able to enter spiracles and enter the hoemocoel through the tracheal wall. In some instances, direct penetration of the cuticle is possible by the IJ. Once inside the hoemocoel the IJ regurgitates or defecates the symbiotic bacteria stored in its gut into the hoemocoel of its host. The bacteria begin to break down host tissues, killing the host through septicemia (Campbell *et al.*, 1995). While breaking down the host tissues, the bacteria release many other secondary compounds that act as preservatives, anti-bacterial agents, anti-fungal agents, and ant-deterrent factors (H. K. Kaya & Gaugler, 1993; Zhou *et al.*, 2002). These compounds keep the host cuticle intact and deter scavengers while the EPN complete their life cycle within the cadaver. Once reproduction is complete, a new generation of IJ burst through the cuticle wall of the cadaver and disperse into the environment in search of new hosts (Campos-Herrera *et al.*, 2008).

Agricultural practices are a large draw of arthropods due to large scale, readily available, resources in the form of regular annual inputs of vegetation, water, and soil amendments. This study assessed different land management practices, organic and conventional production, of beef and wheat in Oklahoma. At least 80% of land in Oklahoma is utilized for agricultural production. Oklahoma is the 2nd largest beef producing state in the nation, with over 56,000 cattle operations, and $2.54 billion annually in gross income from beef. Oklahoma is also the 4th largest wheat producing state in the nation, with over 110.2 million bushels produced annually, and $584 million in sales. These two systems make up the majority of the agricultural practices in Oklahoma. EPN have the potential to increase the profit in these agricultural systems by providing valuable background pest suppression. There is only one certified organic beef and wheat producer in Oklahoma located in Fairview OK.

Beef and wheat production, whether organic or conventional, impacts the biotic and abiotic soil environment in many ways. EPN prevalence can be affected by these biotic and abiotic environmental factors. Soil ecosystems' have complex food webs containing predators, pathogens, parasites, and a wide range of host and non-host arthropods of

EPN. Omnivory is common in these ecosystems, many invertebrates in the soil ecosystem have documented records of nematophagy and exhibit attraction to the presence of EPN-infected cadavers (Greenwood *et al*., 2010). Due to these compounding factors, determining the impact of biotic factors on EPN communities is a difficult task (Stuart *et al.*, 2006).

EPN are have biotic factors to contend with in an agricultural environment including predation, parasitism, and antibiosis. Many soil-dwelling microarthropod pradators, omnivors, and scavengers including mites and collembolans feed on EPN IJs while they are dispersing and host seeking in the soil (Greenwood *et al.*, 2011). Entomophagous fungi, including *Hirsutella rhossiliensis*, can provide strong trophic pressure on EPN IJs in the soil (Harry K. Kaya & Koppenhöfer, 1996). Particularly when considering the highly clumped dispersion pattern of EPN IJs in the environment does the true impact of entomophagous fungi become apparent. Antibiosis occurs when plants in the environment release volatiles from their roots into the soil environment, disrupting EPN IJ's ability to host seek near the roots of the plants releasing the volatiles (Harry K. Kaya & Koppenhöfer, 1996). All of these biotic factors can impact native EPN ability to host seek and successfully infect and reproduce within a host.

Conventional agriculture can involve any and all legal compounds on the market to control pests affecting their crop or animals. This includes, but is not limited to: animal waste, nematicides, insecticides, general pesticides, sewage, and synthetic fertilizers. Swine effluent, synthetic fertilizers, and sewage create soil pH levels that are stressful on EPN. EPN persist well in a soil pH range of 4-8, when approaching a pH of 2 or 10 survival drops dramatically (Shang-Ping *et al.*, 1990). Organic producers commonly fertilize with compost and manure, creating a soil environment that is more porous, more capable of retaining moisture, and maintains a pH comfortably within the EPN preferred range (Karungi *et al.*, 2006). Soil moisture is essential for the continual persistence of EPN in an environment, without an intermediate soil moisture (-10 to -100 kPa) IJs can no longer disperse and host seek successfully. Conventional practices in cattle pasture ecosystems consist of treating cattle with ingestible and systemic nematicides. These compounds are excreted in the feces of the cattle and create a high concentration in the soil underneath the dung pat. These excretions have been shown to inhibit the development of common EPN hosts in dung pats, including *Musca domestica* and certain beetle larvae. Thereby reducing the usable hosts and creating a less favorable environment for EPN (Madsen *et al.*, 1990). Therefore, soil sampling in the pastureland portion of this study targeted the soil habitat directly beneath dung pats.

Organic and conventional management practices promote dissimilar soil environments due to differing fertilizers and pesticides allowed in the two management strategies. Wheat and beef production systems are the two largest agricultural systems in Oklahoma, yielding over three billion dollars in annual sales. The objective of this study is to compare the impacts of organic versus conventional management practices in cattle pasture and wheat agricultural systems in Oklahoma on the prevalence and diversity of native species of EPN in those systems. The results of this comparison will advance our understanding of native EPN conservation agricultural systems. Conservation of native EPN has the potential to be a cost effective strategy for background suppression of natural enemies of soil-dwelling insects, many of which are pests in these agricultural systems.

Methods and Materials

Wheat

2 wheat fields with different management practices were sampled. One certified organic field and one neighboring conventionally managed field. Each field was sampled on 3 different sampling dates (9/3/11, 10/17/11, 11/18/11). On each sampling date, four 100m transects were established at each field site. Transects were a minimum of 150m apart and began 10m from the edge (to avoid edge effects). One 50cm^3 soil core was taken every meter using a handheld soil core, for ten meters. At every 10m interval the soil samples were homogenized in a bucket. A 300 cm^3 subsample was taken from the homogenized sample, placed in a 3.8L ziplock freezer bag and labeled with the collector's code, date, transect, and type of field. The samples were then transported back to the lab in coolers to help moderate the temperature in order to avoid soil sterilization on hot days. A total 40 bagged samples were taken at each field on each sampling date. A total of 240 samples were taken between September 3rd 2011 and October 17th 2012.

Pasture

2 cattle pastures with different management practices were sampled on the same dates the wheat fields were sampled (above). One pasture was managed using certified organic techniques and the other, a neighboring pasture, was conventionally managed using best management practices (BMP). Each pasture was sampled on 3 different sampling dates. During each sampling trip samples were taken from: under fresh dung pats (<2 weeks), under old dung pats (>2 weeks), and away from dung pats. Dung pats were selected randomly through visually canvassing the field. Dung pat age was determined judging

moisture content and breakdown of the pat due to environmental exposure. A professor with extensive experience working in cattle pasture taught collectors how to categorize dung pat age by sight and texture (determined by prodding the dung pat with a hand trowel).When the appropriate dung pat was located, a hand trowel, with the first 7.6cm marked off, was used to take a 300cm^3 from the top 15.2cm soil level. These samples were placed in a 3.8L ziplock freezer bag and labeled with the collector's code, date, dung pat condition, and type of field. The samples were then transported back to the lab in coolers to help moderate the temperature. A total of 18 bagged samples were taken at each pasture on each sampling date. A total of 108 samples were taken between September 3rd 2011 and October 17th 2012. Neighboring farms that use conventional beef and wheat production practices were sampled in conjunction with the organic farm to provide paired landscape comparisons.

Bioassay

Each sample was baited with 6 *Galleria mellonella* and incubated at 25°C for 7 days, consistent with standard bioassay technique described by Lacey & Kaya (1997). *G. mellonella* larvae are the standard insect and stage for bioassays for EPN due to their low LD-50 for all species of EPN species tested (Morris *et al.*, 1990). After five days the bags were removed from the dark and dumped into trays in order to find all of the *G. mellonella* larvae. *G. mellonella* were recovered and their disposition determined. Disposition categories included alive, dead-uninfected, infected with EPN, infected with fungus, or missing. Prevalence was defined as the total number of infections divided by the total number of *G. mellonella* larvae used in the bioassay. Infected insects (cadavers) were placed into a Petri dish with an appropriately sized filter paper lining the bottom. The filter paper was then moistened with a spray bottle to keep humidity high during EPN emergence, filter paper was kept moist while making sure no standing water was present. The cultures were then checked every day to check for IJ emergence. Once EPN began to emerge from a *G. mellonella* cadaver, the filter paper was removed from the dish (while keeping the *G. mellonella* on it), and the Petri dish was flipped over. The Filter paper was then placed back on top of the, now upside-down, Petri dish. The dish was then placed into a larger Petri dish which was filled with a thin layer of water (~10ml). The EPN dispersed sufficiently into the water in the large Petri dish after 24-48 hours. Once the EPN had successfully dispersed, the contents of the large Petri dish were poured into 100ml centrifuge tubes and frozen until the DNA extraction was performed. Before freezing, a sample of the IJs collected were measured with an Amscope MU300 camera, using Amscope ToupView software, to determine average IJ length of the species collected. During the entire time that the infected *G. mellonella* cadaver was in contact with filter paper, the paper was kept moist by lightly misting it with a spray bottle whenever it began to dry out.

DNA extraction and sequencing

Single EPN from each infected cadaver were lysed using EPN lysis buffer (unpublished Adams lab protocol). PCRs were then run using the resulting solution as the DNA template. PCR Primers designed to amplify ITS 18s-26s gene region were used. The resulting PCR product was cleaned with exonuclease and SAP and sent to the CORE facility on the OSU campus for sequencing. The CORE facility used "BigDye™"-

terminated reactions analyzed on an ABI Model 3730 DNA Analyzer to sequence the submitted samples. The resulting sequences were trimmed and cleaned by eye using BioEdit software. The cleaned sequences were identified using the BLASTn program and the NCBI database. Species identification was confirmed based on a low E-value and high Max ident.

Statistical Analysis

Pasture

Prevalence was evaluated as total infections/total *G. mellonella* used to bait. Prevalence data based on infection symptoms *G. mellonella* were subjected to statistical analysis using analysis of variance (ANOVA) techniques (PROC MIXED, PC SAS Version 9.2, SAS Institute, 1996), using the RANDOM option in an LSMEANS statement. Experimental factors in the model included: date, management (Organic or Conventional), cow patty condition, and rep with management*cowpatty*rep considered to be random effects. A test for normality was conducted using PROC UNIVARITE procedure to perform a NORMALTEST. The class included were dates, management, cow patty condition, and rep with management considered to be the variance. P-values of 0.05 or less were considered significant.

Wheat

Prevalence was evaluated as total infections/total *G. mellonella* used to bait. Prevalence data based on infection symptoms *G. mellonella* were subjected to statistical analysis using analysis of variance (ANOVA) techniques (PROC MIXED, PC SAS Version 9.2, SAS Institute, 1996), using the RANDOM option in an LSMEANS statement. Experimental factors in the model included: date, management (Organic or

Conventional), transect, subsample, rep, and response with management*transect and management*transect*subsample considered to be random effects. P-values of 0.05 or less were considered significant.

Results

A total of 6 different species of EPN were identified. The average prevalence of EPN in conventional wheat fields was 6.73%, the average prevalence of EPN in organic wheat fields was 2.06% (Fig. 1). The prevalence of *S. feltiae* in organic wheat was 1.27%. The prevalence *S. glaseri* in organic wheat was 0.32%.The prevalence of *S. diapresi* in organic wheat was 0.48%. The prevalence on *S. feltiae* in conventional wheat was 0.80%. and the prevalence of *S. diapresi* in conventional was 0.32%. No *H. bacteriophora, S. riobrave,* or *S. carpocapsae* was collected from any of the sampled wheat fields (Fig. 1). The average EPN prevalence in all wheat fields sampled was 1.59%. The average EPN collected in all cattle pastures sampled was 7.0% (Fig. 3). The average prevalence of *S. diapresi* in wheat fields sampled was 0.39%. The average prevalence of *S. feltiae* in wheat fields sampled was 1.04%. The average prevalence of *S. glaseri* in wheat fields sampled was 0.16%. The average prevalence of *H. bacteriophora* in cattle pastures sampled was 1.67%. The average prevalence of *S. carpocapsae* in cattle pastures sampled was 3.17%. The average prevalence of *S. feltiae* in cattle pastures sampled was 1.67% The average prevalence of *S. riobrave* in cattle pastures sampled was 0.17% The average prevalence of *S. diapresi* in cattle pastures sampled was 0.33%(Fig. 3). The average EPN prevalence in organic pasture was 7.33%. The average EPN prevalence in conventional pasture was 6.67% (Fig. 2). The averageprevalence of *S. feltiae* in organic pasture was 1.33%.The average prevalence of *S. diapresi* in organic pasture was 0.67%. The prevalence of *S. carpocapsae* in organic pasture was 5.33%. No *H. bacteriophora* was collected from any organic pasture samples. The average prevalence of *S. feltiae* in conventional pasture was 2.0%. and the average prevalence of *S. riobrave* in conventional pasture was 0.33%. The average prevalence of *S. carpocapsae* in conventional pasture was 1.0%. The average prevalence of *H. bacteriophora* in conventional pasture was 3.33% (Fig. 2). All species were confirmed through molecular identification using the 18s-28s gene region of the genome, any samples that were not successfully identified this way were identified by comparing infection symptoms and location with confirmed samples. The soil in the organic wheat field sampled was Sandy Loam (12.5% clay, 55% sand, and 32.5% silt. The soil in the conventional wheat field sampled was Sandy Loam (10% clay, 58.8% sand, and 31.3% silt). The soil in the organic cattle pasture was Sandy Loam (7.5% clay, 64% sand, and 27.5% silt). The soil in the conventional cattle pasture was Loam (16.3% clay, 40% sand, 43.8% Silt) (Table 1).

<u>Discussion</u>

The average prevalence of EPN in organic wheat fields was significantly higher (p=0.0343) than EPN prevalence in conventional wheat fields. This may be due to the type of soil amendments used in organic agriculture (e.g. compost and manure) which create a soil environment with higher porosity and better moisture retention (Karungi *et al.*, 2006). Both of these traits are conducive to successful EPN persistence in an environment. Conventional methods of fertilizing wheat fields include swine affluent, anhydrous ammonia, and the addition of sewage (Bulluck Iii *et al.*, 2002). The previous amendmets utilized in conventional systems change soil pH to limits outside of normal EPN preference and do nothing to increase the soil's porosity or its ability to maintain higher moisture levels. The average prevalence of EPN in organic versus conventional cattle pastures was not significantly different (DF = 30, F value = 0.06, p = 0.8074). However, the diversity of EPN varied between the organic and conventional pastures, most likely due to the nematicides fed to cattle reducing the available hosts for EPN in the local environment. These compounds move through the cattle's systems and are excreted in the dung. These excreted nematicides collect in a high concentration in the soil directly underneath a dung pat, causing selective pressure on the soil ecosystem. A high concentration of these compounds in the soil inhibits the development of arthropods that use the dung pat as a resource, reducing the amount of available hosts for EPN in the area (Römbke *et al.*, 2010). With this pressure in place, *H. bacteriophora* anecdotally have the ability to compete with and displace the more prolific *S. carpocapsae*. When this selective pressure is removed, the *S. carpocapsae* seem to out-compete any *H. bacteriophora* in the area by producing more IJs at the end of a reproduction cycle. The overall prevalence of EPN in the pasture ecosystem is significantly higher than in the agricultural wheat ecosystem (p <.05). Possibly due to the lack of tilling that conventional agricultural crop fields receive, making cattle pastures more stable environments for EPN to persist in. When wheat fields are tilled annually, the populations of available hosts for EPN drop dramatically. In comparison, when cattle are not present on a pasture, high numbers of EPN hosts remain in the pasture making it readily available for EPN to utilize for reproduction year-round (Dennis *et al.*, 1998). All fields sampled had soil texture that is suitable for EPN persistence.Due to organic practices promoting higher EPN prevalence in agricultural crop fields with the benefits to the soil ecosystem (higher porosity and soil moisture retention) these production practices may contribute to promotion of background pest suppression by EPN in these systems. Cattle pastures are not tilled on a yearly basis, creating a more stable environment for EPN then a conventionally tilled agricultural crop field. With added stability comes added persistence of EPN, creating an environment that may allow for higher levels of background suppression then a less stable environment. Conventional nematicides and pesticides given to cattle may create a selective environment for EPN, in which *H. bacteriophora* can out compete, the normally more prolific, *S. carpocapsae*. Producers can conserve the naturally occurring EPN in their agricultural operations by incorporating production practices that promote a healthy soil environment. With successful background suppression, producers can reduce the amount of chemical pest control needed during a production season.

CHAPTER III

COMPARISON OF EPN PREVALENCE AND DIVERSITY ALONG A PRECIPITATION GRADIENT IN CONVENTIONAL CATTLE PASTURES ACROSS THE STATE OF OKLAHOMA

Introduction

Entomopathogenic Nematodes (EPN) are small, soil dwelling, roundworms that exist as obligate pathogens of arthropods. The only free living life stage of the two families, Heterorhabditidae and Steinernematidae, is the Infective Juvenile (IJ) life stage. The IJ uses chemical cues to guide itself through the soil in search of hosts (Gaugler *et al.*, 1989). Once a favorable host is located the IJ enters the hoemocoel through the mouth, anus, or a wound (damaged area of the cuticle) (Kaya & Gaugler, 1993). The IJ then proceeds to regurgitate or defecate symbiotic bacteria from their gut into the host's hoemocoel. Each family of EPN is associated with a specific genus of bacteria. The Heterorhabditidae release *Photorhabdus* spp. into their host, while the Steinernematidae release *Xenorhabdus* spp. (Adams *et al.*, 2006). Most species of EPN have a unique species of symbiotic bacteria that is found nowhere else in the world, except the gut of that EPN (Kaya & Gaugler, 1993). Once introduced to the hoemocoel of an insect, the bacteria begin to break down the tissue of the insect, killing the host through septicemia.

While breaking down the host tissue, the bacteria release preservatives, which include: anti-bacterial agents, anti-fungal agents, and ant repellant compounds known as “ant-deterrent factors”, so that the cadaver can persist in the environment long enough for the EPN to fully utilize all of the available resources (Zhou *et al.*, 2002). The EPN reproduce for 2-3 generations within a host, feeding on a mixture of broken down host tissue and bacterial lawn (Kaya & Gaugler, 1993). Once reproduction is completed, a new generation of IJ burst through the cuticle of the host cadaver and disperse back into the environment in search of their next host (Kaya & Gaugler, 1993).

Naturally-occurring EPN provide valuable background suppression in a variety of systems. Susceptible insect hosts, span over 250 species from over 75 families across 11 different orders (Grewal *et al.*, 2001). The existence of persistent populations of EPN within agricultural systems can provide valuable assistance to producers by cutting costs associated with insect pest management. Any insect that comes in contact with the soil at any point during its development is potentially susceptible to EPN infection. Classifying the environments in which EPN can successfully persist may be the first step in conserving natural populations of these valuable background suppression agents in agricultural systems.

EPN can provide valuable suppression of pests in agricultural settings; however they are still susceptible to trophic pressure from predators, pathogens, and antibiosis themselves. EPN are preyed upon by several common microarthropods including mites and collembolans (Greenwood *et al.*, 2011). Nematophagous fungi (e.g. *Hirsutella rhossiliensis*) thrive on EPN IJs in a moist soil environment (Kaya & Koppenhöfer, 1996). Antibiosis occurs when plants release volatiles from their roots into the soil environment, thereby disrupting the host seeking ability of EPN IJs in the immediate vicinity (Kaya & Koppenhöfer, 1996). The biotic factors listed, with the exception of antibiosis, thrive in similar environments as EPN. While promoting conservation of EPN in the soil environment, a land manager is also promoting predators and pathogens of the EPN, creating trophic pressure that the native EPN must survive.

Soil moisture is a crucial factor to the success of EPN in any environment due to the use of the moisture layer around soil particles for locomotion by the IJ life stage (Kaya & Gaugler, 1993). Oklahoma has 14 different precipitation zones across the state, ranging from 38cm/year in the western most portion of the panhandle, to 145cm/year in the most south eastern area of the state. 4 locations across the state were selected to be sampled for EPN prevalence at precipitation zones 38cm-45cm/year (Goodwell), 61cm-67cm/year (Woodward), 91cm-99cm/year (Stillwater), and 107cm-114cm/year (Haskell) (Fig. 5).

Cattle pastures that are not over-grazed and managed in sustainable manner serve as ideal sampling locations due to the number of available hosts for EPN. These include a variety of Lepidopteran and Coleopteran larvae associated with the diverse vegetation of the pasture, and the multitude of insects associated with cattle, such as hornflies (*Haematobia irritans*), cattle grubs (Hypoderma), stable flies (*Stomoxys calcitrans*), black flies

(Simuliidae), horse and deer flies (Tabanidae), sand flies (Psychodidae), house flies (*Musca domestica*), and hard ticks (Renn, 1998; de Carvalho *et al.*, 2010). The most economically important pests in Oklahoma beef cattle systems include horn flies, stable flies, house flies and ticks; these have all been shown to be successfully infected by EPN. *Steinernema. carpocapsae* alone has been shown to infect more than 250 insect species in over 75 families in 11 different orders(Grewal *et al.*, 2001).

Grazing of livestock can have an impact on the soil environment through compaction, which decreases the pore spaces in the soil, thereby limiting the mobility of IJs. Soil porosity is a significant factor for EPN success in an environment due to their limited mobility when searching for hosts. Clay soils have very small pores between particles, making dispersal more difficult for EPN, whereas moist sandy soils, or soil with manure additions, have large pores between particles and are more ideal for EPN dispersal into the environment (Kaya & Gaugler, 1993). Soil pH is also a significant factor in EPN success. EPN thrive in a soil pH range of 4-8, however when the pH approaches 10, survival drops dramatically (Shang-Ping *et al.*, 1990). The conditions necessary for EPN success in an environment, with the exception of porosity due to intermittent compaction, are supported in a pasture land ecosystem.

Soil moisture levels are a major abiotic influence on the success of EPN in a soil environment. Conventionally managed cattle pastures are relatively stable agricultural environments with a continuous draw of soil-dwelling arthropods. These factors make this system ideal for EPN sampling. Oklahoma has a precipitation gradient running from the western border (38cm) to the eastern border of the state (144cm). This wide variation in precipitation causes drastic differences in soil moisture levels. The objective of this experiment is to determine the impact of different average annual precipitation levels on the prevalence and diversity of native EPN in conventionally managed cattle pasture in Oklahoma.

Method and Materials

Soil Samples

4 sites were selected at Oklahoma state research stations in Goodwell, Woodward, Stillwater, and Haskell for their even distribution within the precipitation gradient across the state and their consistent land management systems (conventionally treated cattle pasture with similar stocking rates). All of the fields sampled contained with porosity and water retention ability that can promote EPN persistence. There was not a significant different between the soil types at the different locations and fields sampled. 2 cattle pastures were selected at each site for sampling. Each site was sampled on 4 different dates within 48 hours of a minimum of 2cm of rainfall. Sampling trips were broken down into Eastern days, when Stillwater and Haskell were sampled, and Western days, when Goodwell and Woodward were sampled. This was necessary do to the long distances between the sites making it impossible to sample all in the same day. Stillwater and Haskell were sampled on 1/3/12, 1/18/12, 4/1/12 (Haskell only), 4/13/12 (Stillwater

only), and 6/7/12. The dates that Haskell and Stillwater were sampled separately was a result of one site getting rain at a time. Goodwell and Woodward were sampled on 2/17/12, 3/18/12, 3/26/12, 10/14/12. Within each cattle pasture samples were taken from underneath fresh (< 2 weeks old) dung pats, old (>2 weeks old) dung pats, and away from dung pats (at least 3m in every direction from a dung pat). During each sampling date 20, 330 cm^3 soil samples were collected from each condition (fresh, old, none) from each pasture at each location, making sure to collect soil from the at least the top 15cm of soil (where most EPN IJ reside). Dung pat age was determined judging moisture content and breakdown of the pat due to environmental exposure. A professor with extensive experience working in cattle pasture taught collectors how to identify the necessary breakdown of dung pats to recognize the 2 week limit by sight and texture (determined by prodding the dung pat with a hand trowel). The soil was sealed in a labeled zip-locking bag and was immediately transported to the lab in coolers to maintain moderate temperatures to avoid sterilization of the soil. A total of 960 samples were taken between June 3rd 2011 and June 7th 2012. OSU research stations graze cattle at rates sustainable to their respective soil types, making them an ideal location to sample.

