

Effects of human actions on four ecological systems,  
with a focus on trophic relationships

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Margaret Candace Kosmala

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Dr. Craig Packer, Dr. G. David Tilman

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*for Ben Hardt,  
my friend, partner, and teammate in life*

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## **Chapter 1 : Introduction**

Humans have touched every corner of the planet. There are, perhaps, no parts of earth remaining that haven't been impacted by human actions, either purposefully or unintentionally. Human agriculture uses 12% of Earth's ice-free land surface and livestock grazing occupies another 26% (Foley et al. 2011). Even in remote, seemingly pristine areas, nitrogen deposition has increased as the result of industrial emissions, and plastic trash washes up on unpopulated beaches. The world's oceans are widely overfished (Pauly et al. 2002), and anthropogenic increases in CO<sub>2</sub> in the atmosphere are leading to ocean acidification, with implications for ocean life. Humans have also accelerated the dispersal rate of many organisms around the planet through horticulture, the pet trade, and unintentional transfers, which has led to unwanted invasions of exotic species and outbreaks of disease.

In order to become better stewards of Earth's biota, we must better understand how our actions affect ecosystems and species. As is increasingly becoming apparent, it is not only the direct actions of people on their environment that matter, but also the indirect effects. Because species are interconnected through their interactions with one another, a decision to manage a landscape or species in a particular way will have consequences for multiple species. One of the most ubiquitous and important links between species is the trophic, or feeding, relationship. When one organism consumes part or all of another organism, both energy and nutrients flow from one to the other. The combination of all trophic relationships across an ecosystem has a great impact on how that system functions, as well as the distribution and dynamics of species within it (Polis et al. 1997).

Here, I examine four ecological systems and the direct and indirect effects of human change on them, with a focus on trophic relationships. In Chapter 2, I investigate the environmental impact of beef production. Pastoralism has been around for thousands of years, and today beef production uses most of the world's 3.8 billion hectares of pasture and rangeland. There are many methods of cattle management, and I compare various methods in terms of land use, water use, energy use, and greenhouse gas

emissions. In particular, I focus on the United States industrial system, which may become a model for other countries as their populations becomes wealthier and demand more meat (Alexandratos and Bruinsma 2012).

In Chapter 3, I present the results of an experiment testing the effects of plant diversity on the biological control of an agricultural pest species. Much of the land in the Midwest consists of monoculture crops, predominantly corn and soybean. An invasive pest of soybean, the soybean aphid (*Aphis glycines*), has spread throughout the region since the 1990's, causing widespread yield losses (Ragsdale et al. 2004). Landscapes with more diverse land use have been shown to increase the abundance of soybean aphid predators and parasitoids compared with less diverse landscapes (Gardiner et al. 2009). I hypothesized that an increase in local plant species diversity in these landscapes leads to an increase in the abundance of insects that prey on and parasitize soybean aphids. Understanding whether increases in local plant diversity around soybean plants would enhance biological control of the soybean aphid has important implications for the management of soybean crops. Professor George Heimpel contributed to this chapter through discussions of experimental design, entomological biology, experimental results and interpretation, and assisting with field work. This experiment was funded by USDA grant 2011-67009-30027 and by an NSF LTER grant funding Cedar Creek Ecosystem Science Reserve.

In Chapter 4, I analyze a twenty-year experiment to better understand the long-term dynamics of plant communities. The experiment began with a large addition of seeds of up to 54 species in each of 60 plots in an already diverse Minnesotan savanna. The seed-addition treatments allow for an analysis of the importance of dispersal limitation in the plant community, while the long-term nature of the experiment highlights the importance of local competition among plants for limiting resources and the local movement of species over time. Understanding the fundamental drivers of plant community dynamics is important for conservation management of grassland and savanna systems. These habitats are being replaced by agriculture and are often

fragmented, which affects dispersal rates and changes the balance of regional and local processes governing long term species dynamics.

In Chapter 5, I use modeling techniques to investigate the dynamics of an emerging wildlife disease in the lion (*Panthera leo*) population in Kruger National Park, South Africa. Bovine tuberculosis (*Mycobacterium bovis*), was inadvertently introduced to southern Africa in the 19th century (Renwick et al. 2006), and has been spreading through the park's African buffalo (*Syncerus caffer*) population for several decades. Lions contract the disease from infected buffalo and from one another and die from it, but the epidemiology of bovine tuberculosis in lions is not well understood, nor easily studied. My model explores the logical space of bovine tuberculosis dynamics in lions, using knowledge about lion demography and behavior, and prevalence rates in lions and buffalo. The results provide important information to park management about the likely long-term dynamics of the disease in lions, lion population size, and the efficacy of intervention approaches. The modeling approach is one that can be used in managing other emerging wildlife diseases. The research in this chapter grew out a 2009 risk assessment workshop facilitated by the Conservation Breeding Specialists Group (CBSG), Apple Valley, Minnesota; many workshop participants shared their expertise. Dr. Phil Miller of CBSG was instrumental in developing early model designs. Models were run at the Minnesota Supercomputer Institute, which provided high performance computing facilities and technical help.

## **Chapter 2 : Environmental impacts of beef production**

Worldwide beef production, which has significant environmental costs, is likely to increase as global population and per capita real incomes grow. Newly industrialized and emerging countries may turn to industrial beef production to maximize production efficiency and profit. Because U.S. beef production may be indicative of the type of industrial system that would be adopted by emerging nations, we conducted a life cycle analysis (LCA) of the full land use, water use, fossil energy use, and greenhouse gas emissions associated with industrial U.S. beef production from calf production through household purchase and consumption. We find that it takes 345 m<sup>2</sup>-yr of land, 1,740 liters water, and 146 MJ fossil energy to produce one kg of beef eaten, while emitting 63 kg CO<sub>2</sub>-eq in greenhouse gases. More than a quarter of fossil energy use and about one third of greenhouse gas emissions occur after the farm gate, emphasizing the need for full-cycle analyses. Additionally, our calculations include wastage, including at the consumer level, which yields higher estimates of beef environmental impact than previous studies. We calculate that feed conversion efficiency for U.S. beef may be as high as 160:1 when all cattle feed is taken into account and all loss from cradle-to-fork is accounted for. In reviewing beef production methods from around the world, we found that no method optimized all environmental impacts; in particular, greenhouse gas emissions were always high due to cattle methane output.

### **Introduction**

The 21st century poses a dual challenge for society – increasing food production to keep pace with a growing human population and its per capita demands and ensuring that food is produced with lower environmental impacts. Demand for meat is expected to more than double by 2050 as world population grows and citizens of transitioning countries become wealthier (Pelletier and Tyedmers 2010). However, global meat production causes environmental harm (Steinfeld et al. 2006). World meat production accounts for 75% of agricultural land use and much of projected future land clearing globally (Foley et al. 2011) and already causes greater global greenhouse gas emissions

(18% of global total) than all forms of transportation combined (14% of total) (Stehfest et al. 2009).

Beef accounts for about a quarter of world meat production (FAO 2009).

Although reductions in beef production and consumption are known to have environmental and human health benefits (McMichael et al. 2007, Foley et al. 2011, Tscharntke et al. 2012), demand for beef is increasing in many areas of the world. Beef supply is 28.2 kg carcass weight/capita/year in industrialized countries, but only 9.6 kg carcass weight/capita/year worldwide (FAO 2009). If everyone alive today consumed as much beef as people in industrialized countries, global production would need to increase 200% (183 million tonnes carcass weight versus 62 million tonnes).

The U.S. is the largest producer of beef in the world, producing 19% of the world's beef, most of which is consumed domestically (FAO 2009). The hallmark of U.S. beef is intensive industrialized production of tender marbled steaks via rapid growth of cattle on high-energy feeds. This system is made economically feasible by a large supply of low-cost crop feeds. In much of the rest of the world, cattle are produced in extensive systems in which cattle graze on pastures or rangeland until slaughtered. These cattle take longer to reach slaughter weight, producing less beef per year. As global demand for beef rises, beef production in developing economies is forecast to switch to industrial systems using concentrate feed (Alexandratos and Bruinsma 2012).

While beef production is often cited as disproportionately burdensome in terms of sustainability (Figure A1-5), studies of the environmental impacts of U.S. industrial beef production have often considered just one type of impact (e.g. Beckett and Oltjen 1993, Pimentel et al. 1997, 2004, Subak 1999, Phetteplace et al. 2001, Hoekstra and Chapagain 2006) or have stopped at the farm-gate, providing only a partial picture of beef production's environmental impacts (Pelletier et al. 2010, Capper 2011). Additionally, many analyses of beef production have not fully accounted for waste and losses within the system (e.g. Cederberg and Stadig 2003, Ogino et al. 2004, 2007, Beauchemin et al. 2010), and results are often reported in functional units that do not compare easily across other meat and food products.



Of the 34 million head of cattle that are slaughtered in the United States annually, 80-90% are fattened in industrial feedlots containing over 1,000 cattle (Lowe and Gereffi 2009). The majority of slaughtered cattle are grown specifically for their meat; however about 20% of animals slaughtered for meat are cull cows from both the beef and dairy industries and dairy calves slaughtered for veal (USDA ERS 2013). In this paper we focus on typical beef cattle that are produced for their meat and are finished in large facilities, as they make up the majority of U.S. beef production.

We investigate the environmental impacts of the U.S. industrial beef system, focusing on land use, water use, fossil energy use, and greenhouse gas emissions. Because it has been suggested that switching from industrially produced beef to pasture beef could mitigate environmental impacts associated with industrial methods (Foley et al. 2011), we also analyze the potential costs and benefits of U.S. grass-fed “green-label” beef. We survey the literature to compare the environmental impacts of beef-production methods throughout the world with U.S. industrial production. We analyze U.S. industrial beef production up to human consumption of beef and include loss and waste within the system. Finally, we calculate cradle-to-grave protein and energy efficiencies for U.S. beef to show how the choice of functional unit affects comparisons of beef with other animal products.

## **Methods**

We used life cycle assessment to calculate the cradle-to-grave land use, water use, fossil energy use, and greenhouse gas emissions of industrial beef production in the U.S. We specifically examined ten stages of beef production: land management and feed production; feed transport, storage, and processing; cow-calf herd management; growth and finishing of cattle; slaughter and carcass processing; beef packaging; beef distribution; food service; retail sales; and household purchasing and consumption. A detailed description of these stages can be found in Appendix 2.

Data were taken from national-level statistics when possible, and otherwise from the peer-reviewed literature, extension publications, and expert beef researchers. Major

sources of national-level data included the U.S. Department of Agriculture (USDA)'s National Agricultural Statistics Service, Agricultural Marketing Service, and Economic Research Service; the U.S. Census Bureau's Commodity Flow Survey; the Federal Highway Administration's Vehicle Inventory and Use Survey and National Household Travel Survey; the Centers for Disease Control and Prevention's National Health and Nutrition Examination Survey; and the U.S. Energy Information Administration's Residential Energy Consumption Survey. When statistics and published data were unavailable, we assumed typical practices for crop and pasture management and cattle husbandry. Detailed descriptions of the data and sources used can be found in Appendix 2.

### ***Impacts Calculations and Functional Units***

We calculated the land use, water use, energy consumption, and greenhouse gas emissions of U.S. industrial beef production from cradle to grave. We excluded from consideration the production of capital goods such as buildings, roads, and vehicles, all packaging materials except for the direct packaging of the beef itself, chemicals used for cleaning slaughterhouses and packinghouses, the lighting, heating, air conditioning, cleaning, etc. associated with maintaining a residential kitchen, and impacts of beef after it has passed through the human digestive system.

To facilitate comparison with other studies, we calculate each impact at four different steps in the production process: per kg live weight of an animal at slaughter; per kg hot carcass, which is the slaughtered animal after removal of hide, head, feet, gastrointestinal tract, internal organs, and blood; per kg boxed beef, which represents the weight of primal cuts; and per kg boneless edible beef. In addition, to determine the impact of food waste, we analyze impacts per kg of eaten beef.

Land is reported as both total land use, as well as arable land use. We used the USDA's definition of "acres irrigated" to calculate water used in growing crops, hay and pastures; rainwater was not included, nor were naturally occurring surface waters. Energy

use was defined as fossil fuel use. Greenhouse gas emissions were determined using standard IPCC methods and conversion factors for methane and nitrous oxide.

We calculated a soil carbon sequestration “opportunity cost” (*sensu* Garnett 2009) to account for the differential ability of land to reduce global warming potential under different management scenarios; we used native vegetation as a baseline and determined the difference in carbon sequestration between the baseline and land used under industrial beef production. We used rates of 0.2, 0.44, 0.3, and 0.5 Mg C/ha/yr for rangeland, alfalfa, other hay, and Southeast grasslands respectively (Watson et al. 2000, Robertson 2000, Schuman et al. 2002) and a rate of 1.01 Mg C/ha/yr for tallgrass prairie (Conant et al. 2001). We assumed no sequestration on cropland used for commodity crops, including those used for wheat pastures, because they are cultivated annually.

### ***Byproduct calculations***

We assume that the winter wheat pastures used to grow stocker (mid-stage) cattle are dual-purpose, producing a wheat crop after cattle are removed. We determined the relative value of grazing versus grain production on a per-hectare basis and apportioned environmental impacts of wheat production to grazing and grain production based on the ratio.

We determined the relative weight and average price of each main product and cattle feed co-product created in the production of corn ethanol, soybean biodiesel, and canola oil. We apportioned environmental impacts to products based on relative value per unit of raw ingredient.

We calculated the fraction of culled cows and bulls associated with each calf, determined the price for each on a weight basis, and apportioned environmental impacts associated with cow-calf herds between calves and cows/bulls according to their relative value.

Cattle manure can be a valuable fertilizer or a waste product. Sometimes owners can sell manure, sometimes they can give it away, and sometimes they must pay to have

it removed (Koelsch et al. 2000). However, because no national statistics are available on beef feedlot manure outcomes, we do not include these in our analyses.

We determined the total economic value of beef byproducts such as hides, tallow, bone meal, blood meal, and organs, and apportioned environmental impacts of beef production to beef and byproducts based on their relative values.

### ***Green-Label Beef***

We modified our LCA to investigate the environmental impacts associated with a “green-label” management approach of intensive rotational grazing on high quality Midwest land. We assumed that only the cattle-production phases changed and that slaughter, processing, distribution, and consumption were the same as for industrially produced beef. In keeping with green-label production practices, we assumed that no hormones, insecticides, or antibiotics were used. Drinking water rates were the same as those used for industrial beef production, but we assumed that all drinking water was diverted from its source. Energy use, greenhouse gas emissions, and byproduct allocations were calculated using the same methods as for industrial beef production. We assumed carbon sequestration on managed pastures was reduced by 10% compared to native prairie (Derner et al. 2006, Derner and Schuman 2007). We excluded energy for on-farm transportation, electric fencing, and shelter lighting.

### ***LCA Survey***

We surveyed published beef life-cycle assessments to investigate whether other beef production methods might have less environmental impact than industrial production. We compiled 29 research articles and technical reports assessing beef production in Great Britain, Europe, Sweden, Australia, Japan, the African Sahel, Brazil, Canada, and the United States (Table A3-1). These studies covered the majority of beef production systems in the world, including subsistence herding, exclusive pasturing on temperate grasslands, intensive feedlot rearing, hybrid pasture-feedlot systems, systems reliant on dairy calves as inputs, and systems with no dairy calf input. We extracted values for greenhouse gas emissions, fossil energy use, land occupation, water use,

acidification potential, and eutrophication potential, although no one study reported all six impacts. In order to make comparisons, we converted all impacts to a functional unit of live weight using conversion factors specific to each study when possible and using standard factors otherwise.

## **Results & Discussion**

### ***Impacts of U.S. Industrial Beef Production***

It takes 345 m<sup>2</sup>-yr of land to produce each kg of U.S. beef eaten. As the average American eats about 22 kg of beef per year (Economic Research Service (ERS) 2010), 7,570 m<sup>2</sup> per capita is perpetually devoted to U.S. beef production. The vast majority of this land (92%) is rangeland used in the cow-calf stage of production. In addition to rangeland, 1 kg of beef eaten requires 9.7 m<sup>2</sup>-yr of high-quality hay and pasture and 16.8 m<sup>2</sup>-yr of cropland.

For each kg of beef that is eaten in the U.S., 1,740 liters (460 gallons) of water have been pumped from surface water and ground water sources, for a total of 38,200 liters per person per year. This is more than a quarter of U.S. annual per capita household water use (Kenny et al. 2009). Over three-quarters of water used in beef production is used to irrigate crops and another 17% is drinking water for cattle. Most (62%) of the irrigation water is groundwater. Corn and alfalfa account for 85% of irrigation water used in beef production. Over half of the water used for beef production is withdrawn in the Midwest, about twenty percent is irrigation in the arid Great Plains and the Southeast, and an additional 12% of water use is drawn from the beef feedlot states of Texas, Nebraska, Kansas, Iowa, and Colorado.

It takes 146 MJ to produce an eaten kg of beef in the U.S., for a yearly per capita energy consumption of 3,200 MJ; this is comparable to U.S. monthly per capita household energy use of 3,070 MJ (Energy Information Administration 2009). Of this amount, 36% is used in the production of corn, mainly for the production of nitrogen fertilizer. Another 13% of energy use derives from the production of hormones, insecticides, and antibiotics used in cattle management. Slaughtering and processing,

food service, retail and household, and transportation also contribute to energy use. About a third of all energy use occurs after the farm gate.

The production and consumption of beef creates 63 kg CO<sub>2</sub>-eq. per kg of beef eaten, equivalent to the greenhouse gas emissions of the average U.S. car driving 1,800 km (1,120 miles). About half of the emissions are directly from cattle and their manure. Another quarter of emissions are due to the food service industry, and particularly the fast-food sector; the decomposition of fast-food packaging and plastic utensils in landfills creates substantial emissions. Another 11% of total emissions occur from management of cropland and pastures. The land used to produce U.S. industrial beef could sequester 13 kg CO<sub>2</sub>-eq. per kg of beef eaten if it reverted to native vegetation. The majority of this sequestration would be on corn land that could sequester carbon as tallgrass prairie and degraded grazed rangeland that, if restored, could sequester additional carbon as shortgrass prairie (Schuman et al. 2002). Over time, these prairies would sequester less carbon annually, reducing the carbon sequestration opportunity cost by ~40% after fifty years (Post et al. 2004).

### ***Green-Label Beef***

We find that green-label beef requires one-sixth the land as industrially produced beef, but that it requires more than two and half times the amount of good quality arable land as industrial beef (Figure 2-1A). Assumptions about forage yield and pasture utilization greatly affect these results; it is conceivable that good management practices and research into increasing forage yields could reduce this land requirement.

Green-label beef requires substantially less water than industrial beef, due to the decreased need for irrigation (Figure 2-1B). Likewise, energy use for green-label beef is significantly less due to the absence of energy-intensive crops and pharmaceuticals, and the fact that animals do not need to be transported between growing phases (Figure 2-1C).

Greenhouse gas emissions are fairly comparable for green-label and industrial beef (Figure 2-1D). While green-label beef does not incur the emissions associated with

crops, cattle used for green-label beef grow more slowly, must live longer and so produce more methane and nitrous oxide per kg of edible beef than industrial cattle.

When the carbon sequestration opportunity cost is included, however, green-label beef performs better than industrial beef (Figure 2-1D). Opportunity costs for green-label beef are 2 kg CO<sub>2</sub>-eq/kg beef.

While we could not tabulate numbers for all environmental impacts, green-label beef would be expected to outperform industrial beef in such areas as air quality and smell, dust, and antibiotic resistant pathogen production. Water pollution and erosion would be expected to decrease, although manure runoff from fields in extreme storm events would continue to be a concern.

### *Literature Survey of Beef LCAs*

Of the 29 beef life-cycle assessments examined, 23 reported greenhouse gas emissions, 11 reported energy use, 8 reported land use, and 7 reported water use. Seven LCAs considered at least one impact past the farm gate.

All together, the LCAs considered 73 scenarios. We scored each scenario on whether or not it used each of four production methods: feedlot; managed grazing, in which pasture fertility and/or composition is actively managed; extensive grazing done on low-quality or non-arable land; and dairy input, in which offspring from dairy cows are grown for meat. Many scenarios involved compound methods, such as employing both managed and extensive grazing, or using extensive grazing for the cow-calf phase and finishing in a feedlot. We extracted land use, water use, fossil energy use, and greenhouse gas emissions results for each scenario and converted the values to a per kg live weight basis at the farm gate, if they were not already reported as such. We used conversion metrics particular to each study when available and otherwise used a standard dressing percentage of 57% for Cederberg et al. (2009) and a carcass-to-beef conversion of 40% for Pimentel et al. (1997, 2004), Cederberg and Stadig (2003), Hoekstra and Chapagain (2006). One scenario involving grazing on organic soils (Edwards-Jones et al. 2009) was excluded due to extremely high greenhouse gas emissions. Two LCAs

(Weidema et al. 2008, Roy et al. 2012) were excluded from this comparison because they did not contain enough information to convert results from beyond the farm gate to a cradle-to-farm-gate scope. Because some LCAs considered multiple similar scenarios, we took means per production method within each publication for each of the environmental impacts as the basis of comparison.

On average across all of the LCAs that were reviewed, greenhouse gas emissions averaged 11.8 kg CO<sub>2</sub>-eq/kg live weight at the farm gate. Emissions were not affected by beef production method, although scenarios including a dairy input tended to have somewhat lower emissions (Figure 2-2A). Most of the greenhouse emissions associated with beef production from cradle to farm-gate (79% in our model) result from direct cattle emissions and manure, suggesting that modification of production methods has limited impact on these emissions.

Research on cattle methane reduction through diet adjustments and animal breeding has shown reduced emissions up to 20%, but may not be able to reduce emissions more than 33% (Grainger and Beauchemin 2011). Further, diet adjustments may not apply to grazing cattle, from which over two-thirds of beef-related methane originates. And although livestock enteric fermentation is the leading cause of methane emissions in the U.S., there has been little effort to develop methods to capture methane from cattle for reuse in the energy sector (Bracmort et al. 2009), likely because of logistic difficulties.

Energy use across the LCAs averaged 21.9 MJ/kg live weight. Variation among studies was high and sample size was relatively low, limiting statistical power. However, feedlot production methods tended to increase energy use, while grazing – and especially extensive grazing – decreased energy use (Figure 2-2B). Scenarios including a dairy input also tended to use less energy.

Land use averaged 46.4 m<sup>2</sup>/kg live weight. Only a handful of studies reported the land base needed for beef production. Managed grazing tended to decrease and extensive grazing increase the amount of land required (Figure 2-2C). Scenarios including a dairy input tended to use less land than those that did not.



Water use is not typically reported in beef production LCAs and in the handful of beef production analyses that report it, it varies widely, depending on whether water use includes rainwater or not (Table A1-2).

Other environmental impacts are reported less often in LCAs, although they should not be neglected when considering the full environmental impact of beef production. Reports of acidification potential range from about 0.05 to 0.4 kg SO<sub>2</sub>-eq/kg cattle live weight (Ogino et al. 2004, 2007, Williams et al. 2006, Weidema et al. 2008, Nguyen et al. 2010) and water eutrophication potential ranges from about 0.01 to 0.18 kg PO<sub>4</sub>-eq/kg cattle live weight (Cederberg and Stadig 2003, Williams et al. 2006, Weidema et al. 2008, Nguyen et al. 2010, Pelletier et al. 2010). The impact of manure in intensive systems is highly variable, depending on manure management. Well-managed manure application to cropland can reduce the amount of synthetic fertilizer used, while poor management practices can cause soil and water pollution and pathogen spread. Environmental impacts that are rarely or never quantified in a manner that allows for scaling up to the national scale include local air quality, local nitrogen deposition, dust, smell, erosion from wind and rain, soil and water contamination by antibiotics and hormones and pesticides, food poisoning, and antibiotic resistance.

### ***Trade-offs and best practices for beef production***

Shifting from grain-fed to pasture-fed beef could reduce the impacts of beef production (Foley et al. 2011). Our model results and analysis of existing LCAs suggest that high greenhouse gas emissions from beef production are inevitable and that different beef production methods exhibit trade-offs in resource use and environmental impact (Table 2-1).

The U.S. industrial method uses less arable land compared to managed grazing methods and produces beef quickly, reducing animal lifespan and thus all the environmental impacts associated with maintaining animals. However, it does so with high energy and water use and by occupying large amounts of non-arable U.S. land.

Green-label beef uses fewer resources than feedlot-based production, but requires 2.8 times the amount of arable land as U.S. industrial beef (Figure 2-1A).

Cattle grown exclusively on non-arable rangeland grow more slowly, reducing the rate of supply. If U.S. beef production were restricted solely to existing rangelands with no crop inputs, 122,000 km<sup>2</sup> of U.S. cropland could be rededicated to other purposes, but national beef production would be halved. World expansion of extensive cattle pasturing typically occurs at the expense of forests and native savannas (McAlpine et al. 2009), resulting in increased greenhouse gas emissions, decreased water quality, and lost biodiversity. Cattle that are not well-managed in these extensive systems increase erosion, pollute waterways, and reduce primary productivity (McAlpine et al. 2009).

A hybrid approach combining managed and extensive grazing could be feasible in the U.S. Production would begin with the already established cow-calf herds of the U.S. industrial system. Then, instead of moving to feedlots, the cattle could be intensively grazed on high-quality arable land. The resulting system would keep energy and water use low, while requiring only 1.5 times the amount of arable land as the U.S. industrial method. Increased costs to growers associated with greater land use could be recouped through the marketplace; grass-fed beef commands a premium of up to 34% from consumers, with additional premiums for traceability and made-in-the-USA labeling (Abidoye et al. 2011). However, the re-dedication of approximately 60,000 km<sup>2</sup> high-quality land away from corn and soybean production could have complicated indirect effects via the worldwide marketplace (Garnett 2009); for example, reduced stocks of U.S. soybeans could prompt increased forest clearing in the tropics for soybean agriculture, thereby counteracting the environmental benefits gained in the U.S.

One beef production method that would lower all environmental impacts is using calves from the dairy industry. Old dairy cows (cull cows) are currently slaughtered for beef and their calves become veal. If dairy calves were grown to a slaughter size instead, this dairy beef production could offset some of the environmental costs due to maintaining beef cows. These sorts of combined dairy-beef systems exist at a small scale throughout Europe (Nguyen et al. 2010), but have not been adopted in the U.S.

Challenges to a combined system include differing genetics of beef cattle versus dairy cattle, added logistics, and historically distinct industries. The number of beef cattle in the U.S. is more than three times the number of dairy cattle, so even a fully combined dairy-beef system would not replace the current beef cattle industry.

### ***Underestimation of environmental impacts***

The results of our analyses from cradle to farm gate are comparable to other published studies, but we have also analyzed environmental impacts from the farm gate to the grave. We find that for industrially produced beef, more than a quarter of energy use (Figure 2-1C) and about a third of greenhouse gas emissions (Figure 2-1D) of U.S. beef production occur after cattle are ready for slaughter. Farm-gate analyses are useful in comparing across beef-production practices and identifying areas for improvement in particular systems. However, comparisons across different food types necessitate analyses from cradle to grave, as food types differ in the extent of processing, need for cooking, and wastage rates.

Waste is frequently ignored in livestock LCAs, causing environmental impacts to be too low as a result (Figures 2-3, A1-1, A1-2, A1-3, A1-4). In our analyses, we accounted for unharvested crops, handling and transport losses, feeding waste, the need to support cows that do not successfully produce a weanable calf, and cattle death from illness. We find that about one-eighth of all food grown to feed cattle is lost in the process of producing a slaughter-ready animal, the majority due to maintaining cows that fail to conceive, unsuccessful pregnancies, and cattle deaths before slaughter. There is also significant waste once an animal is slaughtered. Although 20% of a carcass is recovered for such byproducts as hides, tallow, bone meal, and blood meal, almost half of the carcass weight cannot be used as edible beef or as byproducts and is disposed. Additionally, dining establishments and American consumers waste a significant amount of edible beef. Approximately one fifth of all edible beef in the U.S. ends up uneaten and is disposed of in landfills and down sink garbage disposals, the majority of it in consumers' homes (Parfitt et al. 2010).

To facilitate comparisons across animal products and other food types, food LCA results should be reported in one or more functional units that describe edible food, such as kg of edible food product, kcal of food energy, and kg of edible protein, and should include farm-gate to grave impacts, including loss and waste. Typically, beef LCAs use live weight or carcass weight as the functional unit (e.g. Subak 1999, Phetteplace et al. 2001, Ogino et al. 2004, 2007, Williams et al. 2006, Weidema et al. 2008, Cederberg et al. 2009, Nguyen et al. 2010, Beauchemin et al. 2010, Pelletier et al. 2010, Capper 2011). While these units are appropriate for cross-beef comparisons, conversion fractions from live animal to carcass, carcass meat to retail meat, from retail meat to edible meat, and consumer-level waste vary among animals (Figure 2-4, Table A1-1). As a result, comparisons at the commodity level do not directly translate into comparisons at the ingestion level.

One common metric for comparing across food types is the efficiency of production, described as a ratio of input material to output material. Beef is frequently described as a low-efficiency meat, with ratios ranging from 5:1 to 20:1, indicating that 5 to 20 g of feed product need to be fed to cattle for every 1 g of resulting beef product (e.g. Stehfest et al. 2009, Lesschen et al. 2011). However, these ratios are often calculated based on live weight gain when cattle are fed high-energy finishing diets, and implicitly ignore forage, which makes up the majority of cattle life-time food intake. Further, the unit used is frequently the carcass, which ignores loss and waste after slaughter.

We calculated full feed efficiency ratios for U.S. beef that include all stages of beef production and that are based on consumed beef (Table 2-2), and find that on a dry matter basis, cattle ingest 160 kg of feed for every 1 kg of beef consumed by a person; on a food energy (kcal) basis, the ratio is 96:1; and on a protein basis, it is 24:1. These ratios are triple those that consider just the non-forage portion of the cattle diet.

Other animal products have much higher feed efficiencies: pork and poultry, which are produced using commodity crops almost entirely, have ratios between 3:1 and 6:1; eggs have a ratio between 2.5:1 and 3:1; and milk has a ratio of less than 3:1 (e.g. Stehfest et al. 2009, Wirsenius et al. 2010, Lesschen et al. 2011). Whether considering

just commodity crop input to U.S. beef production or all feed intake, beef is a comparatively inefficient source of animal protein.

As the world's population increases and grows wealthier, global beef production is projected to increase, with developing economies gradually converting from extensive systems to industrial ones like those used in the U.S. (Alexandratos and Bruinsma 2012). The U.S. industrial system maximizes production and minimizes arable land use at the expense of high energy and water use and the large-scale occupation of rangeland. Other methods of beef production exhibit tradeoffs among resource and land use, production efficiency, and local impacts to air, land, and water. Greenhouse gas emissions are large across all types of beef production. Worldwide beef (and other food) production impacts may be even greater than thought due to the lack of studies examining environmental impacts past the farm gate and the necessity of accounting for all types of loss and waste. Reporting results in edible food product functional units not only encourages a full accounting of impacts, but also facilitates comparisons across different food types. The large impact of beef production reported in the literature, despite underestimation, and the inherent tradeoffs associated with different types of beef production suggest that beef production cannot be made environmentally friendly when multiple impacts are considered. The best ways to reduce beef's environmental impact are to produce less of it, especially in the U.S. where beef consumption is 2.5 times the rest of the industrialized world (FAO 2009), and to reduce waste, particularly at the consumer level.

## Tables and Figures

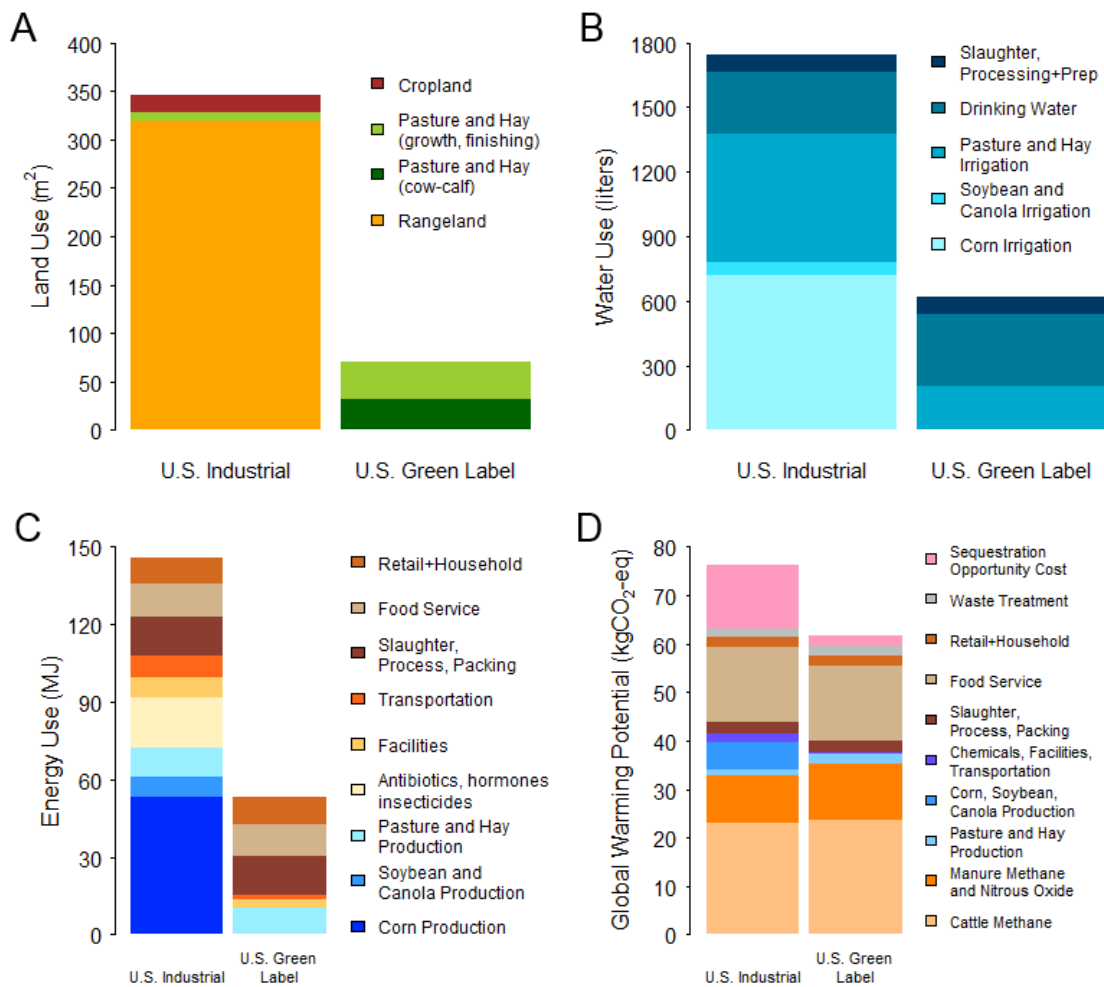
**Table 2-1: Summary of tradeoffs associated with different methods of beef production**

Fast production decreases environmental impacts due to shorter cattle lifetimes. “Land footprint” refers to damaged caused by grazing, such as erosion and waterway pollution. Greenhouse gas emissions are high for all types of beef production.

	<b>Positives</b>	<b>Negatives</b>
<b>U.S. Industrial</b>	Faster production Lower arable land use	Higher energy use Higher water use Higher land footprint
<b>Managed grazing</b>	Lower energy use Lower water use	Higher arable land use
<b>Rangeland only</b>	No arable land use Lower energy use Lower water use	Slower production Higher land footprint
<b>Dairy input</b>	Lower arable land use Lower energy use Lower water use	Slower production Limited by number of dairy cows

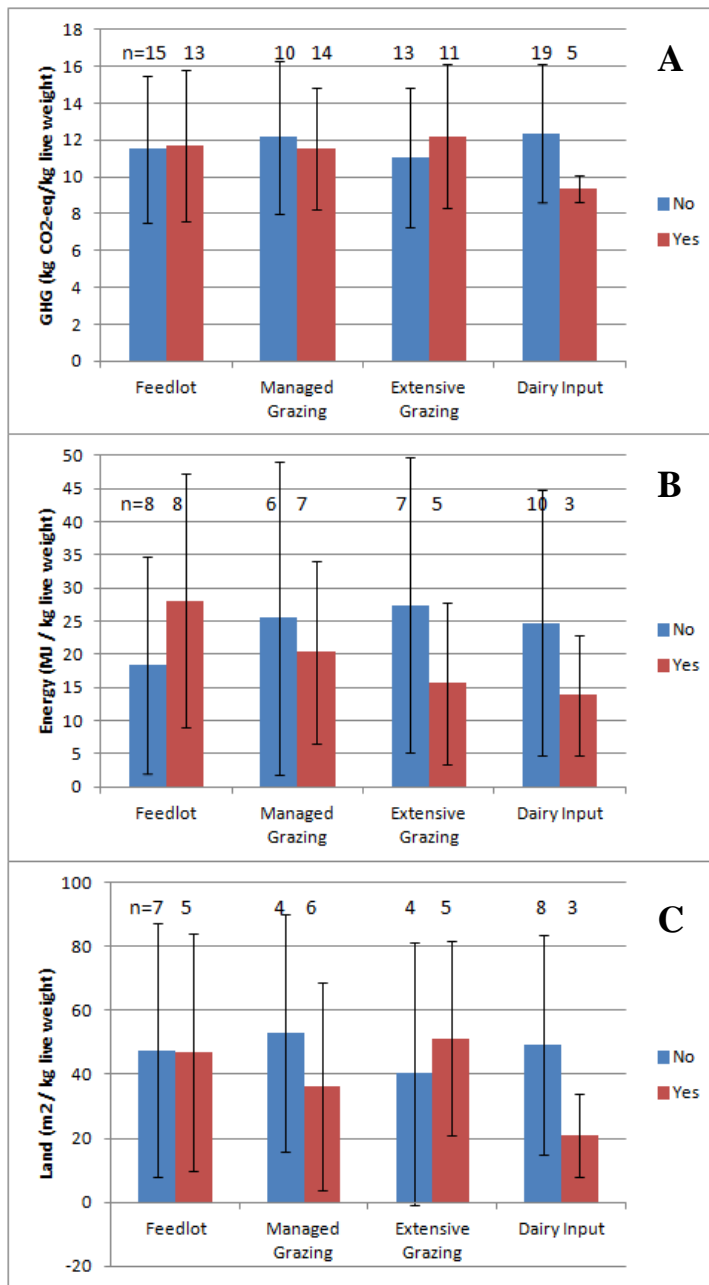
**Table 2-2: Beef efficiency metrics**

<b>Unit</b>	<b>Cattle feed intake considered</b>	<b>Biomass (kg DM feed / kg DM beef)</b>	<b>Energy (MJ feed / MJ beef)</b>	<b>Protein (kg feed protein / kg beef protein)</b>
<b>Eaten Beef</b>	All intake	160	96	24
<b>Eaten Beef</b>	Commodity crops only	43	34	8
<b>Carcass Beef</b>	All intake	54	36	16
<b>Carcass Beef</b>	Commodity crops only	15	13	5



**Figure 2-1: Environmental impacts of U.S. industrial beef production and beef production based on U.S. green-label best practices**

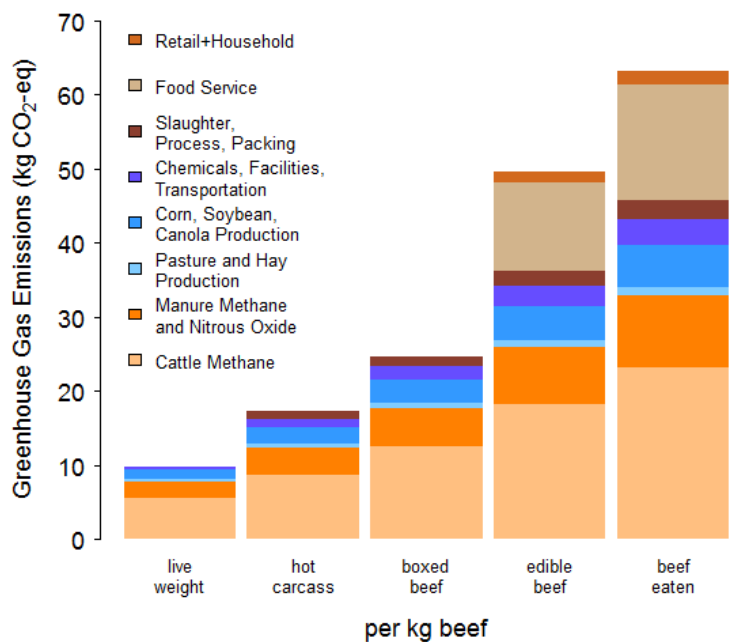
All impacts shown are per kg eaten beef. (A) Land use in the U.S. industrial system is dominated by rangeland; green-label beef uses less land overall, but more high-quality land. (B) Water use in the U.S. industrial system is dominated by irrigation; green-label beef uses much less water. (C) Crop production, industrial processes, and consumer-level activities all contribute to energy use in the U.S. industrial system; green-label beef uses substantially less energy before slaughter. (D) Greenhouse gas emissions similar in both systems and are dominated by methane and nitrous oxide emitted from cattle and their manure, as well as by disposable plastic materials used in the fast-food industry. Use of land for feed crops in the U.S. Industrial system precludes that land sequestering carbon.



**Figure 2-2: Environmental impacts of beef production across beef LCAs**

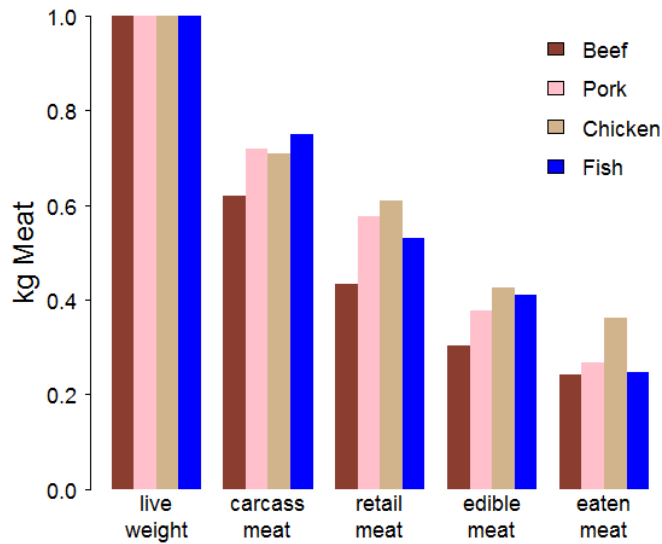
Land use, fossil energy use, and greenhouse gas emissions according to cattle production method across beef LCAs, including this one. “No” indicates LCAs in which the method is not used and “Yes” indicates LCAs in which the method is used. All LCAs are compared as kg live weight at the farm gate. Error bars are standard deviations. Numbers (n=) indicate the number of LCAs represented in each bar.





**Figure 2-3: Analysis of greenhouse gas emissions from U.S. Industrial beef production at five different stages of production and consumption**

Including loss and waste and using cradle-to-grave boundaries increases the estimation of environmental impacts. Here, the difference between successive bars of the same color shows loss of mass in the system; for example, plate waste accounts for the difference between cattle methane emissions per kg beef edible beef and per kg beef eaten. This graph also highlights the difficulty of comparing studies that use different functional units.



**Figure 2-4: Conversion of 1 kg live weight of four meat products into eaten meat, with intermediate units included**

Carcass represents the live animal with standard removal of entrails, head, feet, etc.

Retail represents the “selling-to-consumer” weight and may include bones and fat. Edible meat is lean meat only. Eaten meat is edible meat minus consumer waste. Note that these values are specific to the U.S., and preparations of these animals in other countries would yield somewhat different results.

