AN ABSTRACT OF THE THESIS OF

Shannon B. Andrews for the degree of Master of Science in Soil Science presented on May 10, 2013
Title: Quantifying the Fertilizer Value of Algal Meal: An Evaluation of an Integrated Dairy-Anaerobic Digester-Algae Production Facility

Abstract approved:

David D. Myrold

Algae have shown great potential as a source for renewable fuels. However, current production schemes have not been able to prove a sustainable energy return on investment due in part to the high costs of nutrient addition and the energy required for drying the biomass. Integrated algae-dairy production systems have been posited as a potential solution for algal production barriers as well as a way to capture environmentally problematic nutrients excreted by animals in concentrated animal feeding operations. As the organic food industry grows, so will the need for organic nitrogen (N) fertilizers. Algal meal, the high protein co-product of algal biodiesel production, could help meet this need. This work has two objectives: 1) to quantify the fertilizer value of algal meal relative to an organic N fertilizer, feather meal, and a conventional standard, urea; 2) to show that utilizing the fertilizer value of algal meal at \$800 Mg^{-1} will allow an integrated dairy-anaerobic digester-algae production facility to be an economically viable manure management system.

A laboratory incubation was carried out to characterize carbon (C) and N mineralization of two different algal meal products. The N mineralization rate, C respiration rate, and impact on pH and soluble salts were the same for feather meal and one of the algal meals. A field trial was conducted to assess the overall crop productivity of corn grown with algal meal fertilizer, feather meal, and urea. There was no significant difference in corn ear yield or ear N uptake among any of the algal meal treatment rates (101 kg N ha^{-1}, 146 kg N ha^{-1}, 190 kg N ha^{-1}) and the high rate of feather meal or urea (190 kg N ha^{-1}) application. These results indicate that algal meal is an effective N fertilizer that should be valued relative to feather meal on a price per unit of nutrient basis. Considering environmental benefits and current policy incentives, assigning a fertilizer value of algal meal of \$800 Mg^{-1} for a 7-3-1 product adds enough income to make an integrated dairy-anaerobic digester-algae system an economically viable, sustainable farm-energy production system.

Quantifying the Fertilizer Value of Algal Meal: An Evaluation of an Integrated Dairy-Anaerobic Digester-Algae Production Facility

by
Shannon B. Andrews

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Master of Science

Presented May 10, 2013
Commencement June, 2014

Master of Science thesis of Shannon B. Andrews presented on
May 10, 2013

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Shannon B. Andrews, Author

ACKNOWLEDGEMENTS

I would like to express my sincere gratitude to everyone who I have worked with throughout my life. Each step has led me to this point.

A special thanks goes to:

My parents and family for their constant love, support, and chastisement

My friends all around the world for playing and engaging in meaningful conversations

My co-workers who showed me how to climb many different ropes

My academic committee for giving me the freedom and guidance needed to pursue this path

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Introduction

Nitrogen (N) is a primary limiting factor for plant growth in agro-ecosystems. As the organic food industry grows so will the need for organic N fertilizers. One potential organic fertilizer is algal meal, the high protein portion of algal biomass that is a co-product of algal biodiesel production. Algae are a more attractive biofuel feedstock than other materials as they offer higher biomass yield per acre of cultivation, can be grown on land and water resources that minimize competition with food and feed crops, can utilize waste water and saline waters, can recycle carbon (C) from industrial carbon dioxide (CO_2) emitters, and have more accessible forms of stored C than the lignocelluloses used for cellulosic biofuels (Benemann and Oswald, 1996; Sheehan et al., 1998; McKenzie, 2011). A major limitation to efficient production is nutrient input; namely N and phosphorus (P). In *Sustainable Development of Algal Biofuels* the committee states:

> "R&D is needed to incorporate nutrient recycling into algal biofuel production systems. The potential for combining the use of wastewater in algae cultivation and the production of a fertilizer co-product is worth further investigation."
>
> NAS, 2012

Integrated algae-dairy models have been designed in order to overcome nutrient capture and greenhouse gas (GHG) management issues faced by industrial animal agriculture and in so doing overcome the algae production barrier of

nutrient costs (Lincoln et al., 1996; Craggs et al., 2004; Mulbry et al., 2005; Woertz et al., 2009; Wang et al., 2010; Zhang et al., 2013). I have proposed a dairy-anaerobic digestion-algae system (DADA) (Fig. 1), in which the dairy-housing manure is sent to an anaerobic digester (AD) where a succession of microbes reduce the organic matter into a methane-rich biogas, heat, nutrient-rich liquid effluent, and solid compost. The biogas is used to generate electricity to operate the farm system with any remainder being sold back to the grid. The liquid effluent is diluted and used as the nutrient broth for algae production. The heat is incorporated into drying the algal biomass. The composted solids are used as bedding in the free stalls or sold into the horticultural markets. The algal biomass is extracted for fatty acid methyl esters (FAME biodiesel). From this process glycerin and algal meal are also produced.