EPN Bioassay

Once the samples are unpacked from their transport coolers in the laboratory they are baited with 6 *Galleria mellonella* larvae and stored at room temperature in the dark for 7 days, consistent with standard bioassay technique described by Lacey & Kaya (1997). A

5cm gap is left unzipped at the top of the bags to allow for gas exchange. *G. mellonella* larvae are the standard insect and stage for bioassays for EPN due to their low LD-50 for all species of EPN species tested (Morris *et al.*, 1990). The larvae were collected after being in contact with the soil for 7 days. Disposition of each of the 6 *G. mellonella* larvae was determined. Disposition of the larvae was determined by examining the texture and color of the, normally white, larvae after exposure to the soil. Infected larvae will be soft, but retain their shape, and will be resistant to tearing, due to the preservatives released by the symbiotic bacteria. An infected larva will be a distinctive shade of tan, brown, grey, or red. Dead larvae are normally very soft, don't retain their shape, tear easily, and are very dark and sometimes mottled in color. Prevalence was defined as the total number of infections divided by the total number of *G. mellonella* larvae used in the bioassay. All infected cadavers were gently washed and placed in a clean 9cm petri dish on a 9cm piece of filter paper. The paper was moistened with 1-2ml of DI H_2O. The cadavers were checked for IJ emergence daily. Once the EPN began to emerge, the filter paper was removed and placed on the upside-down lid of a 9cm petri dish, which was then placed into a 14.5cm petri dish with a thin layer of water in it. During the next 24-72hrs, the EPN dispersed across the filter paper and collected in the water of the 14.5cm petri dish, the water was then collected. The EPN were preliminarily identified using symptoms of infection exhibited by the infected *G.mellonella* cadavers according to Field manual of techniques in invertebrate pathology (Lacey, 2008).

Molecular Identification

Single EPN from each infected cadaver were lysed using EPN lysis buffer (unpublished Adams lab protocol). PCRs were then run using the resulting solution as the DNA template. PCR Primers designed to amplify ITS 18s-28s gene region were used. The resulting PCR product was cleaned with exonuclease and SAP and sent to the CORE facility on the OSU campus for sequencing. The CORE facility used "BigDye™"-terminated reactions analyzed on an ABI Model 3730 DNA Analyzer to sequence the submitted samples. The resulting sequences were trimmed and cleaned by eye using BioEdit software. The cleaned sequences were identified using the BLASTn program and the NCBI database. Species identification was confirmed based on a low E-value and high Max ident. Statistical Analysis

Prevalence was evaluated as total infections/total *G. mellonella* used to bait. Prevalence data based on infection symptoms *G. mellonella* were subjected to statistical analysis using analysis of variance (ANOVA) techniques (PROC MIXED, PC SAS Version 9.2, SAS Institute, 1996). Percents were transformed with the arcsine square root function to correct for heterogeneity of variance. Analysis of variance methods were conducted assuming a split plot model with repeated measures. There were no interactions involving DATE, so the main effects of DATE are reported. The LOC by TRT interaction was significant enough to warrant the inspection of simple effects (effect of LOC given TRT and effect of TRT given LOC).

Results

A total of 6 species of EPN were identified. As the average rainfall per year at each site sampled increased, across a west to east gradient, the prevalence and diversity of EPN increased. Samples taken from under fresh dung pats in Goodwell had an average EPN prevalence of 0.833%, samples from underneath fresh dung pats in Woodward had an average EPN prevalence of 0.833%, samples taken from under fresh dung pats in Stillwater had an average EPN prevalence of1.56%, samples from underneath fresh dung pats in Haskell had an average EPN prevalence of 1.35%. Samples taken from under old dung pats in Goodwell had an average EPN prevalence of 0.63%, samples from underneath old dung pats in Woodward had an average EPN prevalence of 0.52%, samples taken from under old dung pats in Stillwater had an average EPN prevalence of 5.83%, samples from underneath old dung pats in Haskell had an average EPN prevalence of 1.97%. Samples taken away dung pats in Goodwell had an average EPN prevalence of 0.10%, samples taken away from dung pats in Woodward had an average EPN prevalence of 0.417%, samples taken away from dung pats in Stillwater had an average EPN prevalence of 1.56%, samples taken away from dung pats in Haskell had an average EPN prevalence of 3.23%. Stillwater had the highest EPN prevalence of any single dung pat type, with 5.83% underneath old dung pats. Goodwell had the lowest EPN prevalence of any single dung pat type, with 0.10% from samples taken away from dung pats. *Steinernema feltiae* was most prevalent in the Stillwater location underneath old dung pats, with a mean prevalence of 1.80%. *Steinernema texanum* was most prevalent at the Haskell location in samples taken away from dung pats, with a mean prevalence of 0.90%. *Steinernema carpocapsae* was most prevalent at the Stillwater location underneath old dung pats, with a prevalence of 3.90%. *Steinernema riobrave* was most prevalent at the Stillwater location underneath old dung pats, with a mean prevalence of 0.10%. *Steinernema glasseri* was most prevalent at the Haskell location underneath old dung pats, with a mean prevalence of 0.20%. *Heterorhabditus bacteriophora* was most prevalent at the Haskell location in samples taken away from dung pats, with a mean prevalence of 0.20%. The average soil moisture for pastures in Goodwell was 8.42%. Woodward pastures had similar soil moisture with 8.30%. The average soil moisture for Stillwater pastures sampled was 18.23%. Haskell pastures had the highest average soil moisture recorded with 22.36% (Fig 2). All species were confirmed through molecular identification using the 18s-28s gene region of the genome, any samples that were not successfully identified this way were identified by comparing infection symptoms and location with confirmed samples. All pastures sampled were a variation of Loam texture. Goodwell pastures contained Gruve clay loam, Woodward pastures contained Devol Fine Silty Loam, Stillwater pastures contained Norge Loam, and Haskell pastures contained Dennis silt loam (Table 1).. There was a significant difference in the average prevalence's along the precipitation gradient from west to east from samples taken away from dung pats (DF = 8.52, F value = 5.42, p = 0.0465). At the Stillwater site there was a significantly higher prevalence of EPN found under old dung pats (p = .0083) versus fresh and no dung pats.

Discussion

EPN prevalence increased from west to east across the state, as the precipitation level increased. It was hypothesized that there would be a steady increase in EPN prevalence as the sites moved up in average rainfall per year, due to the importance of soil moisture to the life cycle of EPN (Kaya & Gaugler, 1993). The results show a much more abrupt change in prevalence of EPN, indicating a potential threshold of centimeters of rainfall per year needed for the EPN to be successful. The threshold of EPN across OK is supported by EPN being most effective at moderate soil moisture levels (-10 to -100 kPa). They become less successful at infection in high moisture content soils (> -1kPa) and lower moisture content soils (< -1000 kPa)(Koppenhöfer & Fuzy, 2007). The south eastern most corner of Oklahoma, where the most rainfall occurs, creates average soil moisture limits below the upper limit of the preferred range for EPN. Stillwater had the highest prevalence of the two most common species of EPN, *S. feltiae* at 1.80% and *S. carpocapsae* at 3.90%, while Haskell had the highest prevalence of other two less common species found, *S. texanum* at 0.90% and *H. bacteriophora* at 0.20%. Stillwater had the highest abundance of the two other uncommon species, *Steinernema riobrave* at 0.10% and *Steinernema glasseri* at 0.20%; both of which were found only underneath old dung pats. This may be attributed a longer life cycle and less drought tolerance by these species, contributing to IJs only being present after an extended period of high soil moisture and available hosts, both of which are provided by the degrading dung pat. Once the threshold of precipitation is reached the prevalence increases and begins to allow less drought tolerant species of EPN to thrive in the environment. This is not surprising, due to the similar overall prevalence of EPN at the Stillwater and Haskell sites, 2.98% and 2.20% respectively as well as the increase in rainfall when moving east toward Haskell. Due to the increase in diversity at the Haskell site, resulting from the increase in precipitation, *S. feltiae* and *S. carpocapsae* have competition from less drought tolerant species (*S. texanum* and *H. bacteriophora*) and thus persist at lower overall prevalence in the environment (Koppenhöfer & Fuzy, 2007). All of the soil types encountered had textures that exibit enough soil porosity and water retention ability to maintain persistent EPN populations.

Once over the soil moisture threshold required by EPN, the increase in soil moisture does not appear to correlate with an increase in EPN prevalence. Further breakdown of the precipitation gradient would give a more accurate idea as to where the actual threshold lies in the precipitation gradient. Other variation in native EPN communities can arise due to varying environmental factors at the different locations. Plant community variation can influence the community composition of EPN hosts in a given environment, there by promoting different EPN species in the local EPN community (De Deyn *et al.*, 2007). Along with variations in EPN host species, differing plant communities can promote different EPN predator and pathogen communities, causing different levels of trophic pressure on the EPN communities at different locations (E. Siemann, 1998). We had the ability to control for land management techniques within out sampling locations, but it was impossible to control for the other variations that are a result of different precipitation levels in the different locations. With the data collected we can state that the threshold for successful EPN populations is between 61cm and 99cm of rainfall a year.

Goodwell and Woodward had higher proportions of Steinernematid species, which agrees with the literature that has shown Steinernematid species to be more drought tolerant then Heterorhabditid species (Somvanshi *et al.*, 2008). Samples from Stillwater and Haskell locations had similar EPN prevalence levels, but different levels of diversity. As the average annual rainfall increased the EPN diversity increased as well, resulting in twice as many EPN species found in Haskell (107cm-114cm/year) than in Stillwater (91cm-99cm/year). The results show that there may be an average rainfall per year tolerance level for EPN populations, above a certain level of average annual rainfall the prevalence of EPN did not increase. However, the diversity continued to increase as the average annual rainfall increased, potentially allowing the drought intolerant species a chance for greater success in those environments.

There were a few samples that we were not able to identify genetically due to contaminant nematodes invading the samples. These samples were grouped with samples that shared identical infection symptoms from the same locations that were successfully identified using the 18s-28s gene region.

All of the pastures sampled had soil types that promote EPN persistence. Soils with high percentages of clay promote poor EPN survival due to low porosity, although it does have high water retention. Loam promotes successful EPN persistence due to high porosity and high water retention. Sand compliments clay due to its high porosity, but low water retention. A mixture of sand and clay has sufficient porosity, due to the pores created by the large grains making up sand, and sufficient water retention, due to the ability of clay to successfully retain moisture, creating a soil environment that allows for successful host seeking and persistence of EPN. The soil at all of the sites sampled had less than 40% clay, showing that the soil type at all locations was capable of promoting EPN persistence.

Old dung pats in Stillwater had a significantly higher prevalence of EPN in the soil underneath them than under the fresh dung pats or away from dung pats. This corresponds with the timing of the EPN life cycle. 2-3 reproduction cycles and a new generation of IJ take from 7-21 days to complete, depending on the species of EPN and the temperature during reproduction. Old dung pats were classified as >2 weeks old. The 2 weeks would allow time for EPN infection of hosts attracted to the input of resources and the successful reproduction and emergence of a new generation of IJs. This timing resulted in a higher prevalence under those conditions due to the recent emergence of IJs under old dung pats.

Overall EPN prevalence was higher in the two eastern sites sampled, showing that the threshold of precipitation level nessesary for successful EPN persistence in a conventionally managed cattle pasture lies between Woodward and Stillwater in Oklahoma. Once the average annual precipitation threshold is reached, there is no significant increase in EPN prevalence as the average annual precipitation level increases, however the diversity of EPN as the average annual precipitation increases also increases. As the annual average precipitation increases above the nessesary levels for successful EPN persistence, more drought intolerant species are capable of successfully persisting at those locations.

Producers that have cattle operations in areas east of the EPN soil moisture threshold in OK may be able to gain successful background pest suppression from EPN in their pastures on a yearly basis. Taking this information into account and monitoring the success of EPN in pastures, may allow for an increase of the economic threshold level of pests of cattle that come into contact with the soil during their life cycles, due to background suppression provided by the persisting EPN. In eastern areas of the state pest control achieved through conservation of indigenous EPN can be invaluable when pest levels are teetering near the economic threshold, reducing the chance that a producer will have to take extra control measures at all, saving both time and money.

CHAPTER IV

SUMMARY

Little is known about native EPN in Oklahoma. Prevalence rates of EPN taxa indicate that EPN are present and diverse throughout the state. Their potential utility in the form of background pest suppression, and potential impact in the form of non-target effects, suggests that more research on native EPN is warranted. This study quantifies the effects of common land use practices in Oklahoma on EPN prevalence, and the effects of a naturally-existing precipitation gradient that runs from east to west across the state. Beef and wheat production constitute the primary agricultural products within Oklahoma. Only one producer in the state is certified in the organic production of both wheat and beef. This producer generously provided access to his fields and pastureland for the purposes of this research.

This study has shown that organic management techniques in agricultural wheat fields can promote EPN prevalence. Organic agriculture prohibits chemical fertilizers and pesticides, resulting in compost and manure being used as soil amendments and more available EPN hosts in the soil (Gruner *et al.*, 2007). Compost and manure increase soil porosity, water retention ability, and organic matter; creating a more suitable and stable environment for EPN (Jabbour & Barbercheck, 2008). Organic practices were not shown to have a significant impact on EPN prevalence; however, they do result in different EPN diversity in those environments. Samples collected from cattle pastures had much higher average prevalence than samples collected from agricultural wheat fields. This makes sense due to the higher stability and more regular inputs of resources of the cattle pasture. Higher stability (e.g. no tillage) results in better persistence of EPN in the environment (Millar & Barbercheck, 2002). With more regular inputs in the cattle pasture environment (e.g. cattle dung pats), and a greater diversity of vegetation, there are areas of the pasture that have very high concentrations of EPN hosts. Concentration of hosts promotes EPN success due to the small distances that IJs are capable of dispersing. Conventionally managed cattle are treated with pesticides, both topically and prophylactically. These additional additives into the environment result in less hosts available for EPN and concentrations of nematicides in the soil underneath cattle dung pats. These addition selective pressures allow more nematicide resistant but less fecund species (e.g *H. bacteriophora*) of EPN to compete with less nematicide resistant but more fecund species (e.g. *S. carpocapsae*) in a conventional environment (Grønvold *et al.*, 2004; Römbke *et al.*, 2010). Conservation of native EPN through organic beef and wheat production systems may contribute to background suppression of pest insects. Since EPN are generalists, though, potential exists for non-target effects. The net effect of EPN conservation in these systems requires more research.

Prevalence of EPN was determined in consistently managed pastureland located at each of four Oklahoma State University Research stations that spanned the gradient from east to west. As the average annual precipitation increases from West to East across

Oklahoma, a threshold is crossed (between 63cm and 99cm of average annual precipitation) allowing successful EPN persistence in the eastern parts of the state. This agrees with the literature, showing that EPN persist most successfully in intermediate soil moisture (Koppenhöfer & Fuzy, 2007). If the soil moisture is too low, the EPN cannot disperse and can potentially desiccate. If the soil moisture is too high EPN cannot host seek successfully and can be washed away in run off. Conservation of EPN for background suppression of pests can be most successfully implemented at or above 99cm of average annual precipitation in Oklahoma.

Further studies should be conducted to elucidate native EPN prevalence in the scenarios described above, and additional scenarios in Oklahoma. The effects of native EPN on both soil-dwelling pest species of insects, and non-target arthropods should be further evaluated as well.

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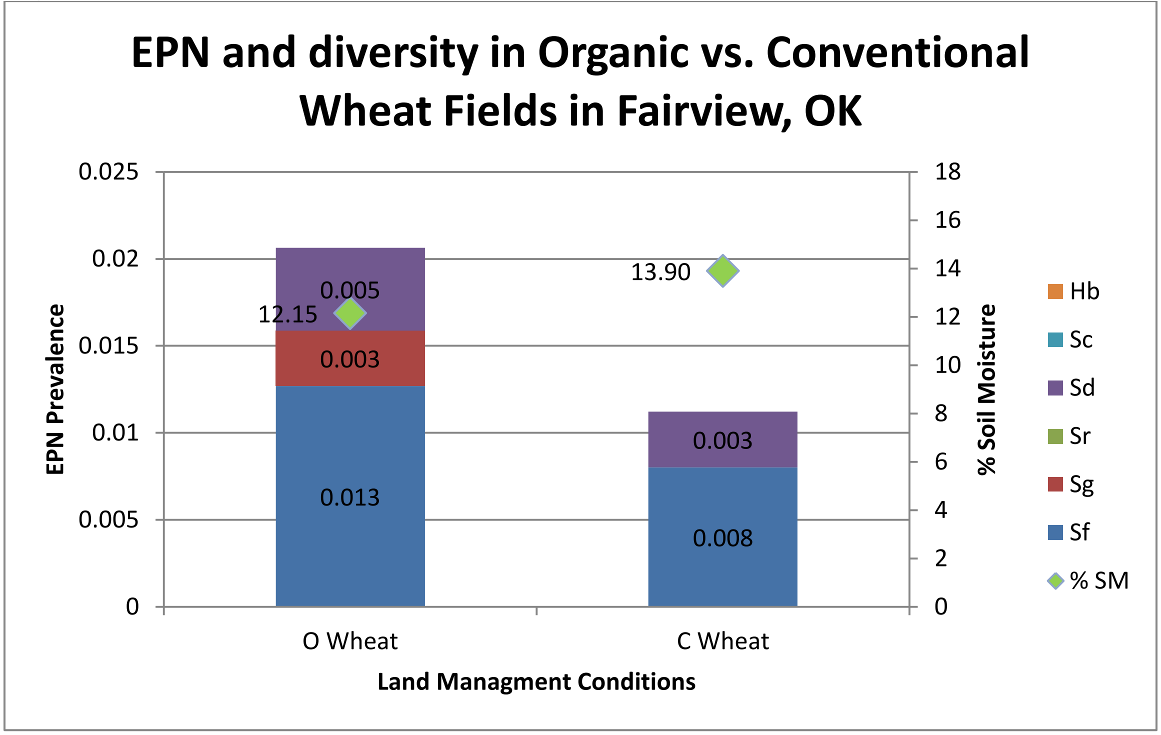
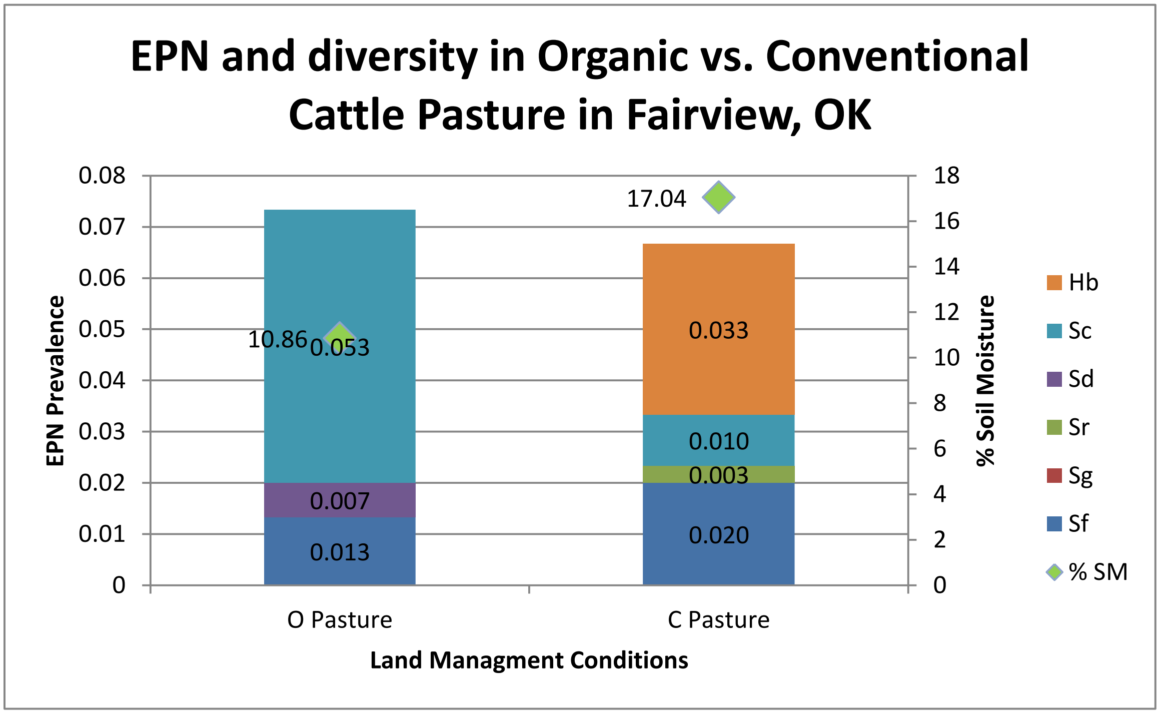
APPPENDICES

Figure 1.



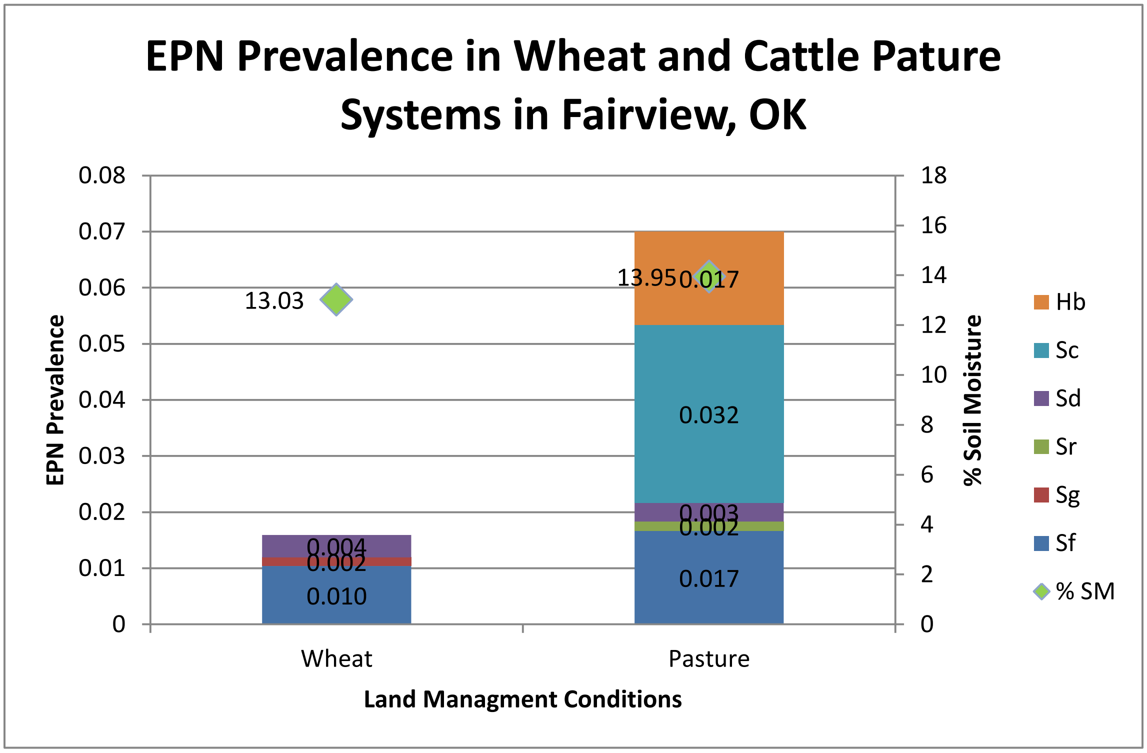
EPN prevalence (based on bioassay infection rate), diversity (Hb= *Heterorhabditis bacteriophora*; Sc = *Steinernema carpocapsae*; Sf = *S. feltiae,* Sg = *S. glaseri, Sr = S. riobrave,* Sd = *S. diapresi*) of EPN, and soil moisture in organic vs. conventional wheat fields in Fairview, OK (data is compiled from 3 sampling dates). DF = 3.06, F-value = 13.33, p = 0.034.

Figure 2.



EPN prevalence (based on bioassay infection rate) and diversity (Hb= *Heterorhabditis bacteriophora*; Sc = *Steinernema carpocapsae*; Sf = *S. feltiae*, Sg = *S. glaseri, Sr = S. riobrave,* Sd = *S. diapresi*) of EPN in organic vs. conventional pastures in Fairview, OK (data is compiled from 3 sampling dates).

Figure 3.