### **Chapter 3 : Local plant diversity does not enhance biological control of soybean aphid in a field experiment**

Farmers wanting to control agricultural pests with minimal pesticide use need guidance on best management practices for supporting natural enemies of pests. Some research has shown that diverse landscapes support greater natural enemy abundance than less diverse ones, leading to an increase in biological control of agricultural pest species. At a local scale, increased plant diversity often increases the abundance of predatory and parasitoid insects. However, it is not clear that increased biological control due to diversity at the landscape scale is caused by local plant diversity *per se*. We conducted an experiment to test whether local prairie plant diversity affected the biological control of soybean aphids (*Aphis glycines*) by growing soybean plants immediately adjacent to 9m x 9m plots in which the number of plant species had been manipulated. We found that plot diversity did not affect the number of insect predators found on soybean plants and was negatively related to the number of parasitized aphids found on them. Natural enemies had a significant impact on aphid population size when populations were compared with and without natural enemy exclusions. However, 23% of plants in 2011 and 47% of plants in 2012 had aphid populations surpassing economic injury levels, indicating that natural enemy action was limited in controlling aphid populations. Plant diversity did not predict aphid population levels or soybean yield. Two biological control agents thought to be most effective at biological control of soybean aphid in U.S. agricultural fields were rare in our experiment, suggesting that agricultural management practices that focus on promoting specific species may be more effective than promoting plant diversity *per se*.

#### **Introduction**

Large-scale agriculture relies on large swathes of crops in monoculture. One of the results is large-scale outbreaks of insect pest species on these crops that can inflict billions of dollars of damage annually. A growing area of research seeks to understand how agricultural ecosystems can be managed to reduce crop pest damage, while minimizing pesticide applications. This integrated pest management approach posits that

natural enemies can effectively control pest species in an effective and sustainable manner.

Recent research has suggested that agricultural landscapes that are more diverse support greater abundances of pest natural enemies, and crops in more diverse landscapes may incur less damage from pest species than landscapes containing only monoculture crops. In these diverse landscapes, crop monocultures are interspersed with areas of more permanent vegetation, such as fields of fallow or native vegetation, woodlots, and hedgerows. A review by Bianchi et al. (2006) found that 74% of observations within 24 studies found higher natural enemy populations in diverse landscapes, and 45% of observations within 10 studies found lower pest pressure in diverse landscapes. Another review focusing on organic agriculture (Letourneau and Bothwell 2008) found that landscape diversity was associated with increased natural enemy diversity and abundance, but not that such diversity led to increased control of pests or crop yield. Likewise, a meta-analysis of 46 studies (Chaplin-Kramer et al. 2011) found strong support for the idea that more diverse landscapes lead to a greater abundance and diversity of natural enemies, but that pest abundance was not related to landscape diversity.

Foundational field research on the effect of plant diversity on crop plants showed that crop plants surrounded by a high diversity of plants tend to have a lower abundance of pests than those in monoculture (Root 1973), whereas those in monocultures are much more likely to experience pest outbreaks (Pimentel 1961). In 209 articles reviewed by Andow (1991), 52% of the 149 herbivore species studied had lower population densities in polycultures. A meta-analysis of 21 studies showed a moderate reduction of herbivores with increased crop diversity (Tonhasca and Byrne 1994).

The increase in natural enemy diversity and abundance and the concurrent decrease in pest abundance or density with plant diversity can be explained by the ‘enemies hypothesis’ (Root 1973). The enemies hypothesis is based on the idea that more diverse vegetation supports a greater variety of food and other resources that natural enemies require. Additionally, these resources are more temporally stable than in

monocultures, as prey find greater refuge in more diverse vegetation and plant resources such as nectar may be provided at different times by different plant species. The result is that natural enemies are more diverse and abundant and their populations are more stable in habitats with higher plant diversity. Experimental tests of the enemies hypothesis have usually found support for it (Russell 1989, Langellotto and Denno 2004).

While there is empirical evidence supporting the enemies hypothesis at landscape and local scales, it is not clear whether plant diversity *per se* is important in supporting biological control. It is possible that increased plant diversity simply increases the likelihood that a particular plant species important for biological control occurs. Studies have shown that plant diversity increases predator and parasitoid control of agricultural pests by (1) providing specific supplemental resources needed for predator/parasitoid development, such as nectar; (2) providing alternative prey/hosts during times when the focal herbivore pest is not available; and (3) providing a more complex or refuge habitat compared to monocultures (Landis et al. 2000). Supplemental and alternative food resources are likely to be species specific, while habitat structure may not depend as much on plant species identity. It is also possible for diversity to decrease the effect of natural enemies on pests through dilution of pest abundances (Andow and Risch 1985) or reduced search efficiency of natural enemies (Sheehan 1986), or to increase the abundance and impact of herbivore pests through the same mechanisms of supplemental resources and alternative habitat (Collins and Johnson 1985).

We tested whether local plant diversity *per se* increased the impact of biological control on soybean aphids (*Aphis glycines*). Native to Asia, the soybean aphid was accidentally introduced to North America in the 1990's, where it rapidly spread throughout the north central United States (Ragsdale et al. 2004). It is now a major pest of American soybeans, causing widespread yield losses (Ragsdale et al. 2004). As many as 48 arthropod species have been identified as natural enemies of soybean aphid in North America (Ragsdale et al. 2011), but inconsistent natural biological control has led the soybean aphid to be the primary driver of soybean pesticide use in the United States (Ragsdale et al. 2011). Integrated pest management practices are being developed to more

sustainably control it (Hodgson 2012), and additional knowledge is needed on how to support control by natural enemies in order to develop cost-effective strategies for managing the soybean aphid.

Various studies have explored using cover crops (intercropping) in soybean fields to increase natural enemy abundance and decrease aphid populations (Schmidt et al. 2007, Koch et al. 2012, Lundgren et al. 2013). Schmidt et al. (2007) found that using alfalfa as a “living mulch” increased natural enemy abundance and decreased aphid abundance compared to soybean grown without alfalfa. Similarly, Lundgren et al. (2013) found that a spring-planted winter rye cover crop reduced aphid load compared to soybean without a cover crop. In both cases, however, soybean yield was substantially reduced (26% and 50% respectively), presumably due to competition between soybean plants and the cover crops. Koch et al. (2012) found evidence for decreased aphid abundance, but not increased predator density, in soybeans planted into fall-planted winter rye cover crops compared with soybean without rye. This study did not find a significant effect of cover cropping on soybean yield.

Landscape diversity can positively affect soybean aphid biological control pressure (Gardiner et al. 2009) and negatively affect aphid abundance (Noma et al. 2010) in the North Central U.S., although a relationship between landscape diversity and the abundance of soybean aphids and their natural enemies is not universal (Schmidt et al. 2011). Both landscape studies and cover-cropping studies suggest that increased plant diversity may increase aphid biological control, but it is unknown whether plant diversity itself accounts for this pattern.

To investigate the impact of local plant diversity on the biological control of soybean aphids, we conducted a replicated field experiment, in which the local plant assemblage was manipulated to contain 1, 4, or 16 prairie species drawn from an 18-species pool. These plots have been shown to have an increased abundance of predators and parasitoids in plots with higher plant species richness (Haddad et al. 2009).

We measured soybean plant growth and bean output, soybean aphid abundance, and natural enemies on the soybean plants. We hypothesized that greater local plant

species richness would increase natural enemy abundance on, reduce aphid abundance on, and increase yield of adjacent soybean plants.

## **Methods**

### ***Site location and preparation***

The experiment was conducted at Cedar Creek Ecosystem Science Reserve (part of the National Science Foundation Long Term Ecological Research Network) within the Biodiversity II experiment (#e120), details of which are described in Tilman et al. (2001). In brief, in 1994, plots were seeded with 1, 2, 4, 8, or 16 species, all randomly drawn from a pool of 18 prairie perennials. The thirty-three 9 m x 9 m plots used in this experiment have been maintained by hand weeding since 1994 to remove non-seeded species. They consist of eleven 1-species plots, eleven 4-species plots, and eleven 16-species plots, chosen randomly from the pool of maintained plots that were not heavily used for other experiments. Annual plot biomass was estimated by cutting strips of vegetation in late July and early August from each plot, drying them, and weighing them.

In May 2011, we dug six 76.2 cm holes using a 20.3 cm auger, immediately adjacent to each of the 33 plots. To limit root competition of soybean plants with prairie vegetation, we sunk 20.3 cm diameter HVAC ducting into each hole so that it protruded approximately 5 cm above ground level. We mixed soil removed from the holes with purchased garden soil at a 1:1 ratio and filled the holes with the mixture.

### ***Soybeans and Soybean Aphids***

We conducted similar experiments in 2011 and 2012. On May 20, 2011 and May 8, 2012, soybeans (Syngenta NK S19-R5) were dipped in soybean inoculant slurry and planted, three beans within each HVAC “pot”. When the plants germinated, 91.4 cm tall cylindrical cages of hardware cloth were positioned around the plants to prevent small-mammal herbivory; the Biodiversity II field is surrounded by a deer fence, and we observed no mammal herbivory in either year.

Three weeks after planting, pots without germination received a transplant from pots with multiple plants germinating; a week later, all pots were thinned to one plant. Soybean plants were watered, as needed, throughout each summer to prevent severe drought stress. Each plant received 0.95 liters of water on watering occasions.

On June 24, 2011, approximately 40 lab-cultured soybean aphids were added to each soybean plant. The plants had an average of 2.5 trifoliates. On June 12, 2012, approximately 50 aphids were added to five of the six plants at each plot; one plant at each plot was left as an aphid-free control. The plants had an average of 2 trifoliates.

For ten weeks each summer, we surveyed each plant once per week, recording the plant size (in trifoliates) and the number of aphids. In 2012, we removed immigrant (winged) aphids from the aphid-free control plants during these surveys. We also surveyed each plant once (2011) or twice (2012) per week, recording the number and identity of all insects found on the plants that were not soybean aphids. In 2011, these insect surveys were done in conjunction with the aphid surveys; the plant was first assessed for mobile insects, and then other insects were noted as aphids were counted. In 2012, insect surveys were done separately from aphid surveys; every plant was searched for the same length of time each survey – from 30 seconds each when the plants were small to 120 seconds each when the plants were large.

We stopped surveys when plants began to senesce. In 2011, we harvested two plants from each plot as soon as the start of senescence was detected to determine aboveground biomass. The plants were dried and weighed. After remaining plants had fully senesced, but before dropping bean pods, we harvested bean pods from remaining plants (4 in 2011, 6 in 2012). Beans were removed from pods and were counted and weighed. Beans were then dried and reweighed.

### ***Natural Enemy Exclusion***

On June 24, 2011, and June 22, 2012, we conducted a natural enemy exclusion experiment. We covered three (2011) or two (2012) plants at each plot with no-see-um mesh by wrapping the mesh around the hardware cloth cage, twisting and clipping mesh



edges together, and burying the bottom edges of the mesh. Unclipping mesh edges allowed for access to the plants to conduct surveys during the natural enemy exclusion. Natural enemies were manually removed from the plants immediately prior to mesh application and during surveys. Mesh was left on for two weeks and was removed on July 6, 2011, and July 6, 2012. To test for microclimate effects of the mesh, we took temperature and humidity readings from two adjacent plants – one with mesh and one without – at each plot twice: once in the morning when the temperature was relatively low and once mid-afternoon when the temperature was approximately at its peak. We also counted winged aphids (alates) on all plants during the aphid surveys while mesh was on and immediately following mesh removal.

### ***Ant Exclusion***

In 2012, we attempted to reduce the presence of aphid-tending ants on the soybean plants. We painted Tangle-Trap coating (Contech Inc.) around the lip of all HVAC pots on June 8, but stickiness was quickly lost after soil splattered on the coating during rain showers. We also used ant-bait traps (Terro Liquid Ant Baits T300) within each HVAC pot, which were placed on June 15 and July 20. During surveys, ants found on the plants were manually removed.

### ***Root Biomass***

The root biomass of the prairie plots has been sampled periodically since 1997. As root biomass per plot does not change appreciably from year to year, we use the most recent root biomass measures – from 2010 – and assume they reasonably represent root biomass of the prairie plots in 2011 and 2012. Root biomass of the soybean plants was determined by collecting twelve 5 cm diameter, 30 cm deep soil cores per plot, washing roots free of soil and other organic material, drying, and weighing.

## ***Data Analyses***

We analyzed data using ‘R’ version 2.15.1 (R Core Team 2012) and its lme4 (Bates et al. 2012), multcomp (Hothorn et al. 2008), plyr (Wickham 2011), and ggplot2 (Wickham 2009) packages.

## ***Soybeans***

We calculated the correlation matrix for the five measures of soybean plant productivity (plant size, plant aboveground biomass, number of soybeans, bean mass at harvest, and bean dry mass) for all plants in 2011.

To evaluate the effect of plot diversity on soybean production, we used linear mixed-effects models with bean dry mass as the response variable, plot diversity as the fixed effect, and plot as a random effect on the intercept. We used a likelihood-ratio test to compare models with and without the plot effect and obtain a p-value.

We investigated the effect of plot root biomass on soybean production using linear mixed-effects models with plot as a random effect on the intercept. We examined whether light competition with plot vegetation was important to soybean yield using linear mixed-effects models with plot as a random effect on the intercept, and whether plants were on the northern or southern side of the plot as a fixed effect. In both analyses, we used likelihood-ratio tests to compare models with and without the plot effect and to obtain p-values.

## ***Aphids***

We analyzed the effect of plot diversity on peak aphid density (aphids/trifoliolate) using linear mixed effects models with plot as a random effect on the intercept. We used a likelihood-ratio test to compare models with and without the diversity effect to obtain a p-value. For both years, we used only aphid density measures from unmeshed plants.

To analyze the effect of peak aphid density on soybean yield, we used linear mixed effects models with plot and treatment as random effects on the intercept. We also calculated cumulative aphid days (CAD) as the summed weekly aphid counts across each

summer. We used linear mixed effects models with plot, treatment, and plot diversity as random effects on the intercept to examine the effect of CAD on soybean yield. For both sets of models, we used a likelihood-ratio test to compare models with and without the density effect to obtain p-values.

To take advantage of the weekly survey data, we created a non-linear mixed effects model to investigate the impact of aphids on soybean plant growth rate and mature size. We assumed plants grew logistically as

$$N_t = \frac{k}{1 + e^{-(r)(t-m)}}$$

where  $N_t$  is the size of the plant at time  $t$ ,  $k$  is the mature size of the plant,  $r$  is the growth rate of the plant, and  $m$  is the time of maximum growth. We created a null mixed effects model with plant identity as a random effect on  $k$ ,  $r$ , and  $m$ .

We then modified the logistic equation by adding terms  $b$ ,  $c$ , and  $d$  to allow aphids ( $A_t$ , log number of aphids per plant at time  $t$ ) to affect growth rate:

$$N_t = \frac{k + cA_t}{1 + e^{-(r+bA_t)(t-(m+dA_t))}}$$

We used a likelihood-ratio test to compare the models with and without  $A_t$  and obtain a p-value. We then tested whether the parameters  $b$ ,  $c$ , and  $d$  were significantly different from zero.

### ***Natural Enemies***

Because of the difficulty of conducting insect surveys within meshed cages, meshed plants were omitted from three of the 18 insect surveys in 2012. We used the mesh treatment as a random effect in models where predator exclusion was not of primary interest, and omitted the data from the two missed surveys for all plants when it was.

Soybean aphid natural enemies were relatively scarce on plants, so we summed occurrences over the full season for each plant to give relative estimates of natural enemy abundance per plant. Because of the numerical dominance of parasitized aphids, we separated all natural enemies into ‘parasitoid mummies’ and ‘predators’ and analyzed them separately. Measures of predators and parasitoid mummies were log-transformed.

Because of the large number of plants on which no parasitoid mummies were observed, log-transformed parasitoid mummy counts still violated normality assumptions. As a result, we analyzed the effects of diversity and plant treatment on the presence or absence of parasitoid mummies on plants and separately analyzed the effects of diversity and plant treatment on parasitoid mummy abundance on plants where parasitoid mummies were found.

We used linear mixed-effects models with plot and treatment as random factors to evaluate the effect of plot diversity on predator abundance, parasitoid mummy occurrence, and parasitoid mummy abundance. We used mixed effects models with plot as a random factor and diversity as a fixed effect to evaluate the effect of plant treatment on the same response variables. When appropriate, we used a post-hoc Tukey test to determine which levels of diversity accounted for significant differences.

We also analyzed the effect of plot diversity on the ratio of predators and parasitoid mummies to aphids. To calculate these ratios, we divided the number of predators and parasitoid mummies by CAD for each plant. These ratios give an estimate of relative pressure of natural enemies on aphids. For analyses, aphid-free control plants in 2012 were excluded, as both aphid and natural enemy numbers were very low. We used linear mixed effects models with plot and treatment as random factors to investigate the effect of diversity as a fixed effect on the predators:aphid and parasitoid mummy:aphid ratios (both log transformed).

To see if greater observed predator pressure resulted in higher soybean yields, we weighted each natural enemy observance by the relative expected number of aphids killed by that natural enemy per day. Values were based on daily feeding rates, which were gleaned from published studies (Table 3-1). These weighting values are not meant to provide a literal estimation of how many aphids were killed per day, but rather provide a way to analyze the effect of predator abundances in a more mechanistic way than simply tallying numbers of natural enemies.

Hover fly and lacewing adults were excluded as not directly killing aphids. Unknown insects and predatory thrips were excluded as having an unknown attack rate;

individuals in these categories accounted for 2.6% and 0.9% of all predator individuals in 2011 and 2012 respectively.

We summed the weighted natural enemy observations over all surveys for each plant in each year as a measure of combined natural enemy pressure. We used linear mixed effects models with plot and treatment as random effects on the intercept and combined predator pressure as a fixed effect to test whether combined natural enemy pressure affects soybean yield. We used a likelihood-ratio test to compare models with and without the fixed effect and to obtain p-values.

### ***Natural enemy exclusion***

To evaluate the effect of natural enemy exclusion on aphid populations, we used the aphid survey data from July 6, 2011 and July 6, 2012, at the time the exclusion mesh was removed. We excluded the aphid-free control plants in 2012. We used linear mixed-effects models with log aphid counts as the response variable, treatment (natural enemies excluded or not) as a fixed effect, and plant size, plot, and surveyor as random effects on the intercept. We used a likelihood-ratio test to compare models with and without the fixed effect and obtain a p-value.

We analyzed the effects of plot diversity on aphid population size separately for natural enemy exclusion plants and plants with natural enemies. We used linear mixed-effects models with log aphid counts as the response variable, plot diversity as a fixed effect, and plot and surveyor as random effects on the intercept. We used a likelihood-ratio test to compare models with and without the fixed effect and obtain a p-value. For models that had a significant effect of plot diversity, we performed a post-hoc Tukey test to determine which levels differed.

We analyzed the effects of mesh on the temperature and relative humidity within cages using linear mixed-effects models with temperature or humidity as the response variable, presence or absence of mesh as a fixed effect, and plot as a random effect on the intercept. We used a likelihood-ratio test to compare models with and without the fixed effect and to obtain p-values.

We investigated the potential effect of changed temperature on aphid populations in meshed cages by modeling expected aphid population size based on the temperature-specific population growth rates of soybean aphids, as reported in McCornack et al. (2004). We fit a quadratic function to the discrete daily growth rate ( $\lambda$ ) at 20°C, 25°C, and 30°C. As our minimum and maximum temperatures are only slightly outside the range of 20-30°C, this is a reasonable approximation. The result is  $\lambda = 1.641 + 0.001 t - 0.00864 (t - 25)^2$  where  $t$  is temperature.

We then used the daily mean temperature recorded at Cedar Creek Ecosystem Science Reserve to calculate  $\lambda$  for each day that mesh covered cages. We also calculated  $\lambda$  for each day using the daily mean temperature adjusted by the temperature difference between meshed and unmeshed cages; we denote the expected discrete daily growth rate in meshed cages as  $\lambda_m$ .

We calculated the expected increase in aphid population after two weeks due to a temperature effect within meshed cages as  $\prod_{days} \frac{\lambda_m}{\lambda}$ . We divided the aphid population counts on each plant by this expected increase to estimate the aphid population on each meshed plant in the absence of microclimate effects. We reanalyzed the effect of natural enemy exclusion on aphid populations using these estimated aphid population sizes for meshed plants and actual aphid population sizes on unmeshed plants.

We tested to see whether mesh enclosures increased the number of alates within a cage, using the survey data from 7/6/2011 and 7/6/2012. We used linear mixed-effects models with log-transformed alates as the response variable, log-transformed aphids, mesh or no mesh, and their interaction as fixed effects, and plot and surveyor as random effects. We used a likelihood-ratio test to compare models with and without the mesh/no-mesh effect and its interaction with log-aphids and to obtain p-values.

### ***Non-aphid herbivores***

To investigate the potential effect of aphids on other soybean herbivores, we compared the season-long summed abundances of these herbivores on the aphid-free plants versus those plants with aphids that remained unmeshed throughout the 2012

season. We also looked at the effect of summed non-aphid herbivore abundance on soybean yield and the season-long summed abundance of non-aphid herbivores combined with soybean aphids on soybean yield. We used linear mixed-effects models with plot as a random effect for these analyses.

## **Results**

### ***Soybeans***

All soybean plants survived the full summer in both years. In 2011, plants reached a mean maximum size of  $54.3 \pm 12.2$  (SD) trifoliates after  $90.4 \pm 5.5$  days, and in 2012, plants reached a mean maximum size of  $32.5 \pm 8.2$  trifoliates after  $91.4 \pm 6.8$  days (Figure 3-1). Plants yielded a mean of  $16.5 \pm 7.5$  g soybean dry matter in 2011 and  $14.0 \pm 5.1$  g soybean dry matter in 2012.

All measures of soybean production (plant size, plant aboveground biomass, number of soybeans, bean mass at harvest, and bean dry mass) were all highly correlated (Figure 3-2). For the remaining analyses we use only bean dry mass as a measure of soybean yield.

Bean dry mass was not significantly affected by plot diversity in 2011 ( $\chi^2=3.09$ ,  $p=0.21$ ), but was in 2012 ( $\chi^2=6.31$ ,  $p=0.04$ ); in 2012, bean dry mass in 16-species plots was less than that in 1-species plots, but 4-species plots were statistically similar to both 1-species and 16-species plots (Figure 3-3; 1-spp: 15.95 g, SE=0.96; 4-spp: 13.59 g, SE=0.96; 16-spp: 12.44 g, SE=0.96).

Plot plant root biomass did not have a significant effect on soybean yield (2011:  $\chi^2=0.248$ ,  $p=0.62$ ; 2012:  $\chi^2=1.16$ ,  $p=0.28$ ). In 2011, plants grown on the north and south sides of plots had statistically similar yields ( $\chi^2=0.057$ ,  $p=0.81$ ); In 2012, plants grown on the north side of plots had statistically higher yields (difference=0.962 g, SE=0.389,  $\chi^2=6.06$ ,  $p=0.013$ ).

## *Aphids*

In each experiment year, soybean aphids were observed on just one plant immediately prior to the addition of lab-cultured aphids. Aphid populations established on all plants to which aphids were added. In 2011, most populations reached their maximum size of  $494 \pm 288$  aphids between July 13 and July 26 (Figure 3-4A). Maximum population size varied between 47 aphids and 1,550 aphids. In 2012, the timing of maximum aphid population size was bimodal, with 75% of populations achieving a maximum of  $900 \pm 637$  aphids between July 5 and July 12 (Figure 3-4B). The remaining populations reached a maximum of  $480 \pm 330$  aphids between August 1 and August 17. All together, maximum population size was  $800 \pm 601$ , and ranged between 34 and 3,850.

Aphid density on soybean plants reached a peak of  $44.8 \pm 33.0$  aphids/trifoliolate between June 29 and July 26 in 2011, and a peak of  $60.0 \pm 43.5$  aphids/trifoliolate between June 28 and July 12 in 2012. Peak aphid density ranged from 2 to 173 aphids/trifoliolate in 2011 and from 8 to 266 aphids/trifoliolate in 2012.

Peak aphid density on unmeshed plants was affected by plot diversity in 2012 (Figure 3-5B,  $\chi^2=8.13$ ,  $p=0.017$ ), but not in 2011 (Figure 3-5A,  $\chi^2=0.78$ ,  $p=0.68$ ).

Including aphids in our model of plant growth increased model fit (2011:  $\chi^2=19.7$ ,  $p=0.0002$ ; 2012:  $\chi^2=190.37$ ,  $p<0.0001$ ). Parameter  $b$  was not significantly different from zero (2011:  $p=0.92$ ; 2012:  $p=0.094$ ), but  $c$  (2011: estimate: -0.395, SE=0.186,  $p=0.017$ ; 2012: estimate: -0.405, SE=0.174,  $p=0.010$ ) and  $d$  (2011: estimate: -0.196, SE=0.090,  $p=0.015$ ; 2012: estimate: -0.767, SE=0.145,  $p<0.0001$ ) were (Figure 3-6).

Higher peak aphid density led to decreased soybean yield in 2011 (Figure 3-7A, param est: -1.77, stderr=0.602,  $\chi^2=8.48$ ,  $p=0.004$ ), but not in 2012 (Figure 3-7B,  $\chi^2=0.720$ ,  $p=0.40$ ).

We did not find a relationship between cumulative aphid days (CAD) and yield in 2011 (slope=-0.00071, SE=0.00069,  $\chi^2=1.043$ ,  $p=0.31$ ) or in 2012 (slope=0.00042, SE=0.00021,  $\chi^2=3.74$ ,  $p=0.053$ ).



### *Natural enemies*

In 2011, we recorded 2,088 aphid natural enemies over 1,782 plant surveys (Table 3-2). In 2012, we recorded 6,868 natural enemies over 3,564 surveys (Table 3-3). Early in the summer, natural enemies had low abundance, with less than one per 10 plants observed (Figure 3-8), but it increased over the summer.

In both years, the number of predators was not significantly affected by plot diversity. In 2012, plants without aphids had about half as many predators as those with aphids (estimates: control plants: 1.00, SE=0.156; mesh plants: 2.18, SE=0.123; unmeshed plants: 1.86, SE=0.110;  $\chi^2 = 45.9$ ,  $p < 0.0001$ ).

In 2011, parasitoid mummies were encountered on 121 of the 198 plants. The presence of mummies was not significantly influenced by plot diversity or by previous mesh application. Plot diversity did affect the number of mummies observed on the plants that had them (Figure 3-9; estimates: 1-spp plots: 1.47, SE=0.137; 4-spp plots: 1.32, SE=0.137; 16-spp plots: 0.931, SE=0.150;  $\chi^2 = 6.74$ ,  $p = 0.034$ ), as did the presence/absence of mesh (estimates: with mesh: 1.33, SE=0.150; without mesh: 1.58, SE=0.147;  $\chi^2 = 4.05$ ,  $p = 0.044$ ).

In 2012, parasitoid mummies were encountered on 157 of the 198 plants. The presence of mummies was not affected by plot diversity, but was affected by treatment (estimates: control: 0.45, SE=0.066; with mesh: 0.76, SE=0.049; without mesh: 0.91, SE=0.041;  $\chi^2 = 36.2$ ,  $p < 0.0001$ ; a post-hoc Tukey test supported all estimates being significantly different at the  $p = 0.05$  level). Plot diversity significantly affected the number of mummies observed on plants that had them (Figure 3-9; estimates: 1-spp plots: 2.18, SE=0.527; 4-spp plots: 1.56, SE=0.533; 16-spp plots: 1.38, SE=0.534;  $\chi^2 = 6.05$ ,  $p = 0.048$ ), as did plant treatment (estimates: control: 0.96, SE=0.369; with mesh: 2.48, SE=0.263; without mesh: 3.00, SE=0.241;  $\chi^2 = 33.2$ ,  $p < 0.0001$ ; a post-hoc Tukey test supported all estimates being significantly different at the  $p = 0.05$  level).

Plot diversity had no significant effect on the full season ratio of predator individuals to aphids, nor the ratio of mummies to aphids in either year (2011 predators:

$\chi^2=0.75$ ,  $p=0.69$ ; 2011 mummies:  $\chi^2=5.12$ ,  $p=0.08$ ; 2012 predators:  $\chi^2=0.285$ ,  $p=0.24$ ; 2012 mummies:  $\chi^2=4.62$ ,  $p=0.10$ ).

Combined natural enemy pressure was not significantly correlated with soybean yield in either year (2011:  $\chi^2=0.205$ ,  $p=0.65$ ; 2012:  $\chi^2=2.59$ ,  $p=0.11$ ).

In first four weeks of 2012, aphid abundance increased rapidly, with enemy:aphid ratios declining (Figure 3-10). Subsequently, natural enemy abundance increased, and there was a negative relationship between the number of natural enemies per aphid and the number of aphids per plant (Figure 3-10).

### *Natural enemy exclusion*

Over the two surveys in 2011 when mesh was applied, we found 16 predator individuals and no wasp mummies in 11 mesh enclosures. During the one insect survey performed in 2012 while mesh covered plants, we found 10 predator individuals and 39 wasp mummies within 14 mesh enclosures.

Aphids increased significantly when natural enemies were excluded (Figure 3-11; 2011: difference in  $\log(\text{aphids}) = 0.673$ ,  $SE=0.173$ ,  $\chi^2=14.4$ ,  $p=0.0001$ ; 2012: difference in  $\log(\text{aphids}) = 0.682$ ,  $SE=0.145$ ,  $\chi^2 = 20.6$ ,  $p<0.0001$ ).

Aphid populations were significantly different between 4-species and 16-species plots in unmeshed plots in 2012 (Figure 3-12; 4-spp plot estimate = 5.98, 16-spp plot estimate = 5.32,  $z$  for difference = -2.99,  $p=0.008$ ). There were no significant differences in diversity in the unmeshed plots or in 2011.

Mesh increased morning temperatures by 1.8 °C ( $SE = 0.10$ ,  $\chi^2=173$ ,  $p<0.0001$ ) and afternoon temperatures by 0.7 °C ( $SE = 0.11$ ,  $\chi^2=40.3$ ,  $p<0.0001$ ). Mesh decreased morning relative humidity by 3.5 percentage points ( $SE=0.37$ ,  $\chi^2=71.6$ ,  $p<0.0001$ ), but did not affect afternoon relative humidity ( $\chi^2=0.315$ ,  $p=0.57$ ).

For a liberal estimate of the effects of temperature on aphid population growth, we calculated  $\lambda_m$  using an increase in temperature of 1.8 °C (Figure 3-13). The expected increase in aphid population size by July 6, 2012 (the day the mesh was removed) was 1.38.

Even when aphid populations were adjusted for the effects of temperature, aphids increased significantly when predators were excluded in 2012 (difference in log(aphids) = 0.365, SE = 0.143,  $\chi^2 = 6.35$ ,  $p=0.01$ ).

The number of alate aphids was significantly higher in meshed cages in 2012 ( $\chi^2 = 33.3$ ,  $p<0.0001$ ), but not in 2011 ( $\chi^2 = 3.81$ ,  $p=0.15$ ). However, in both years the total number of alates present on plants was very low. Unmeshed plants had a mean of 0.0013 (SD 0.0030) alates per aphid in 2011 and 0.0014 (SD 0.0035) alates per aphid in 2012, while meshed plants had a mean of 0.0065 (SD 0.018) alates per aphid in 2011 and 0.0063 (SD 0.0093) alates per aphid in 2012.

### ***Non-aphid herbivores***

Non-aphid herbivores were significantly more abundant on aphid-free plants than on plants with aphids (aphid-free:  $81\pm 4$  herbivores, with aphids:  $58\pm 3$  herbivores,  $\chi^2 = 40.6$ ,  $p<0.0001$ ). Thrips and white flies, the numerically dominant non-aphid herbivores, were both significantly more abundant on aphid-free plants (thrips:  $\chi^2 = 5.15$ ,  $p=0.023$ ; white flies:  $\chi^2 = 29.8$ ,  $p<0.0001$ ), whereas leafhoppers and Lepidoptera were not (leafhoppers:  $\chi^2 = 0.263$ ,  $p=0.26$ ; Lepidoptera:  $\chi^2 = 0.543$ ,  $p=0.46$ ).

Abundance of non-aphid herbivores did not predict soybean yield ( $\chi^2 = 2.21$ ,  $p=0.14$ ). Neither did summed abundance of soybean aphids and non-aphid herbivores ( $\chi^2 = 0.0056$ ,  $p=0.94$ ).

### ***Ants***

Ants were substantially reduced, though not eliminated, in 2012 (Figure 3-14). While surveying time was slightly shorter in 2012, ants are generally large, mobile, and thus easy to see. Longer surveying times would not likely have resulted in much of an increase in 2012 numbers.

## **Discussion**

Within this experiment, we did not detect a difference in biological control among three plant diversity treatments consisting of 1, 4, and 16 prairie species. We did not find

that there was a greater abundance of predators or parasitoids on soybeans next to more diverse plots. We did not see a reduction in aphid abundance next to more diverse plots. Nor did we find greater soybean yield with greater plant diversity.

In 2012, but not in 2011, we found that higher plant diversity led to lower soybean yield. While we tried to control for direct competition between plants in the prairie plots and the soybean plants by separating their roots with HVAC ducting, we cannot eliminate the possibility that root competition affected soybean growth in 2012. Light competition does not appear to have been a factor in soybean yield. Other factors, such as microclimate effects caused by wind screening may have played a role, but were unmeasured.

Aphid populations surpassed economic injury levels of 674 aphids per plant (Ragsdale et al. 2007) on 23% of plants in 2011 and on 47% of plants in 2012. Yet, soybean yield was not affected by aphid populations in 2012, and was only somewhat affected by aphid density in 2011. In particular, while Ragsdale et al. (2007) found a 6.88% reduction in yield per 10,000 cumulative aphid days, we found no yield reductions based on cumulative aphid days in either year. We found that soybean plant growth was only slightly reduced by aphid populations. In particular, the value of  $c$  obtained in our model suggests that aphids reduced mature plant size by at most three trifolates at the highest aphid populations. Therefore, it seems that soybean plants in this experiment were tolerant of aphid herbivory pressure. Such tolerance may result from reduced environmental stress encountered by our experimental soybeans compared with crop field conditions. In particular, we reduced root competition using HVAC ducting and watered soybean plants during dry times. Damage by insect pests can exacerbate drought stress (e.g. Riedell 1989), suggesting that an interaction between environmental stress and soybean aphid abundance may be necessary for plants to exhibit reduced growth and yield.

Our predator exclusion treatments in both years showed that natural enemies reduce aphid population sizes in North America, as has also been shown in other research (Fox et al. 2004, Desneux et al. 2006, Costamagna et al. 2008, Gardiner et al. 2009). We

saw significantly more predators on plants with aphids than on aphid-free plants, suggesting that our observations of predators were a reasonable measure of predator pressure on aphids. However, we did not find more predators on plants next to higher diversity plots.

We found more parasitoid mummies on plants next to low-diversity plots than high-diversity plots in both 2011 and 2012, despite different species being dominant in those two years. In 2011, tan mummies (likely *Lysiphlebus testaceipes*; Ragsdale et al. 2011) were more numerous than black mummies (*Aphelinus* spp. with *A. certus* dominating in 2012; Heimpel et al. 2010), whereas in 2012, the pattern was reversed. When we analyzed whether plot diversity affected which plants had mummies, we found that it did not, suggesting that plot diversity does not affect the ability of female wasps to find soybean plants. However, the number of mummies on a plant depended on both plot diversity and on the number of aphids on the plant. The mechanism underlying the increase in mummies on plants next to low-diversity plants is not clear. It is possible that intraguild predation or ovipositing interference by aphid predators reduces mummy numbers on plants next to high diversity plots. However, plot diversity did not affect the number of aphid predators observed, making this mechanism somewhat unlikely. Other possibilities are that parasitoid wasp females increase oviposition rates on plants next to low diversity plots, perhaps due to visual or olfactory cues, or that the structure of the vegetation in low diversity plots reduced female wasp emigration from these areas (Sheehan 1986).

Parasitoid mummies were abundant in our experiment, unlike in those of Schmidt et al. (2007) and Koch et al. (2012), who experimentally added cover crops to soybean and found very few parasitoid mummies during their surveys. This suggests that parasitoids may benefit from high plant diversity at the hectare scale, while ovipositing larger numbers of eggs in low-diversity patches.

The assemblage of natural enemies in this experiment was diverse, with predators and parasitoids observed across seven arthropod orders (Tables 3-2, 3-3). However, aphid abundance increased rapidly early in the season, when natural enemy numbers were low

(Figure 3-8, 3-10). Lack of soybean aphid suppression by native natural enemies appears to be common in agricultural fields in the north central United States (Noma et al. 2010), and the same lack of suppression has been observed for other aphid agricultural pests (Latham and Mills 2010). Brown (2011) found that the timing of coccinellid arrival was critical to its impact on aphid populations, and the importance of soybean aphid natural enemy abundance early in the season is recognized in China (Wu et al. 2004). Because of the exponential increase of aphid populations early in the season, early predator pressure may be important in determining aphid population trajectories. In our experiment, predator and parasitoid abundances were low early in the season (Figure 3-8), which allowed aphid populations to increase rapidly in the first weeks. Subsequently, natural enemies appear to have responded to the increase in aphid abundance and driven aphid abundance down.

The most abundant and effective biological control agents for soybean aphids in North American soybean fields are thought to be coccinellids and *Orius insidiosus* (Fox et al. 2004, Desneux et al. 2006, Costamagna et al. 2008). Neither Coccinellidae nor *O. insidiosus* were abundant on the plants in this experiment, with fewer than 70 individuals of each being found over all plants over the whole summer in both years. Coccinellidae may search for or stay longer in high aphid density patches (Evans 2003), but because of the experimental design, there were no dense patches of soybean plants in our experiment. While higher abundances of *O. insidiosus* have been found in soybean fields with increased vegetation diversity compared to soybean monoculture (Lundgren et al. 2009), we did not find greater numbers of *O. insidiosus* on soybean plants next to high diversity plots. In China, parasitoids are a major contributor to soybean aphid control, with rates of parasitism up to 50% being observed (Wu et al. 2004). Parasitism rates in our experiment were typically under 20%.

While previous research has shown that predators and parasitoids in our experimental plots were found to be more abundant with higher plant species richness (Haddad et al. 2009), the particular natural enemies that attack soybean aphids may not respond to species richness. A study that evaluated the effect of natural enemies on

soybean aphid on soybean in different habitats found that different natural enemies responded differentially (Brewer and Noma 2010). Some species showed habitat affinity, but were generally in low abundance. The most common parasitoid in their experiment, the generalist aphid parasitoid *L. testaceipes*, showed no preference among soybean, alfalfa, poplar, and early succession vegetation habitats. Similarly, there was no difference in effect of natural enemies on aphid abundances across the four habitats.

One possible reason for the lack of plot diversity effect on predator abundance, aphid abundance, and yield might be that the plot size in this experiment was too small to affect mobile predators. Altieri and Whitcomb (1980) found an effect of plant diversity on *Spodoptera frugiperda* natural enemy abundance and diversity when treatments were separated by 50 meters, but not when they were separated by 8 meters. They suggest that natural enemies can easily traverse short distances between low diversity and high diversity treatments and thus perceive plant diversity as high across all treatments when treatments are close together. Landscape studies have also shown that scale affects the response of natural enemies to landscape diversity (Thies et al. 2003, Gardiner et al. 2009).

However, a meta-analysis of the effect of plant diversity on insect abundance found that effects of plant diversity were more pronounced at smaller plot sizes than larger ones (Bommarco and Banks 2003) and a separate meta-analysis of landscape diversity studies suggests that the response of natural enemies to scale is species-specific (Chaplin-Kramer et al. 2011). In particular, herbivore abundance was reduced more and predator abundance increased more with plant diversity in smaller plots versus larger ones (Bommarco and Banks 2003) and generalist enemies were found to respond at all scales, while specialists responded more at small scales (Chaplin-Kramer et al. 2011). Stronger effects in smaller plots may be due to an increased encounter rate of different diversity treatments, allowing greater insect choice. Soybean aphids in our experiment were added homogenously, so we would not expect aphid choice to affect aphid abundance; results might have been different if we had allowed for natural colonization. Because aphid abundance was relatively high and similar across all diversity treatments,

mobile generalist natural enemies in our experiment may not have experienced much difference in aphid availability from plot to plot.

Our results suggest that natural enemies in diverse plant communities can control soybean aphids below economic injury levels. Plants exposed continuously to natural enemies throughout the summer reached a peak aphid abundance of  $480 \pm 31$  (SE) in 2011 and  $556 \pm 31$  in 2012, below economic injury levels. When natural enemies were excluded, however, economic injury levels were surpassed in 2012 with a peak aphid abundance of  $1,129 \pm 90$ . The lack of significant differences in aphid abundance and natural enemy abundance among the plant diversity treatments suggests that natural enemies may be responding at scales greater than several meters. In particular, mobile natural enemies may disproportionately contribute to aphid biological control.



## Tables and Figures

**Table 3-1: Natural Enemy weights for predator pressure analysis.**

Predator weights are based on aphid feeding and parasitism rates

Natural Enemy	Aphids killed (per day)	References
<b>Araneae</b>	2	(Gavish-Regev et al. 2008)
<b>Coccinellidae adults</b>	62	(Latham and Mills 2010, Atlihan et al. 2010)
<b>Coccinellidae larvae</b>	37	(Shannag and Obeidat 2006, Latham and Mills 2010, Atlihan et al. 2010)
<b>Syrphidae larvae</b>	76	(Hopper et al. 2011)
<b><i>Aphidoletes</i> spp. larvae</b>	3	(Latham and Mills 2010)
<b><i>Orius insidiosus</i></b>	16	(Simonsen et al. 2009)
<b>Other predatory Hemiptera</b>	10	(Fantinou et al. 2009)
<b><i>Aphelinus</i> spp. mummies (black)<sup>1</sup></b>	0.25	(Wu and Heimpel 2007, Frewin et al. 2010)
<b><i>Lysiphlebus testaceipes</i> mummies (tan)<sup>2</sup></b>	0.27	(Hight et al. 1972)
<b>Parasitoid wasp adult</b>	80	(van Steenis 1994, Frewin et al. 2010)
<b>Neuroptera larvae</b>	39	(Latham and Mills 2010)

1. For *Aphelinus* spp., average duration of mummies is 8 days. Adult females engage in host feeding, doubling the number of aphids killed per mummy created.

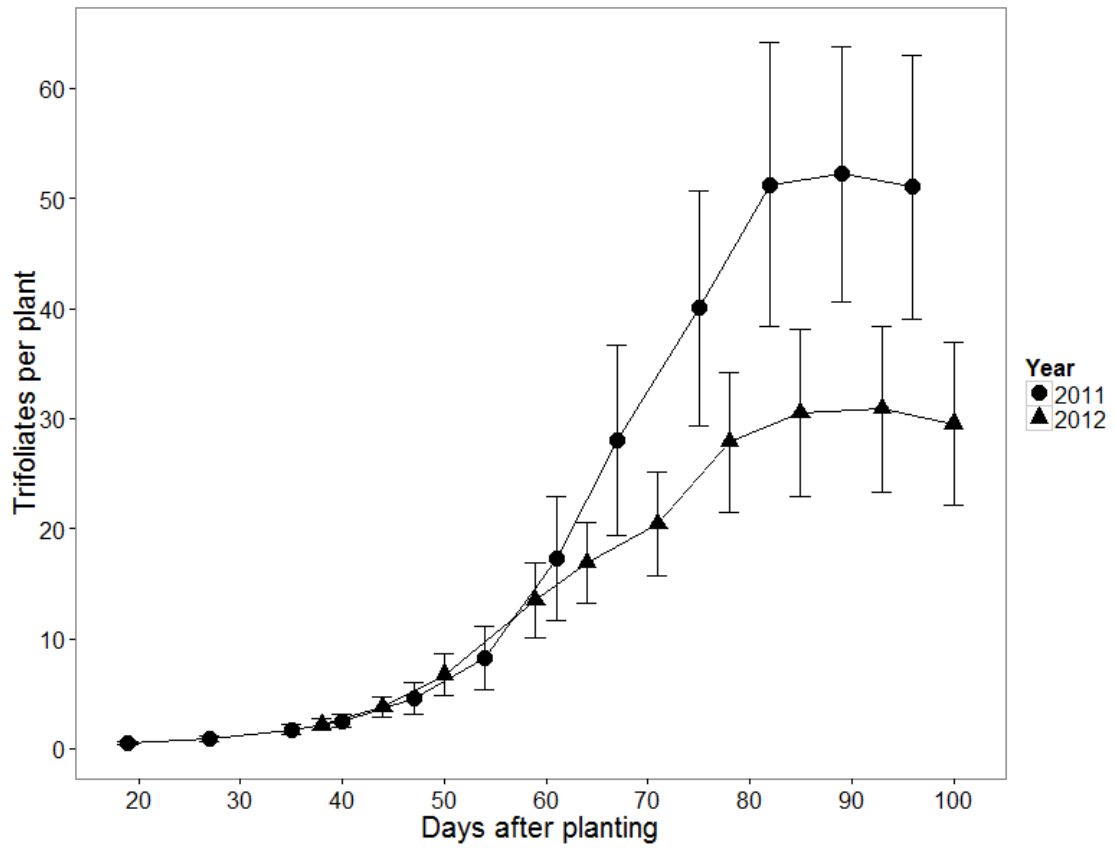
2. For, *Lysiphlebus testaceipes*, average duration of mummies is 3.7 days.