Many agricultural by-products are used for both animal feed and organic fertilizer, often realizing a higher market value as organic fertilizers. I hypothesize that algal meal is as effective as feather meal as an organic fertilizer product on a per unit of N basis. Specifically, I hypothesize that algal meal will have a similar N mineralization rate, microbial respiration rate, and extracellular enzyme production, yield of sweet corn, and not have any negative impacts from the salt concentration (electrical conductivity EC). If this is true, a recalculation of the energy return on investment (EROI) of an integrated DADA production facility with the new value of algal meal priced relative to feather meal has the potential to show an economically viable, sustainable farm-energy production system.

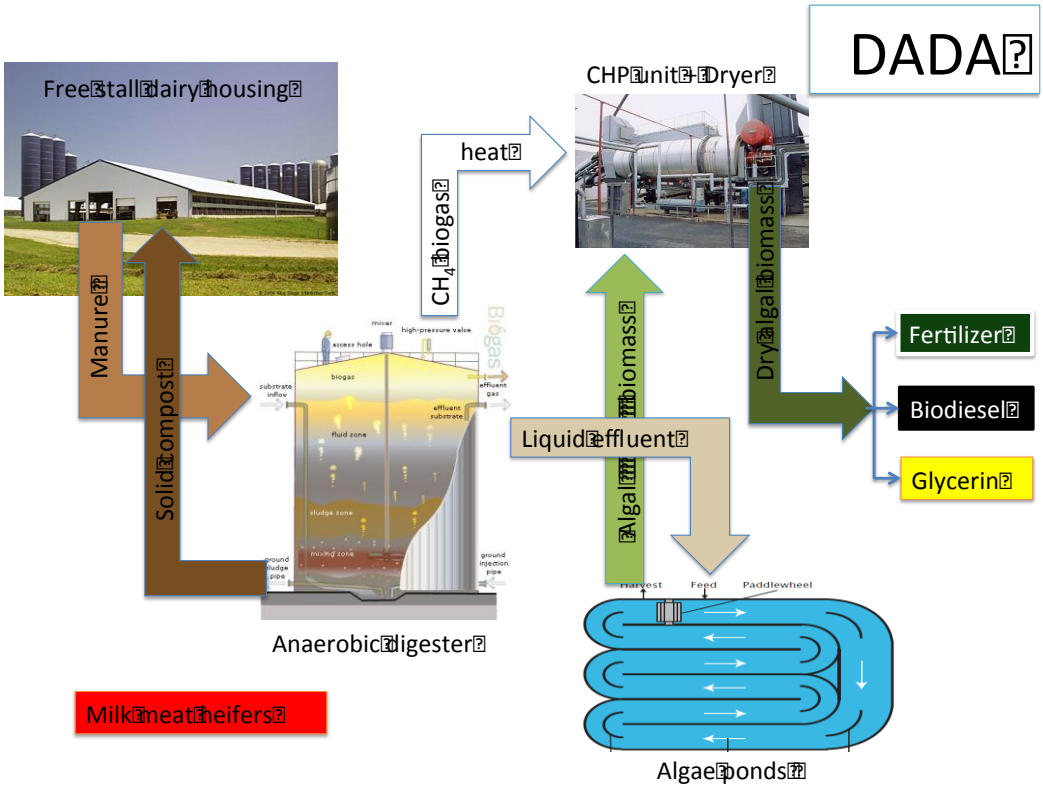

Fig. 1. Integrated Dairy-Anaerobic Digester-Algae Production Schematic. Arrows indicate flow of materials. Rectangular boxes represent saleable products. The barn, anaerobic digester, algal ponds, and CHP unit and dryer are representations of the system components.

CHAPTER 1
LITERATURE REVIEW

Introduction to the Problems

Nitrogen (N) is an essential nutrient required by all known organisms. In agro-ecosystems, N is a primary limiting factor for plant growth. The United States Department of Agriculture (USDA) Economic Research Service (ERS) reported that 11,144,000 Mg of N were applied to fields in 2010, which was roughly three times higher than phosphorus (P) or potassium (K) inputs. In the same year, the organic food industry grew 7.7% to contribute $26.7 billion in sales into the US economy (Organic Trade Association, 2011). Certified organic land reached more than 2,000,000 ha in 2008 growing at a 15% annual rate from 2002 to 2008 (USDA ERS, www.ers.usda.gov/data/organic). As the organic food industry grows, so will the need for organic N fertilizers. One potential organic fertilizer is algal meal, the high protein portion of algal biomass that is a co-product of algal biodiesel production.