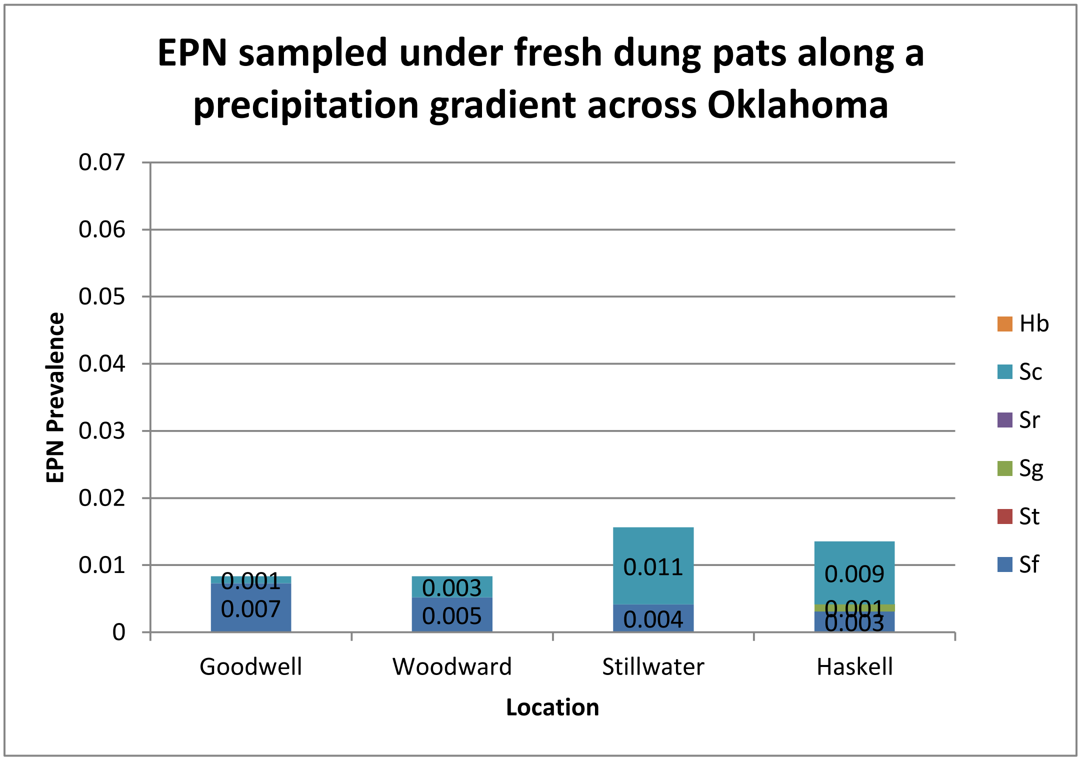


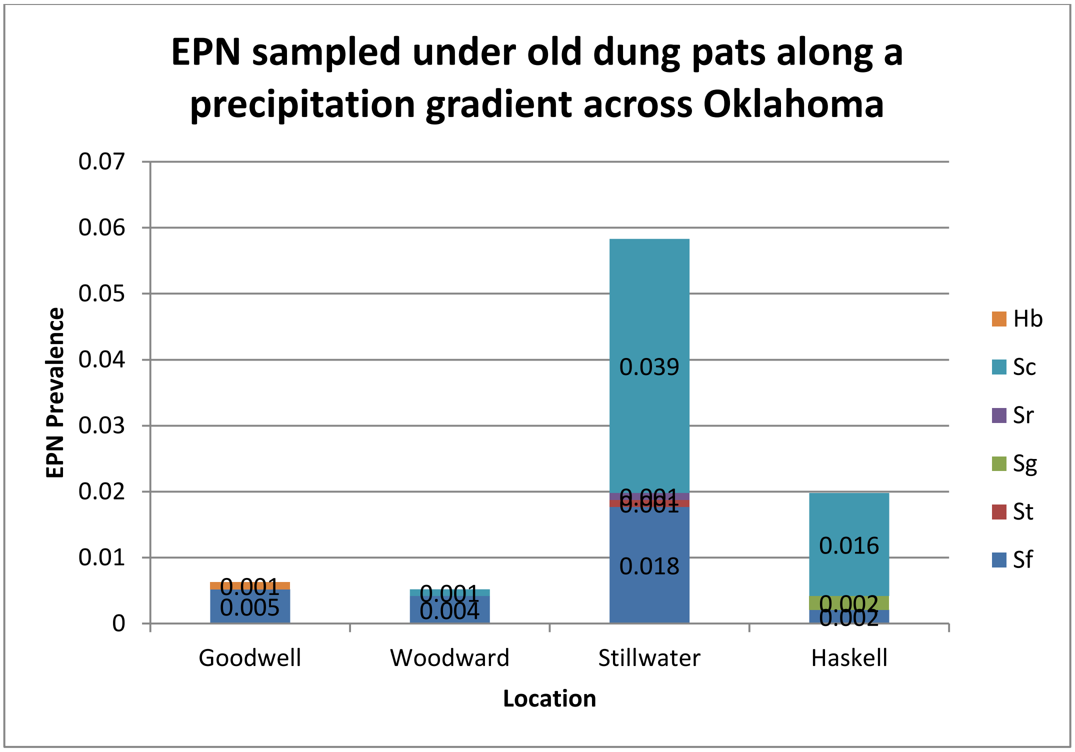
EPN Prevalence (based on bioassay infection rate) and diversity (Hb= *Heterorhabditis bacteriophora*; Sc = *Steinernema carpocapsae*; Sf = *S. feltiae*, Sg = *S. glaseri*, *Sr* = *S. riobrave*, Sd = *S. diapresi*) of EPN in wheat vs. cattle pastures in Fairview, OK (data is compiled from 3 sampling dates).

Figure 4.

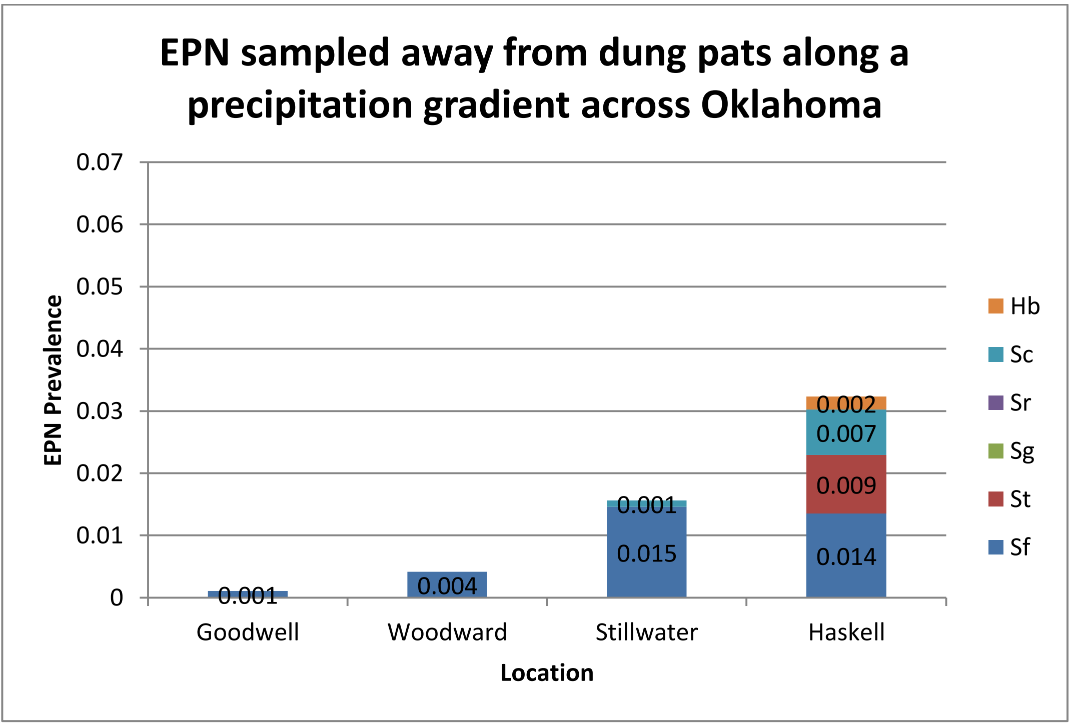
A map showing the precipitation gradient across the state of Oklahoma. The stars are our sampling locations and are labeled with the towns that the sampled cattle pastures were located in. These locations were chosen due to the distinctly different precipitation zones they are located in and their relatively even dispersal throughout the precipitation gradient across the state.

Figure 5.a.


EPN sampled under fresh dung pats along a precipitation gradient across Oklahoma
EPN Prevalence
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0
0.001
0.007
0.003
0.005
0.011
0.004
0.009
0.001
0.003
Goodwell
Woodward
Stillwater
Haskell
Location
Hb
Sc
Sr
Sg
St
Sf


EPN sampled under old dung pats along a precipitation gradient across Oklahoma
EPN Prevalence
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0
0.001
0.005
0.001
0.004
0.039
0.001
0.001
0.018
0.016
0.002
0.002
Goodwell
Woodward
Stillwater
Haskell
Location
Hb
Sc
Sr
Sg
St
Sf

b.


EPN sampled away from dung pats along a precipitation gradient across Oklahoma
EPN Prevalence
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0
0.001
0.004
0.001
0.015
0.002
0.007
0.009
0.014
Goodwell
Woodward
Stillwater
Haskell
Location
Hb
Sc
Sr
Sg
St
Sf

c.

EPN prevalence (based on bioassay infection rate), diversity (Hb = *Heterohhabditus bacteriophora*, Sc = *Steinernema feltiae,* Sr = *Steinernema riobrave,* Sg = *Steinernema glasseri,* St = *Steinernema texanum*, and Sf = *Steinernema feltiae*), and average soil moisture (1 measurement/pasture/sampling date) across Oklahoma ecoregions in conventional cattle pasture systems. a. Samples taken from beneath fresh (< 2 weeks) dung pats b. Samples taken from beneath old (> 2 weeks) dung pats c. Samples taken away from any dung pats.

Table 1.

Field type	Land management	% Soil Moisture	Avg annual Precip	Soil classification	% Clay	% Sand	% Silt
Wheat	Organic	12.15	68-76	Sandy Loam	12.5	55.0	32.5
Wheat	Conventional	13.90	68-76	Sandy Loam	10.0	58.8	31.3
Pasture	Organic	10.86	68-76	Sandy Loam	7.5	65.0	27.5
Pasture	Conventional	17.04	68-76	Loam	16.3	40.0	43.8

Descriptions of the physical soil characteristics and annual precipitation levels at Organic vs Conventional wheat fields and organic vs. conventional pastures over 3 sampling dates (put the dates here)in Fairview, OK.

Table 2

Location	% Soil Moisture	Avg annual Precip (cm/yr)	Soil classification	Particle Size
Goodwell	8.42	38-45	Gruver Clay Loam	35-45% silicate clay

Woodward	8.30	61-68	Devol Fine Silty Loam	Loamy w/ >2% gravel
Stillwater	18.23	91-99	Norge Loam	Fine granular w/ many fine roots
Haskell	22.36	106-114	Dennis Silt Loam	5-25% sand, 37-60% clay

Descriptions of the physical soil characteristics and annual precipitation levels at each of the conventional cattle pastures sampled for the state wide precipitation gradient survey across the state of Oklahoma.

Table 3.

EPN Species	Location	Date	Cadaver Symptoms	GenBank Match Seq #	Total score	E-Value	Max ident
H. bacteriophora	Bessie	5/30	Dark Brown/Red	HM140691.1	996	0.0	86%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	783	0.0	80%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1557	0.0	99%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1288	0.0	94%
S. carpocapsae	Bessie	5/30	Light Brown/Tan	GQ421605.1	1590	0.0	100%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1436	0.0	99%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1067	0.0	88%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1103	0.0	94%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1523	0.0	99%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	HM140691.1	1067	0.0	93%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	HQ225866.1	771	0.0	90%
S. glaseri	Bessie	5/30	Dark Brown/Black	AF122015.1	646	1e-179	79%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1593	0.0	100%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	FJ346826.1	1590	0.0	99%
S. carpocapsae	Bessie	5/30	Light Brown/Tan	GQ421605.1	1438	0.0	99%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	JX403718.1	1299	0.0	100%
H. bacteriophora	Bessie	5/30	Dark Brown/Red	JX403718.1	1297	0.0	100%

Samples from a exploratory sampling trip to Bessie, Oklahoma. All EPN species were confirmed using infection symptoms, GenBank match, and confidence information organic vs conventional comparison. All samples that were not successfully confirmed using molecular techniques were matched with samples that were using infection symptoms and location.

Table 4.

EPN Species	Location	Date	Cadaver Symptoms	GenBank Match Seq #	Total score	E-Value	Max ident
S. diaprepesi	Fairview	9/3	Dark brown/Brown	GU173996.1	1267	0.0	99%
S. riobrave	Fairview	10/17	Dark Brown/Black	DQ835613.1	1180	0.0	97%
H. bacteriophora	Fairview	10/17	Dark Brown/Red	JX164230.1	1303	0.0	100%
S. diaprepesi	Fairview	10/17	Dark brown/Brown	GU173996.1	1267	0.0	99%
S. riobrave	Fairview	10/17	Dark Brown/Black	DQ835613.1	1355	0.0	98%
S. diaprepesi	Fairview	11/18	Light Brown/Tan	GU173996.1	1555	0.0	99%
S. glaseri	Fairview	11/18	Dark Brown/Black	AF122015.1	901	0.0	88%
S. diaprepesi	Fairview	11/18	Light Brown/Tan	GU173996.1	1429	0.0	97%
S. glaseri	Fairview	11/18	Dark Brown/Black	AF122015.1	926	0.0	87%
S. diaprepesi	Fairview	11/18	Light Brown/Tan	GU173996.1	1570	0.0	99%
S. diaprepesi	Fairview	11/18	Light Brown/Tan	GU173995.1	1418	0.0	99%
S. diaprepesi	Fairview	11/18	Dark brown/Brown	GU173995.1	830	0.0	98%

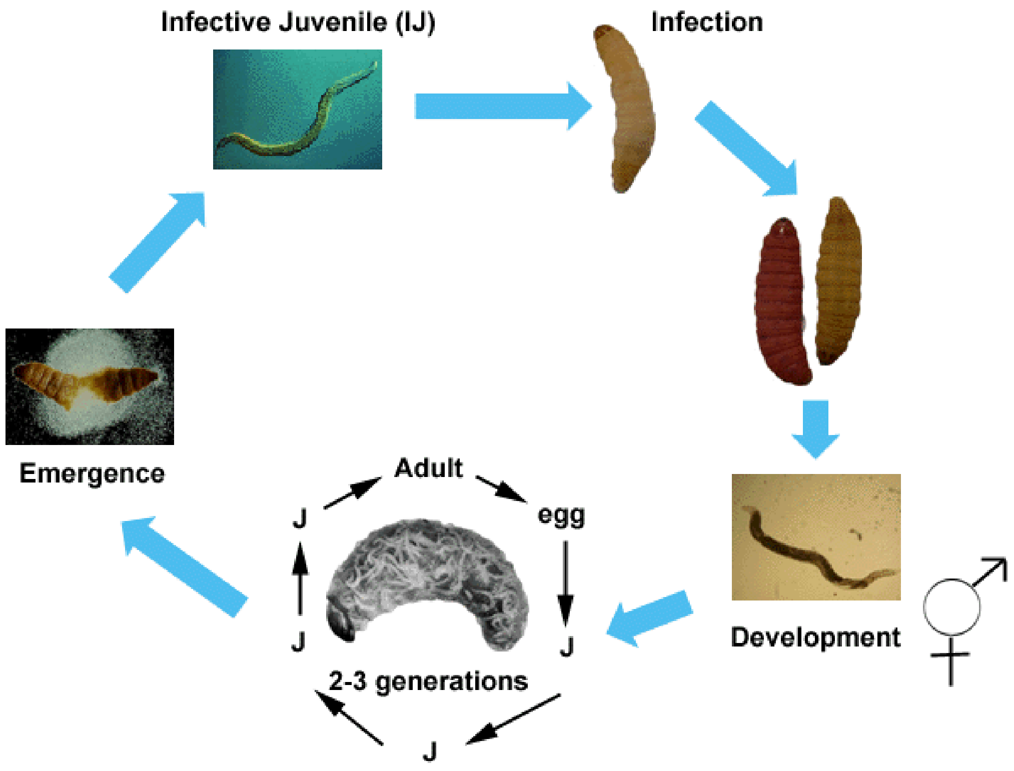
All EPN species were confirmed using infection symptoms, GenBank match, and confidence information for the organic vs conventional comparison. All samples that were not successfully confirmed using molecular techniques were matched with samples that were using infection symptoms and location.

Table 5.

EPN Species	Location	Date	Cadaver Symptoms	GenBank Match Seq #	Total score	E-Value	Max ident
S. feltiae	Stillwater	1/18	Dark Brown/Black	*	*	*	*
S. texanum	Haskell	1/18	Light Brown/Tan	*	*	*	*
S. feltiae	Stillwater	4/13	Dark Brown/Black	*	*	*	*
S. feltiae	Stillwater	4/13	Dark Brown/Black	*	*	*	*
S. feltiae	Stillwater	4/13	Dark Brown/Black	*	*	*	*
S. feltiae	Stillwater	4/13	Dark Brown/Black	*	*	*	*
S. feltiae	Stillwater	4/13	Dark Brown/Black	*	*	*	*
S. feltiae	Stillwater	4/13	Dark Brown/Black	*	*	*	*
S. texanum	Stillwater	4/13	Light Brown/Tan	*	*	*	*
S. riobrave	Stillwater	6/3	Light Brown/Tan	DQ835613.1	1404	0.0	96%
S. glaseri	Haskell	6/7	Dark Brown/Black	GU173998.1	1117	0.0	95%
S. glaseri	Haskell	6/7	Dark Brown/Black	GU173998.1	1018	0.0	95%

S. glaseri	Haskell	6/7	Dark Brown/Black	AF122015.1	1231	0.0	96%
S. feltiae	Stillwater	6/7	Dark Brown/Black	JN886631.1	1150	0.0	99%
S. feltiae	Stillwater	6/7	Dark Brown/Black	JN886631.1	906	0.0	92%
S. feltiae	Stillwater	6/7	Dark Brown/Black	JF728857.1	1294	0.0	99%
S. feltiae	Stillwater	6/7	Dark Brown/Black	JF728857.1	1079	0.0	93%

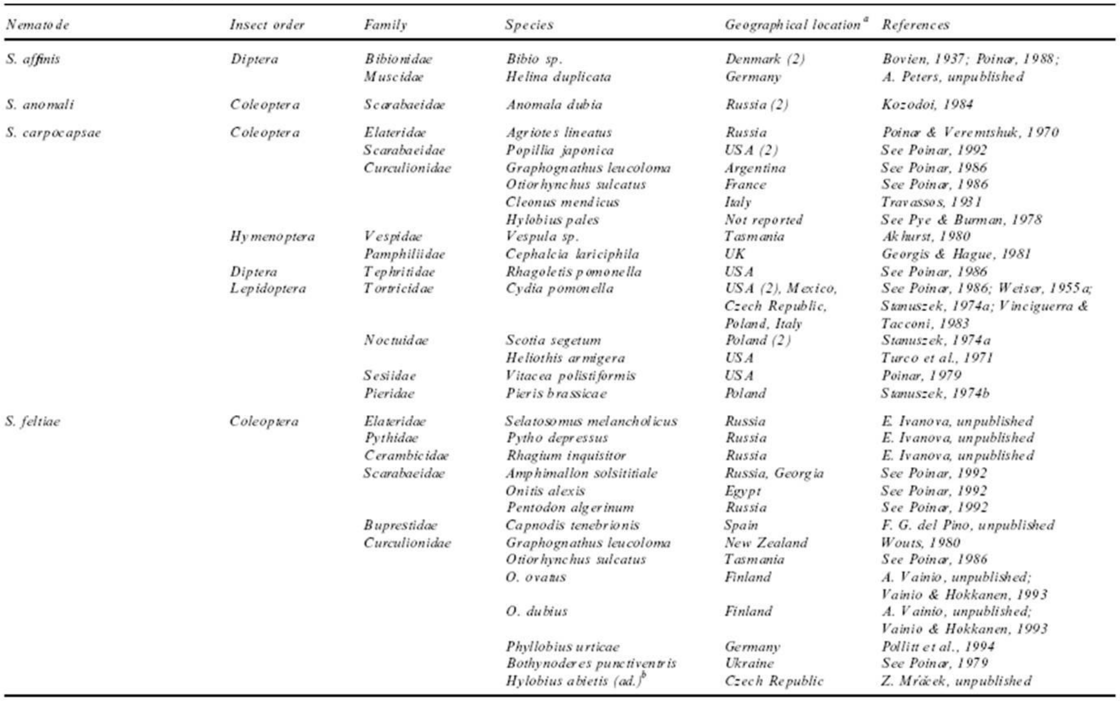
Table 5. Samples and Genbank matches from conventional cattle pastures along a state wide precipitation gradient. All EPN species were confirmed using infection symptoms, GenBank match, and confidence information organic vs conventional comparison. All samples that were not successfully confirmed using molecular techniques were matched with samples that were using infection symptoms and location. * Samples were identified using Geneious to align and edit sequences in Dr. Byron Adams lab at BYU from 5/21-5/24, Genbank match data was not recorded.



EPN lifecycle

Documented naturally occurring EPN infections

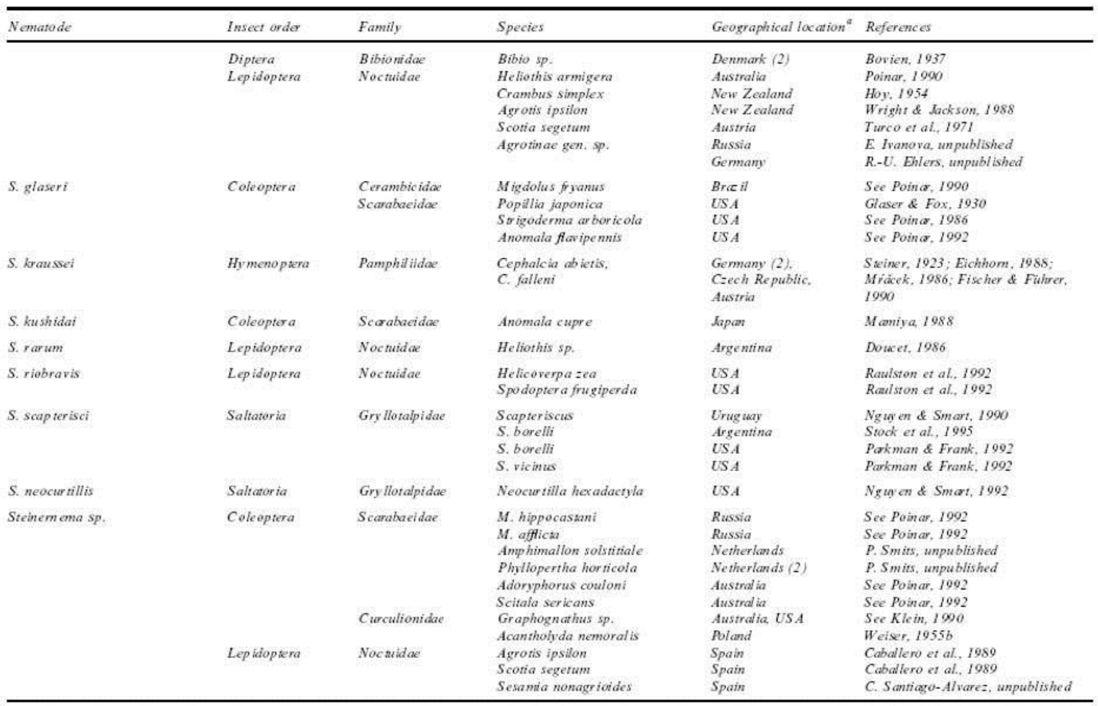
Table 7 - Documented naturally occurring infections of insect with various entompathogenic nematode species (from Peters 1996)



Nematode	Insect order	Family	Species	Geographical location[a]	References
S. affinis	*Diptera*	*Bibionidae*	*Bibio sp.*	*Denmark (2)*	*Bovien, 1937; Poinar, 1988;*
		Muscidae	*Helina duplicata*	*Germany*	*A. Peters, unpublished*
S. anomali	*Coleoptera*	*Scarabaeidae*	*Anomala dubia*	*Russia (2)*	*Kozodoi, 1984*
S. carpocapsae	*Coleoptera*	*Elateridae*	*Agriotes lineatus*	*Russia*	*Poinar & Veremtshuk, 1970*
		Scarabaeidae	*Popillia japonica*	*USA (2)*	*See Poinar, 1992*
		Curculionidae	*Graphognathus leucoloma*	*Argentina*	*See Poinar, 1986*
			Otiorhynchus sulcatus	*France*	*See Poinar, 1986*
			Cleonus mendicus	*Italy*	*Travassos, 1931*
			Hylobius pales	*Not reported*	*See Pye & Burman, 1978*
	Hymenoptera	*Vespidae*	*Vespula sp.*	*Tasmania*	*Akhurst, 1980*
		Pamphiliidae	*Cephalcia lariciphila*	*UK*	*Georgis & Hague, 1981*
	Diptera	*Tephritidae*	*Rhagoletis pomonella*	*USA*	*See Poinar, 1986*
	Lepidoptera	*Tortricidae*	*Cydia pomonella*	*USA (2), Mexico, Czech Republic, Poland, Italy*	*See Poinar, 1986; Weiser, 1955a; Stanuszek, 1974a; Vinciguerra & Tacconi, 1983*
		Noctuidae	*Scotia segetum*	*Poland (2)*	*Stanuszek, 1974a*
			Heliothis armigera	*USA*	*Turco et al., 1971*
		Sesiidae	*Vitacea polistiformis*	*USA*	*Poinar, 1979*
		Pieridae	*Pieris brassicae*	*Poland*	*Stanuszek, 1974b*
S. feltiae	*Coleoptera*	*Elateridae*	*Selatosomus melancholicus*	*Russia*	*E. Ivanova, unpublished*
		Pythidae	*Pytho depressus*	*Russia*	*E. Ivanova, unpublished*
		Cerambicidae	*Rhagium inquisitor*	*Russia*	*E. Ivanova, unpublished*
		Scarabaeidae	*Amphimallon solsitiale*	*Russia, Georgia*	*See Poinar, 1992*
			Onitis alexis	*Egypt*	*See Poinar, 1992*
			Pentodon algerinum	*Russia*	*See Poinar, 1992*
		Buprestidae	*Capnodis tenebrionis*	*Spain*	*F. G. del Pino, unpublished*
		Curculionidae	*Graphognathus leucoloma*	*New Zealand*	*Wouts, 1980*
			Otiorhynchus sulcatus	*Tasmania*	*See Poinar, 1986*
			O. ovatus	*Finland*	*A. Vainio, unpublished; Vainio & Hokkanen, 1993*
			O. dubius	*Finland*	*A. Vainio, unpublished; Vainio & Hokkanen, 1993*
			Phyllobius urticae	*Germany*	*Pollitt et al., 1994*
			Bothynoderes punctiventris	*Ukraine*	*See Poinar, 1979*
			Hylobius abietis (ad.)[b]	*Czech Republic*	*Z. Mráček, unpublished*