**Table 3-2: Insects other than soybean aphids observed in 2011**

Type	Group	Insect	Occurrences	Individuals
herbivore	Hemiptera	Aphidoidea other than <i>A. glycines</i>	2	3
	Other	Other	187	451
	<b>TOTAL</b>		<b>189</b>	<b>454</b>
predator	Araneae		22	24
	Coccinellidae	Adults	56	59
		Larvae	6	8
	Diptera	Predatory adults	20	23
		Predatory larvae	548	1,245
	Hemiptera	Predatory Hemiptera	57	69
	Hymenoptera	<i>Aphelinus</i> spp. mummies	13	18
		<i>Lysiphlebus testaceipes</i> mummies	222	526
	Neuroptera	Adults	46	61
		Eggs	4	7
		Larvae	13	15
	Other	Other	27	33
	<b>TOTAL</b>		<b>1,034</b>	<b>2,088</b>
ants	Formicidae		<b>1,154</b>	<b>7,277</b>
unknown	Other	<b>Other</b>	<b>269</b>	<b>373</b>
<b>OVERALL TOTAL</b>			<b>2,646</b>	<b>10,192</b>

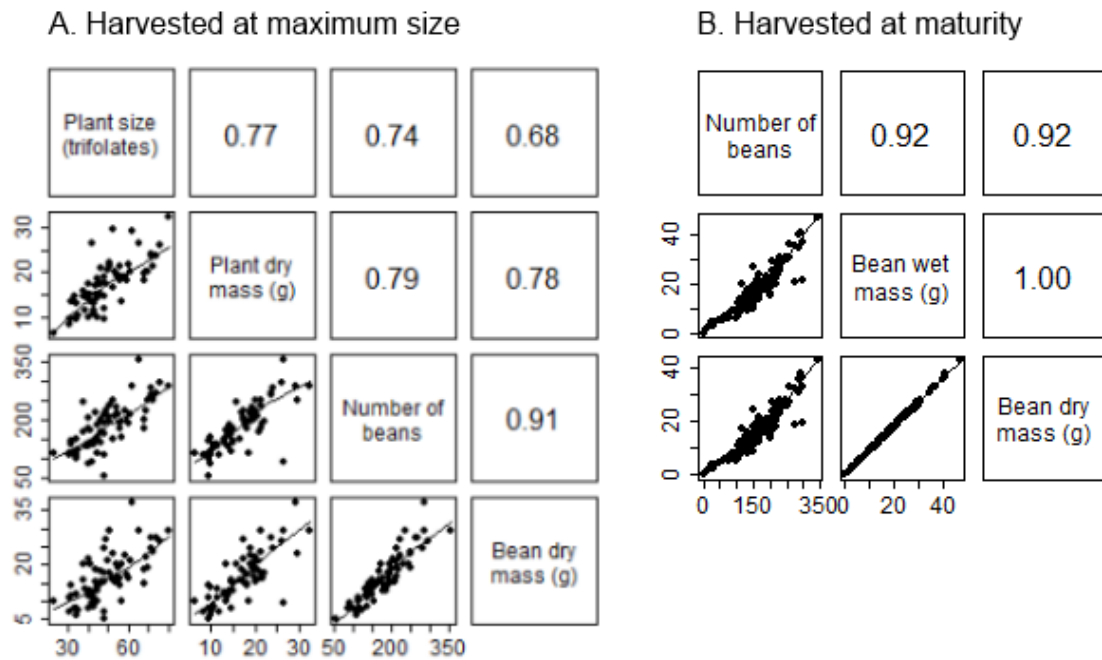
**Table 3-3: Insects other than soybean aphids observed in 2012**

Type	Group	Insect	Occurrences	Individuals	
herbivore	Coleoptera		19	19	
	Diptera		1	1	
	Hemiptera	Cicadellidae		288	328
		Aphidoidea other than <i>A. glycines</i>		8	8
		Aleyrodidae		1,478	5,967
		Other		12	11
	Hymenoptera	Bees		4	4
		Wasps		1	1
	Lepidoptera	Adults		13	14
		Caterpillars		390	644
	Orthoptera			10	10
	Thysanoptera			1,283	3,738
Other	Other		2	3	
<b>TOTAL</b>			<b>3,509</b>	<b>10,748</b>	
predator	Araneae		204	210	
	Coleoptera	Coccinellidae adults	41	44	
		Coccinellidae larvae	16	17	
		Other Coleoptera adults	9	23	
	Diptera	Syrphidae adults	27	33	
		Syrphidae larvae	206	317	
		<i>Aphidoletes aphidimyza</i> larvae	359	961	
		Other predatory Diptera	2	3	
	Hemiptera	Nabidae	18	18	
		<i>Orius insidiosus</i> adults	13	14	
		<i>Orius insidiosus</i> nymphs	30	42	
		Other predatory Hemiptera	2	2	
	Hymenoptera	<i>Aphelinus</i> spp. mummies	698	4,871	
		<i>Lysiphlebus testaceipes</i> mummies	49	102	
		Parasitoid wasp adults	13	13	
	Neuroptera	Adults	2	2	
		Eggs	74	106	
Larvae		48	51		
Thysanoptera		29	39		
<b>TOTAL</b>			<b>1,840</b>	<b>6,868</b>	
ants	Formicidae		<b>886</b>	<b>2,517</b>	
unknown	Coleoptera		7	8	
	Diptera		91	96	
	Thysanoptera		328	955	
	Other		45	57	
	<b>TOTAL</b>			<b>471</b>	<b>1,116</b>
<b>OVERALL TOTAL</b>			<b>6,706</b>	<b>21,249</b>	



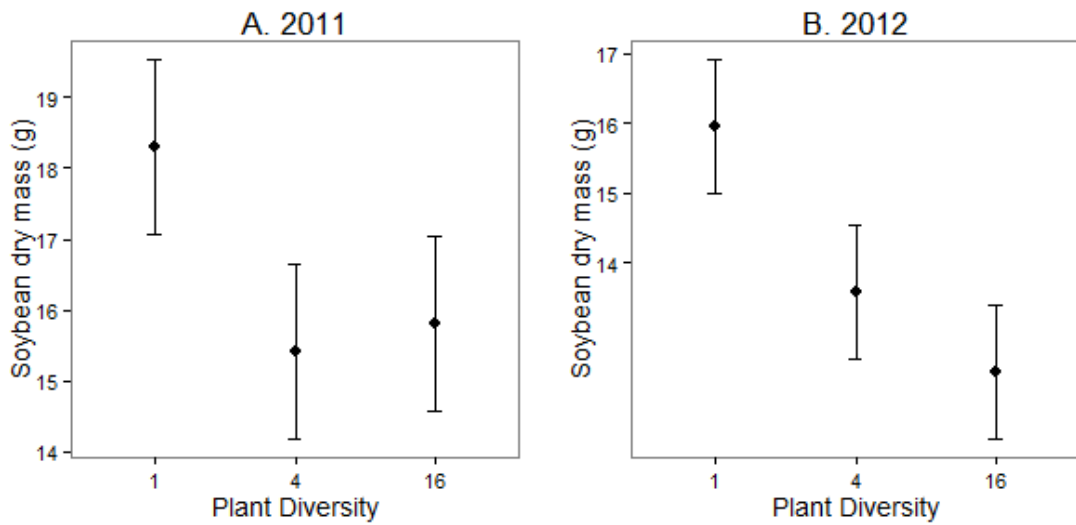
**Figure 3-1: Soybean plant size over time**

Mean number of trifoliates per soybean plant and standard deviations for 2011 (circles) and 2012 (triangles).



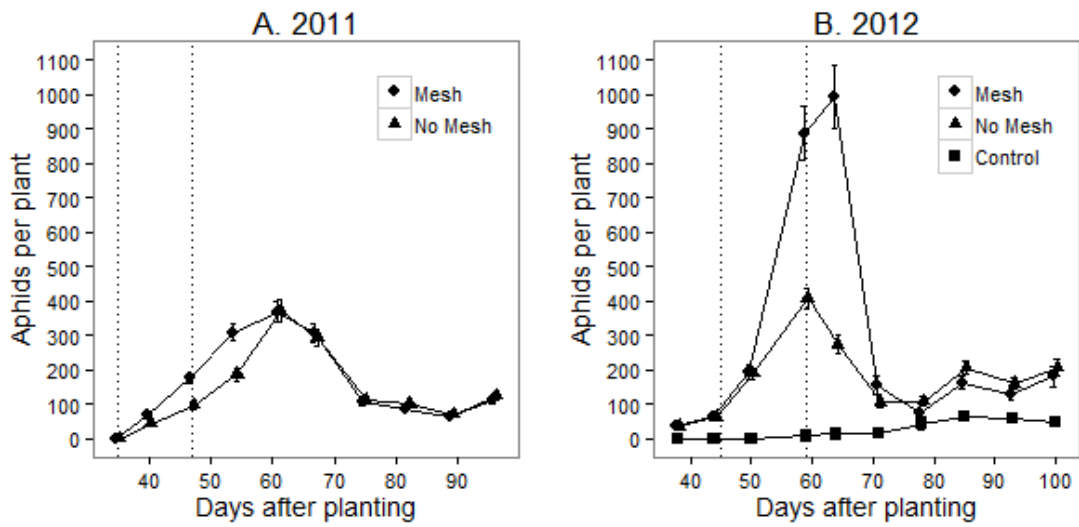
**Figure 3-2: Correlation matrices of measures of soybean plant growth and yield**

(A) Plant maximum size as number of trifoliates, plant (leaf and stem) dry mass, number of soybeans, and soybean dry mass for 66 plants harvested at maximum size in 2011. (B) Number of soybeans, soybean harvested mass, and soybean dry mass for 132 plants harvested at soybean maturity in 2011.



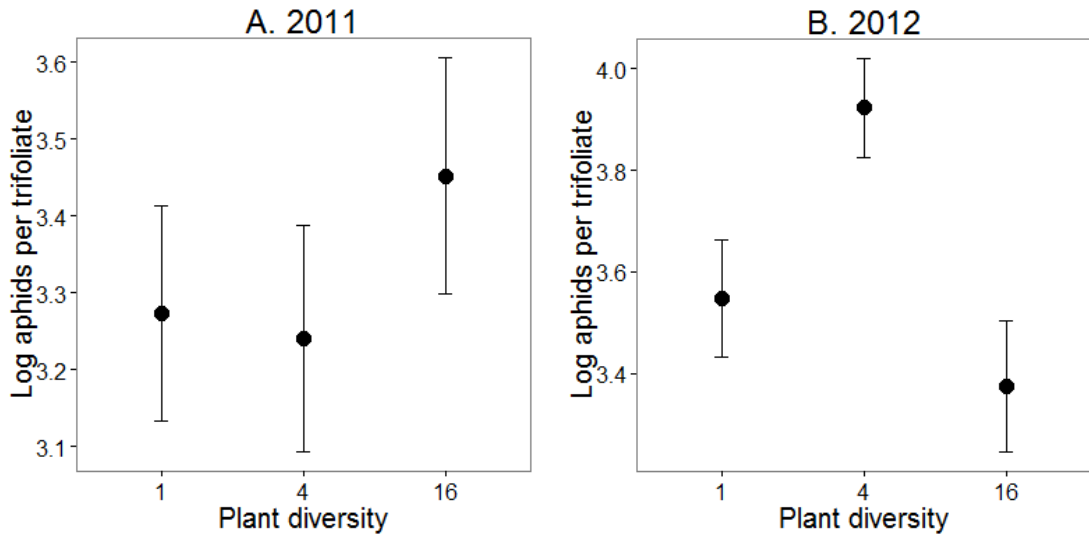
**Figure 3-3: Soybean yield by plant diversity**

Mean soybean dry mass of plants next to 1-, 4-, and 16-species plots. Error bars are standard errors. Shown are the results for all plants each year in a model that incorporates plot identity as a random effect. (A) In 2011, means were not significantly different. (B) In 2012, soybean dry mass was significantly higher in 1-species plots than in 16-species plots.



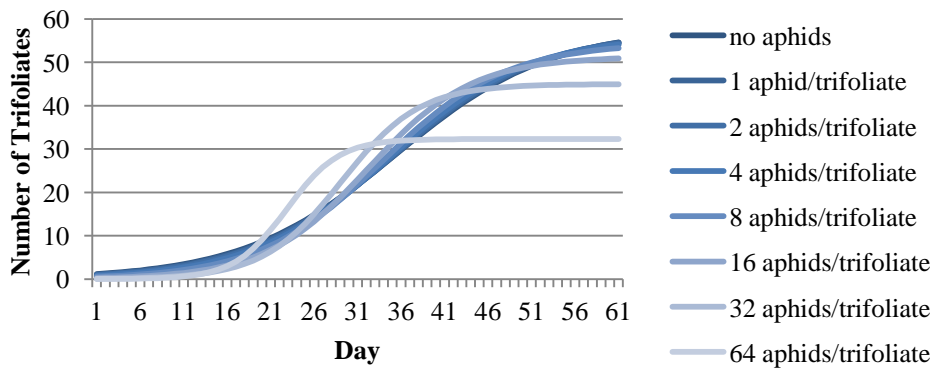
**Figure 3-4: Aphid abundance on plants by plant treatment**

Mean aphid abundance on plants by plant treatment in (A) 2011 and (B) 2012. Error bars are standard error of the mean. Symbols are jittered to aid readability. Vertical dotted lines indicate the beginning and end of natural enemy exclusion for the mesh treatment.



**Figure 3-5: Peak log aphid density per plant by plot diversity**

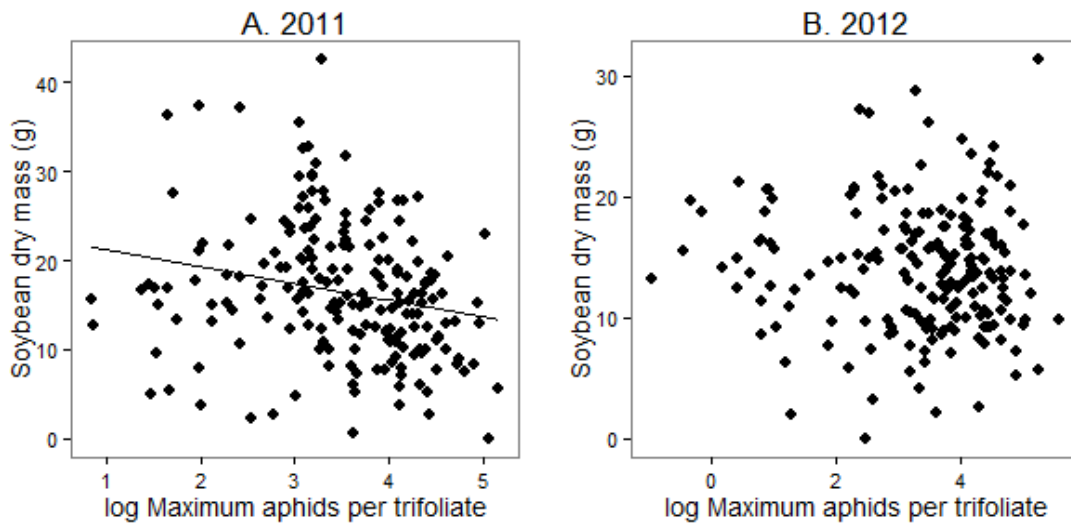
Mean and SEM maximum log aphid density for all plants with aphids added and without mesh. (A) In 2011, means were not statistically different for different plant diversity levels. (B) In 2012, plots with 4 species of plant had statistically more aphids than plots with 1 or 16 species of plant.



**Figure 3-6: Modeled plant growth based on aphid density for 2011**

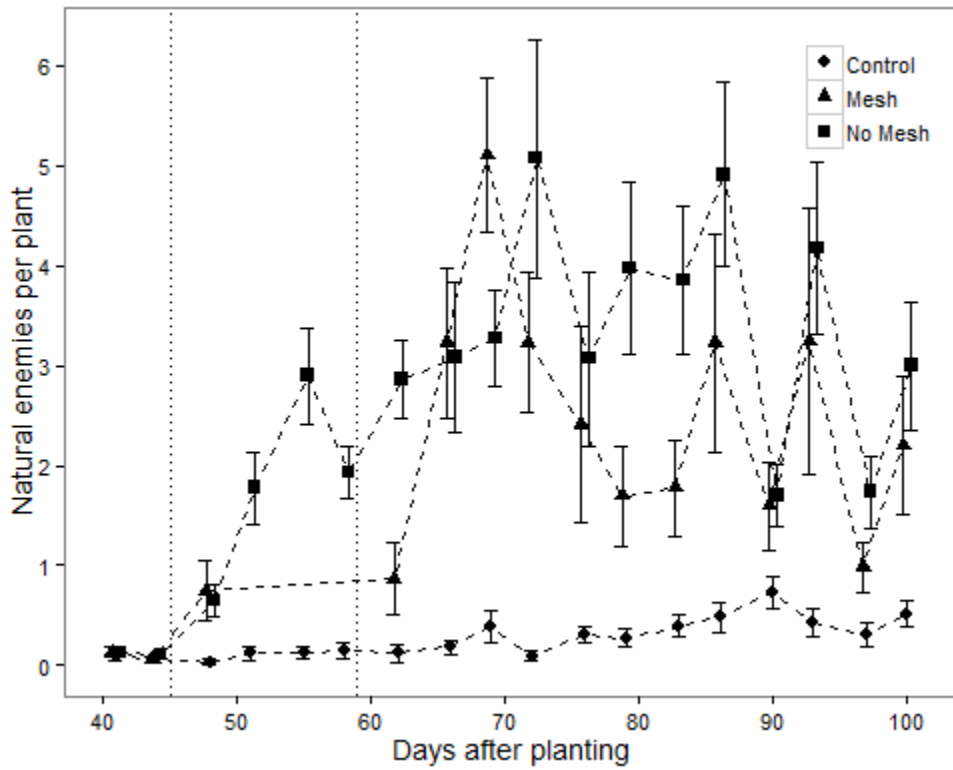
Each curve assumes constant aphid density over the whole growing period.





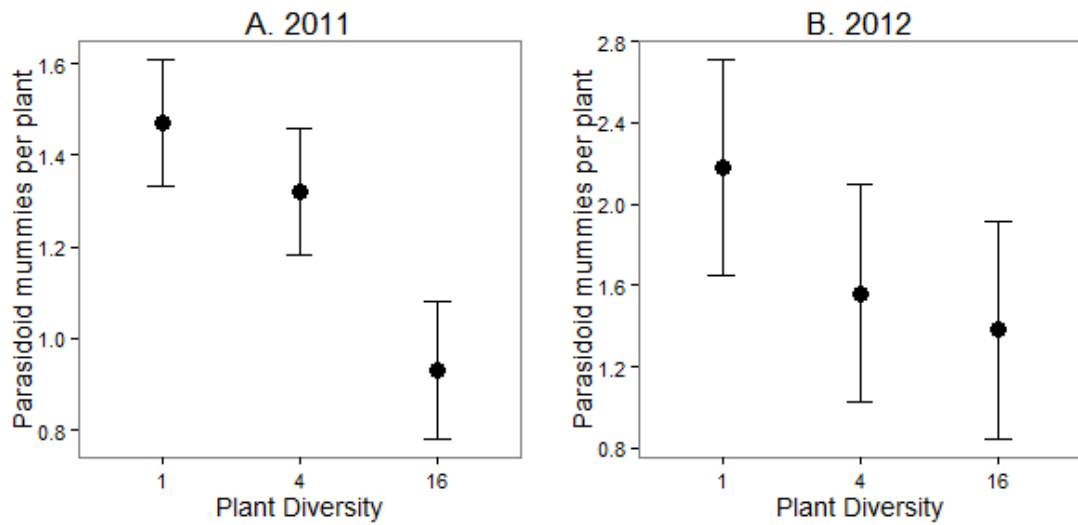
**Figure 3-7: Soybean yield (grams dry matter) by peak log aphid density**

(A) In 2011, higher maximum aphid density led to lower soybean yields ( $p=0.004$ ,  $\chi^2=8.48$ , slope=-1.77). (B) In 2012, there was no significant relationship between maximum aphid density and soybean yield ( $p=0.40$ ).



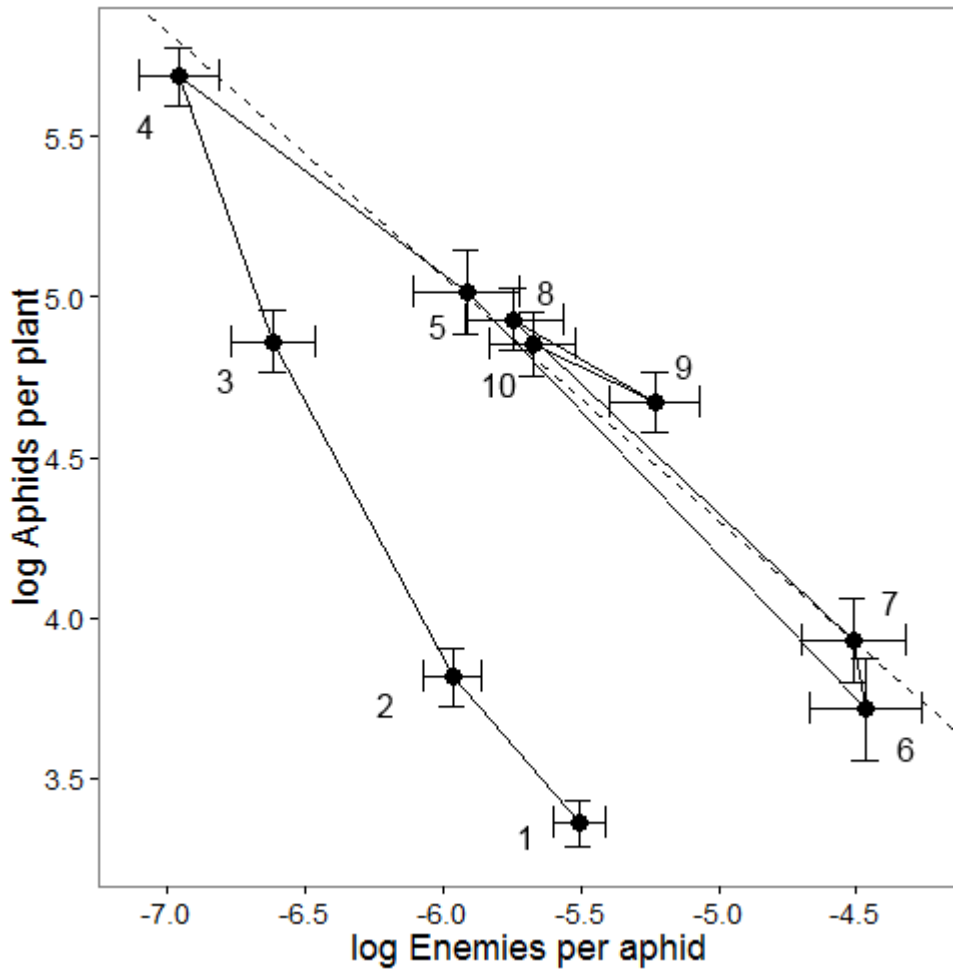
**Figure 3-8: Natural enemy abundance per plant in 2012**

Mean natural enemy abundance on plants by plant treatment in 2012. Error bars are standard error of the mean. Symbols are jittered to aid readability. Vertical dotted lines indicate the beginning and end of natural enemy exclusion for the mesh treatment.



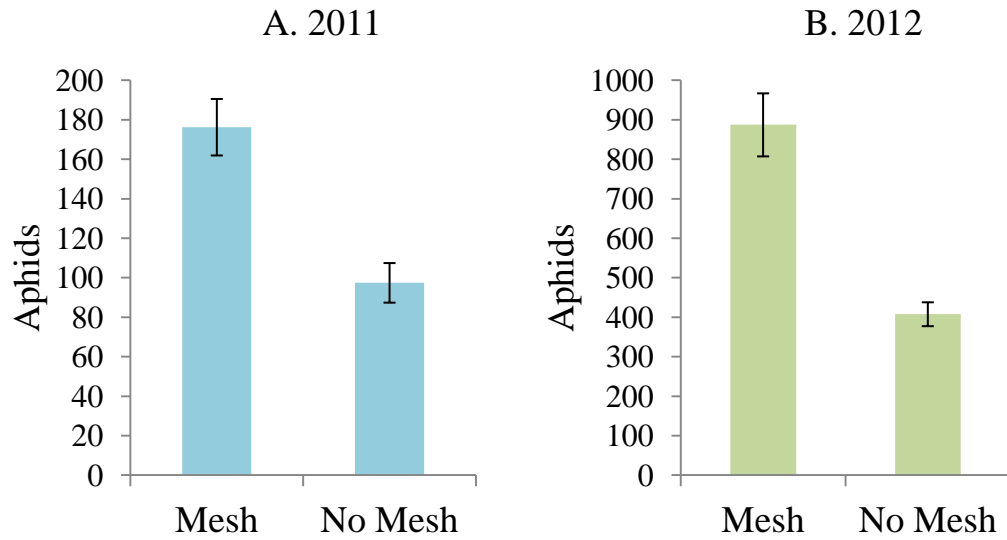
**Figure 3-9: Abundance of parasitoids mummies by plant diversity**

Mean number of parasitoid mummies found per plant by plant diversity. Mummy numbers are summed over each season. Error bars are standard errors of the mean. Plant diversity was significant in both years in a model containing plant treatment, and plot as a random effect.



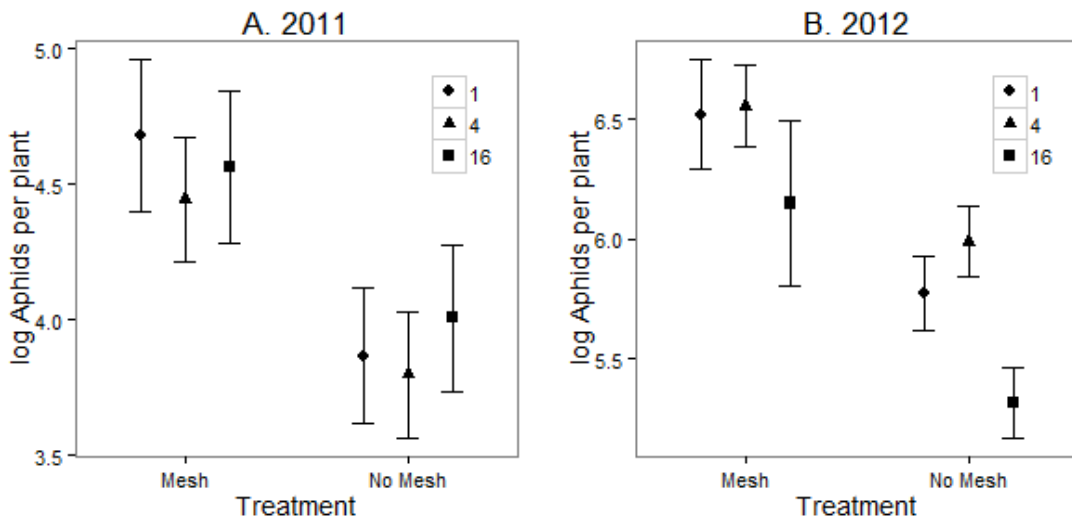
**Figure 3-10: Relationship between the log number of aphids per plant and the log number of natural enemies per aphid in 2012**

Each point represents one week, and solid lines connect weeks in chronological order (indicated by numbers). Error bars are standard errors of the means. Only unmeshed plants that had aphids added were included. The dashed line is a linear regression fit to weeks 4 through 10 (intercept = 0.481, slope = -0.765,  $R^2=0.96$ ,  $p<0.0001$ ).



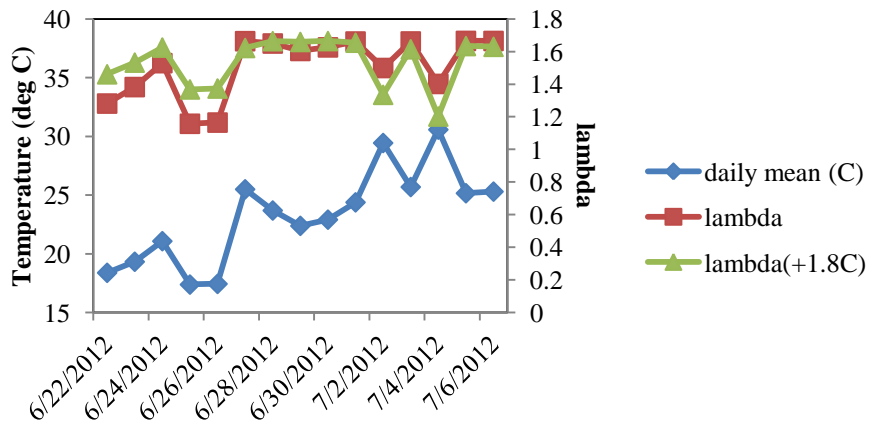
**Figure 3-11: Number of aphids per plant after two weeks of being covered by mesh or left uncovered**

Aphid abundance was significantly reduced in both years when plants were unmeshed. Error bars are standard errors.



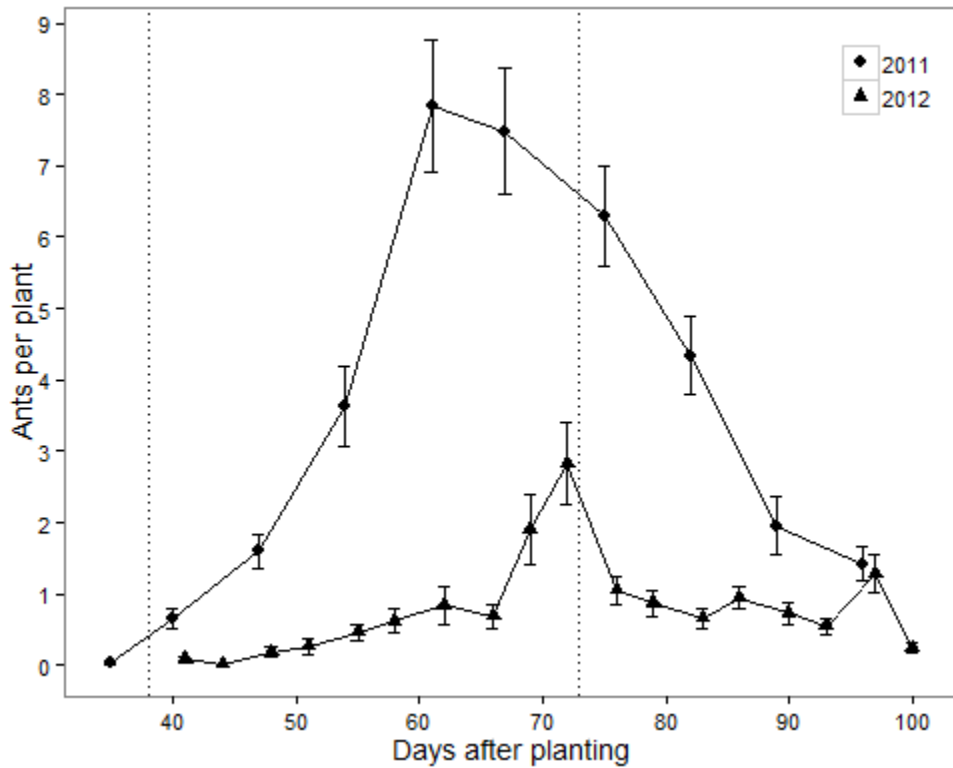
**Figure 3-12: Log number of aphids per plant after two weeks of being covered by mesh or left uncovered, by plant diversity**

Error bars are standard error of the mean.



**Figure 3-13: Daily mean temperature and calculated  $\lambda_m$  values during the two weeks soybean plants were covered with mesh**

Lambda values are for unmeshed (squares) and meshed (triangles) plants.



**Figure 3-14: Ant abundance per plant during the 2011 and 2012 growing seasons**

Mean ant abundance per plant in 2011 (circles) and 2012 (triangles). Error bars are standard error of the mean. Shown are the plants that did not receive mesh and that had aphids added (n=99 plants each year). Vertical dotted lines indicate the timing of ant bait trap deployment in 2012. Ant bait traps were not used in 2011.

## **Chapter 4 : Local competition and dispersal limitation affect community dynamics over twenty years in a seed addition experiment**

Metacommunity theory posits that both local processes, such as competition, predation, and abiotic limitation, and regional processes like dispersal are important in community dynamics. In plant systems, the patch-dynamic paradigm suggests that trade-offs between competition and dispersal can relax the constraints on the number of coexisting species, leading to high levels of diversity and a changing mosaic of species abundances. Seed addition experiments can yield insight into the importance of both local and regional processes under this paradigm. However, the community consequences of most seed addition experiments are followed for only a few years at most. We analyzed data over a twenty-year period following a seed addition experiment that added up to 54 species of seeds to an already species-rich Minnesotan savanna. We found that while mean species richness increased by 25% and total species richness across the experiment increased 29%, local processes such as competition limited local persistence of added species. Twenty years after seeds were added, species richness was no longer proportional the number of species added to a plot, despite a strong relationship in early years. Species that were present before seeding showed a decline in richness proportional to the number of seed species added, a relationship that only became apparent five years after seeds were added. Dispersal limitation was also evident. Two thirds of 33 seeded species that were absent or rare before seeds were added persisted for twenty years, and 10 were found to be increasingly spreading to adjacent areas, suggesting long-term persistence in the savanna.

### **Introduction**

An overarching goal of community ecology is to understand the patterns and processes governing the distribution and abundances of species. Ecological research has traditionally focused on understanding these patterns and processes at a local scale or at a regional scale. Over the past couple decades, the challenge of integrating ecological



processes across scales has been formally identified (Levin 1992) and theoretical advances have been made to link patterns and processes at different scales.

It was recognized forty years ago that species movement among discrete spatial patches could theoretically allow species to coexist even when the patches were identical with regard to limiting resources (Levins and Culver 1971, Armstrong 1976). Since then, increasingly sophisticated models have shown that adding spatial structure can alter expected community dynamics. One important insight has been that species that would go extinct due to being poor competitors can coexist with better competitors in patchy environments by having greater dispersal distances, dispersal rates, and/or establishment rates (Shmida and Ellner 1984, Nee and May 1992, Tilman 1994, Durrett and Levin 1998). As a result, species diversity is less constrained by the number of limiting resources in patch models than in non-spatial, well-mixed ones.

The most recent major advance in spatial community ecology is the metacommunity concept, which incorporates both local and regional processes and the relationships among them (Leibold et al. 2004). Metacommunities consist of multiple discrete local communities that are connected by dispersal of species among those communities. Species interact at the local level through such processes as competition and predation. At the regional level, the rates at which species disperse among local communities affects their abundances in local communities, potentially swamping local processes if dispersal rates are high.

Four non-exclusive conceptual paradigms have been identified within the metacommunity concept (Leibold et al. 2004). The ‘patch-dynamic’ paradigm posits that limitations to species coexistence at a local level can be offset by tradeoffs in dispersal among communities. This paradigm encompasses earlier work showing competition-colonization trade-offs (Hastings 1980, Shmida and Ellner 1984, Tilman 1994) and trade-offs between fecundity and dispersal (Yu et al. 2001). The ‘species-sorting’ paradigm requires that abiotic conditions in local communities vary such that the variation affects local species interactions. Species fare better under certain conditions and are able to “follow” their niche if conditions change via dispersal to other communities. The ‘mass-

effects' paradigm assumes that dispersal happens on the same time scale as local dynamics and focuses on the effects of high dispersal rates on local communities. Finally, the 'neutral' paradigm assumes that all species are equivalent in terms of local interactions and dispersal ability. Such metacommunities may contain many species in a non-equilibrium state for a long time, but eventually result in just one surviving species.

In these spatial models of community structure, dispersal between local communities is an integral component. In plant communities, this dispersal typically occurs as seed movement. Because a plant community that does not respond to increased seed input is considered saturated, spatial models typically require some sort of empty site, gap, or niche be available to allow immigrating seeds to establish. As a result, in some models, species move around in space, persisting at a large scale, but blinking in and out at local sites.

Reviews and meta-analyses of seed addition experiments find that plant communities of many types are seed limited (Turnbull et al. 2000, Clark et al. 2007, Myers and Harms 2009). However, most seed addition experiments are conducted for a relatively brief time – a few years at the most. As a result, conclusions about the effect of seed additions on species' ability to persist in the plant community are limited. For perennial plants, observations of seedling presence may not translate into mature, reproductive plants that are able to at least replace themselves. And for annual plants, the presence of a species in one year is not indicative of long-term population persistence.

We analyze a seed addition experiment over a time period of twenty years to better understand the long-term dynamics of a plant community after a one-time massive addition of seeds.

## **Methods**

The experiment was conducted at Cedar Creek Ecosystem Science Reserve (part of the National Science Foundation Long Term Ecological Research Network), Minnesota, in a ~2 ha stand of oak savanna ("Field D", 45.397°N, 93.181°W). The savanna has never been cultivated and has been free of livestock for >70 years; it has

been maintained by a regime of controlled burning two out of every three springs since 1966. Over the course of this experiment, it was burned in 1991, 1993, 1994, 1996, 1997, 1999, 2000, 2002, 2003, 2005, 2006, 2008, 2009, 2011, and 2012.

In August, 1991, thirty 2 x 2m sites were haphazardly selected across the savanna. Each site was divided into four 1 x 1 m plots that were visually examined to determine percentage cover of all contained plant species and of land area not covered by plants. The next year, in June 1992, mixtures of seeds from 0, 5, 10, 15, 20, 25, 30, 35, 40, or 54 species were selected randomly from a pool of 54 regionally occurring species. Each mixture was a separate random draw of a given level of diversity, with 6 replicates of each level of seeded diversity. Seeds were placed on the surface of the southeast and northwest plots within each site (60 plots total) at a rate of 4.5 g/plot per species. See Tilman (1997) for a detailed description of the seeding treatments and earlier results.

Plots were surveyed between late July and early September in 1992, 1993, 1994, 1995, 1997, 1998, 2004, 2008, and 2012. (Survey data from 1995 could not be located and is not included in our analyses.) Percentage cover of all species and percentage of non-vegetated land area were recorded. Additionally, percentage cover measurements were made in the two 1 x 1 m areas (southwest and northeast) at each site that did not receive any seeds; we refer to these as “adjacent areas.” Adjacent areas were surveyed each year with the exception of 1998 and 2004. In 2012, at each site of seed addition we also searched an 8 m<sup>2</sup> “satellite area” that was separated from the seed addition plots by a 1 m space. In each satellite area, we searched for the presence of 27 seeded species that had been very rare or absent in all plots before seeds had been added (based on our 1991 survey).

Due to changes in nomenclature over the past two decades, species names from all surveys were standardized, using the PLANTS Database (USDA NRCS 2013) as the authority (Table A4-1). For some species groups, a few species were identified to the level of species in some years but only to genus in others. To facilitate comparisons across years, we lumped such species by genera when it was not possible to ascertain species identity from historical field notes for them in every sampling year.

We also standardized percent cover surveys by rescaling such that percentage cover measures sum to 100% in each plot. Analyses using different standardization techniques, such as excluding “bare ground,” yielded similar results.

Data were analyzed using R and its packages MASS, reshape, and ggplot2 (Venables and Ripley 2002, Wickham 2007, 2009, R Core Team 2012). Analyses consist of linear regressions, logistic regressions, and paired t-tests.

Data sets are archived as experiment ‘e093’ and can be obtained on the Cedar Creek Ecosystem Science Reserve website:

<http://www.cbs.umn.edu/explore/field-stations/cedarcreek/research/data>

## **Results**

Across the whole experiment, 103 plant species were recorded in 2012 (Figure 4-1).

Plant species richness (number of species observed in a given plot) initially increased with the number of species added as seeds to plots (Tilman 1997). Five years after seeding, the slope describing this relationship began to decrease (Figure 4-2) and by 2008, sixteen years after seeding, the slope was no longer any statistically significant. Likewise, the difference in species richness between plots with many species added as seed and those with few species added became smaller over time (Figure 4-3). By 2008, there was no statistical difference in species richness between seeded and unseeded plots.

The proportion of plots covered by vegetation initially increased with the number of species added as seeds (Tilman 1997). But by 1997, there was no longer a significant relationship between number of species added as seed and the proportion of plots covered by vegetation (Figure A4-1). We calculated the increase in total cover of seeded species in each plot as the difference between seeded species cover in a given year and 1991. Increase in total cover of seeded species depended initially on the number of species added as seeds, but this relationship was no longer significant by 2004 (Figure A4-2).

Of the 54 seeded species, 36 were found in 5% or fewer of the plots and adjacent areas in 1991, before seeds were sown. These species were each seeded in between 22

and 29 plots. Thirty-three of these 36 species established a plant in at least one plot in which it was seeded, with establishment rates varying from 4% to 100% of seeded plots.

In 2012, 22 of these 33 species (67%) persisted in at least one plot to which they had been added (Table 4-1). Interestingly, of those 22 species, those species that had initially established in the most plots experienced the greatest decrease in plot abundance over time (Figure A4-3, linear regression,  $R^2=0.30$ ,  $p=0.005$ ). Ten of the 22 species occurred in just one or two plots in 2012.

The mean plot species richness increased from 14.6 species in 1991 to 18.1 species in 2012 (Figure 4-4; paired t-test:  $p<0.0001$ ). The mean species richness of seeded species in plots increased from 4.5 in 1991 to 7.1 in 2012 (Figure 4-4; paired t-test:  $p<0.0001$ ). Of the 33 seeded species that were absent or rare in 1991, mean plot species richness in seeded plots increased from 0.3 species in 1991 to 2.2 in 2012 (Figure 4-4; paired t-test:  $p<0.0001$ ).

Of these 33 species, mean plot cover increased from 0.7% in 1991 to 11.7% in 2012 (Figure 4-5, paired t-test:  $p<0.0001$ ). Percent cover of individual species was highly variable in 2012, with a few species covering over a quarter of some plots, but most species covering less than 5% of all plots in which they appeared (Figure A4-4).

There were 18 seeded species that were also relatively common in the 1991 survey (Table A4-2). The addition of their seeds increased the cover of 9 of these species compared to plots in which seeds were not added in at least one year (Table A4-2). One species (*Sorghastrum nutans*), which experienced large declines in cover across all plots over the two decades of this experiment, experience greater decline with seeding (Table A4-2). Despite an increase in cover of half of these 18 seeded species that were relatively common in 1991, mean summed cover of the 18 species was not significantly different between 1991 and 2012 (Figure 4-5, paired t-test,  $p=0.15$ ).

Linear regression showed no significant relationship between initial plot richness and the proportion of seeded species that were found in 2012 ( $R^2=0.026$ ,  $N=54$ ,  $p=0.13$ ). A contingency table analysis using the 33 species that were rare or absent in 1991 showed no significant difference in persistence to 2012 among perennial grass (6

species), perennial legume (7 species), and perennial non-legume forb (17 species) functional groups ( $X^2=0.92$ ,  $df=2$ ,  $p=0.63$ ).

Of the 33 species that were rare or absent before the seeding treatment, 18 species were found in at least one adjacent area by 2012 (Table 4-1). Species were far more likely to be found in adjacent areas next to plots in which the species had been seeded than in areas next to plots that had received seeds, but not of that species (logistic regression,  $z=6.16$ ,  $p<0.0001$ ). Fourteen species had spread to at least one satellite area more than a meter away from seeded plots by 2012 (Table 4-1). One species (*Lupinus perennis*) that was absent from the experiment before the seeding treatment was found to have spread to satellite areas, despite no longer being present in any seeded plot in 2012. Species that persisted in a larger number of plots in 2012 were more likely to be one of these spreading species (logistic regression,  $z=-2.57$ ,  $p=0.01$ ). Ten spreading species showed a statistical increase over time in the number of adjacent areas in which they were found (Figure A4-5).