Eukaryotic microalgae and cyanobacteria are responsible for more than 40% of net primary productivity on Earth (McKenzie, 2011; NAS, 2012). Their high productivity and rapid growth rates led to interest in their development as a renewable fuel source since before World War II (Shelef and Soeder, 1980). After the energy crisis in the early and mid-1970s, algal biofuel production became a topic of national research interest. From 1978 until 1996, the US Department of Energy invested in the Aquatic Species Program, compiling a vast database on algal species

suitable for production, growth efficiency, resource utilization, and possible production, harvesting, and extraction methods (Sheehan et al., 1998). They concluded that algae are a more attractive biofuel feedstock than others as they offer higher biomass yield per acre of cultivation, can be grown on land and water resources that minimize competition with food and feed crops, can utilize waste water and saline waters, can recycle carbon (C) from industrial carbon dioxide (CO_2) emitters, and have more accessible forms of stored C than the lignocelluloses used for cellulosic biofuels (Benemann and Oswald, 1996; Sheehan et al., 1998; US DOE, 2010; McKenzie, 2011). A major limitation to efficient production is nutrient input; namely N and P. In *Sustainable Development of Algal Biofuels* the committee states:

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> NAS, 2012

Manufacture of urea for algal biodiesel production has been demonstrated to be the major contributor to the total greenhouse gas (GHG) emissions of the system (Clarens et al., 2010; NAS, 2012).

Algal fuel production research has focused on species selection, production efficiency, and refining harvesting procedures. Although this work has proven great potential, large-scale production schemes have only realized a 0.13-3.33 Energy Return on Investment (EROI), where a minimum EROI of 3 is considered sustainable (NAS, 2012). These models do not include an accurate market value of the co-product. If any price is offered, it is often priced as the animal feed value, equivalent

to other biofuel co-products, such as dried distillers grains (Chisti, 2007; NAS, 2012).

Many agricultural by-products are used for both animal feed and organic fertilizer: blood meal, feather meal, fish meal, shrimp meal, cottonseed meal, and alfalfa meal, to name a few (Tavoletti, 2013). Animal producers have many feed options and tend to operate on small margins with the feed bill being their largest variable cost. Organic plant producers also manage tight margins but they have limited choices for organic N, a very short fertilization window, and can see severe negative economic consequences if their crops do not receive adequate N. Consequently, organic growers are willing to pay more for high N or protein products than animal producers, particularly for high value crops (Skinner, 2013).

I hypothesize that algal meal is as effective as feather meal as an organic fertilizer product on a per unit of N basis. Specifically, I hypothesize that algal meal will have a similar N mineralization rate, CO_2-C respiration rate, extracellular enzyme production, yield of sweet corn, and not have any negative impacts from high salt concentration (electrical conductivity [EC]). If this is true, a recalculation of the EROI of an integrated algae-dairy production facility with the new value of algal meal priced relative to feather meal has the potential to show an economically viable, sustainable farm-energy production system.

Problems with Land Application of Dairy Waste

The first argument against high-N specialty organic fertilizers is that producers should be using composts to fertilize their land. There is no doubt composts are beneficial for soil tilth and provide a rich C source but by the time the material is composted anywhere from 20% to 40% of the N has volatilized (Eghball et al., 1997). For a period of time after compost application, many soils will experience a period of net N immobilization (Sullivan et al., 1999; Lashermes et al., 2010) as the rush of readily-available C allows soil microbes to become active and reproduce. In order to do this they must take up nutrients, needing one unit of N for every five to ten units of C they take up, thereby stripping available N from the soil solution and into their cells (Havlin et al., 1999; Kissel et al., 2008). While the soil is in this period of net immobilization, many plants will show symptoms of N deficiency: primarily chlorosis, a stunted yellow appearance. Chlorosis results in decreased energy capture leading to reduced yield, grade, vigor, and fruit set (Havlin et al., 1999). Therefore, when applying compost products the timing of available nutrients needs to be carefully assessed.