S. bicornutum, S. caudatum, S cubanum, S. intermedium, S. ritteri, S. longicaudum,[c] *S. serratum*[c]	*No*	*information*	—	—	—
Neosteinernema longicurvicaudum	*Isoptera*	*Rhinotermitidae*	*Reticulitermes flavipes*	*USA*	*Nguyen & Smart, 1994*
Heterorhabditis bacteriophora	*Coleoptera*	*Scarabaeidae*	*Popillia japonica*	*USA*	*See Poinar, 1992*
			Cyclocephala hirta	*USA*	*See Poinar, 1990*
			Phyllophaga sp.	*USA*	*Poinar & Georgis, 1990*
		Chrysomelidae	*Diabrotica balteata*	*USA*	*See Poinar, 1990*
		Curculionidae	*Curculio caryae*	*USA*	*See Poinar, 1990*
			Diaprepes abbreviatus	*USA (2)*	*See Poinar, 1990*
	Lepidoptera	*Noctuidae*	*Heliothis punctigera*	*Australia*	*Poinar, 1975*
			Helicoverpa zea	*USA*	*Poinar, 1975*
		Pyralidae	*Diatrea grandiosella*	*USA*	*See Poinar, 1990*
H. megidis	*Coleoptera*	*Scarabaeidae*	*Popillia japonica*	*USA*	*See Poinar, 1990*
			Phyllopertha horticola	*Netherlands (5)*	*P. Smits, unpublished*
			Amphimallon solstitiale	*Netherlands (2 ×)*	*P. Smits, unpublished*
		Curculionidae	*Otiorhynchus sulcatus*	*Germany*	*R.-U. Ehlers, unpublished*
H. zealandica	*Coleoptera*	*Scarabaeidae*	*Heteronychus arator*	*New Zealand*	*Akhurst, 1987*
Heterorhabditis sp.	*Coleoptera*	*Elateridae*	*Agriotes ponticus*	*Moldavia (2)*	*E. Nesterov, unpublished*
		Scarabaeidae	*Phyllopertha horticola*	*Germany*	*R.-U. Ehlers, unpublished*
			Lepidiota crinita	*Australia*	*Akhurst et al., 1992*
			L. negatoria		*Akhurst et al., 1992*
			L. picticollis		*Akhurst et al., 1992*
			Antitrogus consanguineus		*Akhurst et al., 1992*
		Curculionidae	*Graphognathus leucoloma*	*Australia*	*See Akhurst et al., 1992; Klein, 1990*
			Cylas formicarius	*Cuba*	*Arteaga-Hernandez & Mráček, 1984*
			Pachneus litus	*Cuba*	*Arteaga-Hernandez & Mráček, 1984*
H. indicus, H. hawaiiensis, H. brevicaudis		*No information*	—	—	—

[a] *If occurring on more than one location, number in parentheses.*
[b] *Nematodes were isolated from adult insects instead of larvae.*
[c] *Nomen nudum.*



Nematode	Insect order	Family	Species	Geographical location[a]	References
	Diptera	Bibionidae	Bibio sp.	Denmark (2)	Bovien, 1937
	Lepidoptera	Noctuidae	Heliothis armigera	Australia	Poinar, 1990
			Crambus simplex	New Zealand	Hoy, 1954
			Agrotis ipsilon	New Zealand	Wright & Jackson, 1988
			Scotia segetum	Austria	Turco et al., 1971
			Agrotinae gen. sp.	Russia	E. Ivanova, unpublished
				Germany	R.-U. Ehlers, unpublished
S. glaseri	Coleoptera	Cerambicidae	Migdolus fryanus	Brazil	See Poinar, 1990
		Scarabaeidae	Popillia japonica	USA	Glaser & Fox, 1930
			Strigoderma arboricola	USA	See Poinar, 1986
			Anomala flavipennis	USA	See Poinar, 1992
S. kraussei	Hymenoptera	Pamphiliidae	Cephalcia abietis, C. falleni	Germany (2), Czech Republic, Austria	Steiner, 1923; Eichhorn, 1988; Mráček, 1986; Fischer & Führer, 1990
S. kushidai	Coleoptera	Scarabaeidae	Anomala cupre	Japan	Mamiya, 1988
S. rarum	Lepidoptera	Noctuidae	Heliothis sp.	Argentina	Doucet, 1986
S. riobravis	Lepidoptera	Noctuidae	Helicoverpa zea	USA	Raulston et al., 1992
			Spodoptera frugiperda	USA	Raulston et al., 1992
S. scapterisci	Saltatoria	Gryllotalpidae	Scapteriscus	Uruguay	Nguyen & Smart, 1990
			S. borelli	Argentina	Stock et al., 1995
			S. borelli	USA	Parkman & Frank, 1992
			S. vicinus	USA	Parkman & Frank, 1992
S. neocurtillis	Saltatoria	Gryllotalpidae	Neocurtilla hexadactyla	USA	Nguyen & Smart, 1992
Steinernema sp.	Coleoptera	Scarabaeidae	M. hippocastani	Russia	See Poinar, 1992
			M. afflicta	Russia	See Poinar, 1992
			Amphimallon solstitiale	Netherlands	P. Smits, unpublished
			Phyllopertha horticola	Netherlands (2)	P. Smits, unpublished
			Adoryphorus couloni	Australia	See Poinar, 1992
			Scitala sericans	Australia	See Poinar, 1992
		Curculionidae	Graphognathus sp.	Australia, USA	See Klein, 1990
			Acantholyda nemoralis	Poland	Weiser, 1955b
	Lepidoptera	Noctuidae	Agrotis ipsilon	Spain	Caballero et al., 1989
			Scotia segetum	Spain	Caballero et al., 1989
			Sesamia nonagrioides	Spain	C. Santiago-Alvarez, unpublished

Target pests for entomopathogenic nematodes (from: Lacey, L.A. and H.K. Kaya, eds. 2007. Field Manual of Techniques in Invertebrate Pathology)

Pest insect	Common name	life-stage[2]	Commodity	Nematode sp.[3]
COLEOPTERA				
Curculionidae	Billbugs	L	turf	Sc, Hb
	Root Weevils	L	berries, citrus, forest seedlings, hops, mint, ornamentals, sweet potato, sugar beets	Sc, Sk, Hb,Hi, Hm, Sr

Chrysomelidae	Flea beetles	L	mint, potato, sweet potato, sugar beets	Sc
Scarabeidae	Rootworms	L	corn, peanuts, vegetables	Sc, Sr
	White grubs	L	berries, field crops, ornamentals, turf	Hb, Sg, Hm
DIPTERA				
Agromyzidae	Leaf miners	L	ornamentals, vegetables	Sc
Ephydridae	Shore flies	L	ornamentals, vegetables	Sf
Sciaridae	Fungus gnats	L	ornamentals, vegetables, mushrooms	Sf
Tipulidae	Crane flies	L	turf, ornamentals	Sc, Hm
Muscidae	Filfth flies	A	animal rearing facilities	Sf, Hb
LEPIDOPTERA				
Noctuidae	Cutworms	L/P	corn, cotton, peanuts, turf, vegetables	Sc
	Armyworms	L	corn, cotton, peanuts, turf, vegetables	Sc
Pterophoridae	Plume moths	L	artichoke	Sc
Pyralidae	Webworms	L	cranberries, ornamentals, turf	Sc
Sessiidae	Crown borers	L	berries	Sc
	Stem borers	L	cucurbits, ornamentals, shrubs, fruit trees	Sc
Cossiidae	Carpenter worms	L	ornamentals, shrubs	Sc
	Leopard moth	L	apple, pear	Sc
Carposinidae	Peach borer moth	L	apple	Sc
ORTHOPTERA				
Gryllotalpidae	Mole crickets	N,A	turf, vegetables	Sc, Ss, Sr
BLATTODEA				
Blattellidae	German cockroach	N,A	apartments, structures	Sc
SIPHONAPTERA				
Pulicidae	cat fleas	L/P	pet/vet	Sc
NEMATODA				
Plant-parsitic nematodes	same	L/P	turf	Sc

---

[2] L= larva; P= pupa; N = nymph; A = adult

[3] Sc = Steinernema carpocapsae; Sf = S. feltiae; Sk = S. kraussei; Sr = S. riobrave; Ss = S. scapterisci; Hb = Heterorhabditis bacteriophora; Hi = H. indica; Hm = H. megidis

Characteristics of common EPN and infected host cadavers, taken from Lacey and Kaya (2007).

Nematode species	ij length (µm)	host cadaver color
S. carpocapsae	558 (468-650)	Beige
S. riobrave	622 (561-701)	Beige
S. feltiae	849 (736-950)	Tan/walnut brown
S. glaseri	1130 (864-1448)	Grayish- dark brown
S. kraussei	951 (797-1102)	Tan/walnut brown
H. bacteriophora	588 (512-670)	Brick red to dark purple
H. indica	528 (479-573)	Dark red
H. megidis	768 (736-800)	Orange brown
H. zealandica	685 (570-740)	Pale mint green

SAS code used for statistical analysis.

Pasture

```
data pasture;
input dates management cowpatty rep response;
run;

proc mixed;class dates management cowpatty rep;
model response = management cowpatty management*cowpatty dates
dates*management dates*cowpatty dates*management*cowpatty;
random management*cowpatty*rep;
lsmeans management cowpatty/diff;
lsmeans management*cowpatty/diff slice = (management cowpatty);
lsmeans dates dates*management/diff slice = (dates management);
lsmeans dates*cowpatty/diff slice = (dates cowpatty);
lsmeans dates*management*cowpatty/diff slice = (dates management
cowpatty);
run;

TITLE 'TEST FOR NORMALITY';
PROC UNIVARIATE DATA=pasture NORMALTEST;
CLASS dates management cowpatty rep;
VAR management;
RUN;
```

Wheat

```
data wheat;
```

```
input date management transect subsample rep response;
d=date;
run;

proc mixed;class date management transect subsample;
model response = management date subsample
date*management/ddfm=satterth;
random management*transect management*transect*subsample;
REPEATED/SUBJECT=management*transect*subsample TYPE=UN;
run;

proc mixed;class date management transect subsample;
model response = management date subsample
date*management/ddfm=satterth;
random management*transect management*transect*subsample;
```

```
REPEATED/SUBJECT=management*transect*subsample TYPE=cs;
run;

proc mixed;class date management transect subsample;
model response = management date subsample
date*management/ddfm=satterth;
random management*transect management*transect*subsample;
REPEATED/SUBJECT=management*transect*subsample TYPE=sp(sph);
run;

*pick one;
proc mixed;class date management transect subsample;
model response = management d d*d d*d*d subsample
date*management/htype= 1 ddfm=satterth;
random management*transect management*transect*subsample;
REPEATED/SUBJECT=management*transect*subsample TYPE=UN;
lsmeans management date*management/diff slice = (date management);
run;
```

Precipitation gradient

Wheat SAS Output

```
                                            The SAS System         11:50 Friday, January
13, 2012   1

                                         The Mixed Procedure

                                          Model Information

                        Data Set                     WORK.ARSINCONV
                        Dependent Variable           arsintotinf
                        Covariance Structures        Variance Components,
                                                     Compound Symmetry
                        Subject Effect               management*rep
                        Estimation Method            REML
                        Residual Variance Method     Profile
                        Fixed Effects SE Method      Model-Based
                        Degrees of Freedom Method    Satterthwaite


                                     Class Level Information

                        Class          Levels    Values

                        date                3    17-Oct 18-Nov 3-Sep
                        management          2    C O
                        rep                 4    1 2 3 4


                                           Dimensions

                                 Covariance Parameters             3
                                 Columns in X                     12
                                 Columns in Z                      8
                                 Subjects                          1
                                 Max Obs Per Subject              31


                                     Number of Observations

                             Number of Observations Read              31
```

```
                         Number of Observations Used              31
                         Number of Observations Not Used           0


                                      Iteration History

                 Iteration    Evaluations    -2 Res Log Like       Criterion

                         0              1      -150.52449393
                         1              2      -151.69392611      0.00317245
                         2              1      -151.88981921       .
                         3              1      -151.88984493      0.00000140
                         4              1      -151.88984518      0.00000000


                  Convergence criteria met but final hessian is not positive
                                        definite.

                                        The SAS System          11:50 Friday, January
13, 2012   2

                                       The Mixed Procedure

                                 Covariance Parameter Estimates

                        Cov Parm           Subject           Estimate

                        management*rep                       2.885E-8
                        CS                 management*rep    -0.00003
                        Residual                             0.000135


                                          Fit Statistics

                            -2 Res Log Likelihood          -151.9
                            AIC (smaller is better)        -145.9
                            AICC (smaller is better)       -144.7
                            BIC (smaller is better)        -145.7


                                  Type 3 Tests of Fixed Effects

                                         Num     Den
                     Effect               DF      DF    F Value    Pr > F

                     management            1    3.06      13.33    0.0343
                     date                  2    8.39      15.67    0.0015
                     date*management       2    8.39       2.32    0.1577


                                       Least Squares Means

                                                               Standard
      Effect        date      management    Estimate        Error      DF    t Value    Pr
> |t|

      management              C              0.01389    0.001564    3.06       8.88
0.0028
      management              O              0.02207    0.001606    3.07      13.74
0.0007
      date          17-Oct                   0.01042    0.003661    11.7       2.85
0.0151
      date          18-Nov                   0.03543    0.003661    11.7       9.68
<.0001
      date          3-Sep                   0.008101    0.002297    23.5       3.53
0.0018


                                Differences of Least Squares Means

                                                                 Standard
```

```
   Effect      date    management  _date    _management  Estimate     Error    DF  t Value
Pr > |t|

   management          C                    O           -0.00818  0.002242  3.06    -3.65
0.0343
   date        17-Oct              18-Nov               -0.02501  0.005808  8.12    -4.31
0.0025
   date        17-Oct              3-Sep                0.002316  0.005060   8.5     0.46
0.6586
   date        18-Nov              3-Sep                 0.02733  0.005060   8.5     5.40
0.0005
                                             The SAS System          11:50 Friday, January
13, 2012   3

                                 Obs     id       arsin     converted

                                  1     conv    0.013890     1.38896
                                  2     org     0.022070     2.20682
                                  3     oct     0.010420     1.04198
                                  4     nov     0.035430     3.54226
                                  5     sep     0.008101     0.81009
```