We analyzed the number of species gained and lost in each plot between two successive surveys, with respect to how many species were added as seed. Species gain per number of seeded species was significantly greater than zero in the early years of the experiment, but was not significantly different from zero in 1998-2004, and 2008-2012 (Table 4-2, Figure 4-6). Species loss per number of seeded species initially increased from zero in 1991-1992, reached a peak in 1994-1997, and then decreased (Table 4-2, Figure 4-6).

The species found in plots before the seeding treatment (that were not also seeded) showed decreases in both species richness (Figure 4-7) and cover (Figure A4-6) with increasing number of species seeded starting in 1997 and 1994, respectively. In particular, as the cover of the 33 species that were rare or absent in 1991 increased over time, the fraction of species that were present before seeding decreased (Figure A4-7).

## Discussion

In meta-analyses of seed addition experiments, Clark et al. (2007) and Poulsen et al. (2007) highlight the importance of two separate processes in plant dispersal that are often overlooked or combined in theoretical and empirical studies. In order for a species to successfully establish at a new site, its seeds must first arrive at the new site, and second, those seed must successfully germinate. A limitation in either seed arrival or seed survival reduces the dispersal ability of a species, but each has different underlying mechanisms. Importantly, both types of limitation are likely occurring in many systems.

In this experiment, we find strong evidence for both types of limitation. Seeding rates per m<sup>2</sup> ranged from ~200 (for large-seeded legumes) to over 100,000 (for the tiniest seeds), but only about 50 seedlings per species added were counted in the first year of the experiment. As seeds were purchased from a nursery and had high viability, the fact that most added seeds did not germinate points to other causes of seed mortality; inappropriate abiotic microsite conditions involving available water, light, and nutrients, and/or biotic microsite conditions such as seed predators, pathogens, or competitors may have contributed to seed mortality (Clark et al. 2007).

Seed arrival is also limited in this community. Of the 36 species that were rare or absent before seed addition, 92% germinated at least once where seeded. This indicates the establishment niche exists for these species in this field and that species abundances are constrained by the number of seeds produced and/or dispersal distance. In a review of seed-augmentation experiments, about 30% of species in grasslands were limited by seed arrival (Turnbull et al. 2000). In this experiment, we found that 39% of existing species increased in abundance with seed addition within a year after seeding, and that 50% showed an increased abundance at some point over twenty years. The higher levels of seed limitation for existing species in this experiment compared to others may be explained by the relatively high levels of bare ground that occur due to sandy soil and animal disturbances. Turnbull et al. (2000) found a pattern of increased seed limitation in grasslands with more bare ground and also found that seed limitation was increased in experimental disturbance treatments compared with controls. Presumably, plots with

more bare ground provide more available microsites for colonization than plots with less bare ground. The fact that the fraction of bare ground in plots in this experiment decreased with seeding (Tilman 1997) supports this idea.

This experiment extends the discussion of dispersal limitation to life stages after germination. In order for long-term dispersal of a species to be successful, not only must seeds arrive at a site and germinate, but also, seedlings must persist to an adult state and reproduce at a high enough rate to ensure population viability. This experiment is unique in that it tracks the plant community beyond the few years typical of seed-addition experiments to understand the long-term effects of seed dispersal.

In a review of seed introduction experiments, Turnbull et al. (2000) found that 64% of added species were found as seedlings, but only 23% were found as adults. After twenty years in this experiment, a third of the 33 germinating species that were rare or absent before seed addition were no longer found where seeded, suggesting that these 11 species were unable to reach a mature state, unable to reproduce, or reproduced at too low a level to maintain the species presence. Another third of the 33 germinating species were found in only 1 or 2 plots in 2012, indicating that while these species were able to reach maturity, their rate of reproduction is either too low to maintain species presence or else is so slow that 20 years is not yet enough time to gauge their long-term persistence. Because species loss between 2008 to 2012 was related to the number of seeded species (Table 4-2, Figure 4-6), it seems plausible that seeded species loss is ongoing twenty years after the start of the experiment and that seeded species may continue to disappear from the experiment in future surveys.

In all the experiments reviewed by Turnbull et al. (2000), none looked at nearby areas to see if added species were dispersing and if populations were becoming self-sustaining. In this experiment, only a third of the 33 germinating species were found in more than two plots in 2012. However, these species also showed signs of spreading to nearby areas (Table 4-1, Figure A4-5), suggesting not only that they were able to reach maturity, but also that they reproduced at a high enough rate to ensure local population persistence. The 3 most abundant of these 33 species (*Agastache foeniculum*, *Desmodium*



*canadense*, *Baptisia alba*) comprised, on average, 23%, 22%, and 17% of cover in 2012 in the plots to which they persisted, and thus had become the 4th, 9th, and 10th most abundant species across all plots (out of 103 total species).

While not apparent in the first several years of the experiment, the number of seeded species added to each plot had a significant negative effect on the richness and cover of species that were present before seeds were added (Figures 4-7, A4-6). As seeded species – and especially novel seeded species – established and took up more area, they began to decrease the richness of the species present in plots before experimental seed addition (Figure A4-7). This pattern of displacement continued until 2012, suggesting that pre-experiment species never regained their richness nor cover in seeded plots. Because the relationships between number of seeded species and total species richness, total plant cover, and seeded species plant cover becomes insignificant by 2012 (Figures 4-2, A4-1, A4-2), other species that were neither in the plot before the experiment nor seeded into the plot increased in richness and cover over time to make up the difference.

This long-term view adds to the suite of mechanisms that may limit species dispersal in a meta-community. An absent species may not occur at a site because its seed has not arrive, because its seed has arrived, but not germinated, or because its seed has arrived and germinated, but not reproduced enough to sustain the species locally. Each of these mechanisms is governed by different processes. Arrival requires that seeds be produced elsewhere and disperse far enough. Germination requires the appropriate abiotic germination conditions, as well as escape from granivory, pathogens, and competition. Population persistence also requires the appropriate abiotic conditions, escape from natural enemies, and competition success, as well as suitable conspecific partners for plants requiring pollination. The abiotic conditions necessary for germination are often not the same as those required by adult plants to reproduce (Grubb 1977), however, and the natural enemies of seeds and plants are likewise different.

Because of the different mechanisms limiting the long-term success of dispersal at different life stages, it seems likely that each species makes multiple different trade-offs

among seed dispersal, germination, maturation, and reproduction. These multiple trade-offs can facilitate higher species richness than a two-dimensional tradeoff, as different species are more successful than competitors at different life stages (Kneitel and Chase 2004).

If, for example, there is a strict competition-colonization trade-off hierarchy among species, we would expect species that are the best competitors to increase in abundance in plots to which their seed is added, because they would necessarily be poor colonizers. The result would be domination by one or a few species and a decrease in overall diversity. However, we do not observe this pattern in this experiment; instead, seeding increased mean diversity across plots, and seeded species known to be dominant competitors in tallgrass prairies, such as the perennial grasses *Andropogon gerardii*, *Schizachyrium scoparium*, and *Sorghastrum nutans*, showed no increase or only very slow increase in abundance when their seed was added to plots (Table A4-2).

High levels of species richness may also be maintained by heterogeneity of environmental conditions in space and time (Turnbull et al 2000). The experimental field is a mesic savanna throughout, but at the plot scale, there is heterogeneity: there are mounds and dips and disturbances over time. Gophers (*Geomys bursarius*) and ants (especially *Formica obscuripes*) disturb the soil with mounds and nests. Trees and their branches sometimes fall, changing shading patterns. And the fires that burn the field most years do so unevenly. Because of this heterogeneity, individual plants may no longer be competitive dominants in a particular site as conditions change. We found that within 1m x 1m plots, community composition was quite variable over a twenty-year period, suggesting that changing local conditions are an important component of plant community dynamics.

In total, this long-term seed addition experiment underscores the importance of post-germination success in the dispersal dynamics that structure plant communities. Previous research explored the importance of seed limitation (Poulsen et al 2007) and seed mortality (Clark et al 2007) in plant community dynamics, but seed-addition experiments that followed species communities for more than a few years were

previously lacking. We find both seed limitation and seed mortality to be important components of dispersal limitation in our experiment, and these patterns were readily detected within a few years of seed addition (Tilman 1997). However, following this experiment over 20 years has revealed important long-term patterns. Of the 33 species that were not present before seed was added and that germinated following seeding, two thirds had substantially declined in abundance or had disappeared entirely twenty years later. Plant mortality and/or limited reproduction are responsible for these declines and absences and are major factors determining species abundances two decades after seed sowing. The seeded species that appeared to be maintaining persistent populations after twenty years were associated with the loss of some of the original species from plots, a pattern that was not apparent in the first several years. Finally, there was another suite of species that were not present in any plots before seed was added and that were not added as seed, but that moved into some plots and persisted. Tradeoffs in multiple life-history constraints associated with dispersal (seed arrival, seed survival, plant maturation, and local reproduction) are likely important contributors to the high diversity of plant species in this experiment (103 plants species within 60 m<sup>2</sup>) and their long-term dynamics.

## Tables and Figures

### **Table 4-1: Persistence and spread of the 33 seeded species that were rare in the experiment in 1991 and germinated after seeding.**

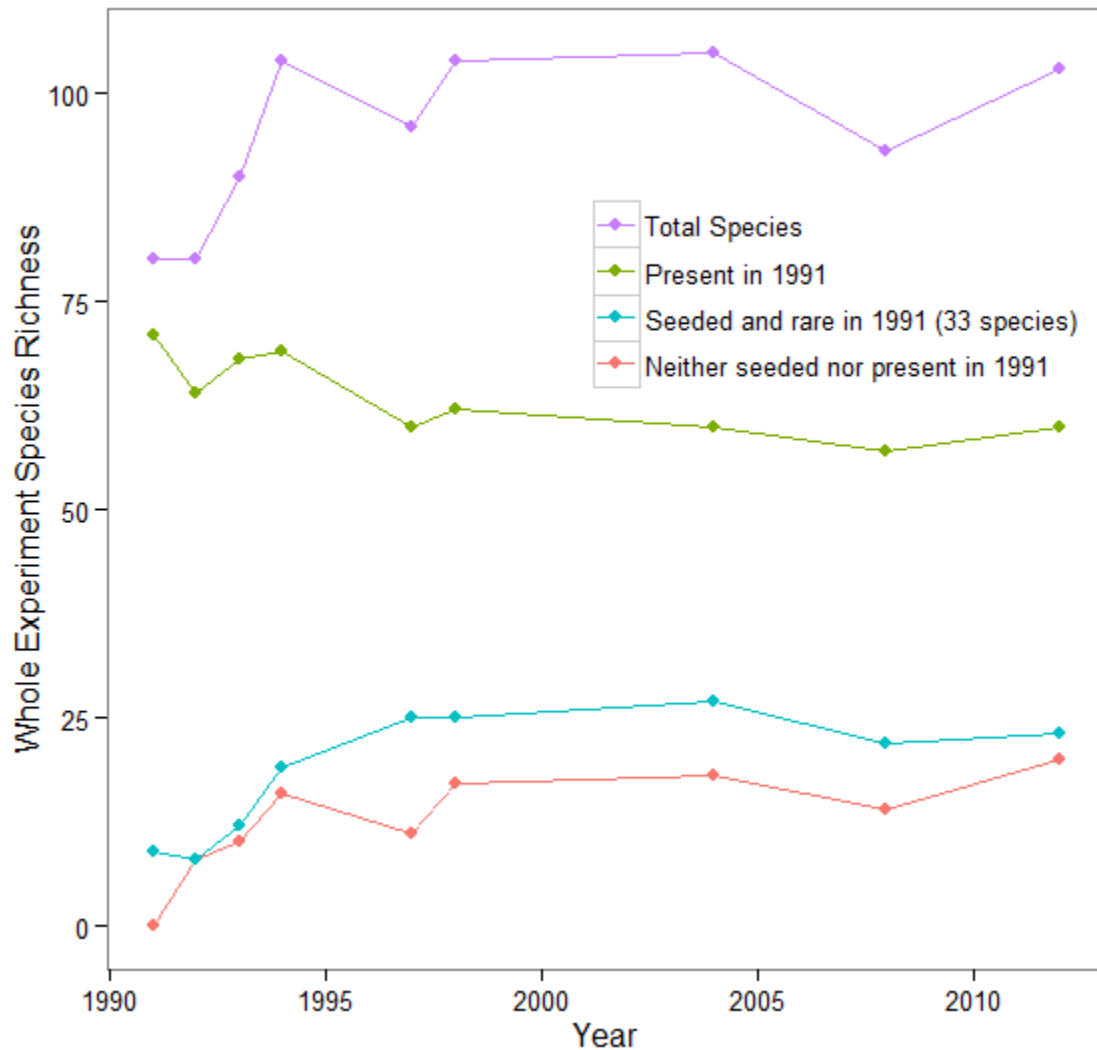
The first column indicates the number of plots and 1m x 1m adjacent areas in which species were found before seeding (out of 120). The second column gives the number of plots (out of 60) to which each species' seed was added. The third column shows the number of seeded plots in which the species was found in 2012. The fourth column shows the number of unseeded plots and adjacent areas in which the species was found in 2012. The fifth column gives the number of satellite areas located at least 1 m from plots (out of 30 sites) in which the species was found in 2012.

	1991	1992	2012	2012	2012
	Found in plots and adjacent areas	Seeds added	Found in plots in which seeded	Found in unseeded plots and adjacent areas	Found in satellite areas
<b>Achillea millefolium</b>	-	29	11	15	4
<b>Agastache foeniculum</b>	-	24	10	17	4
<b>Allium stellatum</b>	-	29	14	10	5
<b>Ambrosia artemisiifolia</b>	2	28	-	-	-
<b>Asclepias syriaca</b>	6	24	-	5	-
<b>Asclepias verticillata</b>	-	27	1	1	-
<b>Astragalus canadensis</b>	-	25	2	3	-
<b>Baptisia alba</b>	-	25	5	2	3
<b>Bouteloua curtipendula</b>	2	23	3	5	1
<b>Coreopsis palmata</b>	5	26	5	11	-
<b>Dalea purpurea</b>	-	26	2	2	-
<b>Dalea villosa</b>	-	22	-	-	-
<b>Dalea candida</b>	-	24	-	-	-
<b>Delphinium carolinianum</b>	-	27	2	-	2
<b>Desmodium canadense</b>	5	28	8	7	4
<b>Elymus canadensis</b>	-	25	-	-	-
<b>Gentiana andrewsii</b>	-	27	-	-	-
<b>Heuchera richardsonii</b>	-	28	-	-	-
<b>Koeleria macrantha</b>	-	23	9	11	6
<b>Lupinus perennis</b>	-	24	-	-	2
<b>Nepeta cataria</b>	-	27	-	-	-
<b>Oligoneuron rigidum</b>	1	28	1	2	-
<b>Panicum virgatum</b>	-	26	8	9	2
<b>Paspalum setaceum</b>	-	25	-	-	-
<b>Penstemon grandiflorus</b>	1	27	2	-	-
<b>Polygonum convolvulus</b>	2	23	5	1	-
<b>Potentilla arguta</b>	6	25	2	3	3
<b>Solidago speciosa</b>	-	29	4	4	2
<b>Sporobolus heterolepis</b>	-	22	2	-	1
<b>Verbena stricta</b>	-	29	4	2	-
<b>Veronicastrum virginicum</b>	-	24	1	-	1
<b>Vicia villosa</b>	-	24	-	-	-
<b>Zizia aptera</b>	-	27	1	-	-

**Table 4-2: Results of linear regressions on the number of species seeded in plots.**

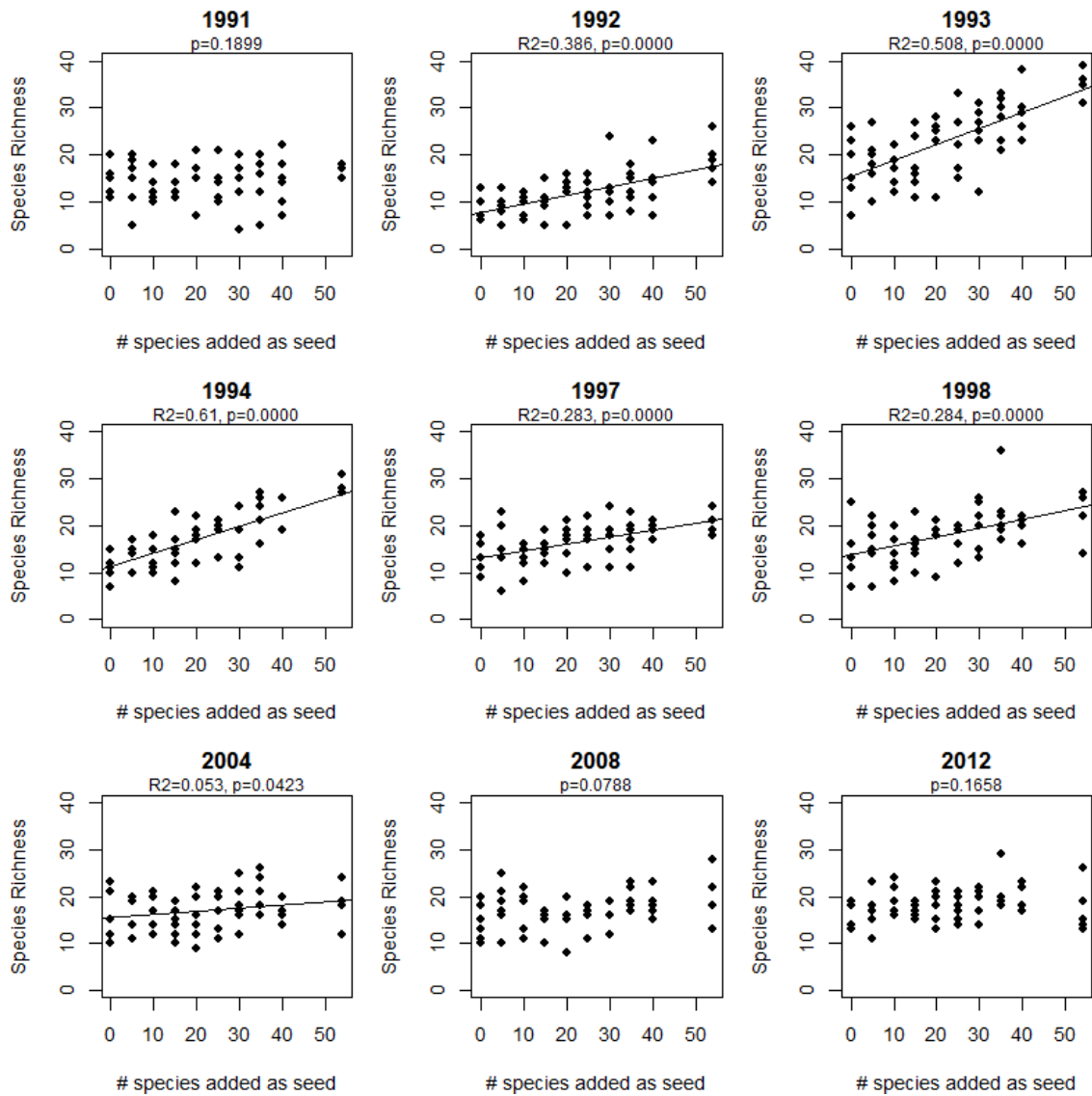
Slopes represent the number of species (gained, lost) per number of species seeded.

	<b>Species Gain</b>	<b>Species Loss</b>
<b>1991-1992</b>	Slope=0.138, R <sup>2</sup> =0.50, p<0.001	R <sup>2</sup> =0, p=0.98
<b>1992-1993</b>	Slope=0.222, R <sup>2</sup> =0.48, p<0.001	Slope=0.060, R <sup>2</sup> =0.29, p<0.001
<b>1993-1994</b>	Slope=0.073, R <sup>2</sup> =0.21, p<0.001	Slope=0.135, R <sup>2</sup> =0.28, p<0.001
<b>1994-1997</b>	R <sup>2</sup> =0.02, p=0.16	Slope=0.168, R <sup>2</sup> =0.44, p<0.001
<b>1997-1998</b>	Slope=0.068, R <sup>2</sup> =0.08, p=0.016	R <sup>2</sup> =0.03, p=0.11
<b>1998-2004</b>	R <sup>2</sup> =0.0, p=0.95	Slope=0.120, R <sup>2</sup> =0.25, p<0.001
<b>2004-2008</b>	Slope=0.042 R <sup>2</sup> =0.05, p=0.052	Slope=0.050, R <sup>2</sup> =0.11, p=0.006
<b>2008-2012</b>	R <sup>2</sup> =0.01, p=0.19	Slope=0.043, R <sup>2</sup> =0.08, p=0.018



**Figure 4-1: Species richness across all plots**

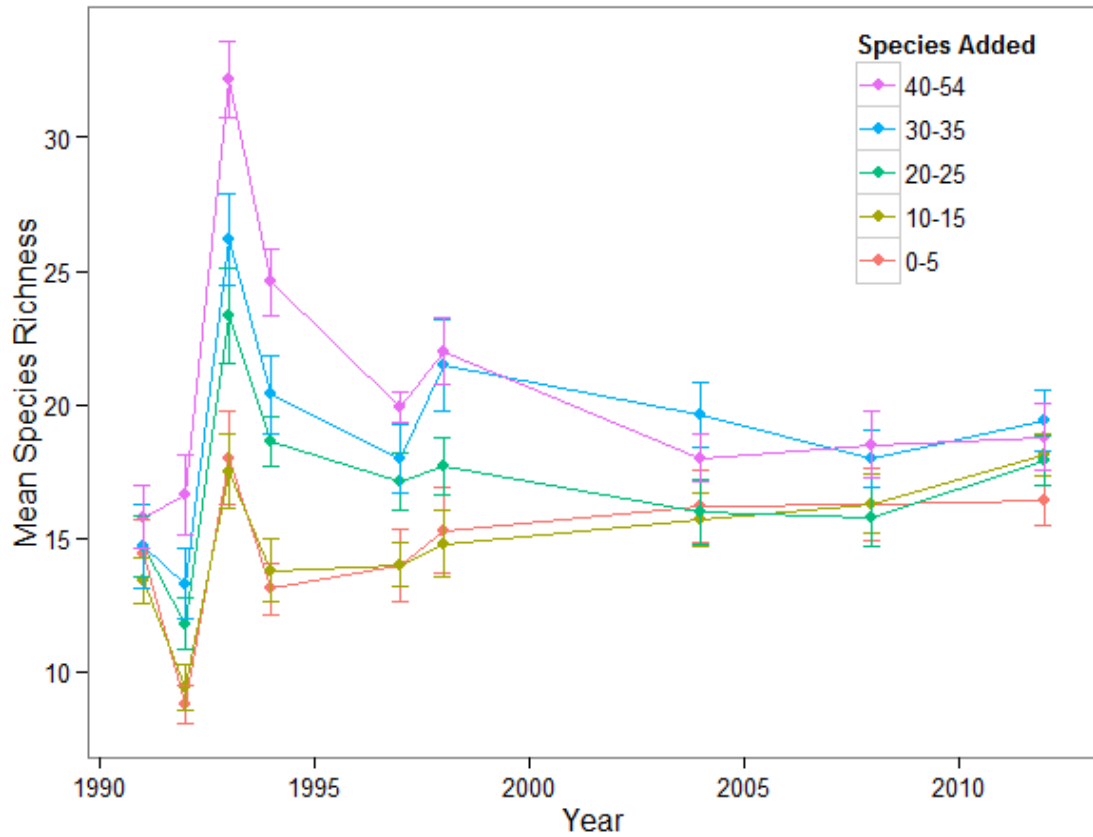
Total number of species, number of species that were present in the plots in 1991, number of the 33 seeded species that were rare in the experiment in 1991, number of species that were neither seeded nor present in 1991. Total numbers of species are (1991) 80, (1992) 80, (1993) 90, (1994) 104, (1997) 96, (1998) 104, (2004) 105, (2008) 93, (2012) 103. The surveys in 1992, 1993, and 1994 covered only the center 0.5 m x 0.5 m center of each plot; in other years, surveys covered the full 1 m x 1 m plot.



**Figure 4-2: The effect of the seeding treatments (number of seeds added to a subplot) by observed species richness**

Significant relationships at the  $p < 0.05$  level are shown as lines. This is the same analysis as Tilman (1997) Figure 2, which shows 1992-1995. By 2008, the positive slope is gone.

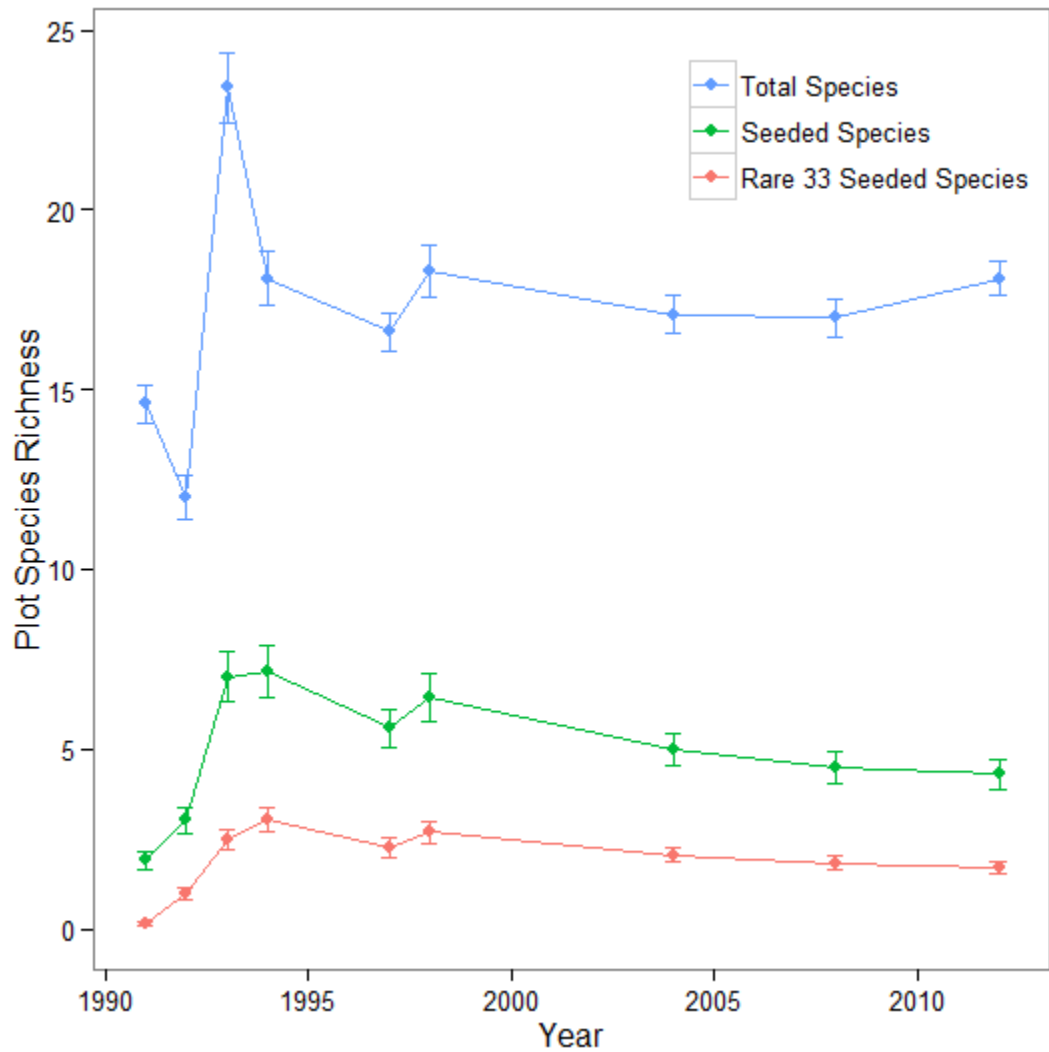




**Figure 4-3: Mean species richness in plots over time by number of species added as seed**

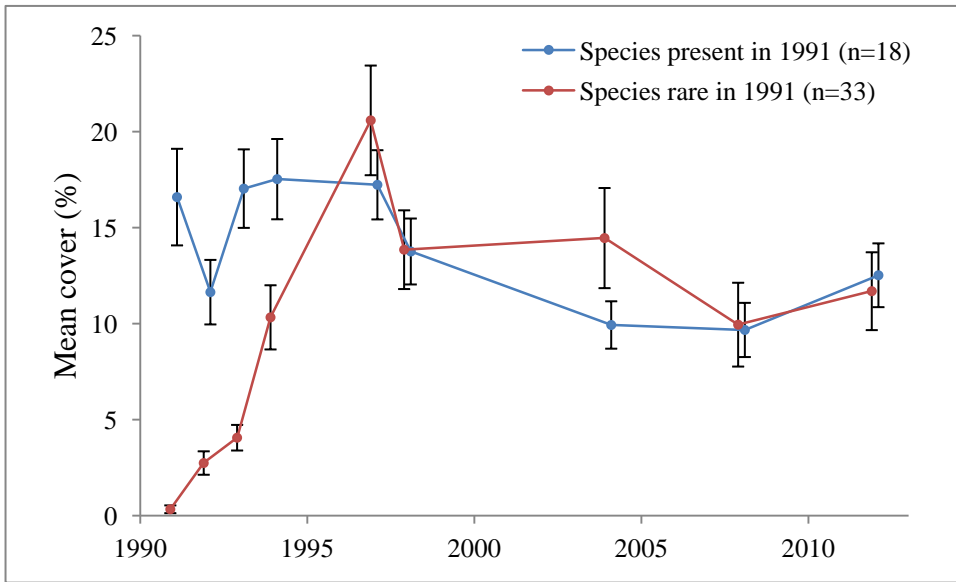
The ten seeding treatments are consolidated into five groups for legibility. Plots with many species added increase greatly and then decrease in species richness. Plots with few species added show a small increase, a decrease, and then a gradual increase over time.

Error bars are standard error.



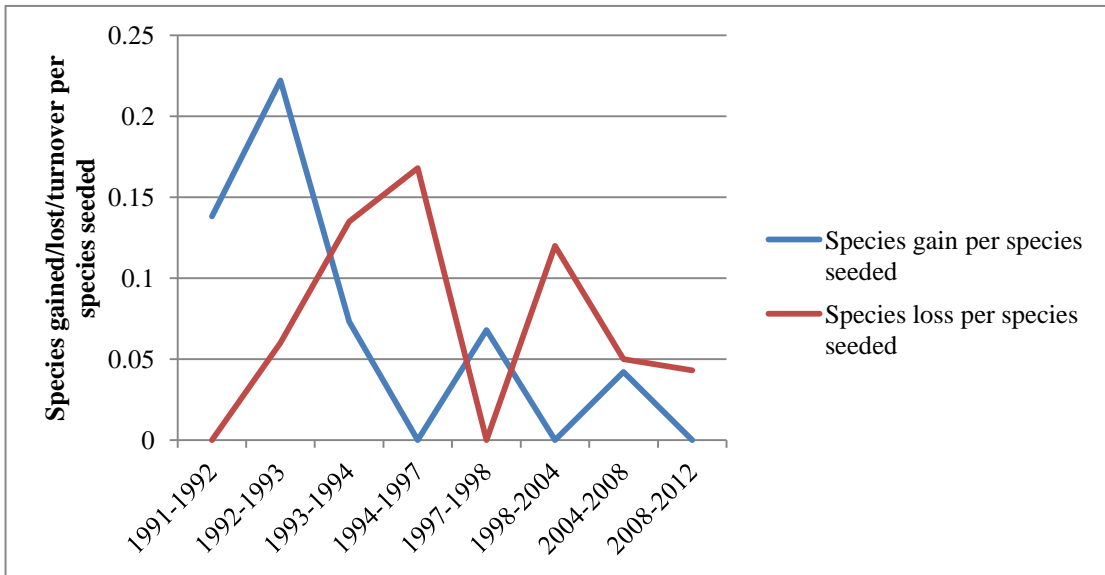
**Figure 4-4: Mean plot species richness, richness of species seeded in plots, and richness of the 33 species that were rare or absent from the survey before seeds were added**

Error bars are standard error.



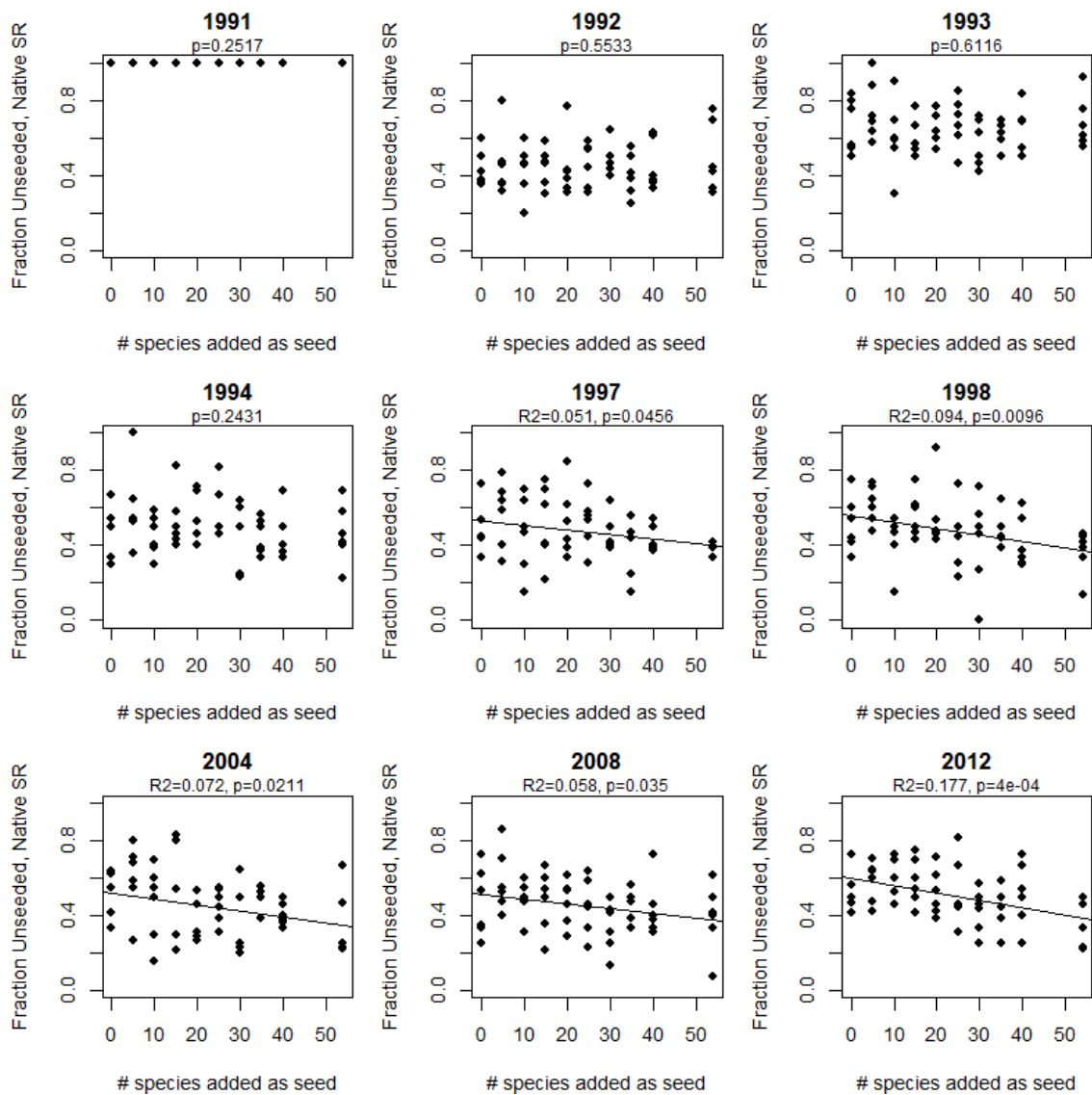
**Figure 4-5: Mean summed cover (%) of all seeded species that were rare in and seeded species that were relatively common in 1991**

Rare in 1991 (n=33). Relatively common in 1991 (n=18). Three seeded species that never germinated are excluded. Error bars are standard error.



**Figure 4-6: Species gained and lost for each period between successive surveys**

Species gain is high in early years, but declines over time. Species loss initially increases, then decreases. Data plotted are slopes in Table 4-2.



**Figure 4-7: Effect of seeding treatment on the proportion of species present in each subplot in 1991 (that were not also seeded)**

There is no observable effect until 1997.

## **Chapter 5 : Estimating wildlife disease dynamics in complex systems using approximate Bayesian computation models**

Emerging infectious diseases of wildlife are an increasing concern to managers and conservation policy-makers, but are often difficult to study and predict due to the complexity of host-disease systems and a paucity of empirical data on them. We demonstrate the use of approximate Bayesian computation (ABC) statistical methods, borrowed from the fields of population genetics and human epidemiology, to reconstruct the disease dynamics of bovine tuberculosis (*Mycobacterium bovis*) in Kruger National Park's lion (*Panthera leo*) population. ABC methods accommodate complex models that incorporate disparate knowledge about host association and movement patterns and heterogeneous disease transmission. They also allow unknown system variables to be left unspecified and search the space of all logical values. The demographics and spatial structure of Kruger National Park's lions have been studied, but it is unknown how bovine tuberculosis spreads through the population and what the average effects are on lion morbidity and mortality. The ABC approach allowed us to leave these disease parameters unspecified and infer the most likely disease parameter values based on a single survey of disease prevalence and knowledge of the lion population size. The modeling results suggest that while a large proportion of the lion population will become infected with bovine tuberculosis, lions are a spill-over host and that long disease latency is common. In the absence of future aggravating factors, bovine tuberculosis is projected to cause the lion population to decline approximately 6% over the next 50 years and stabilize at a new equilibrium.

### **Introduction**

The emergence of exotic diseases often requires wildlife managers to make decisions based on limited information. Disease dynamics are difficult to study in wild populations owing to logistical challenges, expense, and risks of handling, both to individual animals and to veterinary staff. Disease studies also require repeated sampling from the same individuals. Often such sampling it is not feasible, particularly if

endangered species are threatened by disease and intervention may be required before the disease has run its course.

One typical approach to address exotic diseases in wildlife is classical disease modeling, which can provide insight into disease dynamics in some systems (Anderson and May 1991, Keeling and Rohani 2007). However, classical models must make simplifying assumptions about the host and disease in order to be analytically solvable. Host populations must usually be well-mixed and of fixed size, and disease transmission rates between individuals must be uniform. However, many wildlife disease systems do not conform to these assumptions and classical models can give misleading results in these cases. This is especially true for group-living species, where population contact structure has important ramifications for disease spread (Tompkins et al. 2011).

To address the limitations of classical models, more complex wildlife disease models are now being developed that include details about wildlife population spatial structure and contact patterns (e.g. Snäll et al. 2008, Craft et al. 2009, Hamede et al. 2009). These models use simulation and Monte Carlo methods to investigate disease dynamics in heterogeneous networks of individuals. The drawback of these types of models is that they typically require extensive empirical data specific to the study system.

We introduce an approximate Bayesian computation (ABC) modeling approach to the study of wildlife disease as a way to understand complex disease systems without needing to specify all system parameters. The method approximates likelihood functions rather than calculating them directly, allowing for analysis of realistic models that incorporate non-linear dynamics and disparate information about host population structure, movement, behavior, and heterogeneous transmission rates. For system parameters that are not well known, ABC modeling can explore the logically complete space of these parameters, which frees the researcher from needing to specify their means or ranges. This allows for inference of disease dynamics even in cases where empirical data is limited.

Approximate Bayesian Computation (ABC) methods, also known as likelihood-free inference, generate approximate posterior distributions of parameter values when the

likelihood function is analytically intractable. They work by running a large number of simulations of system model while varying input parameters. The results of these simulations are compared to observed data using summary statistics, and those results which are closest to observed data are kept and the rest discarded. Another round of simulations is performed using the kept data with some modification to explore nearby parameter space. The simulation rounds repeat until there is convergence from round to round in the data that are kept.

ABC grew out of simple rejection algorithms that generate samples from probability distributions using large amounts of simulated data and summary statistics (Tavaré et al. 1997, Pritchard et al. 1999). They have since been adopted and refined in the fields of population genetics, systematics, and human epidemiology (Hamilton et al. 2005, Shriener et al. 2006, Tanaka et al. 2006, McKinley et al. 2009). They have also sparked some controversy in the field of phylogeography (Templeton 2009, Beaumont et al. 2010). The main criticisms made by Templeton (2009) surround the choice of model specification, model selection, and unrecognized implicit assumptions. Beaumont et al. (2010) responded by explaining that these criticisms are not specific to ABC, but are true of model-based science in general, and that ABC is simply a statistical tool to generate approximate posterior distributions from a model. We acknowledge that models make assumptions about the systems that they reflect, while embracing the power of ABC to provide meaningful insight in complex systems.

An ABC technique using Sequential Monte Carlo (ABC-SMC) is more computationally efficient than its predecessors (Sisson et al. 2007, Toni et al. 2009, Beaumont et al. 2009, Csilléry et al. 2010, Beaumont 2010), and has recently been used to investigate costs to drug resistance in *Mycobacterium tuberculosis* (Luciani et al. 2009) and to estimate disease parameters for Ebola virus (McKinley et al. 2009), a macroparasite infection of domestic cats (Drovandi and Pettitt 2011b), hospital-acquired staph infections (Drovandi and Pettitt 2011a) and Severe Acute Respiratory Syndrome (SARS) (Walker et al. 2010).



Bovine tuberculosis (*Mycobacterium bovis*) is an emerging disease in lions (*Panthera leo*) in Kruger National Park, South Africa, having been first detected in 1995 (Keet et al. 1997). In response to the threat of bovine tuberculosis to Kruger's lion population, a workshop was convened in 2009 to determine what, if anything, should be done about the situation; it was attended by lion experts, wildlife epidemiologists, and park managers. Despite the expertise present at the workshop, initial models resulting from the workshop predicted an immediate rapid decline in the lion population (Keet et al. 2009), which was not observed empirically.

We demonstrate the use of ABC-SMC to determine the disease dynamics of bovine tuberculosis in lions in Kruger National Park based on spatial data of lions, population size, and disease prevalence across the park. We assumed little knowledge about the disease itself, and investigated the full range of possible disease parameters. In particular, we addressed whether bovine tuberculosis spreads primarily from buffalo to lion or from lion to lion and what the long-term effect of bovine tuberculosis on lion population size and disease prevalence is likely to be.

## **Methods**

Kruger National Park is a 19,485 km<sup>2</sup> wildlife reserve located in the northeastern part of South Africa (22°19' - 25°32'S, 30°52' - 32°03'E.) and is part of the Greater Limpopo Transfrontier Conservation Area. Two major permanent rivers, the Sabie and the Olifants, cut across the park from east to west and reduce animal movements northward and southward, effectively dividing the park into three sectors: the northern, central and southern regions.

Bovine tuberculosis (*Mycobacterium bovis*), exotic to sub-Saharan Africa, was likely introduced to Kruger National park around 1960, when free-roaming African buffalo (*Syncerus caffer*) contracted it from domestic cattle in the southeast part of the park (Bengis et al. 1996, De Vos et al. 2001, Hofmeyr et al. 2006). The park has subsequently been fenced, greatly reducing interactions between buffalo and cattle. Even so, the disease has spread northward through the park via the buffalo population, and has

emerged in the lion population (Keet et al., 1997). Genomic analysis indicated that lions originally contracted bovine tuberculosis from buffalo, an important prey source (Keet et al., 1997). Lions are also suspected of transmitting bovine tuberculosis from one to another via aerosol transmission while in close proximity or via wounds during fights (Kaneene and Pfeiffer, 2006; Keet et al., 1998). Lions exhibiting symptoms of bovine tuberculosis usually die within a few years (Keet et al., 2009). Emergent bovine tuberculosis has raised concerns about the lion population viability in Kruger as diseases have been implicated in declines of lions in protected areas elsewhere (Munson et al. 2008). Information on bovine tuberculosis transmission and progression in lions to evaluate risks to population persistence and guide potential interventions is lacking.