Pasture SAS Output

```
The SAS System          13:59 Monday, October 31, 2011   2

                                         The Mixed Procedure

                                          Model Information

                        Data Set                     WORK.PASTURE
                        Dependent Variable           response
                        Covariance Structure         Variance Components
                        Estimation Method            REML
                        Residual Variance Method     Profile
                        Fixed Effects SE Method      Model-Based
                        Degrees of Freedom Method    Containment


                                       Class Level Information

                        Class         Levels    Values

                        dates              3    17-Oct 18-Nov 3-Sep
                        management         2    C O
                        cowpatty           3    A F O
                        rep                6    1 2 3 4 5 6


                                             Dimensions

                                 Covariance Parameters             2
                                 Columns in X                     48
                                 Columns in Z                     36
                                 Subjects                          1
                                 Max Obs Per Subject             101


                                       Number of Observations

                             Number of Observations Read             101
                             Number of Observations Used             101
                             Number of Observations Not Used           0


                                          Iteration History
```

```
                     Iteration    Evaluations    -2 Res Log Like       Criterion

                             0              1       245.00992216
                             1              2       244.98897843      0.00000000


                                      Convergence criteria met.


                                            The SAS System         13:59 Monday, October
31, 2011   3

                                         The Mixed Procedure

                                   Covariance Parameter Estimates

                                  Cov Parm                 Estimate

                                  managem*cowpatty*rep      0.01304
                                  Residual                   0.7591


                                           Fit Statistics

                                -2 Res Log Likelihood          245.0
                                AIC (smaller is better)        249.0
                                AICC (smaller is better)       249.1
                                BIC (smaller is better)        252.2


                                    Type 3 Tests of Fixed Effects

                                               Num     Den
                      Effect                    DF      DF    F Value    Pr > F

                      management                 1      30       0.06    0.8074
                      cowpatty                   2      30       1.46    0.2483
                      management*cowpatty        2      30       1.82    0.1788
                      dates                      2      53       0.52    0.5948
                      dates*management           2      53       7.15    0.0018
                      dates*cowpatty             4      53       2.14    0.0888
                      dates*manage*cowpatt       4      53       0.43    0.7828


                                         Least Squares Means

                                                                 Standard
   Effect                 dates   management  cowpatty  Estimate     Error    DF  t Value
Pr > |t|

   management                     C                       0.4147    0.1254    30     3.31
0.0025
   management                     O                       0.4586    0.1273    30     3.60
0.0011
   cowpatty                                   A           0.2226    0.1536    30     1.45
0.1577
   cowpatty                                   F           0.5599    0.1536    30     3.64
0.0010
   cowpatty                                   O           0.5275    0.1571    30     3.36
0.0021
   management*cowpatty            C           A           0.1118    0.2173    30     0.51
0.6104
   management*cowpatty            C           F           0.7763    0.2173    30     3.57
0.0012
   management*cowpatty            C           O           0.3559    0.2173    30     1.64
0.1118
   management*cowpatty            O           A           0.3333    0.2173    30     1.53
0.1354
   management*cowpatty            O           F           0.3435    0.2173    30     1.58
0.1243
```

```
   management*cowpatty           O           O           0.6991    0.2269    30     3.08
0.0044
   dates                 17-Oct                          0.4722    0.1464    53     3.22
0.0022
   dates                 18-Nov                          0.5278    0.1464    53     3.60
0.0007
   dates                 3-Sep                           0.3100    0.1637    53     1.89
0.0638
   dates*management      17-Oct  C                       0.7222    0.2071    53     3.49
0.0010
   dates*management      17-Oct  O                       0.2222    0.2071    53     1.07
0.2881
   dates*management      18-Nov  C                      0.05556    0.2071    53     0.27
0.7896

                                            The SAS System          13:59 Monday, October
31, 2011   4

                                         The Mixed Procedure

                                         Least Squares Means

                                                                 Standard
   Effect                dates   management  cowpatty  Estimate     Error    DF  t Value
Pr > |t|

   dates*management      18-Nov  O                       1.0000    0.2071    53     4.83
<.0001
   dates*management      3-Sep   C                       0.4663    0.2269    53     2.06
0.0448
   dates*management      3-Sep   O                       0.1537    0.2361    53     0.65
0.5179
   dates*cowpatty        17-Oct              A          0.08333    0.2537    53     0.33
0.7438
   dates*cowpatty        17-Oct              F           0.9167    0.2537    53     3.61
0.0007
   dates*cowpatty        17-Oct              O           0.4167    0.2537    53     1.64
0.1064
   dates*cowpatty        18-Nov              A           0.5833    0.2537    53     2.30
0.0254
   dates*cowpatty        18-Nov              F           0.1667    0.2537    53     0.66
0.5140
   dates*cowpatty        18-Nov              O           0.8333    0.2537    53     3.29
0.0018
   dates*cowpatty        3-Sep               A         0.001107    0.2779    53     0.00
0.9968
   dates*cowpatty        3-Sep               F           0.5964    0.2779    53     2.15
0.0364
   dates*cowpatty        3-Sep               O           0.3325    0.2947    53     1.13
0.2643
   dates*manage*cowpatt  17-Oct  C           A           0.1667    0.3587    53     0.46
0.6441
   dates*manage*cowpatt  17-Oct  C           F           1.3333    0.3587    53     3.72
0.0005
   dates*manage*cowpatt  17-Oct  C           O           0.6667    0.3587    53     1.86
0.0687
   dates*manage*cowpatt  17-Oct  O           A         1.11E-16    0.3587    53     0.00
1.0000
   dates*manage*cowpatt  17-Oct  O           F           0.5000    0.3587    53     1.39
0.1692
   dates*manage*cowpatt  17-Oct  O           O           0.1667    0.3587    53     0.46
0.6441
   dates*manage*cowpatt  18-Nov  C           A           0.1667    0.3587    53     0.46
0.6441
   dates*manage*cowpatt  18-Nov  C           F         -305E-18    0.3587    53    -0.00
1.0000
   dates*manage*cowpatt  18-Nov  C           O         -833E-18    0.3587    53    -0.00
1.0000
   dates*manage*cowpatt  18-Nov  O           A           1.0000    0.3587    53     2.79
0.0074
```

```
   dates*manage*cowpatt  18-Nov  O           F           0.3333    0.3587    53     0.93
0.3570
   dates*manage*cowpatt  18-Nov  O           O           1.6667    0.3587    53     4.65
<.0001
   dates*manage*cowpatt  3-Sep   C           A         0.002215    0.3929    53     0.01
0.9955
   dates*manage*cowpatt  3-Sep   C           F           0.9956    0.3929    53     2.53
0.0143
   dates*manage*cowpatt  3-Sep   C           O           0.4011    0.3929    53     1.02
0.3120
   dates*manage*cowpatt  3-Sep   O           A         -777E-18    0.3929    53    -0.00
1.0000
   dates*manage*cowpatt  3-Sep   O           F           0.1972    0.3929    53     0.50
0.6178
   dates*manage*cowpatt  3-Sep   O           O           0.2638    0.4393    53     0.60
0.5507


                                  Differences of Least Squares Means


Standard
Effect                dates   management  cowpatty  _dates  _management  _cowpatty
Estimate     Error

management                    C                             O                       -
0.04395    0.1787
cowpatty                                  A                              F           -
0.3373    0.2173
cowpatty                                  A                              O           -
0.3049    0.2197
cowpatty                                  F                              O
0.03242    0.2197
management*cowpatty           C           A                 C            F           -
0.6645    0.3072
management*cowpatty           C           A                 C            O           -
0.2441    0.3072
management*cowpatty           C           A                 O            A           -
0.2215    0.3072
management*cowpatty           C           A                 O            F           -
0.2317    0.3072
management*cowpatty           C           A                 O            O           -
0.5872    0.3141
management*cowpatty           C           F                 C            O
0.4204    0.3072
management*cowpatty           C           F                 O            A
0.4430    0.3072
management*cowpatty           C           F                 O            F
0.4328    0.3072
management*cowpatty           C           F                 O            O
0.07724    0.3141
management*cowpatty           C           O                 O            A
0.02259    0.3072
management*cowpatty           C           O                 O            F
0.01240    0.3072
management*cowpatty           C           O                 O            O           -
0.3431    0.3141
management*cowpatty           O           A                 O            F          -
0.01019    0.3072
management*cowpatty           O           A                 O            O           -
0.3657    0.3141
management*cowpatty           O           F                 O            O           -
0.3555    0.3141
dates                 17-Oct                      18-Nov                            -
0.05556    0.2054
dates                 17-Oct                      3-Sep
0.1622    0.2180
dates                 18-Nov                      3-Sep
0.2178    0.2180
dates*management      17-Oct  C                   17-Oct  O
0.5000    0.2929
```

```
dates*management      17-Oct  C                      18-Nov  C
0.6667    0.2904
dates*management      17-Oct  C                      18-Nov  O                         -
0.2778    0.2929
dates*management      17-Oct  C                      3-Sep   C
0.2559    0.3048
dates*management      17-Oct  C                      3-Sep   O
0.5685    0.3141
dates*management      17-Oct  O                      18-Nov  C
0.1667    0.2929
dates*management      17-Oct  O                      18-Nov  O                         -
0.7778    0.2904
dates*management      17-Oct  O                      3-Sep   C                         -
0.2441    0.3072
dates*management      17-Oct  O                      3-Sep   O
0.06853    0.3118
dates*management      18-Nov  C                      18-Nov  O                         -
0.9444    0.2929
dates*management      18-Nov  C                      3-Sep   C                         -
0.4107    0.3048
dates*management      18-Nov  C                      3-Sep   O                        -
0.09814    0.3141
dates*management      18-Nov  O                      3-Sep   C
0.5337    0.3072
dates*management      18-Nov  O                      3-Sep   O
0.8463    0.3118
dates*management      3-Sep   C                      3-Sep   O
0.3126    0.3274
dates*cowpatty        17-Oct               A         17-Oct               F            -
0.8333    0.3587
dates*cowpatty        17-Oct               A         17-Oct               O            -
0.3333    0.3587
dates*cowpatty        17-Oct               A         18-Nov               A            -
0.5000    0.3557
dates*cowpatty        17-Oct               A         18-Nov               F           -
0.08333    0.3587
dates*cowpatty        17-Oct               A         18-Nov               O            -
0.7500    0.3587
dates*cowpatty        17-Oct               A         3-Sep                A
0.08223    0.3733
dates*cowpatty        17-Oct               A         3-Sep                F            -
0.5131    0.3762
dates*cowpatty        17-Oct               A         3-Sep                O            -
0.2491    0.3888
dates*cowpatty        17-Oct               F         17-Oct               O
0.5000    0.3587
dates*cowpatty        17-Oct               F         18-Nov               A
0.3333    0.3587
dates*cowpatty        17-Oct               F         18-Nov               F
0.7500    0.3557
dates*cowpatty        17-Oct               F         18-Nov               O
0.08333    0.3587
dates*cowpatty        17-Oct               F         3-Sep                A
0.9156    0.3762
dates*cowpatty        17-Oct               F         3-Sep                F
0.3203    0.3733
dates*cowpatty        17-Oct               F         3-Sep                O
0.5842    0.3888
dates*cowpatty        17-Oct               O         18-Nov               A            -
0.1667    0.3587
dates*cowpatty        17-Oct               O         18-Nov               F
0.2500    0.3587
dates*cowpatty        17-Oct               O         18-Nov               O            -
0.4167    0.3557
dates*cowpatty        17-Oct               O         3-Sep                A
0.4156    0.3762
dates*cowpatty        17-Oct               O         3-Sep                F            -
0.1797    0.3762
dates*cowpatty        17-Oct               O         3-Sep                O
0.08419    0.3860
```

```
dates*cowpatty        18-Nov              A         18-Nov               F
0.4167    0.3587
dates*cowpatty        18-Nov              A         18-Nov               O          -
0.2500    0.3587
dates*cowpatty        18-Nov              A         3-Sep                A
0.5822    0.3733
dates*cowpatty        18-Nov              A         3-Sep                F         -
0.01307    0.3762
dates*cowpatty        18-Nov              A         3-Sep                O
0.2509    0.3888
dates*cowpatty        18-Nov              F         18-Nov               O          -
0.6667    0.3587
dates*cowpatty        18-Nov              F         3-Sep                A
0.1656    0.3762
dates*cowpatty        18-Nov              F         3-Sep                F          -
0.4297    0.3733
dates*cowpatty        18-Nov              F         3-Sep                O          -
0.1658    0.3888
dates*cowpatty        18-Nov              O         3-Sep                A
0.8322    0.3762
dates*cowpatty        18-Nov              O         3-Sep                F
0.2369    0.3762
dates*cowpatty        18-Nov              O         3-Sep                O
0.5009    0.3860
dates*cowpatty        3-Sep               A         3-Sep                F          -
0.5953    0.3929
dates*cowpatty        3-Sep               A         3-Sep                O          -
0.3314    0.4050
dates*cowpatty        3-Sep               F         3-Sep                O
0.2639    0.4050
dates*manage*cowpatt  17-Oct  C           A         17-Oct  C            F          -
1.1667    0.5073
dates*manage*cowpatt  17-Oct  C           A         17-Oct  C            O          -
0.5000    0.5073
dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            A
0.1667    0.5073
dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            F          -
0.3333    0.5073
dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            O
9.99E-16    0.5073
dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            A
4.44E-16    0.5030
dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            F
0.1667    0.5073
dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            O
0.1667    0.5073
dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            A          -
0.8333    0.5073
dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            F          -
0.1667    0.5073
dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            O          -
1.5000    0.5073
dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            A
0.1645    0.5280
dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            F          -
0.8289    0.5321
dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            O          -
0.2344    0.5321
dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            A
0.1667    0.5321
dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            F         -
0.03057    0.5321
dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            O         -
0.09717    0.5672
dates*manage*cowpatt  17-Oct  C           F         17-Oct  C            O
0.6667    0.5073
dates*manage*cowpatt  17-Oct  C           F         17-Oct  O            A
1.3333    0.5073
dates*manage*cowpatt  17-Oct  C           F         17-Oct  O            F
0.8333    0.5073
```

```
dates*manage*cowpatt  17-Oct  C           F         17-Oct  O            O
1.1667    0.5073
dates*manage*cowpatt  17-Oct  C           F         18-Nov  C            A
1.1667    0.5073
dates*manage*cowpatt  17-Oct  C           F         18-Nov  C            F
1.3333    0.5030
dates*manage*cowpatt  17-Oct  C           F         18-Nov  C            O
1.3333    0.5073
dates*manage*cowpatt  17-Oct  C           F         18-Nov  O            A
0.3333    0.5073
dates*manage*cowpatt  17-Oct  C           F         18-Nov  O            F
1.0000    0.5073
dates*manage*cowpatt  17-Oct  C           F         18-Nov  O            O            -
0.3333    0.5073
dates*manage*cowpatt  17-Oct  C           F         3-Sep   C            A
1.3311    0.5321
dates*manage*cowpatt  17-Oct  C           F         3-Sep   C            F
0.3378    0.5280
dates*manage*cowpatt  17-Oct  C           F         3-Sep   C            O
0.9322    0.5321
dates*manage*cowpatt  17-Oct  C           F         3-Sep   O            A
1.3333    0.5321
dates*manage*cowpatt  17-Oct  C           F         3-Sep   O            F
1.1361    0.5321
dates*manage*cowpatt  17-Oct  C           F         3-Sep   O            O
1.0695    0.5672
dates*manage*cowpatt  17-Oct  C           O         17-Oct  O            A
0.6667    0.5073
dates*manage*cowpatt  17-Oct  C           O         17-Oct  O            F
0.1667    0.5073
dates*manage*cowpatt  17-Oct  C           O         17-Oct  O            O
0.5000    0.5073
dates*manage*cowpatt  17-Oct  C           O         18-Nov  C            A
0.5000    0.5073
dates*manage*cowpatt  17-Oct  C           O         18-Nov  C            F
0.6667    0.5073
dates*manage*cowpatt  17-Oct  C           O         18-Nov  C            O
0.6667    0.5030
dates*manage*cowpatt  17-Oct  C           O         18-Nov  O            A            -
0.3333    0.5073
dates*manage*cowpatt  17-Oct  C           O         18-Nov  O            F
0.3333    0.5073
dates*manage*cowpatt  17-Oct  C           O         18-Nov  O            O            -
1.0000    0.5073
dates*manage*cowpatt  17-Oct  C           O         3-Sep   C            A
0.6645    0.5321
dates*manage*cowpatt  17-Oct  C           O         3-Sep   C            F            -
0.3289    0.5321
dates*manage*cowpatt  17-Oct  C           O         3-Sep   C            O
0.2656    0.5280
dates*manage*cowpatt  17-Oct  C           O         3-Sep   O            A
0.6667    0.5321
dates*manage*cowpatt  17-Oct  C           O         3-Sep   O            F
0.4694    0.5321
dates*manage*cowpatt  17-Oct  C           O         3-Sep   O            O
0.4028    0.5672
dates*manage*cowpatt  17-Oct  O           A         17-Oct  O            F            -
0.5000    0.5073
dates*manage*cowpatt  17-Oct  O           A         17-Oct  O            O            -
0.1667    0.5073
dates*manage*cowpatt  17-Oct  O           A         18-Nov  C            A            -
0.1667    0.5073
dates*manage*cowpatt  17-Oct  O           A         18-Nov  C            F
4.72E-16    0.5073
dates*manage*cowpatt  17-Oct  O           A         18-Nov  C            O
9.99E-16    0.5073
dates*manage*cowpatt  17-Oct  O           A         18-Nov  O            A            -
1.0000    0.5030
dates*manage*cowpatt  17-Oct  O           A         18-Nov  O            F            -
0.3333    0.5073
```

```
dates*manage*cowpatt  17-Oct  O           A         18-Nov  O            O             -
1.6667    0.5073
dates*manage*cowpatt  17-Oct  O           A         3-Sep   C            A            -
0.00221    0.5321
dates*manage*cowpatt  17-Oct  O           A         3-Sep   C            F             -
0.9956    0.5321
dates*manage*cowpatt  17-Oct  O           A         3-Sep   C            O             -
0.4011    0.5321
dates*manage*cowpatt  17-Oct  O           A         3-Sep   O            A
8.88E-16    0.5280
dates*manage*cowpatt  17-Oct  O           A         3-Sep   O            F             -
0.1972    0.5321
dates*manage*cowpatt  17-Oct  O           A         3-Sep   O            O             -
0.2638    0.5672
dates*manage*cowpatt  17-Oct  O           F         17-Oct  O            O
0.3333    0.5073
dates*manage*cowpatt  17-Oct  O           F         18-Nov  C            A
0.3333    0.5073
dates*manage*cowpatt  17-Oct  O           F         18-Nov  C            F
0.5000    0.5073
dates*manage*cowpatt  17-Oct  O           F         18-Nov  C            O
0.5000    0.5073
dates*manage*cowpatt  17-Oct  O           F         18-Nov  O            A             -
0.5000    0.5073
dates*manage*cowpatt  17-Oct  O           F         18-Nov  O            F
0.1667    0.5030
dates*manage*cowpatt  17-Oct  O           F         18-Nov  O            O             -
1.1667    0.5073
dates*manage*cowpatt  17-Oct  O           F         3-Sep   C            A
0.4978    0.5321
dates*manage*cowpatt  17-Oct  O           F         3-Sep   C            F             -
0.4956    0.5321
dates*manage*cowpatt  17-Oct  O           F         3-Sep   C            O
0.09889    0.5321
dates*manage*cowpatt  17-Oct  O           F         3-Sep   O            A
0.5000    0.5321
dates*manage*cowpatt  17-Oct  O           F         3-Sep   O            F
0.3028    0.5280
dates*manage*cowpatt  17-Oct  O           F         3-Sep   O            O
0.2362    0.5672
dates*manage*cowpatt  17-Oct  O           O         18-Nov  C            A            -
666E-18    0.5073
dates*manage*cowpatt  17-Oct  O           O         18-Nov  C            F
0.1667    0.5073
dates*manage*cowpatt  17-Oct  O           O         18-Nov  C            O
0.1667    0.5073
dates*manage*cowpatt  17-Oct  O           O         18-Nov  O            A             -
0.8333    0.5073
dates*manage*cowpatt  17-Oct  O           O         18-Nov  O            F             -
0.1667    0.5073
dates*manage*cowpatt  17-Oct  O           O         18-Nov  O            O             -
1.5000    0.5030
dates*manage*cowpatt  17-Oct  O           O         3-Sep   C            A
0.1645    0.5321
dates*manage*cowpatt  17-Oct  O           O         3-Sep   C            F             -
0.8289    0.5321
dates*manage*cowpatt  17-Oct  O           O         3-Sep   C            O             -
0.2344    0.5321
dates*manage*cowpatt  17-Oct  O           O         3-Sep   O            A
0.1667    0.5321
dates*manage*cowpatt  17-Oct  O           O         3-Sep   O            F            -
0.03057    0.5321
dates*manage*cowpatt  17-Oct  O           O         3-Sep   O            O            -
0.09717    0.5633
dates*manage*cowpatt  18-Nov  C           A         18-Nov  C            F
0.1667    0.5073
dates*manage*cowpatt  18-Nov  C           A         18-Nov  C            O
0.1667    0.5073
dates*manage*cowpatt  18-Nov  C           A         18-Nov  O            A             -
0.8333    0.5073
```

```
dates*manage*cowpatt  18-Nov  C           A         18-Nov  O            F           -
0.1667    0.5073
dates*manage*cowpatt  18-Nov  C           A         18-Nov  O            O           -
1.5000    0.5073
dates*manage*cowpatt  18-Nov  C           A         3-Sep   C            A
0.1645    0.5280
dates*manage*cowpatt  18-Nov  C           A         3-Sep   C            F           -
0.8289    0.5321
dates*manage*cowpatt  18-Nov  C           A         3-Sep   C            O           -
0.2344    0.5321
dates*manage*cowpatt  18-Nov  C           A         3-Sep   O            A
0.1667    0.5321
dates*manage*cowpatt  18-Nov  C           A         3-Sep   O            F          -
0.03057    0.5321
dates*manage*cowpatt  18-Nov  C           A         3-Sep   O            O          -
0.09717    0.5672
dates*manage*cowpatt  18-Nov  C           F         18-Nov  C            O
5.27E-16    0.5073
dates*manage*cowpatt  18-Nov  C           F         18-Nov  O            A           -
1.0000    0.5073
dates*manage*cowpatt  18-Nov  C           F         18-Nov  O            F           -
0.3333    0.5073
dates*manage*cowpatt  18-Nov  C           F         18-Nov  O            O           -
1.6667    0.5073
dates*manage*cowpatt  18-Nov  C           F         3-Sep   C            A          -
0.00221    0.5321
dates*manage*cowpatt  18-Nov  C           F         3-Sep   C            F           -
0.9956    0.5280
dates*manage*cowpatt  18-Nov  C           F         3-Sep   C            O           -
0.4011    0.5321
dates*manage*cowpatt  18-Nov  C           F         3-Sep   O            A
5.27E-16    0.5321
dates*manage*cowpatt  18-Nov  C           F         3-Sep   O            F           -
0.1972    0.5321
dates*manage*cowpatt  18-Nov  C           F         3-Sep   O            O           -
0.2638    0.5672
dates*manage*cowpatt  18-Nov  C           O         18-Nov  O            A           -
1.0000    0.5073
dates*manage*cowpatt  18-Nov  C           O         18-Nov  O            F           -
0.3333    0.5073
dates*manage*cowpatt  18-Nov  C           O         18-Nov  O            O           -
1.6667    0.5073
dates*manage*cowpatt  18-Nov  C           O         3-Sep   C            A          -
0.00221    0.5321
dates*manage*cowpatt  18-Nov  C           O         3-Sep   C            F           -
0.9956    0.5321
dates*manage*cowpatt  18-Nov  C           O         3-Sep   C            O           -
0.4011    0.5280
dates*manage*cowpatt  18-Nov  C           O         3-Sep   O            A
0    0.5321
dates*manage*cowpatt  18-Nov  C           O         3-Sep   O            F           -
0.1972    0.5321
dates*manage*cowpatt  18-Nov  C           O         3-Sep   O            O           -
0.2638    0.5672
dates*manage*cowpatt  18-Nov  O           A         18-Nov  O            F
0.6667    0.5073
dates*manage*cowpatt  18-Nov  O           A         18-Nov  O            O           -
0.6667    0.5073
dates*manage*cowpatt  18-Nov  O           A         3-Sep   C            A
0.9978    0.5321
dates*manage*cowpatt  18-Nov  O           A         3-Sep   C            F
0.004429    0.5321
dates*manage*cowpatt  18-Nov  O           A         3-Sep   C            O
0.5989    0.5321
dates*manage*cowpatt  18-Nov  O           A         3-Sep   O            A
1.0000    0.5280
dates*manage*cowpatt  18-Nov  O           A         3-Sep   O            F
0.8028    0.5321
dates*manage*cowpatt  18-Nov  O           A         3-Sep   O            O
0.7362    0.5672
```

```
dates*manage*cowpatt  18-Nov  O           F         18-Nov  O            O           -
1.3333    0.5073
dates*manage*cowpatt  18-Nov  O           F         3-Sep   C            A
0.3311    0.5321
dates*manage*cowpatt  18-Nov  O           F         3-Sep   C            F           -
0.6622    0.5321
dates*manage*cowpatt  18-Nov  O           F         3-Sep   C            O          -
0.06777    0.5321
dates*manage*cowpatt  18-Nov  O           F         3-Sep   O            A
0.3333    0.5321
dates*manage*cowpatt  18-Nov  O           F         3-Sep   O            F
0.1361    0.5280
dates*manage*cowpatt  18-Nov  O           F         3-Sep   O            O
0.06949    0.5672
dates*manage*cowpatt  18-Nov  O           O         3-Sep   C            A
1.6645    0.5321
dates*manage*cowpatt  18-Nov  O           O         3-Sep   C            F
0.6711    0.5321
dates*manage*cowpatt  18-Nov  O           O         3-Sep   C            O
1.2656    0.5321
dates*manage*cowpatt  18-Nov  O           O         3-Sep   O            A
1.6667    0.5321
dates*manage*cowpatt  18-Nov  O           O         3-Sep   O            F
1.4694    0.5321
dates*manage*cowpatt  18-Nov  O           O         3-Sep   O            O
1.4028    0.5633
dates*manage*cowpatt  3-Sep   C           A         3-Sep   C            F           -
0.9934    0.5557
dates*manage*cowpatt  3-Sep   C           A         3-Sep   C            O           -
0.3989    0.5557
dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            A
0.002215    0.5557
dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            F           -
0.1950    0.5557
dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            O           -
0.2616    0.5894
dates*manage*cowpatt  3-Sep   C           F         3-Sep   C            O
0.5945    0.5557
dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            A
0.9956    0.5557
dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            F
0.7983    0.5557
dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            O
0.7317    0.5894
dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            A
0.4011    0.5557
dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            F
0.2039    0.5557
dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            O
0.1373    0.5894
dates*manage*cowpatt  3-Sep   O           A         3-Sep   O            F           -
0.1972    0.5557
dates*manage*cowpatt  3-Sep   O           A         3-Sep   O            O           -
0.2638    0.5894
dates*manage*cowpatt  3-Sep   O           F         3-Sep   O            O          -
0.06661    0.5894

                                 Differences of Least Squares Means

  Effect                dates   management  cowpatty  _dates  _management  _cowpatty
DF  t Value

  management                    C                             O
30    -0.25
  cowpatty                                  A                              F
30    -1.55
  cowpatty                                  A                              O
30    -1.39
  cowpatty                                  F                              O
30     0.15
```

```
  management*cowpatty           C           A                 C            F
30    -2.16
  management*cowpatty           C           A                 C            O
30    -0.79
  management*cowpatty           C           A                 O            A
30    -0.72
  management*cowpatty           C           A                 O            F
30    -0.75
  management*cowpatty           C           A                 O            O
30    -1.87
  management*cowpatty           C           F                 C            O
30     1.37
  management*cowpatty           C           F                 O            A
30     1.44
  management*cowpatty           C           F                 O            F
30     1.41
  management*cowpatty           C           F                 O            O
30     0.25
  management*cowpatty           C           O                 O            A
30     0.07
  management*cowpatty           C           O                 O            F
30     0.04
  management*cowpatty           C           O                 O            O
30    -1.09
  management*cowpatty           O           A                 O            F
30    -0.03
  management*cowpatty           O           A                 O            O
30    -1.16
  management*cowpatty           O           F                 O            O
30    -1.13
  dates                 17-Oct                        18-Nov
53    -0.27
  dates                 17-Oct                        3-Sep
53     0.74
  dates                 18-Nov                        3-Sep
53     1.00
  dates*management      17-Oct  C                     17-Oct  O
53     1.71
  dates*management      17-Oct  C                     18-Nov  C
53     2.30
  dates*management      17-Oct  C                     18-Nov  O
53    -0.95
  dates*management      17-Oct  C                     3-Sep   C
53     0.84
  dates*management      17-Oct  C                     3-Sep   O
53     1.81
  dates*management      17-Oct  O                     18-Nov  C
53     0.57
  dates*management      17-Oct  O                     18-Nov  O
53    -2.68
  dates*management      17-Oct  O                     3-Sep   C
53    -0.79
  dates*management      17-Oct  O                     3-Sep   O
53     0.22
  dates*management      18-Nov  C                     18-Nov  O
53    -3.22
  dates*management      18-Nov  C                     3-Sep   C
53    -1.35
  dates*management      18-Nov  C                     3-Sep   O
53    -0.31
  dates*management      18-Nov  O                     3-Sep   C
53     1.74
  dates*management      18-Nov  O                     3-Sep   O
53     2.71
  dates*management      3-Sep   C                     3-Sep   O
53     0.95
  dates*cowpatty        17-Oct              A         17-Oct               F
53    -2.32
  dates*cowpatty        17-Oct              A         17-Oct               O
53    -0.93
```

```
  dates*cowpatty        17-Oct              A         18-Nov               A
53    -1.41
  dates*cowpatty        17-Oct              A         18-Nov               F
53    -0.23
  dates*cowpatty        17-Oct              A         18-Nov               O
53    -2.09
  dates*cowpatty        17-Oct              A         3-Sep                A
53     0.22
  dates*cowpatty        17-Oct              A         3-Sep                F
53    -1.36
  dates*cowpatty        17-Oct              A         3-Sep                O
53    -0.64
  dates*cowpatty        17-Oct              F         17-Oct               O
53     1.39
  dates*cowpatty        17-Oct              F         18-Nov               A
53     0.93
  dates*cowpatty        17-Oct              F         18-Nov               F
53     2.11
  dates*cowpatty        17-Oct              F         18-Nov               O
53     0.23
  dates*cowpatty        17-Oct              F         3-Sep                A
53     2.43
  dates*cowpatty        17-Oct              F         3-Sep                F
53     0.86
  dates*cowpatty        17-Oct              F         3-Sep                O
53     1.50
  dates*cowpatty        17-Oct              O         18-Nov               A
53    -0.46
  dates*cowpatty        17-Oct              O         18-Nov               F
53     0.70
  dates*cowpatty        17-Oct              O         18-Nov               O
53    -1.17
  dates*cowpatty        17-Oct              O         3-Sep                A
53     1.10
  dates*cowpatty        17-Oct              O         3-Sep                F
53    -0.48
  dates*cowpatty        17-Oct              O         3-Sep                O
53     0.22
  dates*cowpatty        18-Nov              A         18-Nov               F
53     1.16
  dates*cowpatty        18-Nov              A         18-Nov               O
53    -0.70
  dates*cowpatty        18-Nov              A         3-Sep                A
53     1.56
  dates*cowpatty        18-Nov              A         3-Sep                F
53    -0.03
  dates*cowpatty        18-Nov              A         3-Sep                O
53     0.65
  dates*cowpatty        18-Nov              F         18-Nov               O