### ***Lion demographic simulation***

We modified an existing individual-based lion demographic simulation model called SimSimba (Whitman et al. 2004, 2007) to incorporate bovine tuberculosis disease dynamics. Individual lions stochastically progressed in half-year time steps through life stages, including birth, maturation, dispersal, reproduction, and death. The modeled lions formed prides and coalitions that mimicked the social patterns of actual lions by moving around on a user-defined spatial lattice of territories and interacting with one another; males fought to compete for access to females and committed infanticide when they took over a pride with cubs. Parameterization of lion demographics in SimSimba used Kruger data when available and was supplemented with data on lions from the Serengeti (Appendix A). Parameterization was validated by running the model without disease and comparing lion age structure, sex ratios, and population size to known demographic values in Kruger.

A landscape map of model territories was created to mimic the geography and lion density of Kruger National Park (Figure 5-1). The lion territories were distributed by dividing the estimated number of actual lions in each of six regions of the park (northeast, northwest, central east, central west, southeast, and southwest) (Ferreira and Funston

2010) by estimated pride sizes, and were arranged in a honeycomb pattern in proportion to the physical dimensions of the park.

### *Disease model*

Each lion existed in one of three disease states: susceptible, exposed, or infectious. All lions began as susceptible and stochastically transitioned to exposed, whereupon they were considered to have a latent form of the disease that had no effect on mortality or fecundity. Exposed lions then stochastically transitioned to infectious, transmitted the disease to susceptibles, and suffered increased mortality. We made the explicit assumption that lions in the latent state could not transmit the disease, based on *Mycobacteria* pathogenesis in human tuberculosis (Bates 1984). We also assumed that exposed lions never returned to the susceptible state nor entered an immune state.

The transition from susceptible to exposed was based on three parameters.  $B$  defined the probability of transmission from the infected buffalo population to a susceptible lion.  $L$  defined the probability of transmission from an infected lion to a susceptible lion.  $O$  defined the probability of an encounter between two lions each time step. Two lions within the same social group had a 100% probability of encounter. Lions in neighboring groups had a probability of encounter between 0 and 1, as did nomadic lions passing through resident lions' territories. Resident lions from non-adjacent territories were assumed not to encounter each other.

The transition from exposed to infectious at each time step was governed by parameter  $E$ , which is comparable to the transition parameter of classical SIR disease models. The increased mortality of infectious lions was modeled with parameter  $I$ , which describes the exponential probability of dying from disease each time step and was additive to the background mortality imposed by demographic specifications. Formally, for each lion in each timestep:

$$\begin{aligned} pr(susceptible \rightarrow exposed) &= B \cdot prev_{buffalo} + [1 - (1 - L)^{i+j \cdot O}] \\ pr(exposed \rightarrow infectious) &= E \\ pr(infectious \rightarrow dead) &= I \end{aligned}$$

where  $prev_{buffalo}$  is the prevalence of bovine tuberculosis in buffalo in the area,  $i$  is the number of infectious lions in the same social group and  $j$  is the number of infectious lions in neighboring groups and local nomads. The disease dynamics parameters –  $B$ ,  $L$ ,  $O$ ,  $E$ , and  $I$  – are summarized in Figure 5-2.

We set the prevalence of bovine tuberculosis in buffalo to follow logistic curves fitted to match the observed prevalence in each region of the park in 1991-2 and 1998, with an asymptote of 0.67 (Figure A6-1) (De Vos et al. 2001, Rodwell et al. 2001). These curves were then used as input to the lion demographic disease model to compute the probability of disease transmission to a lion from infectious buffalo; the rate that lions become exposed to bovine tuberculosis from buffalo in a given region at a given time was the product of the prevalence in buffalo in that region at that time and parameter  $B$ .

### ***Observed field data***

We used ABC-SMC to determine posterior distributions of the disease dynamics parameters in the Kruger lion system. The algorithm searched the space of all possible disease parameters ( $B$ ,  $L$ ,  $O$ ,  $E$ ,  $I$ ) by choosing a set of random parameter values, and then iteratively running the SimSimba-Disease model with those values, comparing model results against field data (Table 5-1) to find the parameter sets with highest likelihood, and then perturbing the parameter values to get new parameter sets for the next round.

The field data used to compare results from the ABC-SMC simulations (Table 5-1) consist firstly of a set bovine tuberculosis prevalence data (Keet et al. 2000) that were obtained by tuberculin testing of 125 “repeat-offender” lions in good condition. These lions have caused park management repeated problem, such as killing cattle or leaving the park, and so are brought to the park veterinarians for euthanizing. While not a perfect random sample, the data on these lions are the best that can be obtained, as tuberculin testing requires repeated handling of lions three days apart. We excluded prevalence data on sick and emaciated lions because they would have biased our data towards high prevalence rates. The tuberculin test for lions has a very high rate of detection in animals in good condition (Keet et al. 2010).

The second set of field data used are lion population surveys, conducted using call-up stations. The first couple were conducted in the 1970's and the most recent in 2005-2006 (Ferreira and Funston 2010). There was no detectable change in lion population size between the 2005-2006 surveys and those conducted in the 1970's (Ferreira and Funston 2010).

### ***ABC-SMC algorithm***

In Bayesian methods, a candidate posterior distribution  $f(\theta|x_0)$  of model parameters  $\theta$  is taken from a parameter space  $\Theta$  given the observed data,  $x_0$ . Through Bayes' theorem,  $f(\theta|x_0)$  is proportional to  $f(x_0|\theta) \pi(\theta)$ , where  $f(x_0|\theta)$  is the likelihood function and  $\pi(\theta)$  is the prior distribution of model parameters.  $f(\theta|x_0)$  is approximated by using Monte Carlo techniques to draw a large number of possible samples from  $f(x_0|\theta) \pi(\theta)$ .

ABC methods circumvent direct calculations of the likelihood function by repeatedly drawing a candidate parameter set,  $\theta^*$ , from the prior density and simulating data,  $x^*$ , with the likelihood function,  $f(x|\theta^*)$ . If the simulated data sufficiently matches the observed data  $x_0$ ,  $\theta^*$  is accepted and becomes part of the sampled posterior distribution (Sisson et al. 2007).

In ABC-SMC, each parameter set is termed a 'particle.' A population of particles  $\theta_1 \dots \theta_N$  is drawn from the parameter space  $\Theta$  according to the parameters' prior distribution. Data  $x^*$  are simulated for each particle, as in all ABC methods, and distance measure  $D$ , a measure of closeness between the simulated data,  $x^*$ , and the observed data,  $x_0$ , is calculated. The tolerance,  $\epsilon$ , is defined as the maximum value of  $D$  that will allow the particle's acceptance. The tolerance is reduced each iteration, improving the fit between the resulting distribution and the posterior distribution. The set of accepted particles is weighted and smoothed to form the prior distribution for the next iteration, whereupon a new set of particles is drawn,  $\epsilon$  is decreased, and the process is repeated until the desired tolerance is reached. This process explores complex parameter spaces

more quickly than previous advances in ABC algorithms in which particles are correlated with one another (Sisson et al. 2007).

We use a modified version of the original ABC-SMC algorithm that selects a fixed fraction of the best particles each round to determine the value of  $\varepsilon$  for that round. This modification speeds up posterior distribution convergence that might otherwise stall with poorly chosen *a priori*  $\varepsilon$  values (Drovandi and Pettitt 2011a).

Our algorithm runs as follows:

1. Our parameter space consists of the disease parameters  $\Theta = \{B, L, O, E, I\}$  and each particle is a point in that space ( $b \in B, l \in L, o \in O, e \in E, i \in I$ ). We assign flat priors for  $B, L, O$ , and  $E$  from uniform distributions on  $[0,1]$ . Observational data suggest that the Kruger lions die within 5 years of showing symptoms of bovine tuberculosis (Keet et al. 2009), so we restrict the prior of parameter  $I$  to be uniform on  $[0.2,1]$ , thereby setting a maximum for  $\frac{1}{I}$ , the average time to death, to 5 time steps, or 2.5 years. We draw 50,000 independent particles from  $\Theta$ .
2. We set the parameter values in SimSimba according to the particle's associated values and run the model from 1960 to 2006 (46 years, 92 time steps).
3. For each run of SimSimba, we sample 22, 39, and 64 lions in the north, central, and south regions respectively in the year 1999. We record the number of exposed or infectious lions in each sample as  $\hat{F}_{north}$ ,  $\hat{F}_{central}$ , and  $\hat{F}_{south}$ . We also record the simulated population size of the north, central, and south region in 1960 and in 2006, denoting the difference in population sizes as  $\hat{N}_{north}$ ,  $\hat{N}_{central}$ , and  $\hat{N}_{south}$  respectively. The outputted data from each run is therefore  $x^* = (\hat{F}_{north}, \hat{F}_{central}, \hat{F}_{south}, \hat{N}_{north}, \hat{N}_{central}, \hat{N}_{south})$ .
4. We compare observed data  $x_0 = (0, 18, 50, 0, 0, 0)$  (from Table 5-1) and  $x^*$  for each run, transforming both  $x_0$  and  $x^*$  with partial least squares regression so as to more equally consider all six components (Wegmann et al. 2009). We then calculate  $D$  as the Euclidean distance between the transformed  $x_0$  and the transformed  $x^*$ .

5. We sort the 50,000 particles by  $D$  and accept the 1,000 particles with the least difference between simulated outcomes and our observed data.
6. For each of the 1,000 accepted particles, we calculate weights  $w^{(i)}$ . In the first round, each particle is weighted equally. Subsequently,  $w_t^{(i)} = \frac{1}{\sum_j w_{t-1}^{(j)} K_t(\theta_{t-1}^{(j)}, \theta_t^{(i)})}$ , where  $t$  represents the current round and  $t-1$  the previous round and  $K_t$  is the kernel, or transition density function, for round  $t$ . This equation computes a current particle's weight based on the distribution of the previous round's particles and the probability that it derived from each of those particles. We then normalize the weights to sum to 1.
7. We generate 50,000 new particles from the weighted distribution of the 1,000 particles accepted from the prior round. For each new particle, we randomly choose an accepted particle and perturb it using the uniform perturbation kernel  $K_t = \alpha U(-1,1)$  with  $\alpha$  as one standard deviation of each parameter value. This perturbation smoothes and spreads the accepted distribution to explore nearby parameter space.
8. We return to Step 2 and repeat until the new distribution no longer departs significantly from the previous iteration.
9. We examine the marginal probabilities of the joint distribution on our five parameters.

We report results as means, medians, and 95% credible intervals of the posterior distributions. Credible intervals are analogous to the confidence intervals of frequentist statistics; there is a 95% certainty that the true value lies within a 95% credible interval (Edwards et al. 1963). SimSimba simulations were run at the University of Minnesota Supercomputing Institute. Transformation, weighting, and perturbation of parameter sets was performed in R on a laptop computer using packages *MASS*, *car*, and *pls* (Venables and Ripley 2002, Fox and Weisberg 2011, Mevik et al. 2011, R Core Team 2012).

We took the 1,000 parameter sets from the posterior distribution of the ABC-SMC algorithm and ran them in SimSimba from 1960 to 2060 (100 years, 200 time steps) to forecast the impact of bovine tuberculosis on disease prevalence and lion population

size. To determine equilibrium impact of the disease, we ran the simulations from 1960 to 2260 (300 years, 600 time steps).

To determine whether lions are maintenance or spillover hosts, we repeated the forecasting procedure while removing all disease from the buffalo from 2010 onwards by eliminating buffalo-to-lion transmission and only allowing the disease to be transmitted from lion to lion.

To see how lion-to-lion transmission affects disease spread and population size, we took the 1,000 parameter sets from the posterior distribution and set  $L$  to zero. We compared the results for simulations with  $L>0$  and  $L=0$  for ten replicates for each parameter set, using t-tests blocked by parameter set number.

## Results

Parameter distributions converged after five rounds of ABC-SMC (Figure A6-2), with observed data values falling into the middle of model summary statistics distributions (Figure A6-3). Mirroring buffalo prevalence patterns, lion prevalence showed a logistic increase in each region of the park (Figure 5-3), while lion population size decreased slightly (Figure 5-4).

Disease dynamics in this system were largely driven by two parameters. The transmission rate between an infected buffalo and a susceptible lion,  $B$ , drove the disease prevalence in lions (Table A6-1, Figures A6-4, A6-5). ABC-SMC converged on a stable posterior distribution for  $B$  with mean and median 0.54 and 95% credible interval of [0.24, 0.82] (Figure 5-5). Assuming a contact rate of one buffalo per six-month period and a stably infected buffalo population, this translates to an average annual exposure rate of 0.59 per individual lion from buffalo consumption (95% CI [0.30, 0.80]).

The rate that lions transition from exposed to infectious,  $E$ , drove changes in lion population size (Table A6-2, Figures A6-6, A6-7). ABC-SMC converged on a stable distribution for  $E$  with mean 0.0074 (median 0.0065, 95% CI [0.0004, 0.0192], Figure 5-5). This is equivalent to an annual per lion rate of transition from exposed to infectious state of 0.015 (95% CI [0.0008, 0.0380]); the mean fraction of exposed individuals who



transitioned to infectious in their lifetimes was 0.057 (95% CI [0.003, 0.151], Figure A6-8). Parameter *I*, the rate at which infectious lions die of bovine tuberculosis, averaged 0.65 (95% CI [0.26, 0.97]). This is equivalent to an average annual disease mortality rate of 88% (95% CI [45%, 100%]). Parameters *L* and *O*, which govern the transmission of bovine tuberculosis from infectious lion to susceptible lion, retained fairly flat posterior distributions (Figure 5-5).

The forecasting simulations suggest that the lion population is not in danger of crashing from the introduction of bovine tuberculosis alone (Figure 5-4). Results suggest a 6% decline over the next fifty years due to increased mortality from bovine tuberculosis (mean, 95% CI [-0.07, 0.21]). The long-term forecast indicates that the lion population will reach a new long-term carrying-capacity at this slightly depressed level.

Mirroring the rise of bovine tuberculosis in buffalo, disease prevalence in lions increased logistically in all three regions. Most lions were exposed to the disease by the mid-2020's, and upwards of 80% of all lions were exposed by 2060 (Figure 5-3, mean 0.83, 95% CI [0.72, 0.91]).

When we simulated a disease-eradication program in buffalo, there was an immediate steep drop in lion prevalence (Figure 5-6), and the disease was eradicated from lions within twenty years. The lion population size declined slightly at first as the disease continued to progress in previously exposed individuals, but then recovered to pre-disease levels (Figure 5-6).

When infectious lions were allowed to transmit the disease to susceptible lions, bovine tuberculosis prevalence was slightly higher than when transmission was strictly from buffalo to lion (south mean difference in 1999 = 0.0389, 95% CI [0.0377,0.0401],  $p < 0.0001$ ) and the total lion population size showed a minor decline (mean difference in 2006 = 5.66 individuals, 95% CI [4.00,7.32],  $p < 0.0001$ ).

## **Discussion**

Using ABC-SMC, we were able to make important inferences about bovine tuberculosis dynamics in Kruger's lion population. In particular, it was necessary to

examine the complete space of logical parameter sets because very little was known about the disease parameters *a priori*.

The workshop model (Keet et al. 2009) that predicted an unobserved crash in the lion population appears to have over-estimated the rate of transition from the exposed, latent state to the infectious state. This overestimation resulted from personal observations of dying lions and one small-scale study, suggesting that expert opinion must be used cautiously when combined with complex models.

By contrast, we assumed nothing about the rate of transition from the exposed to the infectious state ( $E$ ) and instead explored all possible rates. Because only small values of  $E$  resulted in dynamics that matched empirical observations, we can reasonably conclude that  $E$  must be small. In fact, while transition probabilities of tuberculosis are unknown for wildlife, the observed rate in our model is comparable to the rate of 1 out of 10 in humans (Figure A6-8) (Bates 1984).

Our model also suggests that bovine tuberculosis is primarily transmitted from buffalo to lions, whereas transmission from one lion to another is relatively rare. This result is consistent with veterinary findings that infected lions only occasionally show pulmonary lesions, implying infrequent aerosol transmission (Keet et al. 2000). Prior studies have been inconclusive as to whether lions are maintenance or spillover hosts of bovine tuberculosis (Michel et al. 2006), but our model suggests that lions constitute a spillover host.

We forecast that most lions will be exposed to bovine tuberculosis over the next few decades. However, the disease will remain latent in the majority of lions because of the low transition rate from exposed to infectious. The number of sick lions will increase in the central and northern regions, where the disease has not yet reached equilibrium. However, even at equilibrium, we predict that only about a dozen lions will die from bovine tuberculosis across the park each year (Figure 5-4). In total, the lion population size will only decrease by about 6% over the next 50 years, but such a small effect will be difficult to detect from short-term monitoring (Jolles et al. 2005, Ferreira and Funston 2010).

In addition to revealing disease patterns, the results from our model can be used to infer the primary driving variables of a system. In the case of the Kruger lions, we found that parameter  $E$ , the rate of transition from exposed to infectious, was the primary driver of lion population size. With this knowledge, we further infer that changes to the system that increase  $E$ , such as co-infection with new diseases, might cause greater bovine tuberculosis mortality. Likewise, parameter  $B$ , the rate of disease transmission from buffalo to lions was the primary driver of disease prevalence in lions, and so changes in the system that increase  $B$ , such as drought, may increase the incidence of bovine tuberculosis in the lion population.

Information on driving variables also allows for practical management recommendations. Because our model shows that buffalo are the primary source of bovine tuberculosis in lions, recommendations for control of the disease in lions should focus on reducing buffalo-to-lion transmission. In the Kruger system, culling and quarantining buffalo are not feasible, and so the development of a vaccine for buffalo might be one recommendation. However, given the limited impact of the disease on lions, the expense of vaccine development may not be justified.

The strength of the ABC-SMC modeling approach is that it uses available data on population structure and dynamics together with observed spatiotemporal patterns of disease prevalence to constrain the set of all possible combinations of unmeasured variables. In the case of the Kruger lions, we knew little about the disease itself, but were able to infer disease patterns based on lion demographics and social structure and just one set of observations of disease prevalence.

However, we acknowledge that our results rely on the implicit assumptions made in creating the demographic simulation model and the disease model. The demographic simulation model uses a number of parameters (Table A5-1) that describe lion demography and social behavior, and both these parameters and the structure of the model affect our results. It is likely, however, that the model and parameter values provide a reasonable interpretation of Kruger lion dynamics. The model has been shown to realistically mimic Serengeti lion population dynamics, age structure, and sex ratios

observed over decades (Whitman et al. 2004, 2007), was modified to incorporate as much Kruger data as possible, and was validated by comparison with static Kruger demographic data.

Our disease model makes several assumptions. We assume that once lions become infectious, they subsequently die without any chance of developing immunity. We also assume that the timing of infectiousness of bovine tuberculosis in lions corresponds with the timing of their increased mortality from the disease. These assumptions are based on the epidemiology and pathogenesis of tuberculosis in humans and other animals (Bates 1984). If bovine tuberculosis were discovered to manifest significantly differently in lions, these assumptions would need to be reconsidered. We also assumed that there was no feedback between lions and buffalos; in particular, the prevalence of disease in buffalo was not affected by lion population size in our model. Buffalo population size in Kruger is believed to be controlled both by predation and availability of food (Funston and Mills 2006), so a future decrease in lion population size could cause an increase in buffalo population. However, it is unclear whether a change in buffalo population size would have a measurable effect on disease prevalence in buffalo.

Despite these assumptions, we suggest that our demographic simulation and disease model reasonably reflect the dynamics of bovine tuberculosis in lions in Kruger National Park. Our model yields reasonable results that corroborate veterinary findings, that are in accord with similar dynamics in humans, and that predict results that are in agreement with subsequent system observations.

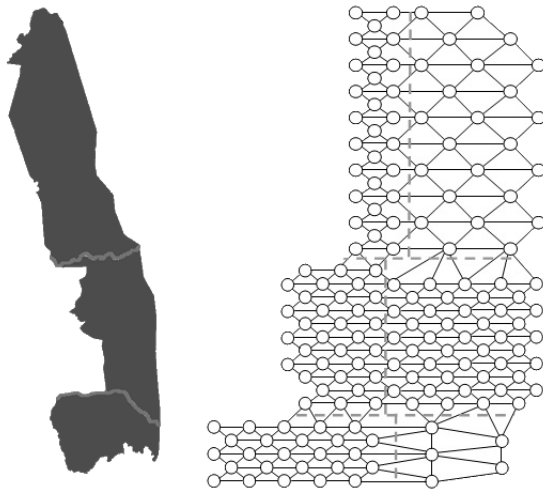
Emerging infectious diseases are an increasing challenge for wildlife management (Daszak 2000) and for international conservation policy. However, wildlife disease dynamics are often difficult to study due to a paucity of data, and the expense and logistical difficulties associated with obtaining them. Classical analytic disease models are of limited use in complex systems where disease dynamics depend on host social structure, behavior, and heterogeneous contact rates. Complex simulation and network models often require large amounts of empirical data and sometimes only examine a fraction of logical parameter space. ABC-SMC models are a next step in wildlife disease

modeling, making it possible to estimate essential disease dynamics in complex systems from relatively limited field data.

## Tables and Figures

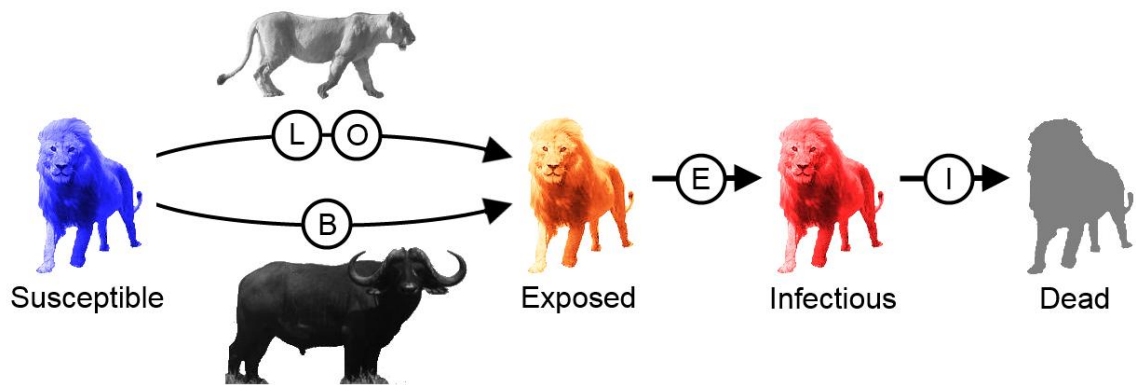
**Table 5-1: Observational data on bovine tuberculosis prevalence and lion population size**

<b>Data</b>	<b>Value</b>	<b>Method</b>	<b>Reference</b>
BTB prevalence in lions in South region (1998-2000)	78.1% (50/64)	Single cervical intradermal testing of repeat offender lions in good condition	(Keet et al. 2000, 2010)
BTB prevalence in lions in Central region (1998-2000)	46.2% (18/39)		
BTB prevalence in lions in North region (1998-2000)	0.0% (0/22)		
Change in lion population size between 1976-1978 and 2005-2006	No detectable change	Full-park survey using call-up stations	(Ferreira and Funston 2010)



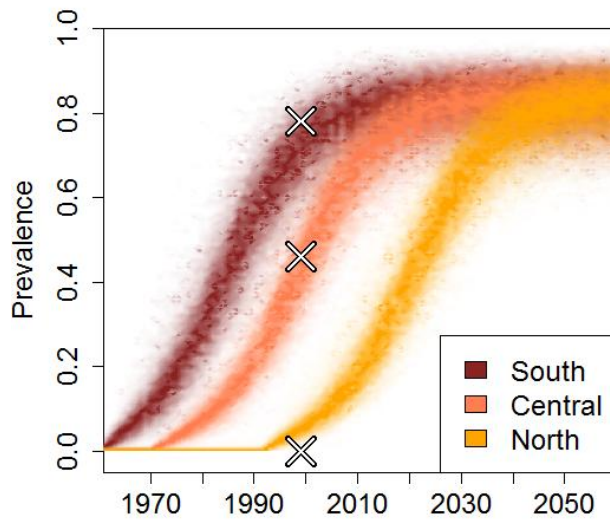
**Figure 5-1: Kruger National Park and model schematic of Kruger National Park**

The geographic shape of Kruger National Park (left), trisected by the Olifants and Sabie Rivers (light lines). Model schematic of Kruger National Park (right); each circle represents a lion territory and potential pride, while lines indicate physical connectance of those territories.



**Figure 5-2: Disease dynamics model parameters**

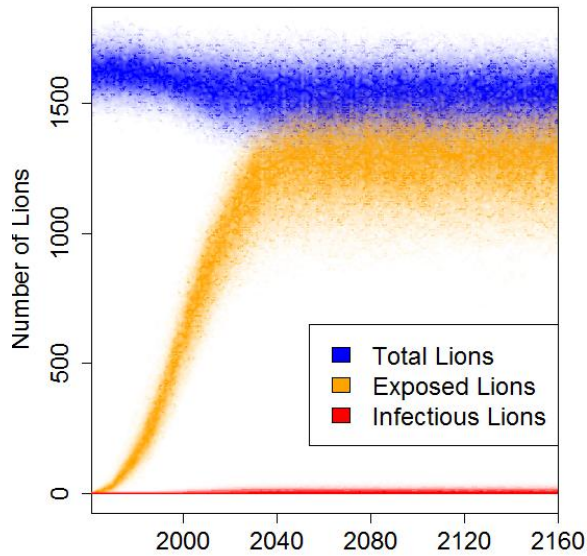
$L$  is the probability of transmission of bovine tuberculosis to a lion from another lion, while  $O$  indicates the rate of contact between lions not in the same pride.  $B$  describes the probability of bovine tuberculosis transmission from the buffalo population.  $E$  is the transition rate of an exposed lion to the infectious state.  $I$  is the mortality rate of infectious lions.



**Figure 5-3: Modeled prevalence of bovine tuberculosis in the lion population**

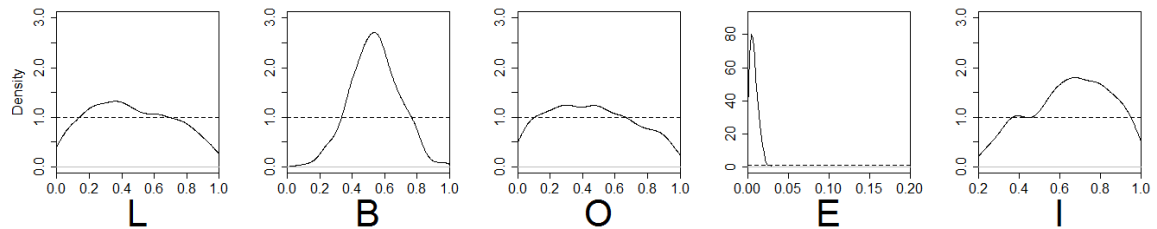
Modeled prevalence of bovine tuberculosis in the lion population for the south (maroon), central (peach), and north (yellow) regions show that prevalence across the park asymptotes at around 80% by 2050. Prevalence values are shaded by likelihood density, with highest likelihood shaded dark and lowest likelihood light. White *X*'s indicate observed prevalence in the south, central, and north regions respectively (Keet et al. 2010).





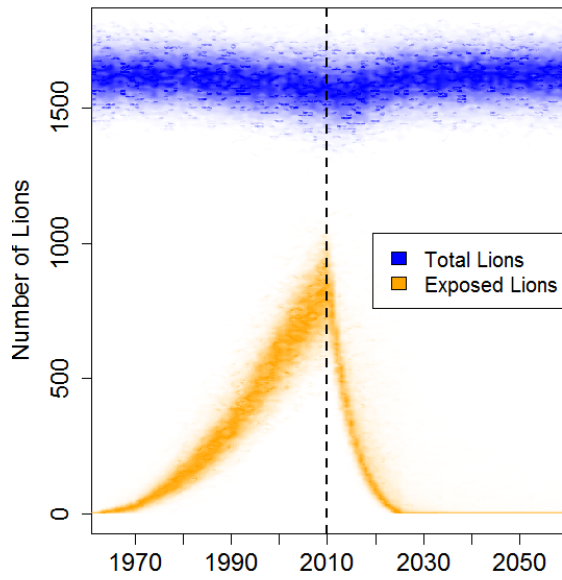
**Figure 5-4: Modeled number of total adult and subadult lions in the population, those that are exposed, and those that are infectious**

Values are shaded by likelihood density, with highest likelihood shaded dark and lowest likelihood light.



**Figure 5-5: Posterior density distributions for the five model disease parameters**

Horizontal dashed lines indicate initial parameter distributions (“priors”). Parameters  $B$  and  $E$  show a peaked distribution that deviates substantially from their priors, indicating that these parameters are most important for model dynamics, whereas parameters  $L$ ,  $O$ , and  $I$  do not deviate as much from their priors.



**Figure 5-6: Modeled number of total adult and subadult lions in the population, and those that are exposed**

The prevalence of bovine tuberculosis in buffalo is set to 0 in the year 2010.

Subsequently, the disease clears from lions and lion population rebounds. Values are shaded by likelihood density, with highest likelihood shaded dark and lowest likelihood light.

## Bibliography

- Aberle, E. D., J. C. Forrest, D. E. Gerrard, and E. W. Mills. 2001. Principles of meat science, 4th edition. Kendall Hunt, Dubuque, Iowa.
- Abidoye, B. O., H. Bulut, J. D. Lawrence, B. Mennecke, and A. M. Townsend. 2011. US Consumers' Valuation of Quality Attributes in Beef Products. *Journal of Agricultural and Applied Economics* 43:1.
- Alexandratos, N., and J. Bruinsma. 2012. World agriculture towards 2030/2050: the 2012 revision. ESA Working paper Rome, FAO.
- Altieri, M. A., and W. H. Whitcomb. 1980. Weed manipulation for insect pest management in corn. *Environmental Management* 4:483–489.
- Anderson, R. M., and R. M. May. 1991. Infectious diseases of humans: dynamics and control. Oxford University Press, Oxford, UK.
- Andow, D. A. 1991. Vegetational diversity and arthropod population response. *Annual review of entomology* 36:561–586.
- Andow, D. A., and S. J. Risch. 1985. Predation in diversified agroecosystems: relations between a coccinellid predator *Coleomegilla maculata* and its food. *Journal of Applied Ecology* 22:357–372.
- Armstrong, R. A. 1976. Fugitive species: experiments with fungi and some theoretical considerations. *Ecology* 57:953–963.
- Atlıhan, R., M. B. Kaydan, A. Yarımbatman, and H. Okut. 2010. Functional response of the coccinellid predator *Adalia fasciatopunctata revelierei* to walnut aphid (*Callaphis juglandis*). *Phytoparasitica* 38:23–29.
- Baldwin, C., N. Wilberforce, and A. Kapur. 2010. Restaurant and food service life cycle assessment and development of a sustainability standard. *The International Journal of Life Cycle Assessment* 16:40–49.
- Barnes, R., C. J. Nelson, K. Moore, and M. Collins (Eds.). 2007. Forages, Volume 2: The Science of Grassland Agriculture, 6th edition. Wiley-Blackwell.
- Bates, D., M. Maechler, and B. Bolker. 2012. lme4: Linear mixed-effects models using S4 classes.
- Bates, J. H. 1984. Transmission, pathogenesis, pathology and clinical manifestations of tuberculosis. Pages 991–1005 *The Mycobacteria: a Sourcebook*. Marcel Dekker Inc., New York.
- Beauchemin, K. A., H. Henry Janzen, S. M. Little, T. A. McAllister, and S. M. McGinn. 2010. Life cycle assessment of greenhouse gas emissions from beef production in western Canada: A case study. *Agricultural Systems* 103:371–379.
- Beaumont, M. A. 2010. Approximate Bayesian computation in evolution and ecology. *Annual Review of Ecology, Evolution, and Systematics* 41:379–406.
- Beaumont, M. A., J.-M. Cornuet, J.-M. Marin, and C. P. Robert. 2009. Adaptive approximate Bayesian computation. *Biometrika* 96:983–990.
- Beaumont, M. A., R. Nielsen, C. Robert, J. Hey, O. Gaggiotti, L. Knowles, A. Estoup, M. Panchal, J. Corander, M. Hickerson, S. A. Sisson, N. Fagundes, L. Chikhi, P. Beerli, R. Vitalis, J.-M. Cornuet, J. Huelsenbeck, M. Foll, Z. Yang, F. Rousset, D.

- Balding, and L. Excoffier. 2010. In defence of model-based inference in phylogeography. *Molecular Ecology* 19:436–446.
- Beckett, J. L., and J. W. Oltjen. 1993. Estimation of the water requirement for beef production in the United States. *Journal of Animal Science* 71:818–826.
- Bengis, R. G., N. P. Kriek, D. F. Keet, J. P. Raath, V. de Vos, and H. F. Huchzermeyer. 1996. An outbreak of bovine tuberculosis in a free-living African buffalo (*Syncerus caffer sparrman*) population in the Kruger National Park: a preliminary report. *The Onderstepoort Journal of Veterinary Research* 63:15–18.
- Bianchi, F. J. J. ., C. J. . Booij, and T. Tscharntke. 2006. Sustainable pest regulation in agricultural landscapes: a review on landscape composition, biodiversity and natural pest control. *Proceedings of the Royal Society B: Biological Sciences* 273:1715–1727.
- Bommarco, R., and J. E. Banks. 2003. Scale as Modifier in Vegetation Diversity Experiments: Effects on Herbivores and Predators. *Oikos* 102:440–448.
- Bracmort, K., J. L. Ramseur, J. E. McCarthy, P. Folger, and D. J. Marples. 2009. Methane Capture: Options for Greenhouse Gas Emission Reduction. U.S. Congressional Research Service.
- Brewer, M. J., and T. Noma. 2010. Habitat Affinity of Resident Natural Enemies of the Invasive *Aphis glycines* (Hemiptera: Aphididae), on Soybean, with Comments on Biological Control. *Journal of Economic Entomology* 103:583–596.
- Brown, M. W. 2011. Importance of early arrival of adult *Harmonia axyridis* for control of *Aphis spiraecola* on apple. *BioControl* 56:65–69.
- Buratti, C., E. Moretti, and F. Fantozzi. 2010. Assessing the GHG emissions of rapeseed and soybean biodiesel in compliance to the EU renewable energy directive methodology for biofuels. 18th European Biomass Conference, Lyon, France.
- Buzby, J. C., H. F. Wells, B. Axtman, and J. Mickey. 2009. Supermarket loss estimates for fresh fruit, vegetables, meat, poultry, and seafood and their use in the ERS loss-adjusted food availability data. Economic Research Service, United States Department of Agriculture.
- Capper, J. L. 2011. The environmental impact of beef production in the United States: 1977 compared with 2007. *Journal of Animal Science* 89:4249–4261.
- CDC. 2008. National Health and Nutrition Examination Survey. Centers for Disease Control and Prevention. <http://www.cdc.gov/nchs/nhanes.htm>.
- Cederberg, C., U. Sonesson, M. Henriksson, V. Sund, and J. Davis. 2009. Greenhouse gas emissions from Swedish production of meat, milk and eggs 1990 and 2005. SIK The Swedish Institute for Food and Biotechnology. ISBN 978-91-7290-284-8.
- Cederberg, C., and M. Stadig. 2003. System expansion and allocation in life cycle assessment of milk and beef production. *The International Journal of Life Cycle Assessment* 8:350–356.
- Chaplin-Kramer, R., M. E. O'Rourke, E. J. Blitzer, and C. Kremen. 2011. A meta-analysis of crop pest and natural enemy response to landscape complexity. *Ecology Letters* 14:922–932.

- Clark, C. J., J. R. Poulsen, D. J. Levey, and C. W. Osenberg. 2007. Are Plant Populations Seed Limited? A Critique and Meta-Analysis of Seed Addition Experiments. *The American Naturalist* 170:128–142.
- Coleman, S., and M. Williams. 2006. Bermudagrass yield and quality through the grazing season. Natural Resources Conservation Service, United States Department of Agriculture.
- Collins, F. L., and S. J. Johnson. 1985. Reproductive response of caged adult velvetbean caterpillar and soybean looper to the presence of weeds. *Agriculture, Ecosystems & Environment* 14:139–149.
- Conant, R. T., K. Paustian, and E. T. Elliott. 2001. Grassland management and conversion into grassland: effects on soil carbon. *Ecological Applications* 11:343–355.
- Costamagna, A. C., D. A. Landis, and M. J. Brewer. 2008. The role of natural enemy guilds in *Aphis glycines* suppression. *Biological Control* 45:368–379.
- Craft, M. E., E. Volz, C. Packer, and L. A. Meyers. 2009. Distinguishing epidemic waves from disease spillover in a wildlife population. *Proceedings of the Royal Society B: Biological Sciences* 276:1777–1785.
- Csilléry, K., M. G. Blum, O. E. Gaggiotti, and O. François. 2010. Approximate Bayesian computation (ABC) in practice. *Trends in Ecology & Evolution* 25:410–418.
- Daszak, P. 2000. Emerging infectious diseases of wildlife: threats to biodiversity and human health. *Science* 287:443–449.
- Delgado, C., L. Barruetabeña, and O. Salas. 2007. Assessment of the environmental advantages and drawbacks of existing and emerging polymers recovery processes. Institute for Prospective Technological Studies, Joint Research Centre, European Commission.
- Derner, J. D., T. W. Boutton, and D. D. Briske. 2006. Grazing and Ecosystem Carbon Storage in the North American Great Plains. *Plant and Soil* 280:77–90.
- Derner, J. D., and G. E. Schuman. 2007. Carbon sequestration and rangelands: a synthesis of land management and precipitation effects. *Journal of Soil and Water Conservation* 62:77–85.
- Desneux, N., R. J. O’Neil, and H. J. S. Yoo. 2006. Suppression of Population Growth of the Soybean Aphid, *Aphis glycines* Matsumura, by Predators: The Identification of a Key Predator and the Effects of Prey Dispersion, Predator Abundance, and Temperature. *Environmental Entomology* 35:1342–1349.
- Drovandi, C. C., and A. N. Pettitt. 2011a. Using approximate Bayesian computation to estimate transmission rates of nosocomial pathogens. *Statistical Communications in Infectious Diseases* 3:1–26.
- Drovandi, C. C., and A. N. Pettitt. 2011b. Estimation of parameters for macroparasite population evolution using approximate Bayesian computation. *Biometrics* 67:225–233.
- Durrett, R., and S. Levin. 1998. Spatial Aspects of Interspecific Competition. *Theoretical Population Biology* 53:30–43.
- Economic Research Service (ERS). 2010. World Agricultural Supply and Demand Estimates (WASDE). U.S. Department of Agriculture, Washington, D.C.

- Edwards, W., H. Lindman, and L. J. Savage. 1963. Bayesian statistical inference for psychological research. *Psychological Review* 70:193–242.
- Edwards-Jones, G., K. Plassmann, and I. M. Harris. 2009. Carbon footprinting of lamb and beef production systems: insights from an empirical analysis of farms in Wales, UK. *The Journal of Agricultural Science* 147:707–719.
- Energy Information Administration. 2005. Residential Energy Consumption Survey (RECS). U.S. Department of Energy, Washington, D.C.
- Energy Information Administration. 2009. Residential Energy Consumption Survey (RECS). U.S. Department of Energy, Washington, D.C.
- Epplin, F. M., I. Hossain, and E. G. Krenzer Jr. 2000. Winter wheat fall–winter forage yield and grain yield response to planting date in a dual-purpose system. *Agricultural Systems* 63:161–173.
- Evans, E. W. 2003. Searching and reproductive behaviour of female aphidophagous ladybirds (Coleoptera: Coccinellidae): a review. *European Journal of Entomology* 100:1–10.
- Fantinou, A. A., D. C. Perdakis, P. D. Labropoulos, and D. A. Maselou. 2009. Preference and consumption of *Macrolophus pygmaeus* preying on mixed instar assemblages of *Myzus persicae*. *Biological Control* 51:76–80.
- FAO. 2009. Food supply. Food and Agriculture Organization of the United Nations. <http://faostat.fao.org>.
- Ferreira, S. M., and P. J. Funston. 2010. Estimating lion population variables: prey and disease effects in Kruger National Park, South Africa. *Wildlife Research* 37:194–206.
- FMI Research. 2009. U.S. grocery shopper trends. Food Marketing Institute, Arlington, Virginia.
- Foley, J. A., N. Ramankutty, K. A. Brauman, E. S. Cassidy, J. S. Gerber, M. Johnston, N. D. Mueller, C. O’Connell, D. K. Ray, P. C. West, C. Balzer, E. M. Bennett, S. R. Carpenter, J. Hill, C. Monfreda, S. Polasky, J. Rockström, J. Sheehan, S. Siebert, D. Tilman, and D. P. M. Zaks. 2011. Solutions for a cultivated planet. *Nature* 478:337–342.
- Fox, J., and S. Weisberg. 2011. *An R Companion to Applied Regression*, 2nd edition. Sage Publications, Thousand Oaks, CA.
- Fox, T. B., D. A. Landis, F. F. Cardoso, and C. D. Difonzo. 2004. Predators Suppress *Aphis glycines* Matsumura Population Growth in Soybean. *Environmental Entomology* 33:608–618.
- Frewin, A. J., Y. Xue, J. A. Welsman, B. A. Broadbent, A. W. Schaafsma, and R. H. Hallett. 2010. Development and Parasitism by *Aphelinus certus* (Hymenoptera: Aphelinidae), a Parasitoid of *Aphis glycines* (Hemiptera: Aphididae). *Environmental Entomology* 39:1570–1578.
- Funston, P. J., and M. G. L. Mills. 2006. The influence of lion predation on the population dynamics of common large ungulates in the Kruger National Park. *South African Journal of Wildlife Research* 36:9–22.

- Funston, P. J., M. G. Mills, P. R. Richardson, and A. S. Jaarsveld. 2003. Reduced dispersal and opportunistic territory acquisition in male lions (*Panthera leo*). *Journal of Zoology* 259:131–142.
- Gardiner, M. M., D. A. Landis, C. Gratton, C. D. DiFonzo, M. O’Neal, J. M. Chacon, M. T. Wayo, N. P. Schmidt, E. E. Mueller, and G. E. Heimpel. 2009. Landscape diversity enhances biological control of an introduced crop pest in the north-central USA. *Ecological Applications* 19:143–154.
- Garnett, T. 2009. Livestock-related greenhouse gas emissions: impacts and options for policy makers. *Environmental Science & Policy* 12:491–503.
- Gavish-Regev, E., R. Rotkopf, Y. Lubin, and M. Coll. 2008. Consumption of aphids by spiders and the effect of additional prey: evidence from microcosm experiments. *BioControl* 54:341–350.
- Grainger, C., and K. A. Beauchemin. 2011. Can enteric methane emissions from ruminants be lowered without lowering their production? *Animal Feed Science and Technology* 166-167:308–320.
- Grubb, P. J. 1977. The Maintenance of Species-Richness in Plant Communities: The Importance of the Regeneration Niche. *Biological Reviews* 52:107–145.
- Haddad, N. M., G. M. Crutsinger, K. Gross, J. Haarstad, J. M. H. Knops, and D. Tilman. 2009. Plant species loss decreases arthropod diversity and shifts trophic structure. *Ecology Letters* 12:1029–1039.
- Hamede, R. K., J. Bashford, H. McCallum, and M. Jones. 2009. Contact networks in a wild Tasmanian devil (*Sarcophilus harrisii*) population: using social network analysis to reveal seasonal variability in social behaviour and its implications for transmission of devil facial tumour disease. *Ecology Letters* 12:1147–1157.
- Hamilton, G., M. Stoneking, and L. Excoffier. 2005. Molecular analysis reveals tighter social regulation of immigration in patrilocal populations than in matrilocal populations. *Proceedings of the National Academy of Sciences of the United States of America* 102:7476–7480.
- Hart, C. R., and B. B. Carpenter. 2005. Stocking rate and grazing management. College Station, TX, USA Texas Agricultural Extension Service E-64.
- Hastings, A. 1980. Disturbance, coexistence, history, and competition for space. *Theoretical Population Biology* 18:363–373.
- Heimpel, G. E., L. E. Frelich, D. A. Landis, K. R. Hopper, K. A. Hoelmer, Z. Sezen, M. K. Asplen, and K. Wu. 2010. European buckthorn and Asian soybean aphid as components of an extensive invasional meltdown in North America. *Biological Invasions* 12:2913–2931.
- Hight, S. C., R. D. Eikenbary, R. J. Miller, and K. J. Starks. 1972. The Greenbug and *Lysiphlebus testaceipes*. *Environmental Entomology* 1:205–209.
- Hill, J., E. Nelson, D. Tilman, S. Polasky, and D. Tiffany. 2006. Environmental, economic, and energetic costs and benefits of biodiesel and ethanol biofuels. *Proceedings of the National Academy of Sciences* 103:11206–11210.
- Hodgson, E. 2012. Management Recommendations for Soybean Aphid (Hemiptera: Aphididae) in the United States. *Journal of Integrated Pest Management* 3:1–10.

- Hoekstra, A. Y., and A. K. Chapagain. 2006. Water footprints of nations: Water use by people as a function of their consumption pattern. *Water Resources Management* 21:35–48.
- Hofmeyr, M. S., P. Buss, L.-M. De Klerk-Lorist, and R. G. Bengis. 2006. Bovine tuberculosis in buffalo and current projects in the Kruger National Park. Pages 1771–1773 *Proceedings of the North American Veterinary Conference*. Orlando, FL.
- Hopper, J. V., E. H. Nelson, K. M. Daane, and N. J. Mills. 2011. Growth, development and consumption by four syrphid species associated with the lettuce aphid, *Nasonovia ribisnigri*, in California. *Biological Control* 58:271–276.
- Hothorn, T., F. Bretz, and P. Westfall. 2008. Simultaneous inference in general parametric models. *Biometrical Journal* 50:346–363.
- IPCC. 2006. Guidelines for National Greenhouse Gas Inventories, prepared by the National Greenhouse Gas Inventories Programme. (S. Eggleston, L. Buendia, K. Miwa, T. Ngara, and K. Tanabe, Eds.). Institute for Global Environmental Strategies, IGES, Japan.
- Jolles, A. E., D. V. Cooper, and S. A. Levin. 2005. Hidden effects of chronic tuberculosis in African buffalo. *Ecology* 86:2358–2364.
- Keeling, M. J., and P. Rohani. 2007. *Modeling infectious diseases in humans and animals*. Princeton University Press, Princeton, NJ.
- Keet, D. F., H. Davies-Mostert, R. G. Bengis, P. Funston, P. Buss, M. Hofmeyr, S. Ferreira, E. Lane, P. Miller, and B. G. Daly. 2009. Disease risk assessment workshop report: African lion (*Panthera leo*) Bovine Tuberculosis. Conservatoin Breeding Specialist Group (CBSG SSC / IUCN) / CBSG Southern Africa. Endangered Wildlife Trust, Skukuza, South Africa.
- Keet, D. F., N. P. J. Kriek, M. Penrith, and A. Michel. 1997. Tuberculosis in lions and cheetahs. Pages 151–156 *Proceedings of a Symposium on Lions and Leopards as Game Ranch Animals*. Onderstepoort, South Africa.
- Keet, D. F., A. L. Michel, R. G. Bengis, P. Becker, D. S. van Dyk, M. van Vuuren, V. P. M. G. Rutten, and B. L. Penzhorn. 2010. Intradermal tuberculin testing of wild African lions (*Panthera leo*) naturally exposed to infection with *Mycobacterium bovis*. *Veterinary Microbiology* 144:384–391.
- Keet, D. F., A. Michel, and D. G. A. Meltzer. 2000. Tuberculosis in free-ranging lions (*Panthera leo*) in the Kruger National Park. Pages 232–241 *Proceedings of the South African Veterinary Association Biennial Congress*. Durban, Kwazulu-Natal.
- Kelling, K. A. 2000. *Alfalfa fertilization*. Cooperative Extension Publishing, University of Wisconsin-Madison.
- Kenny, J. F., J. F. Barber, K. S. Linsey, J. K. Lovelace, and M. A. Maupin. 2009. Estimated use of water in the United States in 2005. U.S. Geological Survey, Reston, Va.
- Kneitel, J. M., and J. M. Chase. 2004. Trade-offs in community ecology: linking spatial scales and species coexistence. *Ecology Letters* 7:69–80.



- Koch, R. L., P. M. Porter, M. M. Harbur, M. D. Abrahamson, K. A. G. Wyckhuys, D. W. Ragsdale, K. Buckman, Z. Sezen, and G. E. Heimpel. 2012. Response of Soybean Insects to an Autumn-Seeded Rye Cover Crop. *Environmental Entomology* 41:750–760.
- Koelsch, R. K., K. Glewen, T. Trehwitt, and D. T. Walters. 2000. Exporting Feedlot Manure to Off-Farm Users. *Beef Cattle Reports*, University of Nebraska - Lincoln.
- Landis, D. A., S. D. Wratten, and G. M. Gurr. 2000. Habitat management to conserve natural enemies of arthropod pests in agriculture. *Annual review of entomology* 45:175–201.
- Langellotto, G. A., and R. F. Denno. 2004. Responses of Invertebrate Natural Enemies to Complex-Structured Habitats: A Meta-Analytical Synthesis. *Oecologia* 139:1–10.
- Latham, D. R., and N. J. Mills. 2010. Quantifying aphid predation: the mealy plum aphid *Hyalopterus pruni* in California as a case study. *Journal of Applied Ecology* 47:200–208.
- Launchbaugh, J. L., and C. E. Owensby. 1978. Kansas rangelands. *Kansas Agricultural Experiment Station Bulletin* 622:56.
- Leibold, M. A., M. Holyoak, N. Mouquet, P. Amarasekare, J. M. Chase, M. F. Hoopes, R. D. Holt, J. B. Shurin, R. Law, D. Tilman, M. Loreau, and A. Gonzalez. 2004. The metacommunity concept: a framework for multi-scale community ecology. *Ecology Letters* 7:601–613.
- Lesschen, J. P., M. van den Berg, H. J. Westhoek, H. P. Witzke, and O. Oenema. 2011. Greenhouse gas emission profiles of European livestock sectors. *Animal Feed Science and Technology* 166–167:16–28.
- Letourneau, D. K., and S. G. Bothwell. 2008. Comparison of Organic and Conventional Farms: Challenging Ecologists to Make Biodiversity Functional. *Frontiers in Ecology and the Environment* 6:430–438.
- Levin, S. A. 1992. The Problem of Pattern and Scale in Ecology: The Robert H. MacArthur Award Lecture. *Ecology* 73:1943–1967.
- Levins, R., and D. Culver. 1971. Regional coexistence of species and competition between rare species. *Proceedings of the National Academy of Sciences* 68:1246–1248.
- Lowe, M., and G. Gereffi. 2009. A value chain analysis of the US beef and dairy industries. Center on Globalization, Governance & Competitiveness, Duke University, Durham, North Carolina.
- Luciani, F., S. A. Sisson, H. Jiang, A. R. Francis, and M. M. Tanaka. 2009. The epidemiological fitness cost of drug resistance in *Mycobacterium tuberculosis*. *Proceedings of the National Academy of Sciences* 106:14711–14715.
- Lundgren, J. G., L. S. Hesler, S. A. Clay, and S. F. Fausti. 2013. Insect communities in soybeans of eastern South Dakota: The effects of vegetation management and pesticides on soybean aphids, bean leaf beetles, and their natural enemies. *Crop Protection* 43:104–118.
- Lundgren, J. G., K. A. G. Wyckhuys, and N. Desneux. 2009. Population responses by *Orius insidiosus* to vegetational diversity. *BioControl* 54:135–142.

- Manitoba Agriculture, Food and Rural Initiatives. 2006, March. Fact Sheet: Fertilizing alfalfa forage.
- McAlpine, C. A., A. Etter, P. M. Fearnside, L. Seabrook, and W. F. Laurance. 2009. Increasing world consumption of beef as a driver of regional and global change: A call for policy action based on evidence from Queensland (Australia), Colombia and Brazil. *Global Environmental Change* 19:21–33.
- McCornack, B. P., D. W. Ragsdale, and R. C. Venette. 2004. Demography of Soybean Aphid (Homoptera: Aphididae) at Summer Temperatures. *Journal of Economic Entomology* 97:854–861.
- McKinley, T., A. R. Cook, and R. Deardon. 2009. Inference in epidemic models without likelihoods. *The International Journal of Biostatistics* 5:1.
- McMichael, A. J., J. W. Powles, C. D. Butler, and R. Uauy. 2007. Food, livestock production, energy, climate change, and health. *The Lancet* 370:1253–1263.
- Mellon, M. G., C. Benbrook, and K. L. Benbrook. 2001. Hogging it: estimates of antimicrobial abuse in livestock. Union of Concerned Scientists.
- Mevik, B.-H., R. Wehrens, and K. H. Liland. 2011. pls: Partial least squares and principal component regression. R package version:2–3.
- Michel, A. L., R. G. Bengis, D. F. Keet, M. Hofmeyr, L. M. de Klerk, P. C. Cross, A. E. Jolles, D. Cooper, I. J. Whyte, P. Buss, and J. Godfroid. 2006. Wildlife tuberculosis in South African conservation areas: Implications and challenges. *Veterinary Microbiology* 112:91–100.
- Munson, L., K. A. Terio, R. Kock, T. Mlengeya, M. E. Roelke, E. Dubovi, B. Summers, A. R. E. Sinclair, and C. Packer. 2008. Climate extremes promote fatal co-infections during canine distemper epidemics in African lions. *PLoS ONE* 3:e2545.
- Muth, M. K., S. A. Karns, S. J. Nielsen, J. C. Buzby, and H. F. Wells. 2011. Consumer-level food loss estimates and their use in the ERS loss-adjusted food availability data. Economic Research Service, United States Department of Agriculture.
- Myers, J. A., and K. E. Harms. 2009. Seed arrival, ecological filters, and plant species richness: a meta-analysis. *Ecology Letters* 12:1250–1260.
- Nee, S., and R. M. May. 1992. Dynamics of metapopulations: habitat destruction and competitive coexistence. *Journal of Animal Ecology* 61:37–40.
- Nguyen, T. L. T., J. E. Hermansen, and L. Mogensen. 2010. Environmental consequences of different beef production systems in the EU. *Journal of Cleaner Production* 18:756–766.
- Noma, T., C. Gratton, M. Colunga-Garcia, M. J. Brewer, E. E. Mueller, K. A. G. Wyckhuys, G. E. Heimpel, and M. E. O’Neal. 2010. Relationship of Soybean Aphid (Homoptera: Aphididae) to Soybean Plant Nutrients, Landscape Structure, and Natural Enemies. *Environmental Entomology* 39:31–41.
- Ogino, A., K. Kaku, T. Osada, and K. Shimada. 2004. Environmental impacts of the Japanese beef-fattening system with different feeding lengths as evaluated by a life-cycle assessment method. *Journal of Animal Science* 82:2115–2122.

- Ogino, A., H. Orito, K. Shimada, and H. Hirooka. 2007. Evaluating environmental impacts of the Japanese beef cow-calf system by the life cycle assessment method. *Animal Science Journal* 78:424–432.
- Packer, C., and A. E. Pusey. 1983. Adaptations of female lions to infanticide by incoming males. *American Naturalist* 121:716–728.
- Packer, C., and A. E. Pusey. 1984. Infanticide in carnivores. Pages 31–42 *in* G. Hausfater and S. B. Hrdy, editors. *Infanticide: comparative and evolutionary perspectives*. Aldine, New York.
- Parfitt, J., M. Barthel, and S. Macnaughton. 2010. Food waste within food supply chains: quantification and potential for change to 2050. *Philosophical Transactions of the Royal Society B: Biological Sciences* 365:3065–3081.
- Pauly, D., V. Christensen, S. Gu nette, T. J. Pitcher, U. R. Sumaila, C. J. Walters, R. Watson, and D. Zeller. 2002. Towards sustainability in world fisheries. *Nature* 418:689–695.
- Pelletier, N., R. Pirog, and R. Rasmussen. 2010. Comparative life cycle environmental impacts of three beef production strategies in the Upper Midwestern United States. *Agricultural Systems* 103:380–389.
- Pelletier, N., and P. Tyedmers. 2010. Forecasting potential global environmental costs of livestock production 2000–2050. *Proceedings of the National Academy of Sciences* 107:18371–18374.
- Phetteplace, H. W., D. E. Johnson, and A. F. Seidl. 2001. Greenhouse gas emissions from simulated beef and dairy livestock systems in the United States. *Nutrient Cycling in Agroecosystems* 60:99–102.
- Pimentel, D. 1961. Species Diversity and Insect Population Outbreaks. *Annals of the Entomological Society of America* 54:76–86.
- Pimentel, D., B. Berger, F. David, M. Newton, B. Wolfe, E. Karabinakis, S. Clark, E. Poon, E. Abbett, and S. Nandagopal. 2004. Water resources: Agricultural and environmental issues. *BioScience* 54:909–918.
- Pimentel, D., J. Houser, E. Preiss, O. White, H. Fang, L. Mesnick, T. Barsky, S. Tariche, J. Schreck, and S. Alpert. 1997. Water resources: Agriculture, the environment, and society. *BioScience* 47:97–106.
- Polis, G. A., W. B. Anderson, and R. D. Holt. 1997. Toward an integration of landscape and food web ecology: the dynamics of spatially subsidized food webs. *Annual review of ecology and systematics* 28:289–316.
- Post, W. M., R. C. Izaurralde, J. D. Jastrow, B. A. McCARL, J. E. Amonette, V. L. Bailey, P. M. Jardine, T. O. West, and J. Zhou. 2004. Enhancement of carbon sequestration in US soils. *Bioscience* 54:895–908.
- Poulsen, J. R., C. W. Osenberg, C. J. Clark, D. J. Levey, and B. M. Bolker. 2007. Plants as Reef Fish: Fitting the Functional Form of Seedling Recruitment. *The American Naturalist* 170:167–183.
- Pritchard, J. K., M. T. Seielstad, A. Perez-Lezaun, and M. W. Feldman. 1999. Population growth of human Y chromosomes: a study of Y chromosome microsatellites. *Molecular Biology and Evolution* 16:1791–1798.

- R Core Team. 2012. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Ragsdale, D. W., D. A. Landis, J. Brodeur, G. E. Heimpel, and N. Desneux. 2011. Ecology and Management of the Soybean Aphid in North America. *Annual Review of Entomology* 56:375–399.
- Ragsdale, D. W., B. P. McCornack, R. C. Venette, B. D. Potter, I. V. MacRae, E. W. Hodgson, M. E. O’Neal, K. D. Johnson, R. J. O’Neil, C. D. DiFonzo, T. E. Hunt, P. A. Glogoza, and E. M. Cullen. 2007. Economic Threshold for Soybean Aphid (Hemiptera: Aphididae). *Journal of Economic Entomology* 100:1258–1267.
- Ragsdale, D. W., D. J. Voegtlin, and R. J. O’Neil. 2004. Soybean Aphid Biology in North America. *Annals of the Entomological Society of America* 97:204–208.
- Redfearn, D. D., and T. G. Bidwell. 1996. Stocking rate: The key to successful livestock production. Oklahoma Cooperative Extension Service.
- Renwick, A. R., P. C. L. White, and R. G. Bengis. 2006. Bovine tuberculosis in southern African wildlife: a multi-species host–pathogen system. *Epidemiology and Infection* 135:529.
- Riedell, W. E. 1989. Effects of Russian wheat aphid infestation on barley plant response to drought stress. *Physiologia Plantarum* 77:587–592.
- Robertson, G. P. 2000. Greenhouse Gases in Intensive Agriculture: Contributions of Individual Gases to the Radiative Forcing of the Atmosphere. *Science* 289:1922–1925.
- Rodwell, T. C., N. P. Kriek, R. G. Bengis, I. J. Whyte, P. C. Viljoen, V. de Vos, and W. M. Boyce. 2001. Prevalence of bovine tuberculosis in African buffalo at Kruger National Park. *Journal of Wildlife Diseases* 37:258–264.
- Root, R. B. 1973. Organization of a plant-arthropod association in simple and diverse habitats: the fauna of collards (*Brassica oleracea*). *Ecological monographs* 43:95–124.
- Roy, P., T. Orikasa, M. Thammawong, N. Nakamura, Q. Xu, and T. Shiina. 2012. Life cycle of meats: An opportunity to abate the greenhouse gas emission from meat industry in Japan. *Journal of Environmental Management* 93:218–224.
- Russell, E. P. 1989. Enemies hypothesis: a review of the effect of vegetational diversity on predatory insects and parasitoids. *Environmental entomology* 18:590–599.
- Schmidt, N. P., M. E. O’Neal, and L. A. Schulte Moore. 2011. Effects of Grassland Habitat and Plant Nutrients on Soybean Aphid and Natural Enemy Populations. *Environmental Entomology* 40:260–272.
- Schmidt, N. P., M. E. O’Neal, and J. W. Singer. 2007. Alfalfa Living Mulch Advances Biological Control of Soybean Aphid. *Environmental Entomology* 36:416–424.
- Schuman, G. E., H. H. Janzen, and J. E. Herrick. 2002. Soil carbon dynamics and potential carbon sequestration by rangelands. *Environmental pollution* 116:391–396.
- Shannag, H. K., and W. M. Obeidat. 2006. Voracity and conversion efficiency by larvae of *Coccinella septempunctata* L. (Coleoptera: Coccinellidae) on *Aphis fabae* Scop. (Homoptera: Aphididae) reared on two faba bean cultivars with different levels of resistance. *Applied Entomology and Zoology* 41:521–527.

- Sheehan, W. 1986. Response by specialist and generalist natural enemies to agroecosystem diversification: a selective review. *Environmental Entomology* 15:456–461.
- Shmida, A., and S. Ellner. 1984. Coexistence of plant species with similar niches. *Vegetatio* 58:29–55.
- Shriner, D., Y. Liu, D. C. Nickle, and J. I. Mullins. 2006. Evolution of intrahost HIV-1 genetic diversity during chronic infection. *Evolution* 60:1165–1176.
- Shroyer, J., K. Dhuyvetter, G. Kuhl, D. Fjell, L. Langemeier, and J. Fritz. 1993, April. Wheat pasture in Kansas. Cooperative Extension Service, Kansas State University.
- Simonsen, M.-L. R., A. Enkegaard, C. N. Bang, and L. Sigsgaard. 2009. Temperature effect on the predation rate of *Anthocoris nemorum* (Het.: Anthocoridae) on cabbage aphids (Hom.: Aphididae). *Journal of Applied Entomology* 133:198–200.
- Sisson, S. A., Y. Fan, and M. M. Tanaka. 2007. Sequential Monte Carlo without likelihoods. *Proceedings of the National Academy of Sciences* 104:1760–1765.
- Smart, A. J. 2005. Forecasting Forage Production. Rapid City, South Dakota.
- Smart, A. J., R. Gates, and B. H. Dunn. 2012. Drought and stocking rate effects on forage yield from western South Dakota rangelands. South Dakota State University Extension.
- Smith, W. K., C. C. Cleveland, S. C. Reed, N. L. Miller, and S. W. Running. 2012. Bioenergy Potential of the United States Constrained by Satellite Observations of Existing Productivity. *Environmental Science & Technology* 46:3536–3544.
- Smuts, G. L. 1976. Population characteristics and recent history of lions in two parts of the Kruger National Park. *Koedoe* 19:153–164.
- Smuts, G. L., J. L. Anderson, and J. C. Austin. 1978a. Age determination of the African lion (*Panthera leo*). *Journal of Zoology* 185:115–146.
- Smuts, G. L., J. Hanks, and I. J. Whyte. 1978b. Reproduction and social organization of lions from the Kruger National Park. *Carnivore* 1:17–28.
- Smuts, G. L., G. A. Robinson, and I. J. Whyte. 1980. Comparative growth of wild male and female lions (*Panthera leo*). *Journal of Zoology* 190:365–373.
- Snäll, T., R. B. O’Hara, C. Ray, and S. K. Collinge. 2008. Climate-driven spatial dynamics of plague among prairie dog colonies. *The American Naturalist* 171:238–248.
- Starfield, A. M., P. R. Furniss, and G. L. Smuts. 1981. A model of lion population dynamics as a function of social behaviour. Pages 121–134 *Dynamics of Large Mammal Populations*. J. Wiley, New York.
- Van Steenis, M. J. 1994. Intrinsic rate of increase of *Lysiphlebus testaceipes* Cresson (Hym.; Braconidae), a parasitoid of *Aphis gossypii* Glover (Hom., Aphididae) at different temperatures. *Journal of Applied Entomology* 118:399–406.
- Stehfest, E., L. Bouwman, D. P. Vuuren, M. G. J. Elzen, B. Eickhout, and P. Kabat. 2009. Climate benefits of changing diet. *Climatic Change* 95:83–102.
- Steinfeld, H., P. Gerber, T. D. Wassenaar, V. Castel, and C. De Haan. 2006. Livestock’s long shadow: environmental issues and options. FAO.

- Subak, S. 1999. Global environmental costs of beef production. *Ecological Economics* 30:79–91.
- Tanaka, M. M., A. R. Francis, F. Luciani, and S. A. Sisson. 2006. Using approximate Bayesian computation to estimate tuberculosis transmission parameters from genotype data. *Genetics* 173:1511–1520.
- Tavaré, S., D. J. Balding, R. C. Griffiths, and P. Donnelly. 1997. Inferring Coalescence Times From DNA Sequence Data. *Genetics* 145:505–518.
- Templeton, A. R. 2009. Statistical hypothesis testing in intraspecific phylogeography: nested clade phylogeographical analysis vs. approximate Bayesian computation. *Molecular Ecology* 18:319–331.
- Thies, C., I. Steffan-Dewenter, and T. Tscharntke. 2003. Effects of Landscape Context on Herbivory and Parasitism at Different Spatial Scales. *Oikos* 101:18–25.
- Tilman, D. 1994. Competition and Biodiversity in Spatially Structured Habitats. *Ecology* 75:2–16.
- Tilman, D. 1997. Community invasibility, recruitment limitation, and grassland biodiversity. *Ecology* 78:81–92.
- Tilman, D., J. Hill, and C. Lehman. 2006. Carbon-Negative Biofuels from Low-Input High-Diversity Grassland Biomass. *Science* 314:1598–1600.
- Tilman, D., P. B. Reich, J. Knops, D. Wedin, T. Mielke, and C. Lehman. 2001. Diversity and Productivity in a Long-Term Grassland Experiment. *Science* 294:843–845.
- Tompkins, D. M., A. M. Dunn, M. J. Smith, and S. Telfer. 2011. Wildlife diseases: from individuals to ecosystems. *Journal of Animal Ecology* 80:19–38.
- Tonhasca, A., and D. N. Byrne. 1994. The effects of crop diversification on herbivorous insects: a meta-analysis approach. *Ecological Entomology* 19:239–244.
- Toni, T., D. Welch, N. Strelkova, A. Ipsen, and M. P. H. Stumpf. 2009. Approximate Bayesian computation scheme for parameter inference and model selection in dynamical systems. *Journal of The Royal Society Interface* 6:187–202.
- Torell, R., W. Riggs, B. Bruce, and B. Kvasnicka. 1999. Wheat pasture grazing: agronomic, cultural and livestock management practices. Nevada Cooperative Extension Fact Sheet 99:39.
- Tscharntke, T., Y. Clough, T. C. Wanger, L. Jackson, I. Motzke, I. Perfecto, J. Vandermeer, and A. Whitbread. 2012. Global food security, biodiversity conservation and the future of agricultural intensification. *Biological Conservation* 151:53–59.
- Turnbull, L. A., M. J. Crawley, and M. Rees. 2000. Are Plant Populations Seed-Limited? A Review of Seed Sowing Experiments. *Oikos* 88:225–238.
- US Census Bureau. 2009. American Community Survey. United States Census Bureau. <http://www.census.gov/acs/www/>.
- US Department of Commerce. 2007. Commodity Flow Survey. <http://www.census.gov/econ/cfs/>.
- USDA AMS. 2006. Annual meat trade review: Meat, livestock, grain & slaughter data. Agricultural Marketing Service, United States Department of Agriculture, Des Moines, Iowa.

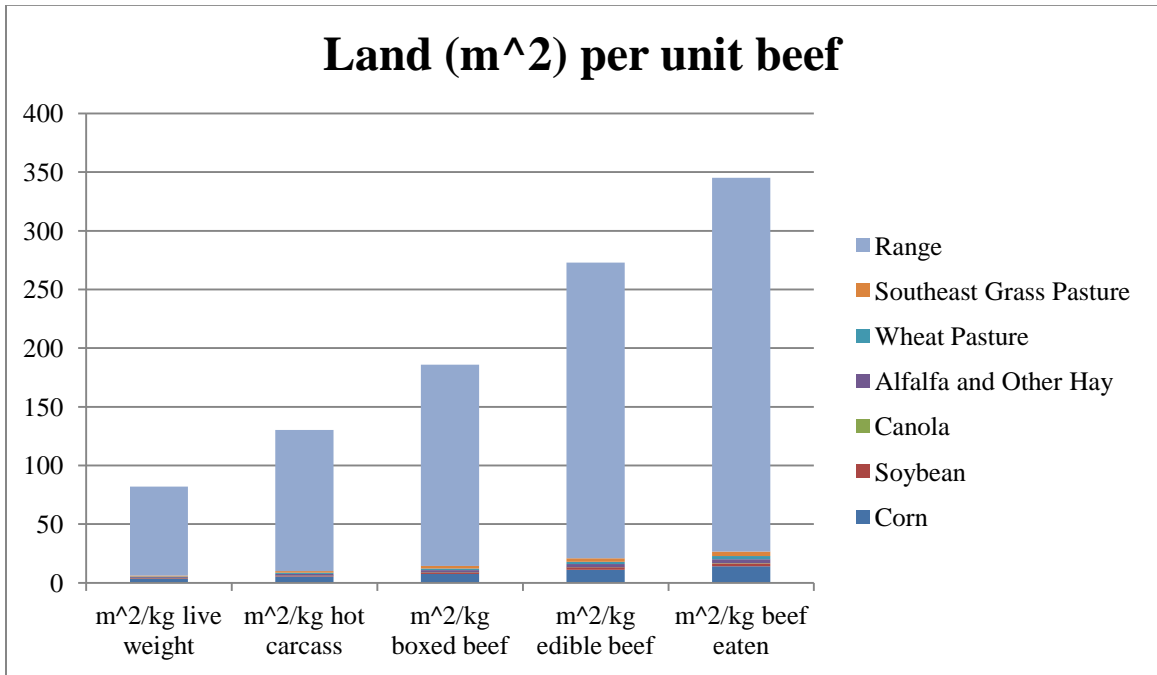
- USDA AMS. 2008. Annual meat trade review: Meat, livestock, grain & slaughter data. Agricultural Marketing Service, United States Department of Agriculture, Des Moines, Iowa.
- USDA AMS. 2010. Annual meat trade review: Meat, livestock, grain & slaughter data. Agricultural Marketing Service, United States Department of Agriculture, Des Moines, Iowa.
- USDA AMS. 2011a. Annual meat trade review: Meat, livestock, grain & slaughter data. Agricultural Marketing Service, United States Department of Agriculture, Des Moines, Iowa.
- USDA AMS. 2011b, April 15. Market News. Agricultural Marketing Service, United States Department of Agriculture.  
<http://www.ams.usda.gov/AMSV1.0/marketnews>.
- USDA ERS. 1997a. Agricultural resources and environmental indicators, 1996-97: land, water, production inputs, production management, technology, conservation & environmental programs Agricultural Handbook No. 712. Economic Research Service, United States Department of Agriculture.
- USDA ERS. 1997b, December. Agricultural Resources and Environmental Indicators: Seed use, costs, and trade. Economic Research Service, United States Department of Agriculture.
- USDA ERS. 2011. Fertilizer use and price. Economic Research Service, United States Department of Agriculture. <http://www.ers.usda.gov/data-products/fertilizer-use-and-price.aspx>.
- USDA ERS. 2013. Livestock & Meat Domestic Data 1921-2012. Economic Research Service, United States Department of Agriculture. <http://www.ers.usda.gov/data-products/livestock-meat-domestic-data.aspx>.
- USDA FSIS. 2009. Meat preparation fact sheet. Food Safety and Inspection Service, United States Department of Agriculture.  
[http://www.fsis.usda.gov/factsheets/beef\\_from\\_farm\\_to\\_table/index.asp](http://www.fsis.usda.gov/factsheets/beef_from_farm_to_table/index.asp).
- USDA NASS. 1999. Agricultural chemical usage: 1997 livestock and general farm summary. National Agricultural Statistics Service, United States Department of Agriculture.
- USDA NASS. 2004. Agricultural chemical usage postharvest applications: Corn and soybeans. National Agricultural Statistics Service, United States Department of Agriculture.
- USDA NASS. 2008a. Farm and Ranch Irrigation Survey, 2007 Census of Agriculture. National Agricultural Statistics Service, United States Department of Agriculture.
- USDA NASS. 2008b. Livestock slaughter 2007 summary. National Agricultural Statistics Service, United States Department of Agriculture.
- USDA NASS. 2010. Cattle Death Loss 2002-2009. National Agricultural Statistics Service, United States Department of Agriculture.  
<http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1017>.

- USDA NASS. 2012a. National Statistics for Cattle 2007-2011. National Agricultural Statistics Service, United States Department of Agriculture. [http://www.nass.usda.gov/Statistics\\_by\\_Subject/index.php](http://www.nass.usda.gov/Statistics_by_Subject/index.php).
- USDA NASS. 2012b. National Statistics for Crops 2007-2011. National Agricultural Statistics Service, United States Department of Agriculture. [http://www.nass.usda.gov/Statistics\\_by\\_Subject/index.php](http://www.nass.usda.gov/Statistics_by_Subject/index.php).
- USDA NASS. 2012c. National Statistics for chemical usage 2007-2011. National Agricultural Statistics Service, United States Department of Agriculture. <http://quickstats.nass.usda.gov>.
- USDA NRCS. 2013, October 11. The PLANTS Database. National Plant Data Team, Greensboro, NC 27401, USA. <http://plants.usda.gov>.
- USDOT FHWA. 2001. Analysis of the vehicle inventory and use survey for trucks with five axles or more. Federal Highway Administration, United States Department of Transportation.
- Venables, W. N., and B. D. Ripley. 2002. MASS: modern applied statistics with S. Springer, New York.
- De Vos, V., R. G. Bengis, N. P. J. Kriek, A. L. Michel, D. F. Keet, J. P. Raath, and H. F. K. A. Huchzermeyer. 2001. The epidemiology of tuberculosis in free-ranging African buffalo (*Syncerus caffer*) in the Kruger National Park, South Africa. Onderstepoort Journal of Veterinary Research 68:119–130.
- Walker, D. M., D. Allingham, H. W. J. Lee, and M. Small. 2010. Parameter inference in small world network disease models with approximate Bayesian computational methods. Physica A: Statistical Mechanics and its Applications 389:540–548.
- Wang, M. 1999. The Greenhouse Gases, Regulated Emissions, and Energy Use in Transportation (GREET) Model. Argonne National Laboratory.
- Wang, Z., R. Zhang, J. Fadel, T. Rumsey, J. Arogo, H. Xin, Y. Liang, G. Mansell, and S. Shepard. 2005. Agricultural Waste Characteristics. Agricultural Waste Management Field Handbook. Soil Conservation Service, US Department of Agriculture.
- Watson, R. T., I. R. Noble, B. Bolin, N. H. Ravindranath, D. Verardo, K. Andrasko, M. Apps, S. Brown, G. Farquhar, D. Goldberg, S. Hamburg, R. Houghton, P. Jarvis, T. Karjalainen, H. Kheshgi, T. Krug, W. Kurz, D. Lashof, B. Lim, W. Makundi, M. Manning, G. Marland, O. Masera, D. Murdiyarso, B. Murray, R. Persson, N. Sampson, J. Sathaye, R. Scholes, B. Schlamadinger, W. Sombroek, S. Prisley, J. Stone, R. Sukumar, and R. Valentini. 2000. Land use, land-use change and forestry: Summary for policymakers. Intergovernmental Panel on Climate Change, Montreal, Canada.
- Wegmann, D., C. Leuenberger, and L. Excoffier. 2009. Efficient Approximate Bayesian Computation Coupled With Markov Chain Monte Carlo Without Likelihood. Genetics 182:1207–1218.
- Weidema, B. P., M. Wesnæs, J. Hermansen, T. Kristensen, and N. Halberg. 2008. Environmental improvement potentials of meat and dairy products. JRC, Institute for Prospective Technological Studies. ISBN 978-92-79-09716-4.

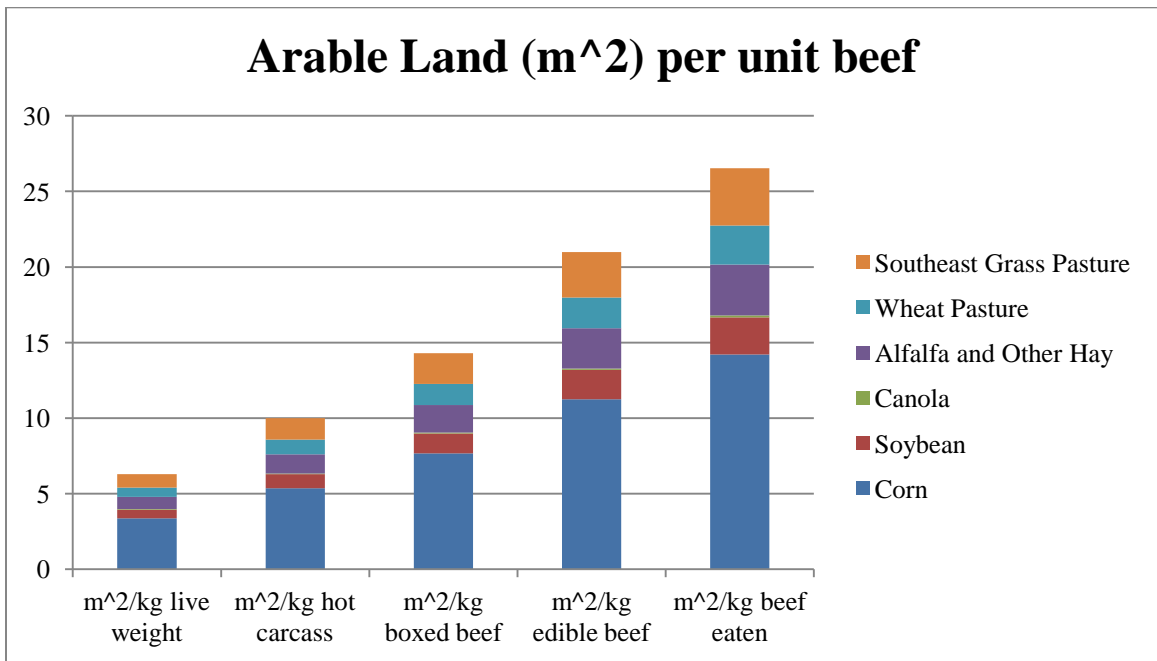


- West, T. O., and G. Marland. 2002a. Net carbon flux from agricultural ecosystems: methodology for full carbon cycle analyses. *Environmental Pollution* 116:439–444.
- West, T. O., and G. Marland. 2002b. A synthesis of carbon sequestration, carbon emissions, and net carbon flux in agriculture: comparing tillage practices in the United States. *Agriculture, Ecosystems & Environment* 91:217–232.
- Whitman, K. L., A. M. Starfield, H. Quadling, and C. Packer. 2007. Modeling the effects of trophy selection and environmental disturbance on a simulated population of African lions. *Conservation Biology* 21:591–601.
- Whitman, K., A. M. Starfield, H. S. Quadling, and C. Packer. 2004. Sustainable trophy hunting of African lions. *Nature* 428:175–178.
- Wickham, H. 2007. Reshaping Data with the reshape Package. *Journal of Statistical Software* 21:1–20.
- Wickham, H. 2009. *ggplot2: elegant graphics for data analysis*. Springer Publishing Company, Incorporated.
- Wickham, H. 2011. The split-apply-combine strategy for data analysis. *Journal of Statistical Software* 40:1–29.
- Williams, A. G., E. Audsley, and D. L. Sandars. 2006. Determining the environmental burdens and resource use in the production of agricultural and horticultural commodities. Main Report. Defra Research Project IS0205, Bedford: Cranfield University and Defra.
- Wilson, A., J. Thorne, and J. Morrill. 2003. *Consumer Guide to Home Energy Savings*, 8th edition. American Council for an Energy Efficient Economy.
- Wirsenius, S., C. Azar, and G. Berndes. 2010. How much land is needed for global food production under scenarios of dietary changes and livestock productivity increases in 2030? *Agricultural Systems* 103:621–638.
- Wu, Z., and G. E. Heimpel. 2007. Dynamic egg maturation strategies in an aphid parasitoid. *Physiological Entomology* 32:143–149.
- Wu, Z., D. Schenk-Hamlin, W. Zhan, D. W. Ragsdale, and G. E. Heimpel. 2004. The Soybean Aphid in China: A Historical Review. *Annals of the Entomological Society of America* 97:209–218.
- Yu, D. W., H. B. Wilson, and N. E. Pierce. 2001. An Empirical Model of Species Coexistence in a Spatially Structured Environment. *Ecology* 82:1761–1771.
- Zhang, H., J. L. Schroder, E. G. Krenzer, O. M. Kachurina, and M. E. Payton. 2004. Yield and Quality of Winter Wheat Forage As Affected by Lime. *Forage and Grazinglands*.

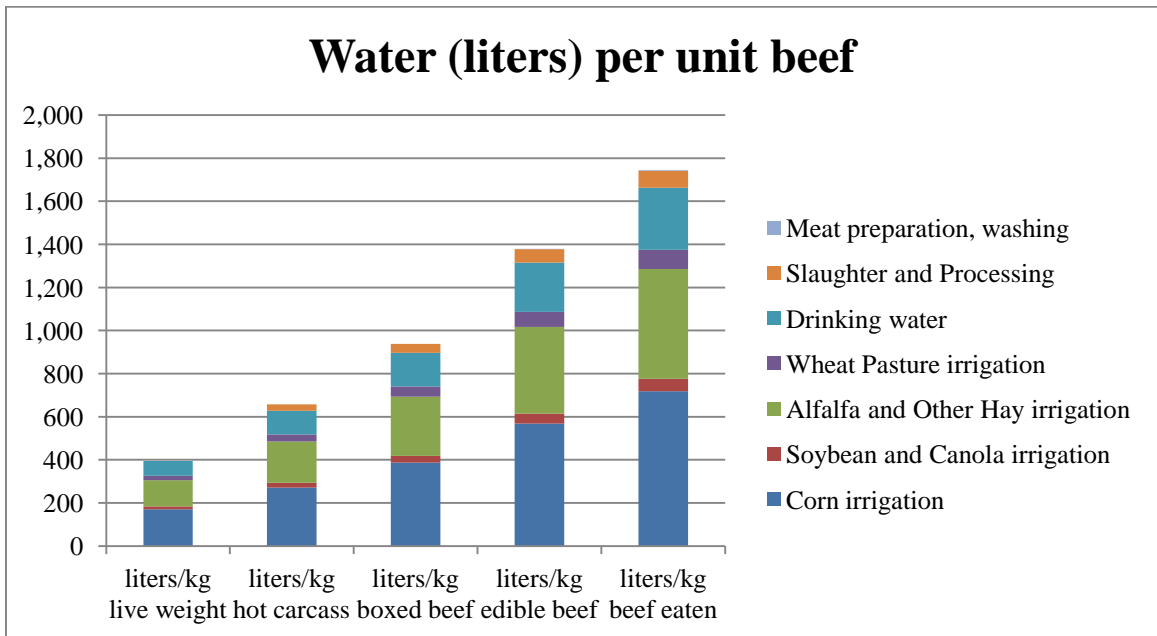
**Appendix 1: Additional Tables and Figures for Chapter 2**



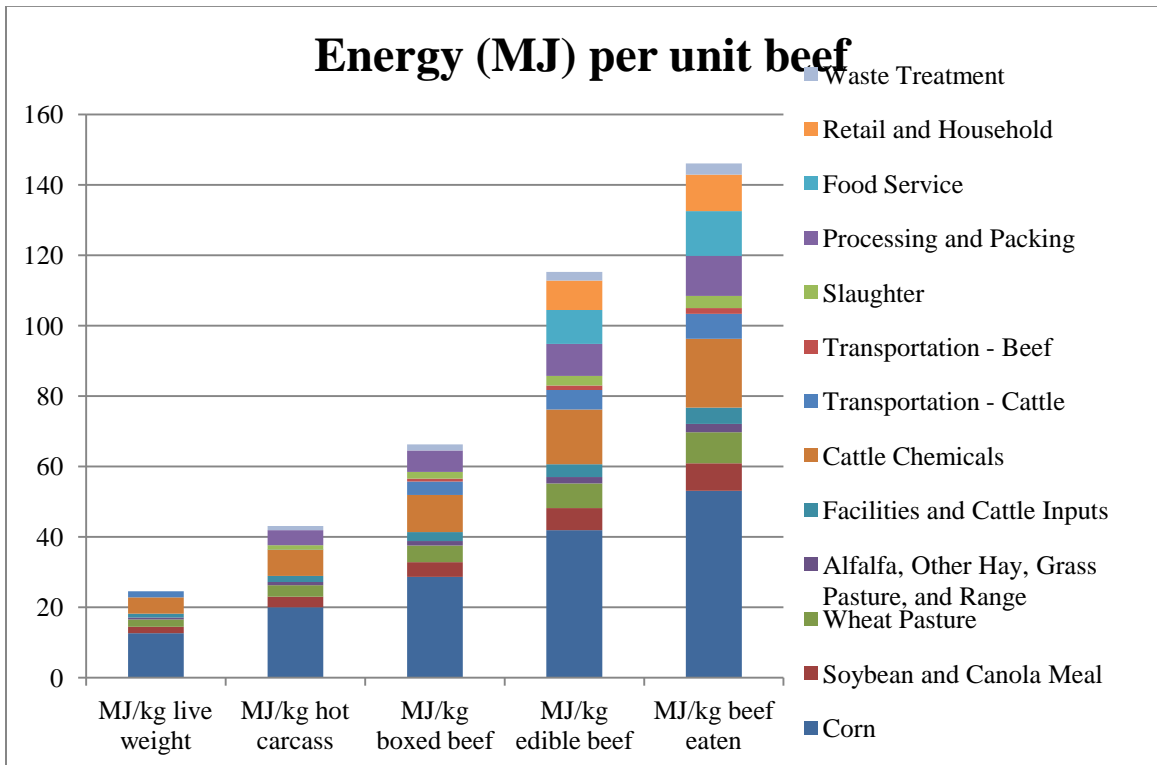
**Figure A1-1: Analysis of land use for U.S. Industrial beef production at five different stages of production and consumption**



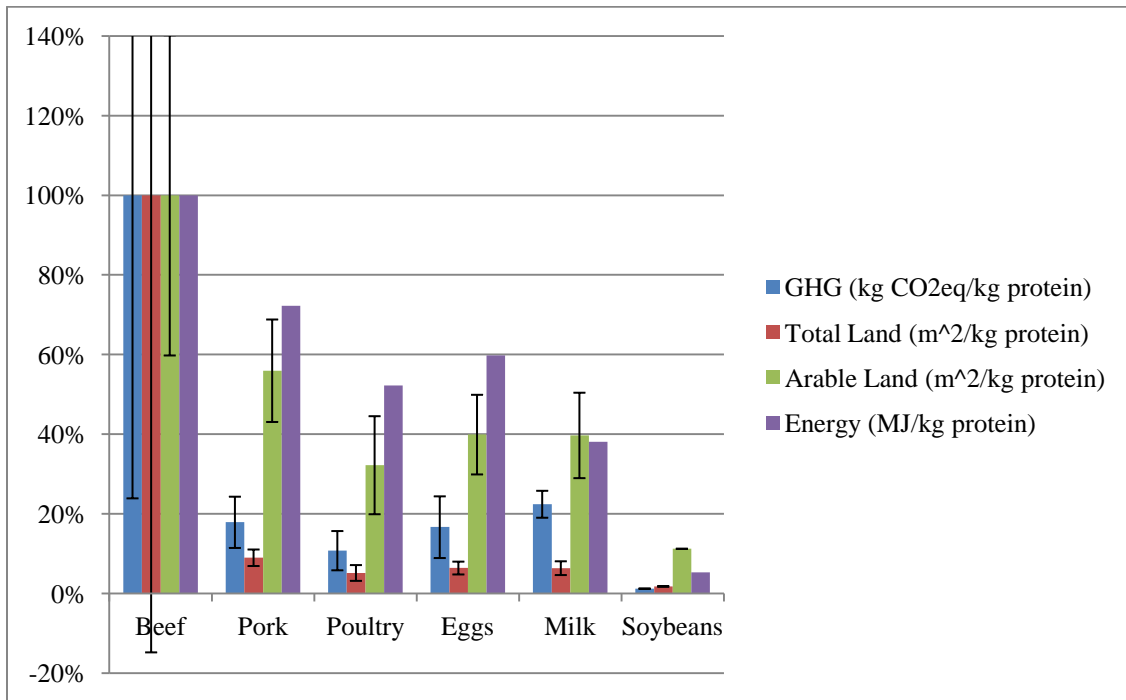
**Figure A1-2: Analysis of arable land use for U.S. Industrial beef production at five different stages of production and consumption**



**Figure A1-3: Analysis of water use for U.S. Industrial beef production at five different stages of production and consumption**



**Figure A1-4: Analysis of fossil energy use for U.S. Industrial beef production at five different stages of production and consumption**



**Figure A1-5: Environmental impacts of different protein sources compared with beef**

Land and GHG numbers are from Nijdam et al 2011 and are calculated on the basis of boneless retail meat from cradle to beef transportation. Energy numbers are from Pelletier et al 2011 and are calculated on the basis of carcass meat from cradle to farm gate. Error bars represent one standard deviation of the mean. Standard deviations are not available for energy estimates. Soybean data is from a single study.