53    -1.86
  dates*cowpatty        18-Nov              F         3-Sep                A
53     0.44
  dates*cowpatty        18-Nov              F         3-Sep                F
53    -1.15
  dates*cowpatty        18-Nov              F         3-Sep                O
53    -0.43
  dates*cowpatty        18-Nov              O         3-Sep                A
53     2.21
  dates*cowpatty        18-Nov              O         3-Sep                F
53     0.63
  dates*cowpatty        18-Nov              O         3-Sep                O
53     1.30
  dates*cowpatty        3-Sep               A         3-Sep                F
53    -1.51
  dates*cowpatty        3-Sep               A         3-Sep                O
53    -0.82
  dates*cowpatty        3-Sep               F         3-Sep                O
53     0.65
  dates*manage*cowpatt  17-Oct  C           A         17-Oct  C            F
53    -2.30
```

```
  dates*manage*cowpatt  17-Oct  C           A         17-Oct  C            O
53    -0.99
  dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            A
53     0.33
  dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            F
53    -0.66
  dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            O
53     0.00
  dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            A
53     0.00
  dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            F
53     0.33
  dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            O
53     0.33
  dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            A
53    -1.64
  dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            F
53    -0.33
  dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            O
53    -2.96
  dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            A
53     0.31
  dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            F
53    -1.56
  dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            O
53    -0.44
  dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            A
53     0.31
  dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            F
53    -0.06
  dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            O
53    -0.17
  dates*manage*cowpatt  17-Oct  C           F         17-Oct  C            O
53     1.31
  dates*manage*cowpatt  17-Oct  C           F         17-Oct  O            A
53     2.63
  dates*manage*cowpatt  17-Oct  C           F         17-Oct  O            F
53     1.64
  dates*manage*cowpatt  17-Oct  C           F         17-Oct  O            O
53     2.30
  dates*manage*cowpatt  17-Oct  C           F         18-Nov  C            A
53     2.30
  dates*manage*cowpatt  17-Oct  C           F         18-Nov  C            F
53     2.65
  dates*manage*cowpatt  17-Oct  C           F         18-Nov  C            O
53     2.63
  dates*manage*cowpatt  17-Oct  C           F         18-Nov  O            A
53     0.66
  dates*manage*cowpatt  17-Oct  C           F         18-Nov  O            F
53     1.97
  dates*manage*cowpatt  17-Oct  C           F         18-Nov  O            O
53    -0.66
  dates*manage*cowpatt  17-Oct  C           F         3-Sep   C            A
53     2.50
  dates*manage*cowpatt  17-Oct  C           F         3-Sep   C            F
53     0.64
  dates*manage*cowpatt  17-Oct  C           F         3-Sep   C            O
53     1.75
  dates*manage*cowpatt  17-Oct  C           F         3-Sep   O            A
53     2.51
  dates*manage*cowpatt  17-Oct  C           F         3-Sep   O            F
53     2.14
  dates*manage*cowpatt  17-Oct  C           F         3-Sep   O            O
53     1.89
  dates*manage*cowpatt  17-Oct  C           O         17-Oct  O            A
53     1.31
  dates*manage*cowpatt  17-Oct  C           O         17-Oct  O            F
53     0.33
  dates*manage*cowpatt  17-Oct  C           O         17-Oct  O            O
53     0.99
```

```
  dates*manage*cowpatt  17-Oct  C           O         18-Nov  C             A
53     0.99
  dates*manage*cowpatt  17-Oct  C           O         18-Nov  C             F
53     1.31
  dates*manage*cowpatt  17-Oct  C           O         18-Nov  C             O
53     1.33
  dates*manage*cowpatt  17-Oct  C           O         18-Nov  O             A
53    -0.66
  dates*manage*cowpatt  17-Oct  C           O         18-Nov  O             F
53     0.66
  dates*manage*cowpatt  17-Oct  C           O         18-Nov  O             O
53    -1.97
  dates*manage*cowpatt  17-Oct  C           O         3-Sep   C             A
53     1.25
  dates*manage*cowpatt  17-Oct  C           O         3-Sep   C             F
53    -0.62
  dates*manage*cowpatt  17-Oct  C           O         3-Sep   C             O
53     0.50
  dates*manage*cowpatt  17-Oct  C           O         3-Sep   O             A
53     1.25
  dates*manage*cowpatt  17-Oct  C           O         3-Sep   O             F
53     0.88
  dates*manage*cowpatt  17-Oct  C           O         3-Sep   O             O
53     0.71
  dates*manage*cowpatt  17-Oct  O           A         17-Oct  O             F
53    -0.99
  dates*manage*cowpatt  17-Oct  O           A         17-Oct  O             O
53    -0.33
  dates*manage*cowpatt  17-Oct  O           A         18-Nov  C             A
53    -0.33
  dates*manage*cowpatt  17-Oct  O           A         18-Nov  C             F
53     0.00
  dates*manage*cowpatt  17-Oct  O           A         18-Nov  C             O
53     0.00
  dates*manage*cowpatt  17-Oct  O           A         18-Nov  O             A
53    -1.99
  dates*manage*cowpatt  17-Oct  O           A         18-Nov  O             F
53    -0.66
  dates*manage*cowpatt  17-Oct  O           A         18-Nov  O             O
53    -3.29
  dates*manage*cowpatt  17-Oct  O           A         3-Sep   C             A
53    -0.00
  dates*manage*cowpatt  17-Oct  O           A         3-Sep   C             F
53    -1.87
  dates*manage*cowpatt  17-Oct  O           A         3-Sep   C             O
53    -0.75
  dates*manage*cowpatt  17-Oct  O           A         3-Sep   O             A
53     0.00
  dates*manage*cowpatt  17-Oct  O           A         3-Sep   O             F
53    -0.37
  dates*manage*cowpatt  17-Oct  O           A         3-Sep   O             O
53    -0.47
  dates*manage*cowpatt  17-Oct  O           F         17-Oct  O             O
53     0.66
  dates*manage*cowpatt  17-Oct  O           F         18-Nov  C             A
53     0.66
  dates*manage*cowpatt  17-Oct  O           F         18-Nov  C             F
53     0.99
  dates*manage*cowpatt  17-Oct  O           F         18-Nov  C             O
53     0.99
  dates*manage*cowpatt  17-Oct  O           F         18-Nov  O             A
53    -0.99
  dates*manage*cowpatt  17-Oct  O           F         18-Nov  O             F
53     0.33
  dates*manage*cowpatt  17-Oct  O           F         18-Nov  O             O
53    -2.30
  dates*manage*cowpatt  17-Oct  O           F         3-Sep   C             A
53     0.94
  dates*manage*cowpatt  17-Oct  O           F         3-Sep   C             F
53    -0.93
```

```
 dates*manage*cowpatt  17-Oct   O           F         3-Sep   C            O
53     0.19
 dates*manage*cowpatt  17-Oct   O           F         3-Sep   O            A
53     0.94
 dates*manage*cowpatt  17-Oct   O           F         3-Sep   O            F
53     0.57
 dates*manage*cowpatt  17-Oct   O           F         3-Sep   O            O
53     0.42
 dates*manage*cowpatt  17-Oct   O           O         18-Nov  C            A
53    -0.00
 dates*manage*cowpatt  17-Oct   O           O         18-Nov  C            F
53     0.33
 dates*manage*cowpatt  17-Oct   O           O         18-Nov  C            O
53     0.33
 dates*manage*cowpatt  17-Oct   O           O         18-Nov  O            A
53    -1.64
 dates*manage*cowpatt  17-Oct   O           O         18-Nov  O            F
53    -0.33
 dates*manage*cowpatt  17-Oct   O           O         18-Nov  O            O
53    -2.98
 dates*manage*cowpatt  17-Oct   O           O         3-Sep   C            A
53     0.31
 dates*manage*cowpatt  17-Oct   O           O         3-Sep   C            F
53    -1.56
 dates*manage*cowpatt  17-Oct   O           O         3-Sep   C            O
53    -0.44
 dates*manage*cowpatt  17-Oct   O           O         3-Sep   O            A
53     0.31
 dates*manage*cowpatt  17-Oct   O           O         3-Sep   O            F
53    -0.06
 dates*manage*cowpatt  17-Oct   O           O         3-Sep   O            O
53    -0.17
 dates*manage*cowpatt  18-Nov   C           A         18-Nov  C            F
53     0.33
 dates*manage*cowpatt  18-Nov   C           A         18-Nov  C            O
53     0.33
 dates*manage*cowpatt  18-Nov   C           A         18-Nov  O            A
53    -1.64
 dates*manage*cowpatt  18-Nov   C           A         18-Nov  O            F
53    -0.33
 dates*manage*cowpatt  18-Nov   C           A         18-Nov  O            O
53    -2.96
 dates*manage*cowpatt  18-Nov   C           A         3-Sep   C            A
53     0.31
 dates*manage*cowpatt  18-Nov   C           A         3-Sep   C            F
53    -1.56
 dates*manage*cowpatt  18-Nov   C           A         3-Sep   C            O
53    -0.44
 dates*manage*cowpatt  18-Nov   C           A         3-Sep   O            A
53     0.31
 dates*manage*cowpatt  18-Nov   C           A         3-Sep   O            F
53    -0.06
 dates*manage*cowpatt  18-Nov   C           A         3-Sep   O            O
53    -0.17
 dates*manage*cowpatt  18-Nov   C           F         18-Nov  C            O
53     0.00
 dates*manage*cowpatt  18-Nov   C           F         18-Nov  O            A
53    -1.97
 dates*manage*cowpatt  18-Nov   C           F         18-Nov  O            F
53    -0.66
 dates*manage*cowpatt  18-Nov   C           F         18-Nov  O            O
53    -3.29
 dates*manage*cowpatt  18-Nov   C           F         3-Sep   C            A
53    -0.00
 dates*manage*cowpatt  18-Nov   C           F         3-Sep   C            F
53    -1.89
 dates*manage*cowpatt  18-Nov   C           F         3-Sep   C            O
53    -0.75
 dates*manage*cowpatt  18-Nov   C           F         3-Sep   O            A
53     0.00
```

```
  dates*manage*cowpatt  18-Nov  C            F         3-Sep   O            F
53    -0.37
  dates*manage*cowpatt  18-Nov  C            F         3-Sep   O            O
53    -0.47
  dates*manage*cowpatt  18-Nov  C            O         18-Nov  O            A
53    -1.97
  dates*manage*cowpatt  18-Nov  C            O         18-Nov  O            F
53    -0.66
  dates*manage*cowpatt  18-Nov  C            O         18-Nov  O            O
53    -3.29
  dates*manage*cowpatt  18-Nov  C            O         3-Sep   C            A
53    -0.00
  dates*manage*cowpatt  18-Nov  C            O         3-Sep   C            F
53    -1.87
  dates*manage*cowpatt  18-Nov  C            O         3-Sep   C            O
53    -0.76
  dates*manage*cowpatt  18-Nov  C            O         3-Sep   O            A
53     0.00
  dates*manage*cowpatt  18-Nov  C            O         3-Sep   O            F
53    -0.37
  dates*manage*cowpatt  18-Nov  C            O         3-Sep   O            O
53    -0.47
  dates*manage*cowpatt  18-Nov  O            A         18-Nov  O            F
53     1.31
  dates*manage*cowpatt  18-Nov  O            A         18-Nov  O            O
53    -1.31
  dates*manage*cowpatt  18-Nov  O            A         3-Sep   C            A
53     1.88
  dates*manage*cowpatt  18-Nov  O            A         3-Sep   C            F
53     0.01
  dates*manage*cowpatt  18-Nov  O            A         3-Sep   C            O
53     1.13
  dates*manage*cowpatt  18-Nov  O            A         3-Sep   O            A
53     1.89
  dates*manage*cowpatt  18-Nov  O            A         3-Sep   O            F
53     1.51
  dates*manage*cowpatt  18-Nov  O            A         3-Sep   O            O
53     1.30
  dates*manage*cowpatt  18-Nov  O            F         18-Nov  O            O
53    -2.63
  dates*manage*cowpatt  18-Nov  O            F         3-Sep   C            A
53     0.62
  dates*manage*cowpatt  18-Nov  O            F         3-Sep   C            F
53    -1.24
  dates*manage*cowpatt  18-Nov  O            F         3-Sep   C            O
53    -0.13
  dates*manage*cowpatt  18-Nov  O            F         3-Sep   O            A
53     0.63
  dates*manage*cowpatt  18-Nov  O            F         3-Sep   O            F
53     0.26
  dates*manage*cowpatt  18-Nov  O            F         3-Sep   O            O
53     0.12
  dates*manage*cowpatt  18-Nov  O            O         3-Sep   C            A
53     3.13
  dates*manage*cowpatt  18-Nov  O            O         3-Sep   C            F
53     1.26
  dates*manage*cowpatt  18-Nov  O            O         3-Sep   C            O
53     2.38
  dates*manage*cowpatt  18-Nov  O            O         3-Sep   O            A
53     3.13
  dates*manage*cowpatt  18-Nov  O            O         3-Sep   O            F
53     2.76
  dates*manage*cowpatt  18-Nov  O            O         3-Sep   O            O
53     2.49
  dates*manage*cowpatt  3-Sep   C            A         3-Sep   C            F
53    -1.79
  dates*manage*cowpatt  3-Sep   C            A         3-Sep   C            O
53    -0.72
  dates*manage*cowpatt  3-Sep   C            A         3-Sep   O            A
53     0.00
```

```
  dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            F
53    -0.35
  dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            O
53    -0.44
  dates*manage*cowpatt  3-Sep   C           F         3-Sep   C            O
53     1.07
  dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            A
53     1.79
  dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            F
53     1.44
  dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            O
53     1.24
  dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            A
53     0.72
  dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            F
53     0.37
  dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            O
53     0.23
  dates*manage*cowpatt  3-Sep   O           A         3-Sep   O            F
53    -0.35
  dates*manage*cowpatt  3-Sep   O           A         3-Sep   O            O
53    -0.45
  dates*manage*cowpatt  3-Sep   O           F         3-Sep   O            O
53    -0.11

                                  Differences of Least Squares Means

     Effect                dates   management  cowpatty  _dates  _management  _cowpatty
Pr > |t|

     management                    C                             O
0.8074
     cowpatty                                  A                              F
0.1310
     cowpatty                                  A                              O
0.1754
     cowpatty                                  F                              O
0.8837
     management*cowpatty           C           A                 C            F
0.0387
     management*cowpatty           C           A                 C            O
0.4332
     management*cowpatty           C           A                 O            A
0.4766
     management*cowpatty           C           A                 O            F
0.4567
     management*cowpatty           C           A                 O            O
0.0714
     management*cowpatty           C           F                 C            O
0.1814
     management*cowpatty           C           F                 O            A
0.1597
     management*cowpatty           C           F                 O            F
0.1692
     management*cowpatty           C           F                 O            O
0.8074
     management*cowpatty           C           O                 O            A
0.9419
     management*cowpatty           C           O                 O            F
0.9681
     management*cowpatty           C           O                 O            O
0.2834
     management*cowpatty           O           A                 O            F
0.9738
     management*cowpatty           O           A                 O            O
0.2535
     management*cowpatty           O           F                 O            O
0.2667
     dates                 17-Oct                        18-Nov
0.7878
```

```
     dates                 17-Oct                   3-Sep
0.4601
     dates                 18-Nov                   3-Sep
0.3224
     dates*management      17-Oct  C                17-Oct  O
0.0937
     dates*management      17-Oct  C                18-Nov  C
0.0257
     dates*management      17-Oct  C                18-Nov  O
0.3472
     dates*management      17-Oct  C                3-Sep   C
0.4049
     dates*management      17-Oct  C                3-Sep   O
0.0759
     dates*management      17-Oct  O                18-Nov  C
0.5717
     dates*management      17-Oct  O                18-Nov  O
0.0098
     dates*management      17-Oct  O                3-Sep   C
0.4304
     dates*management      17-Oct  O                3-Sep   O
0.8269
     dates*management      18-Nov  C                18-Nov  O
0.0022
     dates*management      18-Nov  C                3-Sep   C
0.1835
     dates*management      18-Nov  C                3-Sep   O
0.7559
     dates*management      18-Nov  O                3-Sep   C
0.0881
     dates*management      18-Nov  O                3-Sep   O
0.0089
     dates*management      3-Sep   C                3-Sep   O
0.3441
     dates*cowpatty        17-Oct               A         17-Oct                F
0.0240
     dates*cowpatty        17-Oct               A         17-Oct                O
0.3570
     dates*cowpatty        17-Oct               A         18-Nov                A
0.1656
     dates*cowpatty        17-Oct               A         18-Nov                F
0.8172
     dates*cowpatty        17-Oct               A         18-Nov                O
0.0414
     dates*cowpatty        17-Oct               A         3-Sep                 A
0.8265
     dates*cowpatty        17-Oct               A         3-Sep                 F
0.1784
     dates*cowpatty        17-Oct               A         3-Sep                 O
0.5244
     dates*cowpatty        17-Oct               F         17-Oct                O
0.1692
     dates*cowpatty        17-Oct               F         18-Nov                A
0.3570
     dates*cowpatty        17-Oct               F         18-Nov                F
0.0397
     dates*cowpatty        17-Oct               F         18-Nov                O
0.8172
     dates*cowpatty        17-Oct               F         3-Sep                 A
0.0184
     dates*cowpatty        17-Oct               F         3-Sep                 F
0.3948
     dates*cowpatty        17-Oct               F         3-Sep                 O
0.1389
     dates*cowpatty        17-Oct               O         18-Nov                A
0.6441
     dates*cowpatty        17-Oct               O         18-Nov                F
0.4889
     dates*cowpatty        17-Oct               O         18-Nov                O
0.2467
```

```
     dates*cowpatty        17-Oct              O         3-Sep                A
0.2743
     dates*cowpatty        17-Oct              O         3-Sep                F
0.6348
     dates*cowpatty        17-Oct              O         3-Sep                O
0.8282
     dates*cowpatty        18-Nov              A         18-Nov               F
0.2506
     dates*cowpatty        18-Nov              A         18-Nov               O
0.4889
     dates*cowpatty        18-Nov              A         3-Sep                A
0.1248
     dates*cowpatty        18-Nov              A         3-Sep                F
0.9724
     dates*cowpatty        18-Nov              A         3-Sep                O
0.5216
     dates*cowpatty        18-Nov              F         18-Nov               O
0.0687
     dates*cowpatty        18-Nov              F         3-Sep                A
0.6617
     dates*cowpatty        18-Nov              F         3-Sep                F
0.2549
     dates*cowpatty        18-Nov              F         3-Sep                O
0.6715
     dates*cowpatty        18-Nov              O         3-Sep                A
0.0313
     dates*cowpatty        18-Nov              O         3-Sep                F
0.5316
     dates*cowpatty        18-Nov              O         3-Sep                O
0.2001
     dates*cowpatty        3-Sep               A         3-Sep                F
0.1357
     dates*cowpatty        3-Sep               A         3-Sep                O
0.4169
     dates*cowpatty        3-Sep               F         3-Sep                O
0.5175
     dates*manage*cowpatt  17-Oct  C           A         17-Oct  C            F
0.0254
     dates*manage*cowpatt  17-Oct  C           A         17-Oct  C            O
0.3288
     dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            A
0.7438
     dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            F
0.5140
     dates*manage*cowpatt  17-Oct  C           A         17-Oct  O            O
1.0000
     dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            A
1.0000
     dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            F
0.7438
     dates*manage*cowpatt  17-Oct  C           A         18-Nov  C            O
0.7438
     dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            A
0.1064
     dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            F
0.7438
     dates*manage*cowpatt  17-Oct  C           A         18-Nov  O            O
0.0046
     dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            A
0.7567
     dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            F
0.1252
     dates*manage*cowpatt  17-Oct  C           A         3-Sep   C            O
0.6613
     dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            A
0.7553
     dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            F
0.9544
     dates*manage*cowpatt  17-Oct  C           A         3-Sep   O            O
0.8646
```

```
     dates*manage*cowpatt  17-Oct  C           F         17-Oct  C             O
0.1945
     dates*manage*cowpatt  17-Oct  C           F         17-Oct  O             A
0.0112
     dates*manage*cowpatt  17-Oct  C           F         17-Oct  O             F
0.1064
     dates*manage*cowpatt  17-Oct  C           F         17-Oct  O             O
0.0254
     dates*manage*cowpatt  17-Oct  C           F         18-Nov  C             A
0.0254
     dates*manage*cowpatt  17-Oct  C           F         18-Nov  C             F
0.0106
     dates*manage*cowpatt  17-Oct  C           F         18-Nov  C             O
0.0112
     dates*manage*cowpatt  17-Oct  C           F         18-Nov  O             A
0.5140
     dates*manage*cowpatt  17-Oct  C           F         18-Nov  O             F
0.0539
     dates*manage*cowpatt  17-Oct  C           F         18-Nov  O             O
0.5140
     dates*manage*cowpatt  17-Oct  C           F         3-Sep   C             A
0.0155
     dates*manage*cowpatt  17-Oct  C           F         3-Sep   C             F
0.5251
     dates*manage*cowpatt  17-Oct  C           F         3-Sep   C             O
0.0855
     dates*manage*cowpatt  17-Oct  C           F         3-Sep   O             A
0.0153
     dates*manage*cowpatt  17-Oct  C           F         3-Sep   O             F
0.0374
     dates*manage*cowpatt  17-Oct  C           F         3-Sep   O             O
0.0648
     dates*manage*cowpatt  17-Oct  C           O         17-Oct  O             A
0.1945
     dates*manage*cowpatt  17-Oct  C           O         17-Oct  O             F
0.7438
     dates*manage*cowpatt  17-Oct  C           O         17-Oct  O             O
0.3288
     dates*manage*cowpatt  17-Oct  C           O         18-Nov  C             A
0.3288
     dates*manage*cowpatt  17-Oct  C           O         18-Nov  C             F
0.1945
     dates*manage*cowpatt  17-Oct  C           O         18-Nov  C             O
0.1907
     dates*manage*cowpatt  17-Oct  C           O         18-Nov  O             A
0.5140
     dates*manage*cowpatt  17-Oct  C           O         18-Nov  O             F
0.5140
     dates*manage*cowpatt  17-Oct  C           O         18-Nov  O             O
0.0539
     dates*manage*cowpatt  17-Oct  C           O         3-Sep   C             A
0.2172
     dates*manage*cowpatt  17-Oct  C           O         3-Sep   C             F
0.5391
     dates*manage*cowpatt  17-Oct  C           O         3-Sep   C             O
0.6171
     dates*manage*cowpatt  17-Oct  C           O         3-Sep   O             A
0.2157
     dates*manage*cowpatt  17-Oct  C           O         3-Sep   O             F
0.3816
     dates*manage*cowpatt  17-Oct  C           O         3-Sep   O             O
0.4807
     dates*manage*cowpatt  17-Oct  O           A         17-Oct  O             F
0.3288
     dates*manage*cowpatt  17-Oct  O           A         17-Oct  O             O
0.7438
     dates*manage*cowpatt  17-Oct  O           A         18-Nov  C             A
0.7438
     dates*manage*cowpatt  17-Oct  O           A         18-Nov  C             F
1.0000
```

```
     dates*manage*cowpatt  17-Oct  O           A         18-Nov  C            O
1.0000
     dates*manage*cowpatt  17-Oct  O           A         18-Nov  O            A
0.0520
     dates*manage*cowpatt  17-Oct  O           A         18-Nov  O            F
0.5140
     dates*manage*cowpatt  17-Oct  O           A         18-Nov  O            O
0.0018
     dates*manage*cowpatt  17-Oct  O           A         3-Sep   C            A
0.9967
     dates*manage*cowpatt  17-Oct  O           A         3-Sep   C            F
0.0668
     dates*manage*cowpatt  17-Oct  O           A         3-Sep   C            O
0.4543
     dates*manage*cowpatt  17-Oct  O           A         3-Sep   O            A
1.0000
     dates*manage*cowpatt  17-Oct  O           A         3-Sep   O            F
0.7123
     dates*manage*cowpatt  17-Oct  O           A         3-Sep   O            O
0.6437
     dates*manage*cowpatt  17-Oct  O           F         17-Oct  O            O
0.5140
     dates*manage*cowpatt  17-Oct  O           F         18-Nov  C            A
0.5140
     dates*manage*cowpatt  17-Oct  O           F         18-Nov  C            F
0.3288
     dates*manage*cowpatt  17-Oct  O           F         18-Nov  C            O
0.3288
     dates*manage*cowpatt  17-Oct  O           F         18-Nov  O            A
0.3288
     dates*manage*cowpatt  17-Oct  O           F         18-Nov  O            F
0.7417
     dates*manage*cowpatt  17-Oct  O           F         18-Nov  O            O
0.0254
     dates*manage*cowpatt  17-Oct  O           F         3-Sep   C            A
0.3537
     dates*manage*cowpatt  17-Oct  O           F         3-Sep   C            F
0.3559
     dates*manage*cowpatt  17-Oct  O           F         3-Sep   C            O
0.8533
     dates*manage*cowpatt  17-Oct  O           F         3-Sep   O            A
0.3516
     dates*manage*cowpatt  17-Oct  O           F         3-Sep   O            F
0.5688
     dates*manage*cowpatt  17-Oct  O           F         3-Sep   O            O
0.6788
     dates*manage*cowpatt  17-Oct  O           O         18-Nov  C            A
1.0000
     dates*manage*cowpatt  17-Oct  O           O         18-Nov  C            F
0.7438
     dates*manage*cowpatt  17-Oct  O           O         18-Nov  C            O
0.7438
     dates*manage*cowpatt  17-Oct  O           O         18-Nov  O            A
0.1064
     dates*manage*cowpatt  17-Oct  O           O         18-Nov  O            F
0.7438
     dates*manage*cowpatt  17-Oct  O           O         18-Nov  O            O
0.0043
     dates*manage*cowpatt  17-Oct  O           O         3-Sep   C            A
0.7585
     dates*manage*cowpatt  17-Oct  O           O         3-Sep   C            F
0.1252
     dates*manage*cowpatt  17-Oct  O           O         3-Sep   C            O
0.6613
     dates*manage*cowpatt  17-Oct  O           O         3-Sep   O            A
0.7553
     dates*manage*cowpatt  17-Oct  O           O         3-Sep   O            F
0.9544
     dates*manage*cowpatt  17-Oct  O           O         3-Sep   O            O
0.8637
```

```
     dates*manage*cowpatt  18-Nov  C           A          18-Nov  C            F
0.7438
     dates*manage*cowpatt  18-Nov  C           A          18-Nov  C            O
0.7438
     dates*manage*cowpatt  18-Nov  C           A          18-Nov  O            A
0.1064
     dates*manage*cowpatt  18-Nov  C           A          18-Nov  O            F
0.7438
     dates*manage*cowpatt  18-Nov  C           A          18-Nov  O            O
0.0046
     dates*manage*cowpatt  18-Nov  C           A          3-Sep   C            A
0.7567
     dates*manage*cowpatt  18-Nov  C           A          3-Sep   C            F
0.1252
     dates*manage*cowpatt  18-Nov  C           A          3-Sep   C            O
0.6613
     dates*manage*cowpatt  18-Nov  C           A          3-Sep   O            A
0.7553
     dates*manage*cowpatt  18-Nov  C           A          3-Sep   O            F
0.9544
     dates*manage*cowpatt  18-Nov  C           A          3-Sep   O            O
0.8646
     dates*manage*cowpatt  18-Nov  C           F          18-Nov  C            O
1.0000
     dates*manage*cowpatt  18-Nov  C           F          18-Nov  O            A
0.0539
     dates*manage*cowpatt  18-Nov  C           F          18-Nov  O            F
0.5140
     dates*manage*cowpatt  18-Nov  C           F          18-Nov  O            O
0.0018
     dates*manage*cowpatt  18-Nov  C           F          3-Sep   C            A
0.9967
     dates*manage*cowpatt  18-Nov  C           F          3-Sep   C            F
0.0648
     dates*manage*cowpatt  18-Nov  C           F          3-Sep   C            O
0.4543
     dates*manage*cowpatt  18-Nov  C           F          3-Sep   O            A
1.0000
     dates*manage*cowpatt  18-Nov  C           F          3-Sep   O            F
0.7123
     dates*manage*cowpatt  18-Nov  C           F          3-Sep   O            O
0.6437
     dates*manage*cowpatt  18-Nov  C           O          18-Nov  O            A
0.0539
     dates*manage*cowpatt  18-Nov  C           O          18-Nov  O            F
0.5140
     dates*manage*cowpatt  18-Nov  C           O          18-Nov  O            O
0.0018
     dates*manage*cowpatt  18-Nov  C           O          3-Sep   C            A
0.9967
     dates*manage*cowpatt  18-Nov  C           O          3-Sep   C            F
0.0668
     dates*manage*cowpatt  18-Nov  C           O          3-Sep   C            O
0.4508
     dates*manage*cowpatt  18-Nov  C           O          3-Sep   O            A
1.0000
     dates*manage*cowpatt  18-Nov  C           O          3-Sep   O            F
0.7123
     dates*manage*cowpatt  18-Nov  C           O          3-Sep   O            O
0.6437
     dates*manage*cowpatt  18-Nov  O           A          18-Nov  O            F
0.1945
     dates*manage*cowpatt  18-Nov  O           A          18-Nov  O            O
0.1945
     dates*manage*cowpatt  18-Nov  O           A          3-Sep   C            A
0.0663
     dates*manage*cowpatt  18-Nov  O           A          3-Sep   C            F
0.9934
     dates*manage*cowpatt  18-Nov  O           A          3-Sep   C            O
0.2654
```

```
     dates*manage*cowpatt  18-Nov  O           A         3-Sep   O            A
0.0637
     dates*manage*cowpatt  18-Nov  O           A         3-Sep   O            F
0.1373
     dates*manage*cowpatt  18-Nov  O           A         3-Sep   O            O
0.1999
     dates*manage*cowpatt  18-Nov  O           F         18-Nov  O            O
0.0112
     dates*manage*cowpatt  18-Nov  O           F         3-Sep   C            A
0.5364
     dates*manage*cowpatt  18-Nov  O           F         3-Sep   C            F
0.2187
     dates*manage*cowpatt  18-Nov  O           F         3-Sep   C            O
0.8991
     dates*manage*cowpatt  18-Nov  O           F         3-Sep   O            A
0.5337
     dates*manage*cowpatt  18-Nov  O           F         3-Sep   O            F
0.7976
     dates*manage*cowpatt  18-Nov  O           F         3-Sep   O            O
0.9029
     dates*manage*cowpatt  18-Nov  O           O         3-Sep   C            A
0.0029
     dates*manage*cowpatt  18-Nov  O           O         3-Sep   C            F
0.2127
     dates*manage*cowpatt  18-Nov  O           O         3-Sep   C            O
0.0210
     dates*manage*cowpatt  18-Nov  O           O         3-Sep   O            A
0.0028
     dates*manage*cowpatt  18-Nov  O           O         3-Sep   O            F
0.0079
     dates*manage*cowpatt  18-Nov  O           O         3-Sep   O            O
0.0159
     dates*manage*cowpatt  3-Sep   C           A         3-Sep   C            F
0.0796
     dates*manage*cowpatt  3-Sep   C           A         3-Sep   C            O
0.4760
     dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            A
0.9968
     dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            F
0.7270
     dates*manage*cowpatt  3-Sep   C           A         3-Sep   O            O
0.6589
     dates*manage*cowpatt  3-Sep   C           F         3-Sep   C            O
0.2896
     dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            A
0.0789
     dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            F
0.1567
     dates*manage*cowpatt  3-Sep   C           F         3-Sep   O            O
0.2199
     dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            A
0.4736
     dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            F
0.7152
     dates*manage*cowpatt  3-Sep   C           O         3-Sep   O            O
0.8167
     dates*manage*cowpatt  3-Sep   O           A         3-Sep   O            F
0.7241
     dates*manage*cowpatt  3-Sep   O           A         3-Sep   O            O
0.6562
     dates*manage*cowpatt  3-Sep   O           F         3-Sep   O            O
0.9104


                                       Tests of Effect Slices

                                                                           Num      Den
    Effect                    dates     management     cowpatty      DF       DF    F Value
Pr > F
```

```
    management*cowpatty               C                            2      30       2.39
0.1086
    management*cowpatty               O                            2      30       0.87
0.4307
    management*cowpatty                             A              1      30       0.52
0.4766
    management*cowpatty                             F              1      30       1.98
0.1692
    management*cowpatty                             O              1      30       1.19
0.2834
    dates*management        17-Oct                                 1      53       2.91
0.0937
    dates*management        18-Nov                                 1      53      10.40
0.0022
    dates*management        3-Sep                                  1      53       0.91
0.3441
    dates*management                  C                            2      53       2.68
0.0781
    dates*management                  O                            2      53       4.96
0.0106
    dates*cowpatty          17-Oct                                 2      53       2.73
0.0741
    dates*cowpatty          18-Nov                                 2      53       1.76
0.1815
    dates*cowpatty          3-Sep                                  2      53       1.15
0.3238
    dates*cowpatty                                  A              2      53       1.50
0.2324
    dates*cowpatty                                  F              2      53       2.24
0.1168
    dates*cowpatty                                  O              2      53       1.05
0.3569
    dates*manage*cowpatt    17-Oct                                 5      53       1.85
0.1191
    dates*manage*cowpatt    18-Nov                                 5      53       3.49
0.0084
    dates*manage*cowpatt    3-Sep                                  5      53       0.89
0.4975
    dates*manage*cowpatt              C                            8      53       1.75
0.1090
    dates*manage*cowpatt              O                            8      53       2.28
0.0356
    dates*manage*cowpatt                            A              5      53       1.17
0.3385
    dates*manage*cowpatt                            F              5      53       1.93
0.1043
    dates*manage*cowpatt                            O              5      53       2.80
0.0256
```