**Table A1-1: U.S. conversion rates for different meats**

Dressing percentage is the portion of the whole animal that forms the carcass. Retail percentage is the portion of the carcass that is typically sold at retail establishments and consists of meat, bones, and fat.

	Dressing percentage	Retail percentage (of carcass)	Edible meat / retail meat	Eaten meat / Edible meat
<b>Beef</b>	62%	70%	70%	80%
<b>Pork</b>	72%	80%	66%	71%
<b>Chicken</b>	71%	86%	70%	85%
<b>Fish</b>	75%	71%	77%	60%

**Table A1-2: Water use, showing wide range of water use estimates**

“Blue” water refers to surface and ground water. “Green” water is rainfall.

<b>Publication</b>	<b>Functional Unit</b>	<b>Management Type</b>	<b>Scope</b>	<b>Type of Water</b>	<b>Water (mean liters)</b>
Peters et al 2010b	kg carcass weight	extensive	Cow-calf only	blue	34
Eady et al 2011	kg live weight	extensive	Cradle-to-farm-gate	blue	153
Peters et al 2010b	kg carcass weight	intensive	Weaner through processing	blue	502
<i>This paper</i>	<i>kg boneless beef</i>	<i>intensive</i>	<i>Cradle-to-grave</i>	<i>blue</i>	<i>1,379</i>
Capper 2011	kg carcass weight	intensive	Cradle-to-farm-gate	blue	1,763
Beckett and Oltjen 1993	kg boneless beef	intensive	Cradle-to-market	blue	3,682
Peters et al 2010b	kg carcass weight	extensive	Cow-calf only	green+blue	7,684
Peters et al 2010b	kg carcass weight	intensive	Weaner through processing	green+blue	9,647
Hoekstra and Chapagain 2007	kg boneless beef	intensive	Cradle-to-farm-gate	green+blue	13,193
Hoekstra and Chapagain 2007	kg boneless beef	mixed	Cradle-to-farm-gate	green+blue	15,497
Eady et al 2011	kg live weight	extensive	Cradle-to-farm-gate	green+blue	16,600
Pimentel et al 2004	unspecified ("kg beef")	intensive	Cradle-to-farm-gate	green+blue	43,000
Pimentel et al 1997	unspecified ("kg beef")	intensive	Cradle-to-farm-gate	green+blue	105,400
Pimentel et al 2004	unspecified ("kg beef")	extensive	Cradle-to-farm-gate	green+blue	160,000
Pimentel et al 1997	unspecified ("kg beef")	extensive	Cradle-to-farm-gate	green+blue	200,000

## **Appendix 2: Detailed Methods for Chapter 2**

The retail value of the U.S. beef industry is \$73 billion, producing 11.9 million tonnes of beef (carcass weight) per year (USDA ERS 2013).

In the U.S., a typical calf is born in a cow-calf herd on rangeland. It is weaned at around six months and transported by truck to a growing facility. The animal might be stockered on a high-quality pasture for several months; it might be backgrounded in a feeding facility where it is fed primarily harvested forage; or it might be sent directly to a feedlot to be fed high-energy feed. Regardless of the mid-stage growing method, the animal is eventually transported to a feedlot to be finished on high-energy feeds.

When the animal has reached an average of 604 kg live weight for steers or 557 kg for heifers (derived from (USDA NASS 2012a), it is slaughtered, and the carcass butchered into large segments called primals. These primals are then either sold whole as “boxed beef” to retail outlets and food service establishments or further prepared at the packing house (cutting, grinding, etc.) and sold as “retail-ready” beef. At retail outlets, boxed beef is prepared and packaged for sale and retail-ready beef is sold without further processing. Consumers consume beef at food service establishments or purchase beef products from stores to cook at home.

### **LCA Methods Details**

#### ***Land, crop and pasture inputs, crop storage***

The land used to support the U.S. cattle industry primarily consists of relatively dry rangelands, high-quality wheat pastures in the Great Plains and Southeast grass pastures, hayed land for alfalfa and other hay, and cropland used to grow corn, soybeans, and canola. We used fertilizer, lime, pesticide, seeding, and irrigation rates from national USDA statistics, published literature, and extension service recommendations. We assumed that pastures and alfalfa hay land are rejuvenated on average once every six years, and that rangeland and non-alfalfa hayfields are never rejuvenated. We assumed that one third of southeast grass pasture land is hayed for winter feeding. We calculated the average amount of insecticides and fungicides used for the storage of corn grain and soybeans using average rates from USDA statistics (USDA NASS 2004).

**Table A2-1. Calculation of land use for each feed input**

	Yield (kg/ha DM)	Allocation	Allocated Land Use (ha/kg DM)
<b>Corn Grain</b>	8,272	100%	1.21E-04
<b>Corn Silage</b>	14,399	100%	6.95E-05
<b>Soy</b>	2,548	100%	3.93E-04
<b>Canola</b>	1,575	100%	6.35E-04
<b>Alfalfa Hay</b>	6,775	100%	1.48E-04
<b>Other Hay</b>	3,886	100%	2.57E-04
<b>Wheat Pasture</b>	2,242	17%	7.42E-05
<b>Southeast Grass Pasture</b>	9,191	100%	1.09E-04
<b>Range</b>	2,802	100%	3.57E-04
<b>Midwest Mixed Pasture</b>	10,393	100%	9.62E-05

Notes: Raw yield data is taken from USDA statistics (USDA NASS 2012b) and is the mean from years 2007-2011 for corn, corn silage, soy, canola, alfalfa hay, and other hay. Winter wheat biomass was approximated as 1 dry matter ton/acre (Epplin et al. 2000, Zhang et al. 2004). We used the yield of coastal bermudagrass (Coleman and Williams 2006) for southeast grass pasture yield. We estimate average range yield at 1.25 dry matter tons/acre (Launchbaugh and Owensby 1978, Redfearn and Bidwell 1996, Hart and Carpenter 2005, Smart 2005, Smith et al. 2012, Smart et al. 2012). Grain weights used were 56 lbs/bushel for corn and 60 lbs/bushel for soy and winter wheat. Dry matter fractions are from the IPCC (IPCC 2006).

**Table A2-2. Calculation of water use for each feed input**

	Average water application rate (cubic meters/ha)	Allocation	Allocated water usage (liters/kg feed)
<b>Corn Grain</b>	422	100%	51
<b>Corn Silage</b>	1725	100%	120
<b>Soy</b>	234	100%	92
<b>Canola</b>	46	100%	29
<b>Alfalfa Hay</b>	2143	100%	316
<b>Other Hay</b>	461	100%	119
<b>Wheat Pasture</b>	344	17%	26
<b>Southeast Grass Pasture</b>	29	100%	3
<b>Range</b>	0	100%	0
<b>Midwest Mixed Pasture</b>	29	100%	3

Notes: Irrigation rates are from the USDA's 2007 Census of Agriculture (USDA NASS 2008a).

**Table A2-3: Calculation of energy use for each feed input**

(all columns: MJ per dry matter kg of feed produced)

	<b>Inputs</b>	<b>Production</b>	<b>TOTAL Energy</b>	<b>Allocation</b>	<b>Allocated Energy</b>
<b>Corn Grain</b>	3.3672	0.5999	3.9671	100%	3.9671
<b>Corn Silage</b>	1.9345	0.3446	2.2792	100%	2.2792
<b>Soy</b>	9.0220	0.8159	9.8378	100%	9.8378
<b>Canola</b>	4.9464	2.2430	7.1893	100%	7.1893
<b>Alfalfa Hay</b>	0.3641	0.2989	0.6630	100%	0.6630
<b>Other Hay</b>	0.0009	0.1672	0.1681	100%	0.1681
<b>Wheat Pasture</b>	14.3315	0.9899	15.3214	17%	2.5491
<b>Southeast Grass Pasture</b>	0.2053	0.0389	0.2442	100%	0.2442
<b>Range</b>	0.0032	0.0000	0.0032	100%	0.0032
<b>Midwest Mixed Pasture</b>	0.1032	0.0344	0.1376	100%	0.1376

Notes: Energy figures for nutrient and pesticide production and application, planting, cultivation, and harvesting are from are from (West and Marland 2002a, Hill et al. 2006, Tilman et al. 2006, Buratti et al. 2010). Mean input transport distances are from (US Department of Commerce 2007).



**Table A2-4: Calculation of greenhouse gas emissions for each feed input**(all columns: kg CO<sub>2</sub>-eq per dry matter kg of feed produced).

	<b>Input Production and Transport</b>	<b>Lime Application</b>	<b>Farm Manage- ment</b>	<b>Irrigation electricity</b>	<b>N<sub>2</sub>O on Managed Lands</b>
<b>Corn Grain</b>	0.17623	0.01013	0.02343	0.01006	0.23076
<b>Corn Silage</b>	0.10125	0.00582	0.01346	0.01295	0.12228
<b>Soy</b>	0.18427	0.02788	0.06365	0.01260	0.05078
<b>Canola</b>	0.22706	0.01065	0.11733	0.00387	0.45267
<b>Alfalfa Hay</b>	0.02333	0.01516	0.01648	0.03735	0.00783
<b>Other Hay</b>	0.00006	0.00000	0.02752	0.00751	0.00000
<b>Wheat Pasture</b>	0.22974	0.00748	0.04556	0.02484	0.32069
<b>Southeast Grass Pasture</b>	0.00970	0.01117	0.00542	0.00022	0.01521
<b>Range</b>	0.00021	0.00000	0.00548	0.00000	0.00000
<b>Midwest Mixed Pasture</b>	0.00451	0.00988	0.00673	0.00019	0.00885

	<b>TOTAL</b>	<b>Allocation</b>	<b>Allocated TOTAL</b>
<b>Corn Grain</b>	0.45062	100%	0.45062
<b>Corn Silage</b>	0.25576	100%	0.25576
<b>Soy</b>	0.33918	100%	0.33918
<b>Canola</b>	0.81157	100%	0.81157
<b>Alfalfa Hay</b>	0.10014	100%	0.10014
<b>Other Hay</b>	0.03509	100%	0.03509
<b>Wheat Pasture</b>	0.62831	17%	0.10454
<b>Southeast Grass Pasture</b>	0.04172	100%	0.04172
<b>Range</b>	0.00568	100%	0.00568
<b>Midwest Mixed Pasture</b>	0.03016	100%	0.03016

Notes: Emissions from nutrient and pesticide production and application, planting, cultivation, and harvesting are from (West and Marland 2002a, 2002b). Input application rates are from USDA statistics (Shroyer et al. 1993, USDA ERS 1997a, 1997b, 2011, Torell et al. 1999, USDA NASS 2012c, 1999, 2012b, Kelling 2000, Manitoba Agriculture, Food and Rural Initiatives 2006, Barnes et al. 2007). Greenhouse gas emissions from electricity use are from (Wang 1999). Nitrous oxide emissions (direct and indirect) are calculated using IPCC methodology (IPCC 2006).

### ***Cow-calf herd management***

Calves are born in cow-calf herds on rangelands and feed exclusively on milk from their mothers. Because cows give birth once a year, we calculate the yearly maintenance budget of a cow, part of a bull, and part of a replacement heifer that are required to produce one calf. We use USDA statistics, published literature and expert opinion to ascertain typical herd demographics, calving rate, diet, manure production, and mortality rates. Calves are weaned at 204 kg (450 lb). All animals in cow-calf herds wear insecticide ear tags.

**Table A2-5: Cow-calf herd demographics**

Cow replacement rate	15%
Cow reproduction rate	89.4%
Bulls per cow	35
Bull reproductive period	6 years
Calf mortality rate	6.3%
Calf weaning weight	204 kg
Cow slaughter weight	587 kg
Bull slaughter weight	799 kg
Manure production rate (adult)	10,430 kg/AU/yr
Manure production rate (calf)	8,477 kg/AU/yr

Cow reproduction rate and calf mortality rate calculated from (USDA NASS 2010). Cow and bull slaughter weights are derived from (USDA NASS 2012a). Manure rates are from (Wang et al. 2005).

### ***Growth and finishing of cattle***

After weaning, calves enter one of three production channels. We assumed that 40% of calves enter a feedlot directly. Of the remaining 60%, three quarters are stockered on pasture on the Great Plains and in the Southeast and the remainder are backgrounded on high-forage rations. The stockered and backgrounded animals are then sent to the feedlot at an average of 340 kg (750 lb). Regardless of growing method, all animals are finished at the feedlot on high-energy diets at an average of 578 kg (1,275 lb) (USDA NASS 2008b). Diets, average daily gain, and manure production, and mortality for each growth and finishing production channel were determined from USDA statistics, published literature, and expert opinion. We calculated direct insecticide application, growth hormone use, and fed subtherapeutic antibiotics from USDA statistics, vendor ingredient lists, and published literature.

**Table A2-6: Cattle diets**

	Total required before feeding loss (kg/animal)				
	Stocker	Background	Direct to feedlot	Finishing diet 1 (67%)	Finishing diet 2 (33%)
Corn, dry rolled		414	376		1,367
Corn, fine ground			29	39	
Corn, steam flaked				692	
Corn, high moisture				387	
Corn silage			174	68	80
Dry distiller grains (DDGS)		59	101	320	
Soybean meal	215				19
Canola meal					20
Alfalfa hay		284	197	75	80
Other hay		425	10		
Pasture	2,268				

**Table A2-7: Insecticide, hormone, and antibiotic use**

	kg/animal
Direct insecticides	0.0066
Hormones	0.00017
Subtherapeutic antibiotics	2.0

Insecticide use from (USDA NASS 1999); hormone use based on market research; antibiotic use derived from (Mellon et al. 2001).

### ***Slaughter and processing***

We assume that slaughtering and processing facilities are located close to feedlots and do not include transportation of cattle and carcasses in our analyses. Cattle are slaughtered at an average of 578 kg (1,275 lb) (USDA NASS 2008b), and processed with a dressing percent of 62.9% (USDA AMS 2011b) for a hot carcass weight of 364 kg. We assume that 70% of hot carcass weight is retained as wholesale boxed beef, consisting of beef, bone, and fat (Aberle et al. 2001), and that 49.3% of hot carcass weight is ready-to-eat boneless beef (USDA AMS 2006, 2008, 2010, 2011a).

### ***Beef packaging and distribution***

We used the National Health and Nutrition Examination Survey (NHANES) to determine the form (e.g. cut, ground, smoked, encased) and venue (home or away) of beef eaten in the U.S. (CDC 2008). We calculated impacts of beef packaging based on typical production chain pathways for these different types of beef, using energy and greenhouse gas emissions associated with packaging materials (Delgado et al. 2007).

**Table A2-8: Beef packaging**

		kg beef /carcass	Polyethylene (kg/carcass)	PVC wrap (kg/carcass)	Polystyrene foam trays / carcass
Vacuum-packed	boxed beef	178	3.1		
Vacuum-packed	retail beef	48			
PVC wrap with foam tray	retail beef	9		0.036	9.3

***Food service***

Beef arrives at food service establishments as wholesale cuts of beef (in primarily sit-in restaurants) or ground beef (in primarily fast-food restaurants). We calculated the percentage of food service food that is beef from the NHANES survey, and multiply the total environmental impacts of food service establishments by this percentage. We include environmental impacts from all on-going facets of the food service, including lighting, heating and air conditioning, refrigeration, air conditioning, cooking, dishwashing, as well as production and disposal of disposable trays and utensils, paper liners, and packaging (Baldwin et al. 2010).

***Retail sales***

Beef arrives at grocery stores and supermarkets as retail-ready beef and as wholesale beef that is processed and repackaged at the store. We calculate the percentage of store food that is beef from the NHANES survey and multiply this percentage by the total environmental impacts of stores to determine the beef portion. We assume a 4.3% loss rate of beef in supermarkets (Buzby et al. 2009).

***Household purchasing and consumption***

We assumed the beef is stored in the home in an 18-cubic-foot upright refrigerator-freezer unit that is, on average, half full. We assumed that one quarter of the beef is frozen for an average of 30 days and the remainder is refrigerated for an average of 2 days. We used national surveys and USDA recommendations to determine impacts of cooking beef. We assumed 20% of bought beef was lost as spoilage and beef thrown away during preparation and post-eating (Muth et al. 2011).

**Table A2-9: Calculation of beef cooking impacts**

	Percent of beef	Energy use (MJ/kg beef)	GHG emissions (CO <sub>2</sub> -eq/kg beef)
Cut of beef in electric oven	37%	7.94	1.49
Cut of beef in gas oven	23%	13.02	0.13
Ground beef on electric stove	24%	1.65	0.31
Ground beef on gas stove	16%	1.09	0.01

Percentage of beef based on numbers of electric and gas ovens and stoves owned in the U.S. (Energy Information Administration 2005) and approximately 60% cuts and 40% ground beef (CDC 2008). Cuts are assumed to be cooked at 350 °F with cooking times based on weight (USDA FSIS 2009). Ground beef is assumed to be cooked on the stovetop for five minutes per four 4-oz. patties. Energy use is from (Wilson et al. 2003) and greenhouse gas emissions are from (Wang 1999).

### ***Transportation***

We calculated average travel distances for fertilizers, lime, pesticides, seeds, cattle feed, cattle, and beef based on the U.S. Commodity Flow Survey (US Department of Commerce 2007). We do not include any distances trucks travel empty to return to their starting point. We assume feedlots and biofuel facilities are local to feed production and that tractor-plus-semitrailer trucks are used for hauling. We assume that all beef travels from the packinghouse to a distributor warehouse and from there to a store or restaurant, and that 90% of the total distance, from packinghouse to warehouse, is in 40-foot refrigerated trucks, and the remaining 10% of the distance, from the warehouse to the retail outlet, is in 20-foot refrigerated trucks. We assumed beef spends an average of one day in the warehouse. We use truck specifications from the Federal Highway Administration (USDOT FHWA 2001). We used national surveys (USDOT FHWA 2001, CDC 2008, FMI Research 2009, US Census Bureau 2009) to calculate the environmental impacts of driving to the grocery store to purchase beef.

**Table A2-10: Travel distances**

	Distance traveled by truck (km)
N, Phosphate, Potash	324
Lime	72
Pesticides	669
Seeds	464
Hormones and antibiotics	655
Cattle feed	288
Calf to growing facility	1,044
Cattle to finishing facility	1,044
Finished cattle to slaughter	1,044
Beef from Packer to Distributor	1,237
Beef from Distributor to Retailer	137
Household to grocery store	11

## Appendix 3: Tables of Beef LCAs for Chapter 2

**Table A3-1: Characteristics of beef LCA studies used in the literature survey**

Publication	Location	Number Scenarios	Management Type	Scope	Functional Unit
Beauchemin et al 2010	Western Canada	1	intensive	Cradle-to-farm-gate	kg live weight
Beckett and Oltjen 1993	USA	1	intensive	Cradle-to-market	kg boneless beef
Capper 2011	USA	1	intensive	Cradle-to-farm-gate	kg carcass weight
Casey and Holden 2006a	Ireland	6	pasture	Cradle-to-farm-gate	kg live weight / year <sup>1</sup>
Casey and Holden 2006b	Ireland	3	pasture	Cradle-to-farm-gate	kg live weight / year <sup>1</sup>
Cederberg and Stadig 2003	Sweden	1	extensive	Cradle-to-farm-gate	kg boneless beef
Cederberg et al 2009a	Brazil	1	extensive	Cradle-to-market	kg boneless beef
Cederberg et al 2009b	Sweden	1	intensive	Cradle-to-farm-gate	kg carcass weight
Eady et al 2011	Queensland, Australia	1	extensive	Cradle-to-farm-gate	kg live weight
Edwards-Jones et al 2009	Wales, UK	1	intensive	Cradle-to-farm-gate	kg live weight
	Wales, UK	1	extensive	Cradle-to-farm-gate	kg live weight
Foley et al 2011	Ireland	3	pasture	Cradle-to-farm-gate	kg carcass weight
Hoekstra and Chapagain 2007	USA	1	intensive	Cradle-to-farm-gate	kg boneless beef
	World	1	mixed	Cradle-to-farm-gate	kg boneless beef
Liebig et al 2010	Great Plains, USA	1	pasture	Stockering only	kg live weight gain <sup>2</sup>
	Great Plains, USA	2	extensive	Stockering only	kg live weight gain <sup>2</sup>
Nguyen et al 2010	Europe	3	intensive	Cradle-to-farm-gate	kg carcass weight
	Europe	1	pasture	Cradle-to-farm-gate	kg carcass weight
Ogino et al 2004	Japan	1	intensive	Weaner to farm-gate	kg live weight
Ogino et al 2007	Japan	1	intensive	Cow-calf only	kg live weight

Publication	Location	Number Scenarios	Management Type	Scope	Functional Unit
Pelletier et al 2010	Upper Midwest, USA	2	intensive	Cradle-to-farm-gate	kg live weight
	Upper Midwest, USA	1	pasture	Cradle-to-farm-gate	kg live weight
Peters et al 2010a	Victoria Australia	2	extensive	Cow-calf only	kg carcass weight
	New South Wales, Australia	2	intensive	Weaner through processing	kg carcass weight
Peters et al 2010b	Victoria Australia	2	extensive	Cow-calf only	kg carcass weight
	New South Wales, Australia	2	intensive	Weaner through processing	kg carcass weight
Phetteplace et al 2001	USA	1	intensive	Cradle-to-farm-gate	kg live weight
Pimentel et al 1997	USA	1	intensive	Cradle-to-farm-gate	? <sup>3</sup>
	USA	1	extensive	Cradle-to-farm-gate	? <sup>3</sup>
Pimentel et al 2004	USA	1	intensive	Cradle-to-farm-gate	? <sup>3</sup>
	USA	1	extensive	Cradle-to-farm-gate	? <sup>3</sup>
Roy et al 2012	Japan	1	intensive	Cradle-to-grave	kg boneless beef
Subak 1999	Africa	1	extensive	Cradle-to-farm-gate	kg carcass weight
	USA	1	intensive	Cradle-to-farm-gate	kg carcass weight
Vergé et al 2008	Canada	1	intensive	Cradle-to-farm-gate	kg live weight
Veysset et al 2010	Charolais, France	3	extensive	Cow-calf only	kg live weight
		2	pasture	Cradle-to-farm-gate	kg live weight
Weidema et al 2008	Europe	1	mixed	Cradle-to-grave	kg live weight
Williams et al 2006	UK	3	pasture	Cradle-to-farm-gate	kg carcass weight
Xue and Landis 2010	USA	1	intensive	Cradle-to-market	kg boneless beef

<sup>1</sup> Because “kg live weight per year” is not comparable with “kg live weight”, we exclude the results from the two Casey and Holden papers.

<sup>2</sup> Because “kg live weight gain” is not comparable with “kg live weight”, we exclude the results from Leibig et al 2010.

<sup>3</sup> We were unable to determine the functional unit used in the two Pimentel papers.



**Table A3-2: Values from beef LCA studies used in the literature survey**

Means are shown for each category when reported in a study. Standard deviations are provided for studies that reported multiple values per category.

Publication	Management Type	GHG		Energy Mean (MJ)	Energy SD (MJ)	Land Mean (m <sup>2</sup> )	Land SD (m <sup>2</sup> )	Water Mean (liters)	Water SD (liters)
		Mean (kg CO <sub>2</sub> -eq)	GHG SD (kg CO <sub>2</sub> -eq)						
Beauchemin et al 2010	intensive	13.04							
Beckett and Oltjen 1993	intensive							3,682	
Capper 2011	intensive	17.95		9.64		61.06		1,763	
Casey and Holden 2006a	managed pasture	10.38	2.58						
Casey and Holden 2006b	managed pasture	11.65	2.54			34.48	13.56		
Cederberg and Stådig 2003	extensive	22.30		25.90		78.00			
Cederberg et al 2009a	extensive	41.00		17.50		250.00			
Cederberg et al 2009b	intensive	15.53							
Eady et al 2011	extensive	27.58	5.22					153	
Edwards-Jones et al 2009	intensive	15.50							
	extensive	47.60							
Foley et al 2011	managed pasture	21.60	1.73						
Hoekstra and Chapagain 2007	intensive							13,193	
	mixed							15,497	
Liebig et al 2010	managed pasture	0.50	37.48						
	extensive	-145.00							
Nguyen et al 2010	intensive	17.93	1.95	43.73	3.87	18.63	3.52		
	managed pasture	27.30		59.20		42.90			
Ogino et al 2004	intensive	8.25		45.43					
Ogino et al 2007	intensive	19.85		70.23					

Publication	Management Type	GHG		Energy Mean (MJ)	Energy SD (MJ)	Land Mean (m2)	Land SD (m2)	Water Mean (liters)	Water SD (liters)
		Mean (kg CO2-eq)	SD (kg CO2-eq)						
Pelletier et al 2010	intensive	15.51	0.95	41.56	4.82	91.05	9.62		
	managed pasture	19.19		48.40		120.06			
Peters et al 2010a	extensive	9.85	2.33	26.55	3.18				
	intensive	10.00	0.28	28.60	1.27				
Peters et al 2010b	extensive							34	9
	intensive							502	54
Phetteplace et al 2001	intensive	6.68	1.24						
Pimentel et al 1997	intensive							105,400	
	extensive							200,000	
Pimentel et al 2004	intensive							43,000	
	extensive							160,000	
Roy et al 2012	intensive	35.60							
Subak 1999	extensive	6.90							
	intensive	9.10							
Vergé et al 2008	intensive	10.37							
Veysset et al 2010	extensive	16.00	1.03	30.38	1.14				
	managed pasture	16.55	2.44	28.05	1.84				
Weidema et al 2008	mixed	28.70		276.00		58.90			
Williams et al 2006	managed pasture	19.77	4.94	28.83	11.34	34.63	9.98		
Xue and Landis 2010 <sup>1</sup>	intensive								

<sup>1</sup> Xue and Landis 2010 report values for eutrophication and acidification, but not GHG, Energy, Water, or Land Use.

## Appendix 4: Additional Tables and Figures for Chapter 4

**Table A4-1: Standardization of plant species and non-plant descriptors over surveys in all years**

<b>Recorded Name</b>	<b>USDA Standardized Name</b>
<b>Acer negundo</b>	Acer negundo
<b>Achillea millefolium(lanulosa)</b>	Achillea millefolium
<b>Achillea millefolium(Lanulosa)</b>	Achillea millefolium
<b>Agastache foeniculum</b>	Agastache foeniculum
<b>Agropyron repens</b>	Elymus repens
<b>Agrostis alba</b>	Agrostis gigantea
<b>Agrostis scabra</b>	Agrostis scabra
<b>Allium stellatum</b>	Allium stellatum
<b>Ambrosia artemisiifolia elatior</b>	Ambrosia artemisiifolia
<b>Ambrosia coronopifolia</b>	Ambrosia psilostachya
<b>Amelanchier humilis</b>	Amelanchier humilis
<b>Amelanchier sp.</b>	Amelanchier humilis
<b>Amorpha canescens</b>	Amorpha canescens
<b>Amphicarpa bracteata</b>	Amphicarpaea bracteata
<b>Andropogon gerardi</b>	Andropogon gerardii
<b>Anemone cylindrica</b>	Anemone cylindrica
<b>Antennaria neglecta</b>	Antennaria howellii
<b>Aristida basiramea</b>	Aristida basiramea
<b>Aristida tuberculosa</b>	Aristida tuberculosa
<b>Artemisia (Caudata) campestris</b>	Artemisia campestris
<b>Artemisia ludoviciana</b>	Artemisia ludoviciana
<b>Asclepias ovalifolia</b>	Asclepias ovalifolia
<b>Asclepias syriaca</b>	Asclepias syriaca
<b>Asclepias tuberosa</b>	Asclepias tuberosa
<b>Asclepias verticillata</b>	Asclepias verticillata
<b>Asclepias viridiflora</b>	Asclepias syriaca
<b>Aster azureus</b>	Symphyotrichum oolentangiense
<b>Aster ericoides</b>	Symphyotrichum ericoides
<b>Aster junciformis</b>	Symphyotrichum boreale
<b>Aster novae-angliae</b>	Symphyotrichum novae-angliae
<b>Aster simplex</b>	Symphyotrichum lanceolatum
<b>Aster sp.</b>	Aster sp.
<b>Astragalus canadensis</b>	Astragalus canadensis
<b>Baptisia leucantha</b>	Baptisia alba

<b>Bare ground</b>	Non-plant Bare ground
<b>Betula papyrifera</b>	Betula papyrifera
<b>Bouteloua curtipendula</b>	Bouteloua curtipendula
<b>Bouteloua hirsuta</b>	Bouteloua hirsuta
<b>Bouteloua sp.</b>	Bouteloua hirsuta
<b>Bromus inermis</b>	Bromus inermis
<b>bucket</b>	Non-plant bucket
<b>Calamagrostis canadensis</b>	Calamagrostis canadensis
<b>Calamagrostis inexpansa</b>	Calamagrostis canadensis
<b>Calamovilfa longifolia</b>	Calamovilfa longifolia
<b>Campanula rotundifolia</b>	Campanula rotundifolia
<b>Carex muhlenbergii</b>	Carex sp.
<b>carex sp.</b>	Carex sp.
<b>Carex sp.</b>	Carex sp.
<b>Carex stricta</b>	Carex sp.
<b>Ceanothus americanus</b>	Ceanothus americanus
<b>Celastrus scandens</b>	Celastrus scandens
<b>Chenopodium album</b>	Chenopodium album
<b>Chenopodium leptophyllum</b>	Chenopodium leptophyllum
<b>Cirsium arvense</b>	Cirsium arvense
<b>Comandra richardsiana</b>	Comandra umbellata
<b>Coreopsis palmata</b>	Coreopsis palmata
<b>Cornus racemosa</b>	Cornus racemosa
<b>Cornus sp.</b>	Cornus racemosa
<b>Corylus americanus</b>	Corylus americana
<b>Crepis tectorum</b>	Crepis tectorum
<b>Cyperus filiculmis</b>	Cyperus sp.
<b>Cyperus schweinitzii</b>	Cyperus sp.
<b>Cyperus sp.</b>	Cyperus sp.
<b>Delphinium virescens</b>	Delphinium carolinianum
<b>Desmodium canadense</b>	Desmodium canadense
<b>Digitaria ischaemum</b>	Digitaria ischaemum
<b>Digitaria sanguinalis</b>	Digitaria sanguinalis
<b>Disturbance</b>	Non-plant Disturbance
<b>Elymus canadensis</b>	Elymus canadensis
<b>Equisetum laevigatum</b>	Equisetum laevigatum
<b>Equisetum sp.</b>	Equisetum laevigatum
<b>Eragrostis spectabilis</b>	Eragrostis spectabilis
<b>Erechtites hieracifolia</b>	Erechtites hieraciifolia
<b>Erigeron canadensis</b>	Conyza canadensis

<b>Erigeron strigosus</b>	Erigeron strigosus
<b>Euphorbia corollata</b>	Euphorbia corollata
<b>Euphorbia geyeri</b>	Chamaesyce glyptosperma
<b>Euphorbia glyptosperma</b>	Chamaesyce glyptosperma
<b>Forb seedlings</b>	Miscellaneous forbs
<b>Fragaria sp.</b>	Fragaria virginiana
<b>Fragaria virginiana</b>	Fragaria virginiana
<b>Fungi</b>	Non-plant Fungi
<b>Galium boreale</b>	Galium boreale
<b>Gentiana andrewsii</b>	Gentiana andrewsii
<b>Gnaphalium obtusifolium</b>	Pseudognaphalium helleri
<b>gopher mound</b>	Non-plant gopher mound
<b>grass seedling</b>	Miscellaneous grasses
<b>Habenaria viridis</b>	Platanthera lacera
<b>Helianthemum bicknellii</b>	Helianthemum bicknellii
<b>Helianthus giganteus</b>	Helianthus giganteus
<b>Helianthus laetiflorus</b>	Helianthus pauciflorus
<b>Helianthus sp.</b>	Helianthus pauciflorus
<b>Heuchera richardsonii</b>	Heuchera richardsonii
<b>Hieracium aurantiacum</b>	Hieracium aurantiacum
<b>Hieracium longipilum</b>	Hieracium longipilum
<b>Koeleria cristata</b>	Koeleria macrantha
<b>Lactuca canadensis</b>	Lactuca sp.
<b>Lactuca sp.</b>	Lactuca sp.
<b>Lathyrus venosus</b>	Lathyrus venosus
<b>Lechea stricta</b>	Lechea stricta
<b>Leptoloma cognatum</b>	Digitaria cognata
<b>Lespedeza capitata</b>	Lespedeza capitata
<b>Liatris aspera</b>	Liatris aspera
<b>Lithospermum canescens</b>	Lithospermum canescens
<b>Lithospermum caroliniense</b>	Lithospermum caroliniense
<b>Lithospermum sp.</b>	Lithospermum sp.
<b>Lobelia spicata</b>	Lobelia spicata
<b>Lupinus perennis</b>	Lupinus perennis
<b>Lychnis alba</b>	Silene latifolia
<b>Lysimachia ciliata</b>	Lysimachia ciliata
<b>Mimulus ringens</b>	Mimulus ringens
<b>Miscellaneous grasses</b>	Miscellaneous grasses
<b>Miscellaneous herbs</b>	Miscellaneous forbs
<b>Miscellaneous legumes</b>	Miscellaneous legumes

<b>Miscellaneous litter</b>	Non-plant Miscellaneous litter
<b>Miscellaneous sedges</b>	Miscellaneous sedges
<b>Miscellaneous woody plants</b>	Miscellaneous woody plants
<b>Mollugo verticillata</b>	Mollugo verticillata
<b>Monarda fistulosa</b>	Monarda fistulosa
<b>Mosses &amp; lichens</b>	Non-plant Mosses & lichens
<b>Muhlenbergia racemosa</b>	Muhlenbergia glomerata
<b>Nepeta cataria</b>	Nepeta cataria
<b>Oenothera biennis</b>	Oenothera biennis
<b>Osmunda claytonii</b>	Osmunda claytoniana
<b>Oxalis sp.</b>	Oxalis sp.
<b>Oxybaphus hirsutus</b>	Mirabilis hirsuta
<b>Panicum capillare</b>	Panicum capillare
<b>Panicum lanuginosum</b>	Dichanthelium villosissimum
<b>Panicum oligosanthes</b>	Dichanthelium oligosanthes
<b>Panicum perlongum</b>	Dichanthelium linearifolium
<b>Panicum praecocious</b>	Dichanthelium villosissimum
<b>Panicum sp.</b>	Dichanthelium sp.
<b>Panicum virgatum</b>	Panicum virgatum
<b>Parthenocissus inserta</b>	Parthenocissus quinquefolia
<b>Parthenocissus quinquefolia</b>	Parthenocissus quinquefolia
<b>Paspalum ciliatifolium</b>	Paspalum setaceum
<b>Penstemon gracilis</b>	Penstemon grandiflorus
<b>Penstemon grandiflorus</b>	Penstemon grandiflorus
<b>Petalostemum candidum</b>	Dalea candida
<b>Petalostemum purpureum</b>	Dalea purpurea
<b>Petalostemum sp.</b>	Dalea sp.
<b>Petalostemum villosum</b>	Dalea villosa
<b>Phlox pilosa</b>	Phlox pilosa
<b>Physalis heterophylla</b>	Physalis heterophylla
<b>Physalis virginiana</b>	Physalis hispida
<b>Plantago major</b>	Plantago major
<b>Platanthera lacera</b>	Platanthera lacera
<b>Poa compressa</b>	Poa pratensis
<b>Poa palustris</b>	Poa pratensis
<b>Poa pratensis</b>	Poa pratensis
<b>Polygala polygama</b>	Polygala polygama
<b>Polygala sanguinea</b>	Polygala sanguinea
<b>Polygonatum biflorum</b>	Maianthemum sp.
<b>Polygonum convolvulus</b>	Polygonum convolvulus

<b>Polygonum tenue</b>	Polygonum tenue
<b>Populus deltoides</b>	Populus deltoides
<b>Populus tremuloides</b>	Populus tremuloides
<b>Portulaca oleracea</b>	Portulaca oleracea
<b>Potentilla argentea</b>	Potentilla arguta
<b>Potentilla arguta</b>	Potentilla arguta
<b>Potentilla recta</b>	Potentilla recta
<b>Potentilla simplex</b>	Potentilla simplex
<b>Potentilla sp.</b>	Potentilla sp.
<b>Prunus pumila</b>	Prunus sp.
<b>Prunus serotina</b>	Prunus sp.
<b>Prunus sp.</b>	Prunus sp.
<b>Pteridium aquilinum</b>	Pteridium aquilinum
<b>Pycnanthemum virginianum</b>	Pycnanthemum virginianum
<b>Quercus borealis</b>	Quercus ellipsoidalis
<b>Quercus borealis-ellipsoidalis</b>	Quercus ellipsoidalis
<b>Quercus ellipsoidalis</b>	Quercus ellipsoidalis
<b>Quercus macrocarpa</b>	Quercus macrocarpa
<b>Ranunculus rhomboideus</b>	Ranunculus rhomboideus
<b>Rhus glabra</b>	Rhus glabra
<b>Rhus radicans</b>	Toxicodendron rydbergii
<b>Rosa arkansana</b>	Rosa arkansana
<b>Rubus allegheniensis</b>	Rubus sp.
<b>Rubus sp.</b>	Rubus sp.
<b>Rudbeckia hirta</b>	Rudbeckia hirta
<b>Rudbeckia serotina</b>	Rudbeckia hirta
<b>Rumex acetosella</b>	Rumex acetosella
<b>Salix humilis</b>	Salix humilis
<b>Salix sp.</b>	Salix humilis
<b>Sapinaria officinalis</b>	Gentiana andrewsii
<b>Schizachyrium scoparium</b>	Schizachyrium scoparium
<b>Scleria triglomerata</b>	Carex sp.
<b>Scutellaria galericulata</b>	Scutellaria parvula
<b>Scutellaria lateriflora</b>	Scutellaria parvula
<b>Scutellaria parvula</b>	Scutellaria parvula
<b>Secale cereale</b>	Elymus canadensis
<b>Seedlings</b>	Miscellaneous vegetation
<b>Setaria lutescens (Glauc)</b>	Setaria pumila
<b>Setaria viridis</b>	Setaria viridis
<b>Sisyrinchium campestre</b>	Sisyrinchium campestre

<b>Smilacina racemosa</b>	Maianthemum sp.
<b>Smilacina stellata</b>	Maianthemum sp.
<b>Solidago altissima</b>	Solidago speciosa
<b>Solidago gigantea</b>	Solidago missouriensis
<b>Solidago graminifolia</b>	Euthamia graminifolia
<b>Solidago missouriensis</b>	Solidago missouriensis
<b>Solidago nemoralis</b>	Solidago nemoralis
<b>Solidago rigida</b>	Oligoneuron rigidum
<b>Solidago sp.</b>	Solidago sp.
<b>Solidago speciosa</b>	Solidago speciosa
<b>Sorghastrum nutans</b>	Sorghastrum nutans
<b>Sporobolus cryptandrus</b>	Sporobolus cryptandrus
<b>Sporobolus heterolepis</b>	Sporobolus heterolepis
<b>Stachys palustris</b>	Stachys palustris
<b>Stellaria longifolia</b>	Stellaria longifolia
<b>Stellaria media</b>	Stellaria media
<b>Stipa spartea</b>	Hesperostipa spartea
<b>Stipa spicata</b>	Hesperostipa spartea
<b>Taraxicum officinalis</b>	Taraxacum officinale
<b>thatch ant nest</b>	Non-plant thatch ant nest
<b>Tradescantia bracteata</b>	Tradescantia occidentalis
<b>Tradescantia occidentalis</b>	Tradescantia occidentalis
<b>Trifolium sp.</b>	Trifolium sp.
<b>Unknown</b>	Miscellaneous vegetation
<b>unknown seedling</b>	Miscellaneous vegetation
<b>Vaccinium angustifolium</b>	Vaccinium angustifolium
<b>Verbena stricta</b>	Verbena stricta
<b>Veronicastrum virginicum</b>	Veronicastrum virginicum
<b>Vicia villosa</b>	Vicia villosa
<b>Viola pedata</b>	Viola pedatifida
<b>Viola pedatifida</b>	Viola pedatifida
<b>Viola sagittata</b>	Viola sagittata
<b>Viola sp.</b>	Viola pedatifida
<b>Woody</b>	Non-plant Woody debris
<b>Woody debris</b>	Non-plant Woody debris
<b>Zizia aptera</b>	Zizia aptera
<b>Zizia aurea</b>	Zizia aurea



**Table A4-2: Effect of seeding species that were already relatively common in 1991**

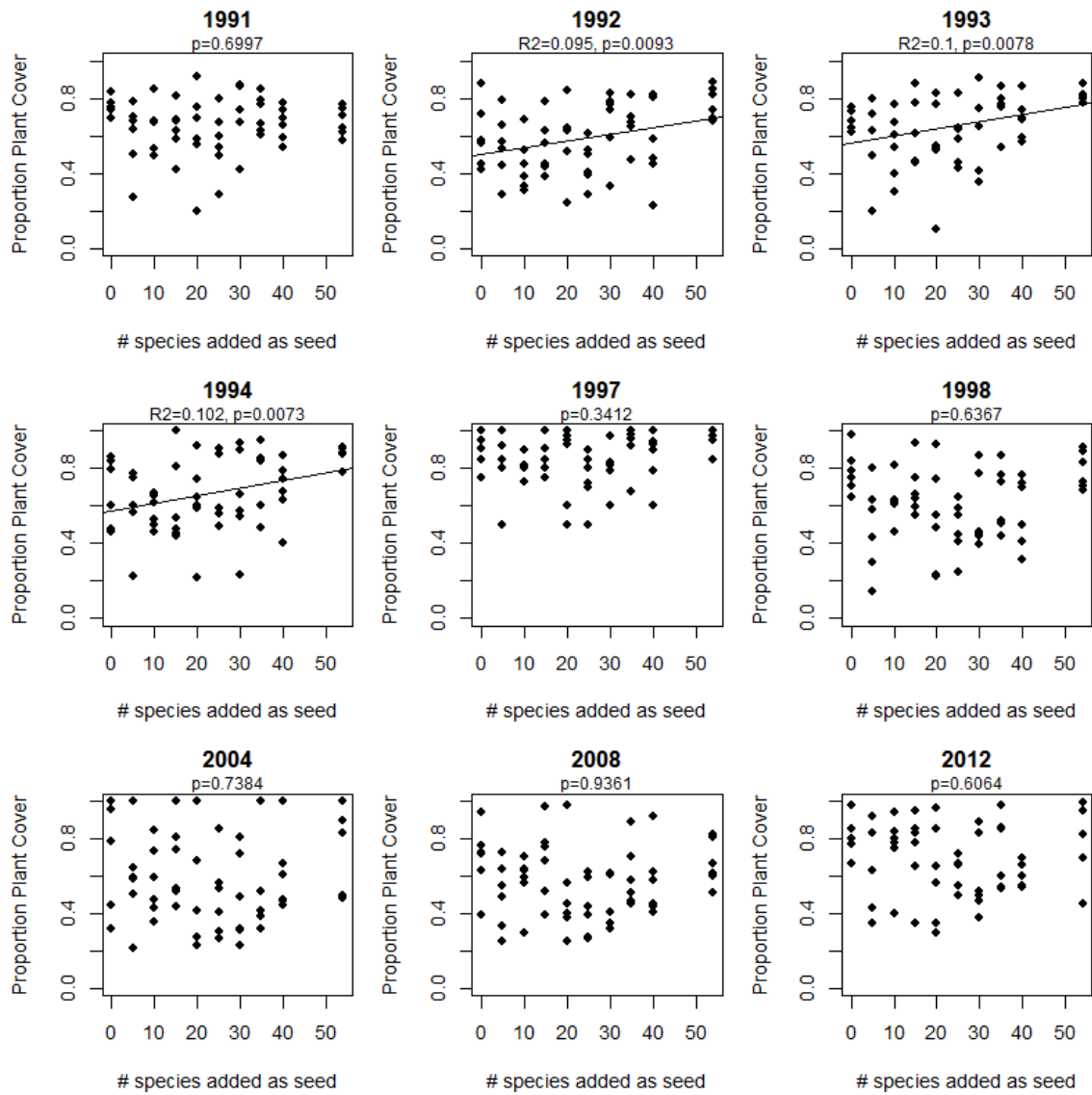
Differences were calculated between the cover of a given species in the given year and its cover in 1991. Data were split into two groups: those plots that had received the species as seed and those that had not. A two-tailed t-test was performed to determine whether there was a significant difference in change in cover between the two. A plus (+) indicates that species cover significantly increased with seeding; a minus (-) indicates that cover significantly decreased with seeding.

	1992	1993	1994	1997	1998	2004	2008	2012
<b>Amorpha canescens</b>								
<b>Andropogon gerardii</b>								
<b>Anemone cylindrica</b>	+	+	+	+	+	+	+	+
<b>Asclepias tuberosa</b>		+	+	+	+	+	+	+
<b>Hesperostipa spartea</b>								
<b>Lespedeza capitata</b>	+	+	+	+	+			
<b>Liatris aspera</b>		+	+	+	+			
<b>Monarda fistulosa</b>	+	+	+	+	+	+	+	+
<b>Oenothera biennis</b>		+	+					
<b>Panicum capillare</b>								
<b>Rosa arkansana</b>								
<b>Rudbeckia hirta</b>		+	+		+			
<b>Schizachyrium scoparium</b>							+	+
<b>Setaria pumila</b>			+					
<b>Solidago nemoralis</b>								
<b>Sorghastrum nutans</b>						-		-
<b>Symphotrichum ericoides</b>								
<b>Symphotrichum oolentangiense</b>								

**Table A4-3: Diversity measures of seeded plots and over all plots**

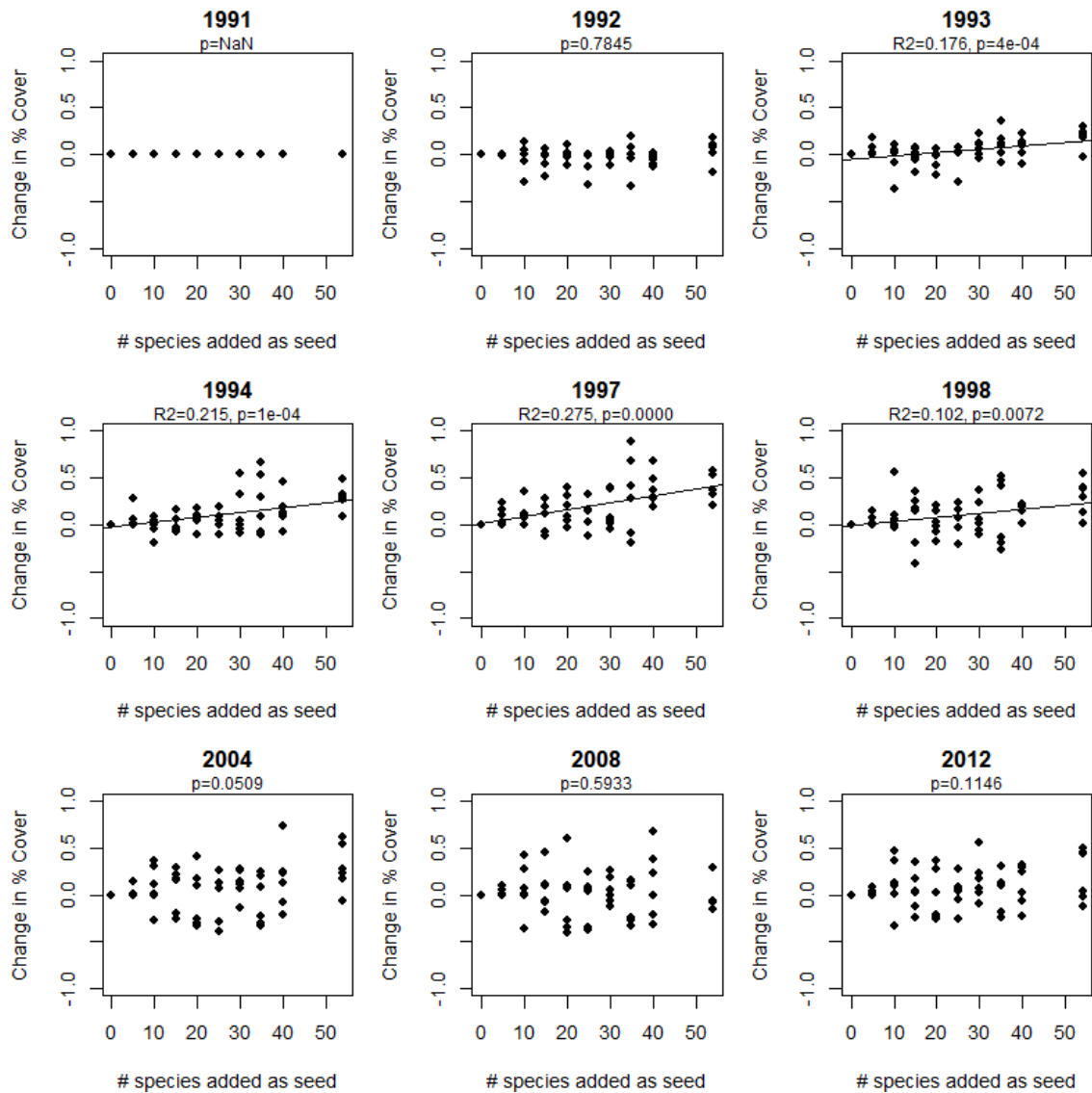
Shannon diversity is calculated as  $e^{H'}$  where  $H' = -\sum p \ln p$ , the summation is over all species, and  $p$  is a species' proportion of cover.

<b>Year</b>	<b>Plot Richness (Mean ± SD)</b>	<b>Plot Shannon Diversity (Mean ± SD)</b>	<b>Species Richness Over All Plots</b>	<b>Shannon Diversity Over All Plots</b>
<b>1991</b>	14.6 ± 4.2	6.2 ± 2.6	80	18.7
<b>1992</b>	12.0 ± 4.7	6.2 ± 2.7	80	25.0
<b>1993</b>	23.4 ± 7.7	10.8 ± 4.5	90	34.2
<b>1994</b>	18.1 ± 5.8	8.2 ± 2.8	104	30.4
<b>1997</b>	16.6 ± 4.3	9.0 ± 3.2	96	33.2
<b>1998</b>	18.3 ± 5.5	6.4 ± 2.4	104	25.5
<b>2004</b>	17.1 ± 4.1	9.0 ± 3.2	105	36.5
<b>2008</b>	17.0 ± 4.1	9.3 ± 3.5	93	32.3
<b>2012</b>	18.2 ± 3.6	8.7 ± 2.4	103	30.7



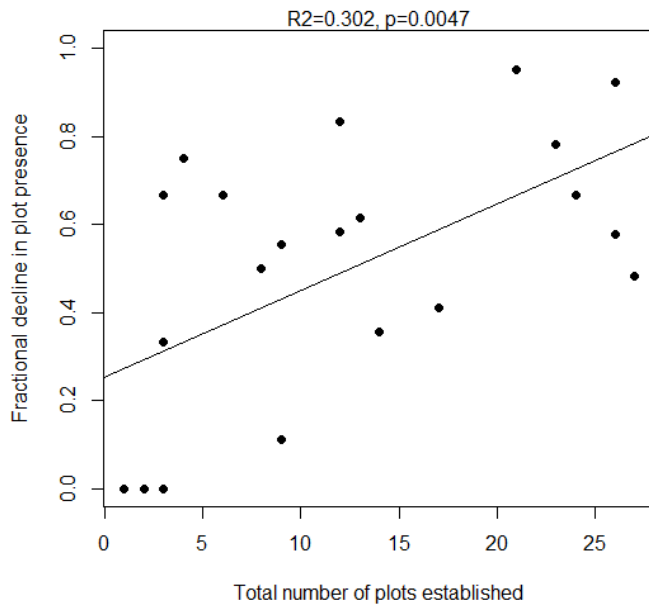
**Figure A4-1: The effect of seeding treatments on total plant cover**

Seeding treatments initially increase total plant cover. The year 1995 also showed a positive relationship (Tilman 1997 Figure 3A). The relationship disappears by 1997.



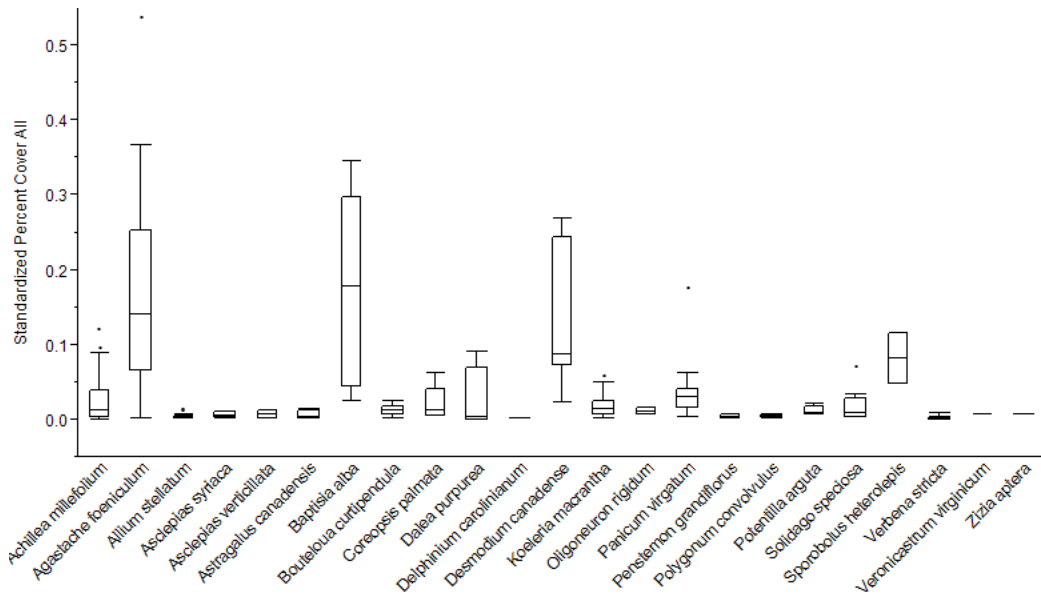
**Figure A4-2: Effect of seeding treatment on the change in percent cover of seeded species between the given year and 1991**

There was a positive relationship in 1995 (Tilman 1997, Figure 3B). The positive relationship disappears by 2004.



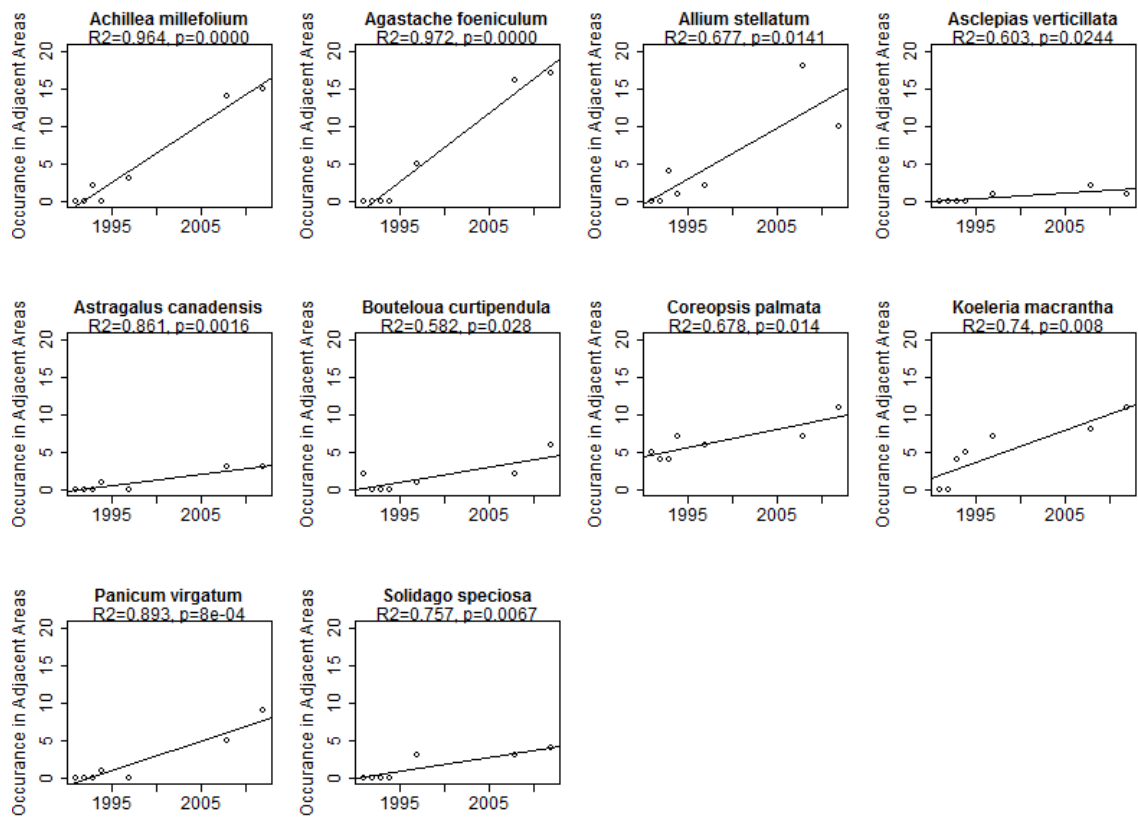
**Figure A4-3: Species that established in more seeded plots had a greater rate of decline in the number of plots in which they were found by 2012**

Species which were no longer seen in any seeded plot in 2012 are excluded.

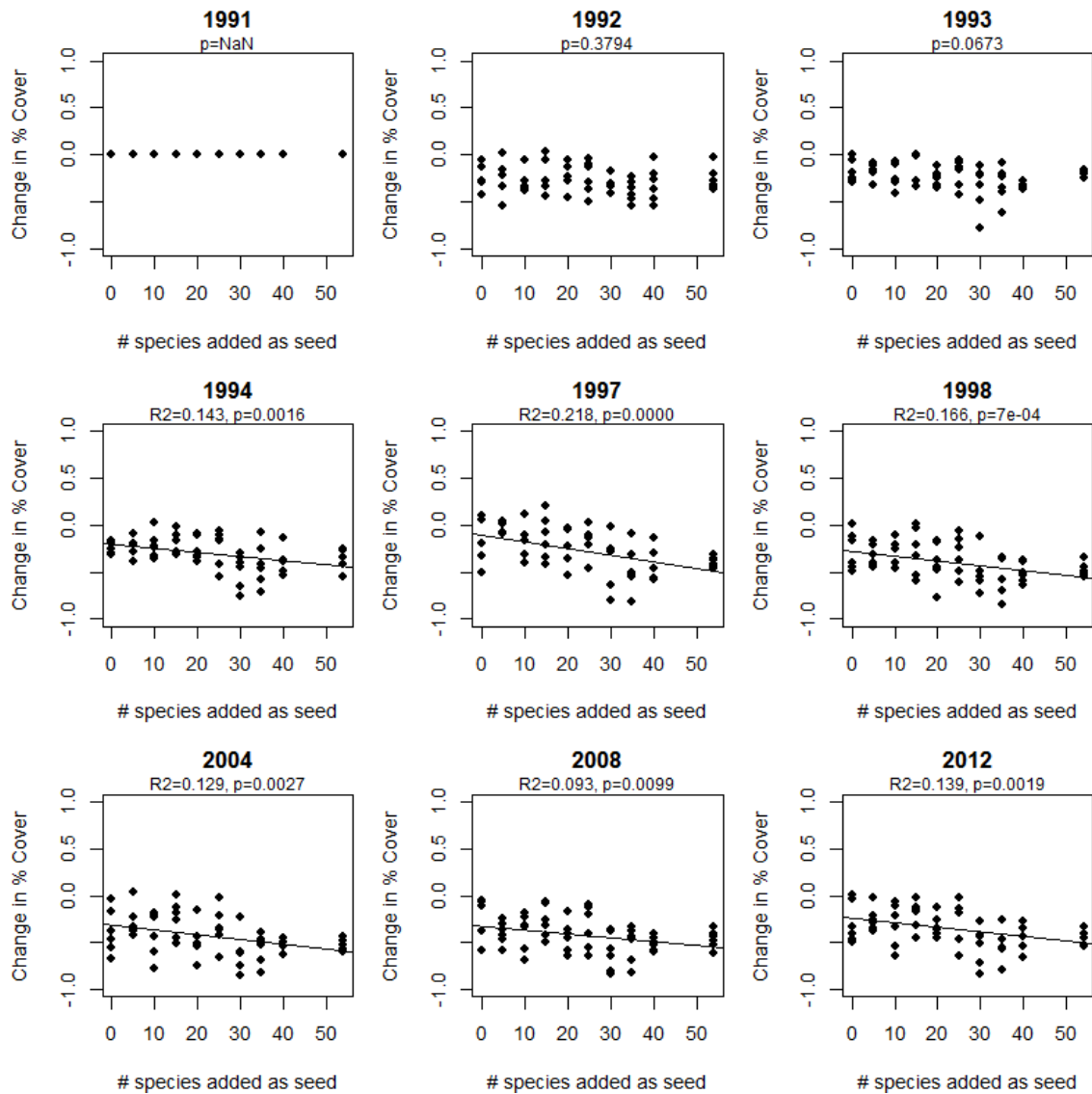


**Figure A 4-4: Cover in 2012 of persisting seeded species that were absent or rare in 1991**

Boxes show medians and quartiles. Whiskers reach at most 1.5 times the interquartile range.



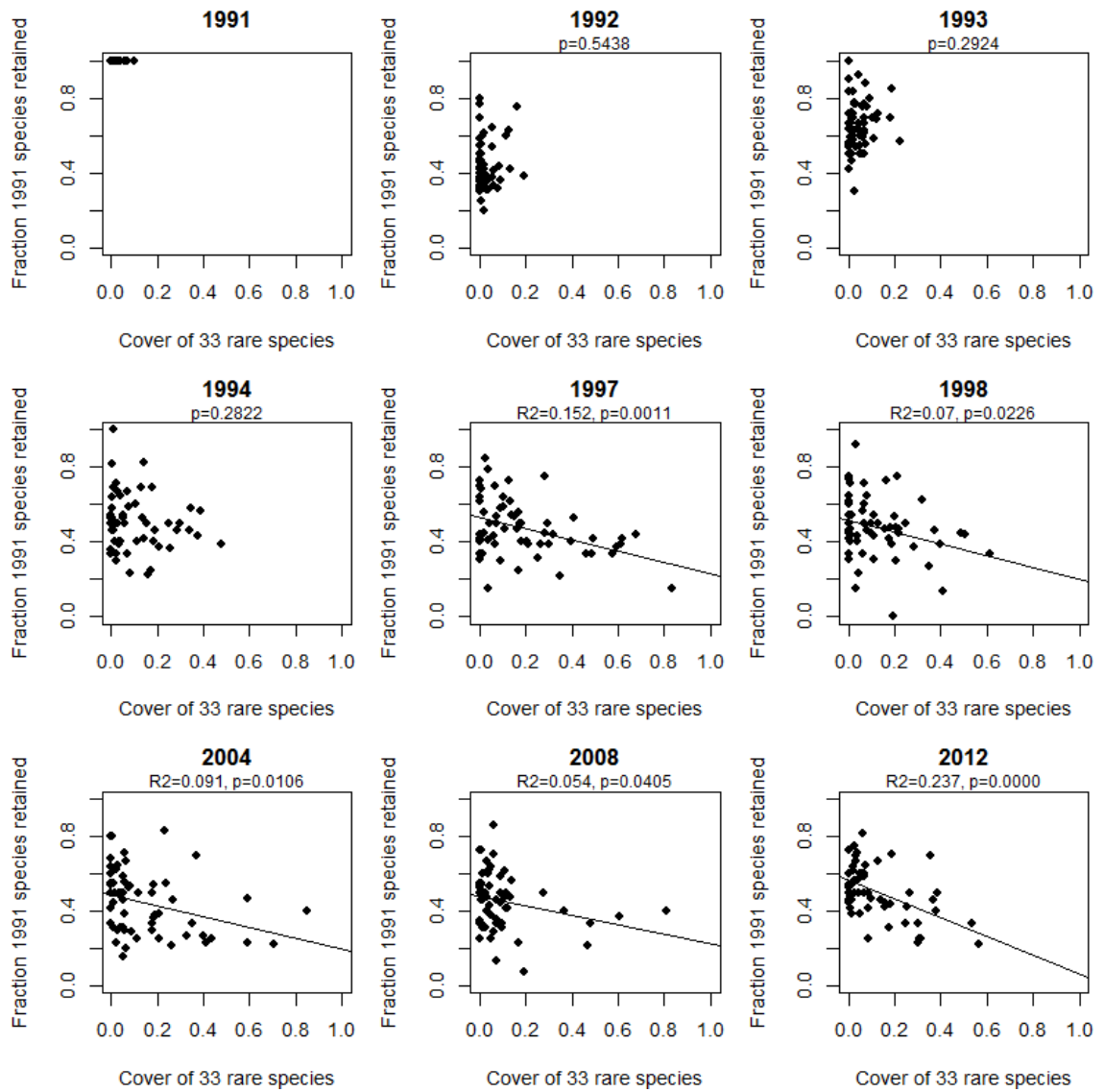
**Figure A4-5: Ten species showed increases over time in the number of adjacent areas in which they were found**



**Figure A4-6: Effect of seeding treatment on change in percent cover of species that were present in 1991 (and not also seeded)**

The change in cover is calculated as between the given year and 1991. Cover includes both plants and abiotic measures (such as ‘bare ground’), so the cover of species in 1991 would not necessarily decline with the presence of new species if total plant cover increased.





**Figure A4-7: Summed cover of initially rare species (n=33) causes declines in the species richness of species that were present in 1991 and were not seeded**

## Appendix 5: SimSimba Parameters for Chapter 5

	Parameter	Value	References
Survivorship	Cub under 6 months	0.707 / half-year	(Smuts et al. 1978b, Funston et al. 2003, Ferreira and Funston 2010)
	Cub 6-12 months	0.775 / half-year	
	Cub 12-24 months	0.906 / half-year	
	Subadult male (2-5 years)	0.877 / half-year	
	Subadult female (2-5 years)	0.964 / half-year	
	Adult male	0.949 / half-year	
	Adult resident female	0.929 / half-year	(Whitman et al. 2004)
	Orphan cub 0-24 months	0.5 / half-year	
	Adult homeless female	0.5 / half-year	
	Coalition fight, winning male	0.97 / event	
	Coalition fight, defending losing male	0.4 / event	
	Coalition fight, attacking losing male	0.5 / event	
	Coalition takeover, cub under 6 months	0.01 / event	(Packer and Pusey 1983)
	Coalition takeover, cub 6-12 months	0.25 / event	
	Coalition takeover, cub 12-24 months	0.65 / event	
Coalition takeover, defending mother	0.9 / event		
	Maximum age	16 years	(Smuts et al. 1978a, 1980, Ferreira and Funston 2010)
	Reproductive age	3.5 years	(Smuts et al. 1978b)
	Maximum reproductive females per pride	7	(Smuts 1976, Smuts et al. 1978b, Funston et al. 2003)
Litter Size	1 cub	0.05 / litter	(Smuts 1976, Smuts et al. 1978b, Funston et al. 2003)
	2 cubs	0.05 / litter	
	3 cubs	0.75 / litter	
	4 cubs	0.15 / litter	
	Cub abandoned if no pride littermates within 1 year of age	0.7 at birth	(Packer and Pusey 1984)
	Nomadic male traveling rate	3 territories / half-year	(Funston et al. 2003)
	Subadult male traveling rate	1 territory / half-year	
	Coalition (2 males) takes second territory	0.0001 / half-year	(Whitman et al. 2004)
	Coalition (3+ males) takes second territory	0.33 / half-year	
	Coalition (3+ males) takes third territory	0.01 / half-year	
	Nomadic male joins solitary resident male	0.5 / encounter	
	Nomadic male joins solitary nomadic male	1.0 / encounter	
	Nomadic male joins pair of nomadic males	0.2 / encounter	
Subadult female dispersal	1 territory away, if available	1.0	
	2 territories away, if available	0.8	
	3 territories away, if available	0.3	
	4 territories away, if available	0.15	
	5 territories away, if available	0.10	
	6 or more territories away	0	

Male Coalition Fights: The Competition Matrix specifies the fighting abilities of subadult and adult males (Starfield et al. 1981). During each fight, the strength of the coalition relative the residents (R) is determined by summing up the Q value of all pairs of combatants, using the table below, and dividing by the number of pairs. A fight value (F) is determined as (Number of attackers) / [R \* (Number of defenders)]. If F is less than 1.0, the attackers surrender without a fight. If  $1.0 \leq F \leq 1.1$ , there is a fight and the defenders win. If  $F > 1.1$ , the attackers win and take over the territory from the residents.

		Age of Attackers						
		2-3	4	5	6	7	8	9+
Age of Defenders	2-3	1.40	0.90	0.65	0.70	0.75	0.70	1.00
	4	1.65	1.40	1.00	0.85	0.75	0.70	1.00
	5	2.20	1.65	1.50	1.00	0.90	1.00	1.00
	6	2.20	2.20	2.00	1.40	1.50	1.80	1.00
	7	2.50	2.20	2.00	1.40	1.50	1.80	1.00
	8	1.65	1.40	1.30	1.20	1.25	1.40	1.00
	9	1.30	1.30	1.25	1.00	1.05	1.35	1.00
	10+	1.00	1.00	1.90	1.40	1.40	1.00	1.00

## Appendix 6: Additional Table and Figures for Chapter 5

**Table A6-1: Parameter ‘B’ is the primary control on the prevalence of bovine tuberculosis in lions**

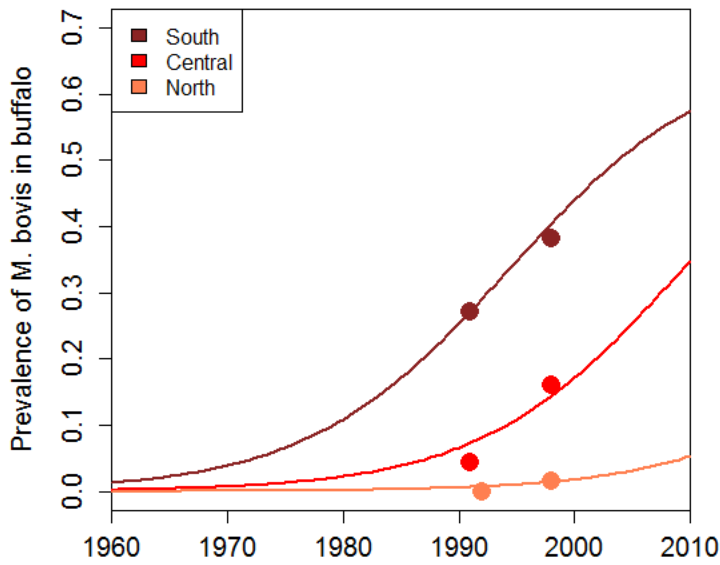
Results from multiple regression on the prevalence of bovine tuberculosis in lions in 1999 on particles in the second round ( $R^2 = 0.65$ ,  $F_{5,49994}=0.00019$ ,  $p<0.0001$ ). The second round was chosen because it had the maximum coverage of the parameter space after round one. Round one was not used because over 70% of particles in round one caused extinction in at least one region.

Parameter	Estimate	Std. Error	t value	Pr(> t )
intercept	0.316247	0.002025	156.16	<0.0001
L	0.203557	0.001724	118.06	<0.0001
B	0.468672	0.001778	263.61	<0.0001
O	0.104695	0.001660	63.08	<0.0001
E	2.705313	0.029762	90.90	<0.0001
I	-0.342846	0.002176	-157.58	<0.0001

**Table A6-2: Parameter ‘E’ is the primary driver of lion population size**

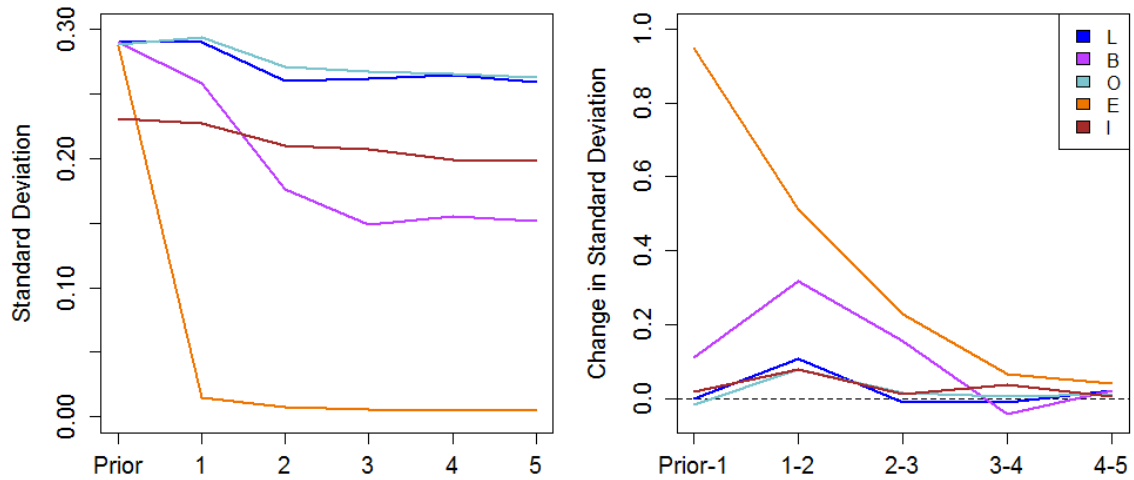
Results from multiple regression on the change in population size between 1960 and 2006 on particles in the second round ( $R^2 = 0.57$ ,  $F_{5,49994}=0.00013$ ,  $p<0.0001$ ). The second round was chosen because it had the maximum coverage of the parameter space after round one. Round one was not used because over 70% of particles in round one caused extinction in at least one region.

Parameter	Estimate	Std. Error	t value	Pr(> t )
intercept	90.299	2.295	39.35	<0.0001
L	-109.336	1.954	-55.97	<0.0001
B	-143.342	2.015	-71.15	<0.0001
O	-57.299	1.881	-30.47	<0.0001
E	-8594.821	33.723	-254.86	<0.0001
I	128.402	2.465	52.08	<0.0001



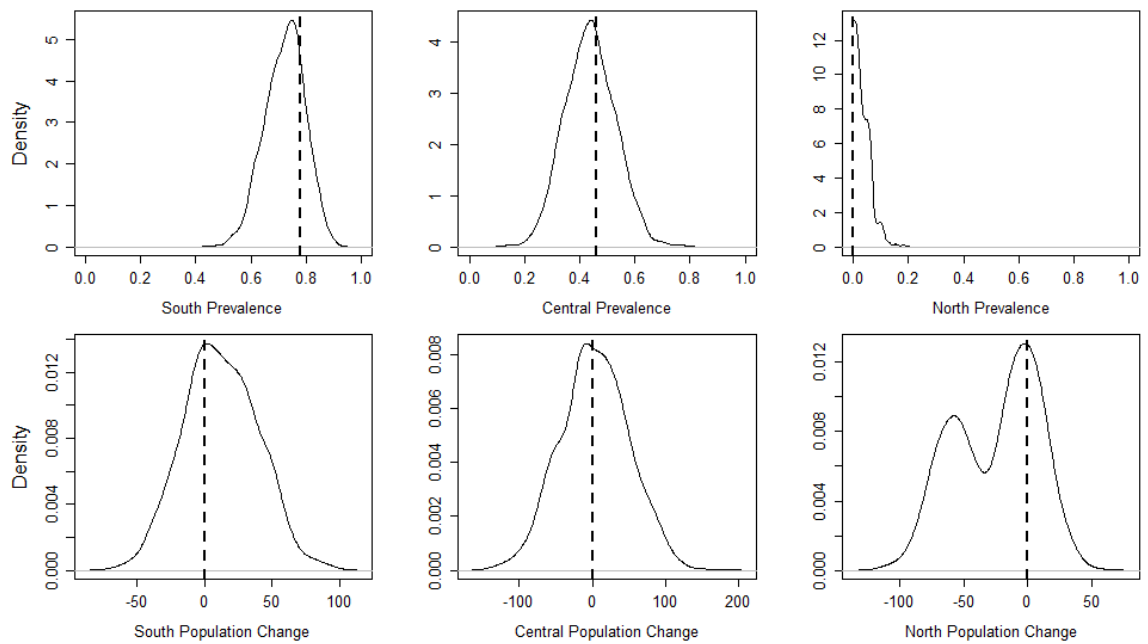
**Figure A6-1: Prevalence of bovine tuberculosis in buffalo over time**

Points indicate prevalence in the south, central, and north regions of the park; data are from (Rodwell et al. 2001). Curves show logistic fits to the data assuming the same logistic increase in each region offset by time; they were fit using equation:  $P_{r,t} = \frac{0.67}{1+e^{-[m(t-lag_r)+b]}}$  where  $P_{r,t}$  represents the prevalence of bovine tuberculosis in buffalo in region  $r$  at time  $t$ , and  $lag_r$  is the lag in years between the arrival of the disease in the south and in each successive region; by definition,  $lag_{south} = 0$ . We used least squares to solve for  $m$ ,  $b$ , and  $lag_r$ . Fitted values are  $m=0.1145$ ,  $b=-228.3475$ ,  $lag_{central}=15$ ,  $lag_{north}=37$ .



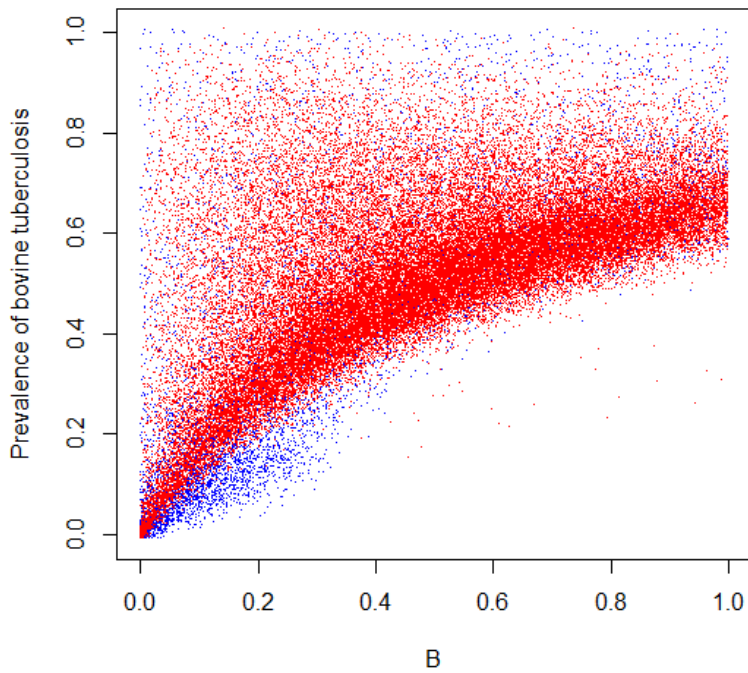
**Figure A6-2: Convergence of SMC-ABC rounds**

(left) The standard deviation of each parameter after each round of SMC-ABC. As standard deviations level off, there will be no further refinement of the parameter estimates. The parameters showing the most change, E and B, have the most impact on model outcomes. (right) The change in standard deviation of each parameter from round to round. As the change in standard deviation converge to 0, there will be no further refinement of the parameter estimates.



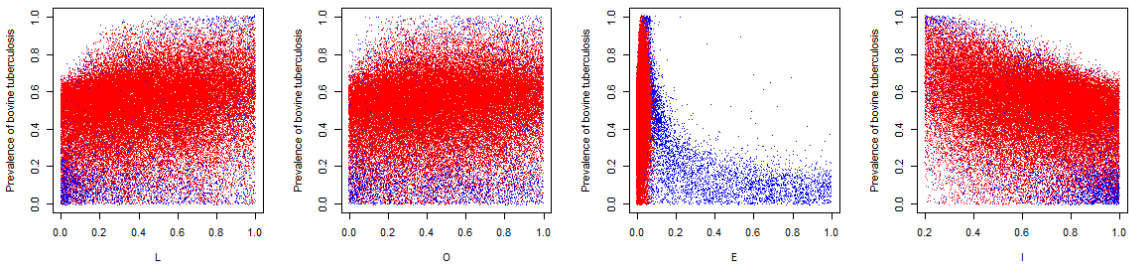
**Figure A6-3: Density distributions of the summary statistics from the final 1,000 accepted particles.**

Vertical dashed lines indicate the observed data,  $x_0$ .



**Figure A6-4: Prevalence of bovine tuberculosis in the lion population and parameter  $B$**

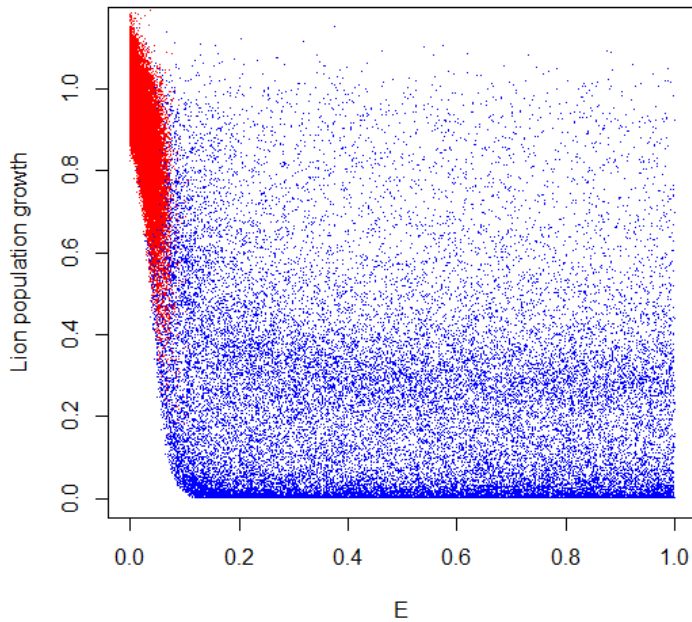
Prevalence of bovine tuberculosis in the lion population as a consequence of parameter  $B$  (the rate of transmission of the disease from buffalo) for the 7,656 particles in round 1 (blue) in which lions did not go extinct in any of the three regions and all 50,000 particles in round 2 (red).  $B$  strongly controls the prevalence of bovine tuberculosis.



**Figure A6-5: Prevalence of bovine tuberculosis in the lion population and parameters  $L$ ,  $O$ ,  $E$ , and  $I$**

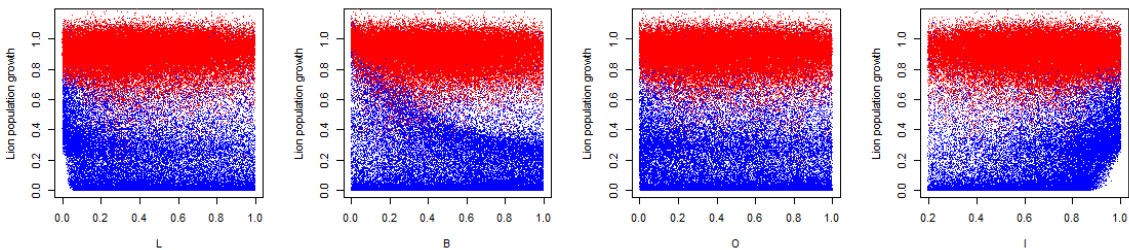
Prevalence of bovine tuberculosis in the lion population as a consequence of parameters  $L$ ,  $O$ ,  $E$ , and  $I$  for the 7,656 particles in round 1 (blue) in which lions did not go extinct in any of the three regions and all 50,000 particles in round 2 (red).





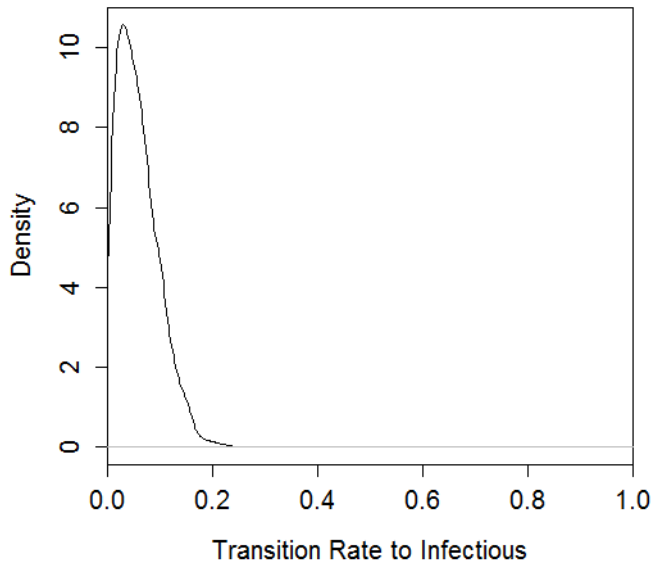
**Figure A6-6: Lion population growth between 1960 and 2006 and parameter  $E$**

Lion population growth between 1960 and 2006 as a consequence of parameter  $E$  (rate of transition from the exposed to the infectious state) for all 100,000 particles in round 1 (blue) and round 2 (red) of the ABC-SMC algorithm.  $E$  controls the lion population size; only at small values ( $E < 0.1$ ) can the lion population persist.



**Figure A6-7: Lion population growth between 1960 and 2006 and parameters  $L$ ,  $B$ ,  $O$ , and  $I$**

Lion population growth between 1960 and 2006 as a consequence of parameters  $L$ ,  $B$ ,  $O$ , and  $I$  for all 100,000 particles in round 1 (blue) and round 2 (red) of the ABC-SMC algorithm.



**Figure A6-8: Rate of transition from the exposed to the infectious state**

Median = 0.050, 95% CI = [0.003,0.151].

## **Appendix 7: Acknowledgements (continued)**

Many Cedar Creek summer interns helped with this experiment in both 2011 and 2012; I am especially thankful to Geoff Bazuin, Kylee Ehlers, Courtney Jones, Alecia Mienert, Andy Richter, Jenny Soltys, Ellen Squires, Bridget Steele, and Amanda Thompson for conducting surveys week after week. I sincerely appreciate the volunteer time and expertise that members of the Heimpel lab gave in assisting with surveys and identifying insects: Nico Desneux, Jonathan Dregni, Megan Carter, Joe Kaser, Julie Peterson, Stefanie Wolf, and George Heimpel himself. Dan Bahauddin provided invaluable technical and data support throughout.

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