Precipitation gradient contrasts

```
Contrasts



                                      Num     Den

               Label                   DF      DF     F Value     Pr > F



               EAST V. WEST TRT=F       1    8.52        0.05     0.8210
```

```
EAST V. WEST TRT=N         1     8.52         2.31     0.1644

EAST V. WEST TRT=O         1     8.52         5.42     0.0465
```

Organic vs. Conventional wheat field and cattle pasture comparisons in Fairview, OK.

Date	O or C	Field Type	T	sample #	D	L	E	Sf/g	Sc	Hb	TI
17-Oct	O	Wheat	A	1	0	4	2	0	0	0	0
17-Oct	O	Wheat	A	2	0	6	0	0	0	0	0
17-Oct	O	Wheat	A	3	0	6	0	0	0	0	0
17-Oct	O	Wheat	A	4	0	6	0	0	0	0	0
17-Oct	O	Wheat	A	5	0	6	0	0	0	0	0
17-Oct	O	Wheat	A	6	0	6	0	0	0	0	0
17-Oct	O	Wheat	A	7	0	5	0	0	0	0	0
17-Oct	O	Wheat	A	8	0	6	0	0	0	0	0
17-Oct	O	Wheat	A	9	0	5	0	0	0	0	0
17-Oct	O	Wheat	A	10	1	5	0	0	0	0	0
17-Oct	O	Wheat	B	1	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	2	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	3	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	4	0	5	0	0	0	0	1
17-Oct	O	Wheat	B	5	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	6	1	5	0	0	0	0	0
17-Oct	O	Wheat	B	7	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	8	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	9	0	6	0	0	0	0	0
17-Oct	O	Wheat	B	10	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	1	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	2	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	3	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	4	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	5	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	6	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	7	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	8	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	9	0	6	0	0	0	0	0
17-Oct	O	Wheat	C	10	0	5	0	0	0	0	1
17-Oct	O	Wheat	D	1	0	6	0	0	0	0	0
17-Oct	O	Wheat	D	2	0	6	0	0	0	0	0
17-Oct	O	Wheat	D	3	0	6	0	0	0	0	0
17-Oct	O	Wheat	D	4	0	6	0	0	0	0	0

17-Oct	O	Wheat	D	5	0	6	0	0	0	0	0
17-Oct	O	Wheat	D	6	1	5	0	0	0	0	0
17-Oct	O	Wheat	D	7	0	6	0	0	0	0	0
17-Oct	O	Wheat	D	8	0	5	1	0	0	0	0
17-Oct	O	Wheat	D	9	0	5	1	0	0	0	0
17-Oct	O	Wheat	D	10	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	1	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	2	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	3	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	4	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	5	0	5	0	0	0	0	0
17-Oct	C	Wheat	A	6	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	7	0	6	0	0	0	0	0
17-Oct	C	Wheat	A	8	0	5	0	0	0	0	1
17-Oct	C	Wheat	A	9	1	5	0	0	0	0	0
17-Oct	C	Wheat	A	10	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	1	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	2	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	3	0	5	0	0	0	0	1
17-Oct	C	Wheat	B	4	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	5	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	6	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	7	2	4	0	0	0	0	0
17-Oct	C	Wheat	B	8	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	9	0	6	0	0	0	0	0
17-Oct	C	Wheat	B	10	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	1	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	2	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	3	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	4	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	5	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	6	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	7	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	8	0	6	0	0	0	0	0
17-Oct	C	Wheat	C	9	0	5	1	0	0	0	0
17-Oct	C	Wheat	C	10	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	1	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	2	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	3	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	4	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	5	0	5	0	0	0	0	1

17-Oct	C	Wheat	D	6	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	7	1	5	0	0	0	0	0
17-Oct	C	Wheat	D	8	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	9	0	6	0	0	0	0	0
17-Oct	C	Wheat	D	10						0	
17-Oct	O	Cow	F	1						0	
17-Oct	O	Cow	F	2	0	4	1	0	0	0	1
17-Oct	O	Cow	F	3	0	5	0	0	0	0	1
17-Oct	O	Cow	F	4						0	
17-Oct	O	Cow	F	5	0	5	0	0	1	0	1
17-Oct	O	Cow	F	6	0	5	0	0	0	0	0
17-Oct	O	Cow	O	1	0	6	0	0	0	0	0
17-Oct	O	Cow	O	2	0	5	0	0	0	0	1
17-Oct	O	Cow	O	3	0	6	0	0	0	0	0
17-Oct	O	Cow	O	4	0	6	0	0	0	0	0
17-Oct	O	Cow	O	5	1	4	1	0	0	0	0
17-Oct	O	Cow	O	6	0	5	1	0	0	0	0
17-Oct	O	Cow	A	1	0	6	0	0	0	0	0
17-Oct	O	Cow	A	2	0	6	0	0	0	0	0
17-Oct	O	Cow	A	3						0	
17-Oct	O	Cow	A	4	0	6	0	0	0	0	0
17-Oct	O	Cow	A	5	0	6	0	0	0	0	0
17-Oct	O	Cow	A	6	0	6	0	0	0	0	0
17-Oct	C	Cow	F	1	0	4	0	0	0	1	1
17-Oct	C	Cow	F	2	0	3	0	0	0	3	3
17-Oct	C	Cow	F	3	0	6	0	0	0	0	0
17-Oct	C	Cow	F	4	1	5	0	0	0	0	0
17-Oct	C	Cow	F	5	0	2	0	0	0	4	4
17-Oct	C	Cow	F	6	1	5	0	0	0	0	0
17-Oct	C	Cow	O	1	0	6	0	0	0	0	0
17-Oct	C	Cow	O	2	0	5	0	1	0	0	1
17-Oct	C	Cow	O	3	0	5	0	0	0	1	1
17-Oct	C	Cow	O	4							
17-Oct	C	Cow	O	5	1	3	0	0	0	1	1
17-Oct	C	Cow	O	6	0	3	2	0	0	0	1
17-Oct	C	Cow	A	1	0	6	0	0	0	0	0
17-Oct	C	Cow	A	2	0	6	0	0	0	0	0
17-Oct	C	Cow	A	3	1	5	0	0	0	0	0
17-Oct	C	Cow	A	4	0	6	0	0	0	0	0
17-Oct	C	Cow	A	5	1	5	0	0	0	0	0
17-Oct	C	Cow	A	6	0	5	0	0	0	1	1

18-Nov	C	Wheat	A	1	0	4	0	0	0	0	0
18-Nov	C	Wheat	A	2	1	4	0	0	0	0	0
18-Nov	C	Wheat	A	3	1	5	0	0	0	0	0
18-Nov	C	Wheat	A	4	1	5	0	0	0	0	0
18-Nov	C	Wheat	A	5	0	5	0	0	1	0	1
18-Nov	C	Wheat	A	6	2	2	0	0	0	0	0
18-Nov	C	Wheat	A	7	1	4	0	0	0	0	0
18-Nov	C	Wheat	A	8	0	5	0	0	0	0	0
18-Nov	C	Wheat	A	9	0	4	0	0	0	0	0
18-Nov	C	Wheat	A	10	0	6	0	0	0	0	0
18-Nov	C	Wheat	B	1	1	5	0	0	0	0	0
18-Nov	C	Wheat	B	2	1	5	0	0	0	0	0
18-Nov	C	Wheat	B	3	3	2	0	0	0	0	0
18-Nov	C	Wheat	B	4	2	4	0	0	0	0	0
18-Nov	C	Wheat	B	5	0	0	0	0	0	0	0
18-Nov	C	Wheat	B	6	0	5	0	1	0	0	1
18-Nov	C	Wheat	B	7	4	2	0	0	0	0	0
18-Nov	C	Wheat	B	8	0	5	0	1	0	0	1
18-Nov	C	Wheat	B	9	1	4	1	0	0	0	0
18-Nov	C	Wheat	B	10	1	5	0	0	0	0	0
18-Nov	C	Wheat	C	1	0	5	0	0	0	0	0
18-Nov	C	Wheat	C	2	0	5	0	0	0	0	0
18-Nov	C	Wheat	C	3	1	5	0	0	0	0	0
18-Nov	C	Wheat	C	4	0	6	0	0	0	0	0
18-Nov	C	Wheat	C	5	2	2	0	0	0	0	0
18-Nov	C	Wheat	C	6	0	6	0	0	0	0	0
18-Nov	C	Wheat	C	7	0	6	0	0	0	0	0
18-Nov	C	Wheat	C	8	0	5	0	1	0	0	1
18-Nov	C	Wheat	C	9	1	5	0	0	0	0	0
18-Nov	C	Wheat	C	10	2	2	1	0	0	0	0
18-Nov	C	Wheat	D	1	2	3	0	1	0	0	1
18-Nov	C	Wheat	D	2	0	6	0	0	0	0	0
18-Nov	C	Wheat	D	3	1	5	0	0	0	0	0
18-Nov	C	Wheat	D	4	0	6	0	0	0	0	0
18-Nov	C	Wheat	D	5	2	4	0	0	0	0	0
18-Nov	C	Wheat	D	6	1	4	0	0	1	0	1
18-Nov	C	Wheat	D	7	0	6	0	0	0	0	0
18-Nov	C	Wheat	D	8	1	5	0	0	0	0	0
18-Nov	C	Wheat	D	9	1	5	0	0	0	0	0
18-Nov	C	Wheat	D	10	2	5	0	0	0	0	0
18-Nov	O	Wheat	A	1	4	1	0	0	0	0	0

18-Nov	O	Wheat	A	2	4	1	0	0	0	0	0
18-Nov	O	Wheat	A	3	3	2	0	1	0	0	1
18-Nov	O	Wheat	A	4	5	1	0	0	0	0	0
18-Nov	O	Wheat	A	5	1	5	0	0	0	0	0
18-Nov	O	Wheat	A	6	1	3	2	0	0	0	0
18-Nov	O	Wheat	A	7	3	1	1	0	1	0	1
18-Nov	O	Wheat	A	8	5	1	0	0	0	0	0
18-Nov	O	Wheat	A	9	5	1	0	0	0	0	0
18-Nov	O	Wheat	A	10	1	5	0	0	0	0	0
18-Nov	O	Wheat	B	1	2	1	0	0	0	0	0
18-Nov	O	Wheat	B	2	1	0	3	0	1	0	1
18-Nov	O	Wheat	B	3	0	0	1	0	0	0	0
18-Nov	O	Wheat	B	4	2	1	0	0	0	0	0
18-Nov	O	Wheat	B	5	3	1	0	0	0	0	0
18-Nov	O	Wheat	B	6	4	0	0	0	1	0	1
18-Nov	O	Wheat	B	7	6	0	0	0	0	0	0
18-Nov	O	Wheat	B	8	5	1	0	0	0	0	0
18-Nov	O	Wheat	B	9	3	0	0	2	0	0	2
18-Nov	O	Wheat	B	10	1	4	0	0	0	0	0
18-Nov	O	Wheat	C	1	2	3	0	0	0	0	0
18-Nov	O	Wheat	C	2	1	0	0	0	0	0	0
18-Nov	O	Wheat	C	3	0	0	1	2	0	0	2
18-Nov	O	Wheat	C	4	2	1	0	0	0	0	0
18-Nov	O	Wheat	C	5	0	5	1	0	0	0	0
18-Nov	O	Wheat	C	6	1	3	0	0	0	0	0
18-Nov	O	Wheat	C	7	2	1	1	0	0	0	0
18-Nov	O	Wheat	C	8	1	2	1	0	0	0	0
18-Nov	O	Wheat	C	9	3	2	0	0	0	0	0
18-Nov	O	Wheat	C	10	1	4	0	0	0	0	0
18-Nov	O	Wheat	D	1	1	2	0	0	0	0	0
18-Nov	O	Wheat	D	2	2	4	0	0	0	0	0
18-Nov	O	Wheat	D	3	0	6	0	0	0	0	0
18-Nov	O	Wheat	D	4	0	4	1	1	0	0	1
18-Nov	O	Wheat	D	5	1	4	0	0	0	0	0
18-Nov	O	Wheat	D	6	1	2	1	0	0	0	0
18-Nov	O	Wheat	D	7	0	2	1	0	0	0	0
18-Nov	O	Wheat	D	8	0	0	0	0	0	0	0
18-Nov	O	Wheat	D	9	0	2	0	2	0	0	2
18-Nov	O	Wheat	D	10	0	1	0	0	0	0	0
18-Nov	C	Cow	A	1	0	0	0	1	0	0	1
18-Nov	C	Cow	A	2	1	5	0	0	0	0	0

18-Nov	C	Cow	A	3	1	3	2	0	0	0	0
18-Nov	C	Cow	A	4	0	0	0	0	0	0	0
18-Nov	C	Cow	A	5	0	6	0	0	0	0	0
18-Nov	C	Cow	A	6	4	2	0	0	0	0	0
18-Nov	C	Cow	O	1	0	6	0	0	0	0	0
18-Nov	C	Cow	O	2	0	6	0	0	0	0	0
18-Nov	C	Cow	O	3	2	4	0	0	0	0	0
18-Nov	C	Cow	O	4	2	4	0	0	0	0	0
18-Nov	C	Cow	O	5	0	5	1	0	0	0	0
18-Nov	C	Cow	O	6	1	4	1	0	0	0	0
18-Nov	C	Cow	F	1	1	5	0	0	0	0	0
18-Nov	C	Cow	F	2	0	0	0	0	0	0	0
18-Nov	C	Cow	F	3	1	5	0	0	0	0	0
18-Nov	C	Cow	F	4	2	4	0	0	0	0	0
18-Nov	C	Cow	F	5	0	6	0	0	0	0	0
18-Nov	C	Cow	F	6	0	6	0	0	0	0	0
18-Nov	O	Cow	A	1	0	5	0	0	1	0	1
18-Nov	O	Cow	A	2	0	6	0	0	0	0	0
18-Nov	O	Cow	A	3	0	2	0	0	4	0	4
18-Nov	O	Cow	A	4	0	0	0	0	0	0	0
18-Nov	O	Cow	A	5	0	5	0	0	0	0	0
18-Nov	O	Cow	A	6	0	4	1	1	0	0	1
18-Nov	O	Cow	O	1	0	3	0	0	3	0	3
18-Nov	O	Cow	O	2	2	4	0	0	0	0	0
18-Nov	O	Cow	O	3	1	5	0	0	0	0	0
18-Nov	O	Cow	O	4	2	3	1	0	0	0	0
18-Nov	O	Cow	O	5	2	1	0	0	3	0	3
18-Nov	O	Cow	O	6	0	2	0	0	4	0	4
18-Nov	O	Cow	F	1	6	0	0	0	0	0	0
18-Nov	O	Cow	F	2	3	3	0	0	0	0	0
18-Nov	O	Cow	F	3	0	6	0	1	0	0	1
18-Nov	O	Cow	F	4	0	5	0	1	0	0	1
18-Nov	O	Cow	F	5	0	5	0	0	0	0	0
18-Nov	O	Cow	F	6	1	5	0	0	0	0	0
3-Sep	O	Wheat	A	1	0	6	0	0	0	0	0
3-Sep	O	Wheat	A	2	0	6	0	0	0	0	0
3-Sep	O	Wheat	A	3	0	6	0	0	0	0	0
3-Sep	O	Wheat	A	4	0	6	0	0	0	0	0
3-Sep	O	Wheat	A	5	0	6	0	0	0	0	0
3-Sep	O	Wheat	A	6	0	6	0	0	0	0	0
3-Sep	O	Wheat	A	7	0	6	0	0	0	0	0

3-Sep	O	Wheat	B	1	1	5	0	0	0	0	0
3-Sep	O	Wheat	B	2	0	6	0	0	0	0	0
3-Sep	O	Wheat	B	3	0	6	0	0	0	0	0
3-Sep	O	Wheat	B	4	0	6	0	0	0	0	0
3-Sep	O	Wheat	B	5	0	5	1	0	0	0	0
3-Sep	O	Wheat	B	6	0	6	0	0	0	0	0
3-Sep	O	Wheat	C	1	0	6	0	0	0	0	0
3-Sep	O	Wheat	C	2	0	5	0	1	0	0	1
3-Sep	O	Wheat	C	3	0	6	0	0	0	0	0
3-Sep	O	Wheat	C	4	0	6	0	0	0	0	0
3-Sep	O	Wheat	C	5	0	6	0	0	0	0	0
3-Sep	O	Wheat	C	6	1	5	0	0	0	0	0
3-Sep	O	Wheat	D	1	0	6	0	0	0	0	0
3-Sep	O	Wheat	D	2	0	6	0	0	0	0	0
3-Sep	O	Wheat	D	3	0	6	0	0	0	0	0
3-Sep	O	Wheat	D	4	0	6	0	0	0	0	0
3-Sep	O	Wheat	D	5	1	4	1	1	0	0	1
3-Sep	O	Wheat	D	6	0	6	0	0	0	0	0
3-Sep	C	Wheat	A	1	0	6	0	0	0	0	0
3-Sep	C	Wheat	A	2	0	6	0	0	0	0	0
3-Sep	C	Wheat	A	3	0	6	0	0	0	0	0
3-Sep	C	Wheat	A	4	0	6	0	0	0	0	0
3-Sep	C	Wheat	A	5	0	6	0	0	0	0	0
3-Sep	C	Wheat	A	6	0	5	0	0	0	0	0
3-Sep	C	Wheat	B	1	0	6	0	0	0	0	0
3-Sep	C	Wheat	B	2	1	5	0	0	0	0	0
3-Sep	C	Wheat	B	3	0	6	0	0	0	0	0
3-Sep	C	Wheat	B	4	0	6	0	0	0	0	0
3-Sep	C	Wheat	B	5	0	6	0	0	0	0	0
3-Sep	C	Wheat	B	6	0	6	0	0	0	0	0
3-Sep	C	Wheat	C	1	0	5	1	0	0	0	0
3-Sep	C	Wheat	C	2	0	6	0	0	0	0	0
3-Sep	C	Wheat	C	3	1	5	0	0	0	0	0
3-Sep	C	Wheat	C	4	0	6	0	0	0	0	0
3-Sep	C	Wheat	C	5	0	5	0	1	0	0	1
3-Sep	C	Wheat	C	6	0	6	0	0	0	0	0
3-Sep	C	Wheat	D	1	0	6	0	0	0	0	0
3-Sep	C	Wheat	D	2	0	6	0	0	0	0	0
3-Sep	C	Wheat	D	3	0	6	0	0	0	0	0
3-Sep	C	Wheat	D	4	0	5	1	0	0	0	0
3-Sep	C	Wheat	D	5	0	6	0	0	0	0	0

3-Sep	C	Wheat	D	6	0	6	0	0	0	0	0
3-Sep	O	Cow	A	1	0	4	2	0	0	0	0
3-Sep	O	Cow	A	2	0	6	0	0	0	0	0
3-Sep	O	Cow	A	3	0	6	0	0	0	0	0
3-Sep	O	Cow	A	4	0	4	2	0	0	0	0
3-Sep	O	Cow	A	5	0	6	0	0	0	0	0
3-Sep	O	Cow	F	1	0	4	1	1	0	0	1
3-Sep	O	Cow	F	2	0	6	0	0	0	0	0
3-Sep	O	Cow	F	3	0	6	0	0	0	0	0
3-Sep	O	Cow	F	4	0	6	0	0	0	0	0
3-Sep	O	Cow	F	5	0	6	0	0	0	0	0
3-Sep	O	Cow	O	1	1	2	2	1	0	0	1
3-Sep	O	Cow	O	2	1	3	2	0	0	0	0
3-Sep	O	Cow	O	3	0	6	0	0	0	0	0
3-Sep	O	Cow	O	4	0	5	1	0	0	0	0
3-Sep	C	Cow	A	1	0	6	0	0	0	0	0
3-Sep	C	Cow	A	2	0	5	1	0	0	0	0
3-Sep	C	Cow	A	3	0	6	0	0	0	0	0
3-Sep	C	Cow	A	4	1	5	0	0	0	0	0
3-Sep	C	Cow	A	5	0	6	0	0	0	0	0
3-Sep	C	Cow	F	1	0	4	0	2	0	0	2
3-Sep	C	Cow	F	2	1	0	2	3	0	0	3
3-Sep	C	Cow	F	3	2	4	0	0	0	0	0
3-Sep	C	Cow	F	4	2	4	0	0	0	0	0
3-Sep	C	Cow	F	5	0	5	1	0	0	0	0
3-Sep	C	Cow	O	1	4	0	0	0	2	0	2
3-Sep	C	Cow	O	2	0	4	2	0	0	0	0
3-Sep	C	Cow	O	3	1	4	1	0	0	0	0
3-Sep	C	Cow	O	4	0	6	0	0	0	0	0
3-Sep	C	Cow	O	5	0	6	0	0	0	0	0

State wide precipitation gradient results.

Date	Location	Field	trt	sample #	TI	NI
1	Woodward	A	F	1	0	6
1	Woodward	A	F	2	0	6
1	Woodward	A	F	3	0	6
1	Woodward	A	F	4	0	6

1	Woodward	A	F	5	0	6
1	Woodward	A	F	6	0	6
1	Woodward	A	F	7	1	5
1	Woodward	A	F	8	0	6
1	Woodward	A	F	9	0	6
1	Woodward	A	F	10	0	6
1	Woodward	A	F	11	0	6
1	Woodward	A	F	12	0	6
1	Woodward	A	F	13	0	6
1	Woodward	A	F	14	0	6
1	Woodward	A	F	15	0	6
1	Woodward	A	F	16	0	6
1	Woodward	A	F	17	0	6
1	Woodward	A	F	18	0	6
1	Woodward	A	F	19	0	6
1	Woodward	A	F	20	0	6
1	Woodward	A	O	1	0	6
1	Woodward	A	O	2	0	6
1	Woodward	A	O	3	0	6
1	Woodward	A	O	4	0	6
1	Woodward	A	O	5	0	6
1	Woodward	A	O	6	0	6
1	Woodward	A	O	7	0	6
1	Woodward	A	O	8	0	6
1	Woodward	A	O	9	0	6
1	Woodward	A	O	10	0	6
1	Woodward	A	O	11	0	6
1	Woodward	A	O	12	0	6
1	Woodward	A	O	13	0	6
1	Woodward	A	O	14	0	6
1	Woodward	A	O	15	0	6
1	Woodward	A	O	16	0	6
1	Woodward	A	O	17	0	6
1	Woodward	A	O	18	0	6
1	Woodward	A	O	19	0	6
1	Woodward	A	O	20	0	6
1	Woodward	A	N	1	0	6
1	Woodward	A	N	2	0	6
1	Woodward	A	N	3	0	6
1	Woodward	A	N	4	0	6
1	Woodward	A	N	5	0	6

1	Woodward	A	N	6	0	6
1	Woodward	A	N	7	0	6
1	Woodward	A	N	8	0	6
1	Woodward	A	N	9	0	6
1	Woodward	A	N	10	0	6
1	Woodward	A	N	11	0	6
1	Woodward	A	N	12	0	6
1	Woodward	A	N	13	0	6
1	Woodward	A	N	14	0	6
1	Woodward	A	N	15	0	6
1	Woodward	A	N	16	0	6
1	Woodward	A	N	17	0	6
1	Woodward	A	N	18	0	6
1	Woodward	A	N	19	0	6
1	Woodward	A	N	20	0	6
1	Woodward	B	F	1	0	6
1	Woodward	B	F	2	0	6
1	Woodward	B	F	3	0	6
1	Woodward	B	F	4	0	6
1	Woodward	B	F	5	0	6
1	Woodward	B	F	6	0	6
1	Woodward	B	F	7	0	6
1	Woodward	B	F	8	0	6
1	Woodward	B	F	9	0	6
1	Woodward	B	F	10	0	6
1	Woodward	B	F	11	0	6
1	Woodward	B	F	12	0	6
1	Woodward	B	F	13	0	6
1	Woodward	B	F	14	0	6
1	Woodward	B	F	15	0	6
1	Woodward	B	F	16	0	6
1	Woodward	B	F	17	0	6
1	Woodward	B	F	18	0	6
1	Woodward	B	F	19	0	6
1	Woodward	B	F	20	0	6
1	Woodward	B	O	1	0	6
1	Woodward	B	O	2	0	6
1	Woodward	B	O	3	0	6
1	Woodward	B	O	4	0	6
1	Woodward	B	O	5	0	6
1	Woodward	B	O	6	0	6

1	Woodward	B	O	7	0	6
1	Woodward	B	O	8	0	6
1	Woodward	B	O	9	0	6
1	Woodward	B	O	10	0	6
1	Woodward	B	O	11	0	6
1	Woodward	B	O	12	0	6
1	Woodward	B	O	13	0	6
1	Woodward	B	O	14	0	6
1	Woodward	B	O	15	0	6
1	Woodward	B	O	16	0	6
1	Woodward	B	O	17	0	6
1	Woodward	B	O	18	0	6
1	Woodward	B	O	19	0	6
1	Woodward	B	O	20	0	6
1	Woodward	B	N	1	0	6
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1	Woodward	B	N	3	0	6
1	Woodward	B	N	4	0	6
1	Woodward	B	N	5	0	6
1	Woodward	B	N	6	0	6
1	Woodward	B	N	7	0	6
1	Woodward	B	N	8	0	6
1	Woodward	B	N	9	0	6
1	Woodward	B	N	10	0	6
1	Woodward	B	N	11	0	6
1	Woodward	B	N	12	0	6
1	Woodward	B	N	13	0	6
1	Woodward	B	N	14	0	6
1	Woodward	B	N	15	0	6
1	Woodward	B	N	16	0	6
1	Woodward	B	N	17	0	6
1	Woodward	B	N	18	0	6
1	Woodward	B	N	19	0	6
1	Woodward	B	N	20	0	6
1	Goodwell	A	F	1	0	6
1	Goodwell	A	F	2	0	6
1	Goodwell	A	F	3	0	6
1	Goodwell	A	F	4	1	5
1	Goodwell	A	F	5	0	6
1	Goodwell	A	F	6	0	6
1	Goodwell	A	F	7	0	6

1	Goodwell	A	F	8	0	6
1	Goodwell	A	F	9	0	6
1	Goodwell	A	F	10	0	6
1	Goodwell	A	F	11	0	6
1	Goodwell	A	F	12	0	6
1	Goodwell	A	F	13	0	6
1	Goodwell	A	F	14	0	6
1	Goodwell	A	F	15	0	6
1	Goodwell	A	F	16	0	6
1	Goodwell	A	F	17	0	6
1	Goodwell	A	F	18	0	6
1	Goodwell	A	F	19	0	6
1	Goodwell	A	F	20	0	6
1	Goodwell	A	O	1	0	6
1	Goodwell	A	O	2	0	6
1	Goodwell	A	O	3	0	6
1	Goodwell	A	O	4	0	6
1	Goodwell	A	O	5	0	6
1	Goodwell	A	O	6	0	6
1	Goodwell	A	O	7	1	5
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1	Goodwell	A	O	9	0	6
1	Goodwell	A	O	10	0	6
1	Goodwell	A	O	11	0	6
1	Goodwell	A	O	12	0	6
1	Goodwell	A	O	13	0	6
1	Goodwell	A	O	14	0	6
1	Goodwell	A	O	15	0	6
1	Goodwell	A	O	16	0	6
1	Goodwell	A	O	17	0	6
1	Goodwell	A	O	18	0	6
1	Goodwell	A	O	19	0	6
1	Goodwell	A	O	20	0	6
1	Goodwell	A	N	1	0	6
1	Goodwell	A	N	2	0	6
1	Goodwell	A	N	3	0	6
1	Goodwell	A	N	4	0	6
1	Goodwell	A	N	5	0	6
1	Goodwell	A	N	6	0	6
1	Goodwell	A	N	7	0	6
1	Goodwell	A	N	8	0	6

1	Goodwell	A	N	9	0	6
1	Goodwell	A	N	10	0	6
1	Goodwell	A	N	11	0	6
1	Goodwell	A	N	12	0	6
1	Goodwell	A	N	13	0	6
1	Goodwell	A	N	14	0	6
1	Goodwell	A	N	15	0	6
1	Goodwell	A	N	16	0	6
1	Goodwell	A	N	17	0	6
1	Goodwell	A	N	18	0	6
1	Goodwell	A	N	19	0	6
1	Goodwell	A	N	20	0	6
1	Goodwell	B	F	1	0	6
1	Goodwell	B	F	2	0	6
1	Goodwell	B	F	3	0	6
1	Goodwell	B	F	4	0	6
1	Goodwell	B	F	5	0	6
1	Goodwell	B	F	6	0	6
1	Goodwell	B	F	7	0	6
1	Goodwell	B	F	8	0	6
1	Goodwell	B	F	9	0	6
1	Goodwell	B	F	10	0	6
1	Goodwell	B	F	11	0	6
1	Goodwell	B	F	12	0	6
1	Goodwell	B	F	13	0	6
1	Goodwell	B	F	14	0	6
1	Goodwell	B	F	15	0	6
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1	Goodwell	B	F	17	0	6
1	Goodwell	B	F	18	0	6
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1	Goodwell	B	O	5	0	6
1	Goodwell	B	O	6	0	6
1	Goodwell	B	O	7	0	6
1	Goodwell	B	O	8	0	6
1	Goodwell	B	O	9	0	6

1	Goodwell	B	O	10	0	6
1	Goodwell	B	O	11	0	6
1	Goodwell	B	O	12	0	6
1	Goodwell	B	O	13	0	6
1	Goodwell	B	O	14	0	6
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1	Goodwell	B	O	19	0	6
1	Goodwell	B	O	20	0	6
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1	Goodwell	B	N	16	0	6
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1	Goodwell	B	N	18	0	6
1	Goodwell	B	N	19	0	6
1	Goodwell	B	N	20	0	6
1	Stillwater	A	O	1	0	6
1	Stillwater	A	O	2	1	5
1	Stillwater	A	O	3	0	6
1	Stillwater	A	O	4	2	4
1	Stillwater	A	O	5	0	6
1	Stillwater	A	O	6	0	6
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1	Stillwater	A	O	8	0	6
1	Stillwater	A	O	9	0	6
1	Stillwater	A	O	10	0	6

1	Stillwater	A	O	11	6	0
1	Stillwater	A	O	12	0	6
1	Stillwater	A	O	13	0	6
1	Stillwater	A	O	14	0	6
1	Stillwater	A	O	15	0	6
1	Stillwater	A	O	16	0	6
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1	Stillwater	A	O	18	0	6
1	Stillwater	A	O	19	0	6
1	Stillwater	A	O	20	0	6
1	Stillwater	A	F	1	0	6
1	Stillwater	A	F	2	0	6
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1	Stillwater	A	F	18	0	6
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1	Stillwater	A	N	7	0	6
1	Stillwater	A	N	8	0	6
1	Stillwater	A	N	9	0	6
1	Stillwater	A	N	10	0	6
1	Stillwater	A	N	11	0	6

1	Stillwater	A	N	12	0	6
1	Stillwater	A	N	13	0	6
1	Stillwater	A	N	14	0	6
1	Stillwater	A	N	15	0	6
1	Stillwater	A	N	16	0	6
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1	Stillwater	A	N	19	0	6
1	Stillwater	A	N	20	0	6
1	Stillwater	B	O	1	0	6
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1	Stillwater	B	O	3	0	6
1	Stillwater	B	O	4	0	6
1	Stillwater	B	O	5	0	6
1	Stillwater	B	O	6	0	6
1	Stillwater	B	O	7	0	6
1	Stillwater	B	O	8	0	6
1	Stillwater	B	O	9	0	6
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1	Stillwater	B	O	14	0	6
1	Stillwater	B	O	15	0	6
1	Stillwater	B	O	16	0	6
1	Stillwater	B	O	17	0	6
1	Stillwater	B	O	18	0	6
1	Stillwater	B	O	19	0	6
1	Stillwater	B	O	20	0	6
1	Stillwater	B	F	1	0	6
1	Stillwater	B	F	2	0	6
1	Stillwater	B	F	3	0	6
1	Stillwater	B	F	4	0	6
1	Stillwater	B	F	5	0	6
1	Stillwater	B	F	6	0	6
1	Stillwater	B	F	7	0	6
1	Stillwater	B	F	8	0	6
1	Stillwater	B	F	9	0	6
1	Stillwater	B	F	10	0	6
1	Stillwater	B	F	11	0	6
1	Stillwater	B	F	12	0	6

1	Stillwater	B	F	13	0	6
1	Stillwater	B	F	14	0	6
1	Stillwater	B	F	15	0	6
1	Stillwater	B	F	16	0	6
1	Stillwater	B	F	17	0	6
1	Stillwater	B	F	18	0	6
1	Stillwater	B	F	19	0	6
1	Stillwater	B	F	20	0	6
1	Stillwater	B	N	1	0	6
1	Stillwater	B	N	2	0	6
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1	Stillwater	B	N	4	0	6
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1	Stillwater	B	N	6	0	6
1	Stillwater	B	N	7	0	6
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1	Stillwater	B	N	9	0	6
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1	Stillwater	B	N	12	0	6
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1	Stillwater	B	N	16	0	6
1	Stillwater	B	N	17	0	6
1	Stillwater	B	N	18	0	6
1	Stillwater	B	N	19	0	6
1	Stillwater	B	N	20	0	6
1	Haskell	A	O	1	0	6
1	Haskell	A	O	2	0	6
1	Haskell	A	O	3	0	6
1	Haskell	A	O	4	0	6
1	Haskell	A	O	5	0	6
1	Haskell	A	O	6	0	6
1	Haskell	A	O	7	0	6
1	Haskell	A	O	8	0	6
1	Haskell	A	O	9	0	6
1	Haskell	A	O	10	0	6
1	Haskell	A	O	11	0	6
1	Haskell	A	O	12	0	6
1	Haskell	A	O	13	0	6

1	Haskell	A	O	14	1	5
1	Haskell	A	O	15	0	6
1	Haskell	A	O	16	0	6
1	Haskell	A	O	17	0	6
1	Haskell	A	O	18	0	6
1	Haskell	A	O	19	0	6
1	Haskell	A	O	20	0	6
1	Haskell	A	F	1	0	6
1	Haskell	A	F	2	2	4
1	Haskell	A	F	3	0	6
1	Haskell	A	F	4	0	6
1	Haskell	A	F	5	0	6
1	Haskell	A	F	6	0	6
1	Haskell	A	F	7	0	6
1	Haskell	A	F	8	0	6
1	Haskell	A	F	9	0	6
1	Haskell	A	F	10	0	6
1	Haskell	A	F	11	0	6
1	Haskell	A	F	12	0	6
1	Haskell	A	F	13	0	6
1	Haskell	A	F	14	0	6
1	Haskell	A	F	15	0	6
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1	Haskell	A	F	19	0	6
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1	Haskell	A	N	4	5	1
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1	Haskell	A	N	6	3	3
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1	Haskell	A	N	9	0	6
1	Haskell	A	N	10	1	5
1	Haskell	A	N	11	0	6
1	Haskell	A	N	12	0	6
1	Haskell	A	N	13	0	6
1	Haskell	A	N	14	0	6

1	Haskell	A	N	15	1	5
1	Haskell	A	N	16	0	6
1	Haskell	A	N	17	0	6
1	Haskell	A	N	18	0	6
1	Haskell	A	N	19	0	6
1	Haskell	A	N	20	0	6
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1	Haskell	B	O	3	0	6
1	Haskell	B	O	4	0	6
1	Haskell	B	O	5	0	6
1	Haskell	B	O	6	0	6
1	Haskell	B	O	7	0	6
1	Haskell	B	O	8	0	6
1	Haskell	B	O	9	0	6
1	Haskell	B	O	10	0	6
1	Haskell	B	O	11	0	6
1	Haskell	B	O	12	0	6
1	Haskell	B	O	13	0	6
1	Haskell	B	O	14	0	6
1	Haskell	B	O	15	0	6
1	Haskell	B	O	16	0	6
1	Haskell	B	O	17	0	6
1	Haskell	B	O	18	0	6
1	Haskell	B	O	19	0	6
1	Haskell	B	O	20	0	6
1	Haskell	B	F	1	0	6
1	Haskell	B	F	2	0	6
1	Haskell	B	F	3	0	6
1	Haskell	B	F	4	0	6
1	Haskell	B	F	5	0	6
1	Haskell	B	F	6	0	6
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1	Haskell	B	F	8	0	6
1	Haskell	B	F	9	0	6
1	Haskell	B	F	10	0	6
1	Haskell	B	F	11	0	6
1	Haskell	B	F	12	0	6
1	Haskell	B	F	13	0	6
1	Haskell	B	F	14	0	6
1	Haskell	B	F	15	0	6

1	Haskell	B	F	16	0	6
1	Haskell	B	F	17	0	6
1	Haskell	B	F	18	0	6
1	Haskell	B	F	19	0	6
1	Haskell	B	F	20	0	6
1	Haskell	B	N	1	0	6
1	Haskell	B	N	2	0	6
1	Haskell	B	N	3	0	6
1	Haskell	B	N	4	0	6
1	Haskell	B	N	5	0	6
1	Haskell	B	N	6	0	6
1	Haskell	B	N	7	0	6
1	Haskell	B	N	8	0	6
1	Haskell	B	N	9	0	6
1	Haskell	B	N	10	0	6
1	Haskell	B	N	11	0	6
1	Haskell	B	N	12	0	6
1	Haskell	B	N	13	0	6
1	Haskell	B	N	14	0	6
1	Haskell	B	N	15	0	6
1	Haskell	B	N	16	0	6
1	Haskell	B	N	17	0	6
1	Haskell	B	N	18	0	6
1	Haskell	B	N	19	0	6
1	Haskell	B	N	20	0	6
2	Stillwater	A	O	1	0	6
2	Stillwater	A	O	2	1	5
2	Stillwater	A	O	3	0	6
2	Stillwater	A	O	4	2	4
2	Stillwater	A	O	5	0	6
2	Stillwater	A	O	6	0	6
2	Stillwater	A	O	7	0	6
2	Stillwater	A	O	8	0	6
2	Stillwater	A	O	9	0	6
2	Stillwater	A	O	10	0	6
2	Stillwater	A	O	11	6	0
2	Stillwater	A	O	12	0	6
2	Stillwater	A	O	13	0	6
2	Stillwater	A	O	14	0	6
2	Stillwater	A	O	15	0	6
2	Stillwater	A	O	16	0	6

2	Stillwater	A	O	17	0	6
2	Stillwater	A	O	18	0	6
2	Stillwater	A	O	19	0	6
2	Stillwater	A	O	20	0	6
2	Stillwater	A	F	1	0	6
2	Stillwater	A	F	2	0	6
2	Stillwater	A	F	3	0	6
2	Stillwater	A	F	4	0	6
2	Stillwater	A	F	5	0	6
2	Stillwater	A	F	6	0	6
2	Stillwater	A	F	7	0	6
2	Stillwater	A	F	8	0	6
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VITA

Kyle Joseph Risser

Candidate for the Degree of

Master of Science

Thesis: COMPARISON OF ENTOMOPATHOGENIC NEMATODES (EPN) PREVALENCE AND DIVERSITY IN ORGANIC AND CONVENTIONAL BEEF AND WHEAT SYSTEMS AND ACROSS A STATE WIDE PRECIPITATION GRADIENT IN OKLAHOMA

Major Field: Entomology

Biographical:

Education:

Completed the requirements for the Master of Science in Entomology at Oklahoma State University, Stillwater, Oklahoma in December, 2012.

Completed the requirements for the Bachelor of Science in Entomology at University of California Riverside, Riverside, California in 2009.

Experience:

Senior Laboratory assistant for the Dhanukar Laboratory during its start up at UCR. Ordered and installed all necessary equipment. 07/09-08/10

Emesinae (Reduviidae) systematics under Dr. Weirauch at UCR. Re-arranged phylogony of the tribe through for-tarsi comparison using electon microscopy and genetic phylogeny. 01/07-01/08

Triatominae infection rate with *T. cruzi* in southern California, under Dr. Weirauch, was determined through dissecting out hind guts of field collected specimens and preforming PCR on positive samples to confirm species of trypanosome as *T. cruzi*. 01/07-01/08

Professional Memberships:

ESA: Entomological Society of America

ESA: Ecological Society of America

Name: Kyle Risser Date of Degree: December, 2012

Institution: Oklahoma State University Location: Stillwater, Oklahoma

Title of Study: COMPARISON OF ENTOMOPATHOGENIC NEMATODE (EPN) PREVALENCE AND DIVERSITY IN ORGANIC AND CONVENTIONAL BEEF AND WHEAT PRODUCTION SYSTEMS AND ACROSS A STATE WIDE PRECIPITATION GRADIENT IN OKLAHOMA

Pages in Study: 148 Candidate for the Degree of Master of Science

Major Field: Entomology

Findings and Conclusions:

Entomopathogenic nematodes in the families Steinernematidae and Heterorhabditidae are obligate parasites of arthropods, exist naturally in soils worldwide, and have been used to suppress soil-dwelling insect pests. Little is known about EPN diversity within Oklahoma. Ranging from east to west, Oklahoma is home to 11 different ecoregions, 9 precipitation zones and 7 soil orders. This study aimed to characterize EPN communities throughout these diverse habitats. An additional objective of this study was to compare EPN communities in organic versus conventional beef and wheat production systems within the same ecoregion. A combination of bioassay technique and molecular identification was used to identify EPN species . Soil samples were subjected to bioassay using *G. mellonella* to ascertain infection rates by EPN. EPN were identified, initially by infected *G. mellonella* symptoms. Based on these symptoms, appropriate primers were chosen to amplify regions of the ITS gene. These regions were then sequenced to confirm identification. The *Heterorhabditis* species identified was *Heterorhabditis bacteriophora*. *The Steinernema* species of EPN identified included: *Steinernema feltiae*, *S. texanum*, *S. glaseri*. *S. carpocapsae,* and *S. reiobrave*. Within the organic and conventional fields, overall infection rates were 2.06% in organic wheat, 6.73% in conventional wheat, 7.33% in organic pasture, and 6.67% in conventional pasture. This study showed a higher incidence of EPN in organic wheat fields than conventional wheat fields; pastures than agricultural fields; and a positive correlation with the increase in soil moisture as you move eastward across the state.

ADVISER'S APPROVAL: Dr. Carmen M. Greenwood