Composts and manures are often applied using rates calculated to the most limiting nutrient, typically N, using Liebig's Law of the Minimum (Hart et al., 1997; Bary et al., 2000). They are normally relatively balanced in NPK ranging from 1-1-1 to 4-4-3, leading to excess P and K additions (Hart et al., 1997). Phosphorus is known to accumulate in soil when it is applied in excess of the crop harvest, especially in areas of high-density livestock confinement operations (Mozaffari and

Sims, 1994; Simard et al., 1995; Whalen and Chang, 2001). Through time this can increase the risk of P transport to water bodies through leaching, erosion, and runoff (Sims et al., 1998; Sharpley and Tunney, 2000; Hooda et al., 2001). In a study on the soil P dynamics after ten annual applications of mineral fertilizers and liquid dairy manure, Zheng et al. (2004) found that repeated P additions (from manure treatments) in excess of plant removal elevated soil test P and the potential of P transfer from the soil to surface waters. They also found that labile P fractions were significantly higher in the liquid dairy manure treatments than in the mineral fertilizer treatments, further increasing chance of downstream eutrophication events from overland flow.

Potassium poses little threat for waterway contamination but is a concern for dairy operators. Animal nutritionists recommend that dietary K not exceed 3% of a dairy cow ration to reduce the risk of milk fever, hypocalcemia, downer cow syndrome, and even death (Hart et al., 1997). Perennial grasses repeatedly fertilized with manure accumulate K in excess of growth requirements. One study from British Columbia showed grass forage K increased from 2.7% in 1983 to 3.6% in 1992 with repeated dairy manure application (Schmidt, 1994).

In addition to contributing to the buildup of P and K in soils, manure decomposition produces the GHGs: nitrous oxide (N_2O) (global warming potential relative to CO_2 [CO_2e] is 310), methane (CH_4) (CO_2e is 24), and CO_2, as well as the particulate forming ammonia (NH_3). Manure storage systems are second only to enteric fermentation gases in the total contribution of GHG coming from animal

production operations (Sneath et al. 2006; Pitesky et al., 2009; Leytem et al., 2011). In an exhaustive study of GHG emissions from an open-lot dairy in Idaho, Leytem et al. (2011) found CH_4 ranged from 19.4 to 231 g CH_4 m^{-2} d^{-1} with a mean of 103 g CH_4 m^{-2} d^{-1} from the lagoon manure storage. Emissions increased as wind and temperature increased with variability depending on volatile solids content. Others reported mean CH_4 emissions from lagoons ranging from 2 to 203 g CH_4 m^{-2} d^{-1} (Kaharabata et al., 1998; Sneath et al., 2006; Bjorneberg et al., 2009). Carbon dioxide emission rates ranged from 289 to 855 g CO_2 m^{-2} d^{-1} with a mean of 637 g CO_2 m^{-2} d^{-1}, the NH_3 emission rate from the lagoon was 2 g NH_3 m^{-2} d^{-1}, and N_2O showed low emission rates from the lagoons, averaging 0.49 g N_2O m^{-2} d^{-1} (Leytem et al., 2011). Flesch et al. (2009) reported 2.3 to 3.5 g NH_3 m^{-2} d^{-1} from lagoons.

Ammonia losses from land-applied dairy slurry range from 10% to 50% of applied N depending on application method (Moal et al., 1995). With more rapid and complete incorporation of material resulting in lower volatilization rates (Wulf and Clemens, 2000; Sullivan et al., 2010). Injection of slurry into soil is one method to reduce NH_3 losses but this can lead to increased N_2O production (Wulf and Clemens, 2000). Nitrous oxide production is a result of incomplete microbial denitrification in anaerobic soils with available NO_3^-. Dairy slurry sprayed onto a field in a virtual sheet of nutrient-laden, high-viscosity liquid provides a prime habitat for incomplete denitrification. Myrold et al. (1992) found denitrification losses were increased with higher rates of manure applications and soils with higher clay

content. Denitrification losses ranged from 5-16% of applied N resulting in N_2O losses ranging from 33 to 108 kg N ha^{-1} y^{-1}.

High-N Alternatives

Feather meal, a ground, hydrolyzed, and dried by-product of large-scale poultry operations, has become a popular organic N source. Every day companies such as Pilgrims Pride and Fosters produce upwards of 75 Mg of this material (Skinner, 2013). It has long been used as a feed ingredient in cattle and hog rations as it contains about 90% protein. The commodity value has historically been calculated relative to other protein meals, such as soybean meal and canola meal, with a discount for reduced palatability. However, around 2007 the value of feather meal started to exceed its value relative to other protein meals as more companies started producing pelletized products and recommending it as an organic fertilizer. Comparing prices from a major producer in Texas; March 2007 feather meal prices were \$350 Mg^{-1} freight on board (FOB). By May of the same year, prices had jumped to \$580 Mg^{-1} FOB. March 2008 prices were \$550 Mg^{-1} FOB and by May they were about \$725 Mg^{-1} FOB. In 2012 traders reported purchasing feather meal in the spring for as high as \$1,100 Mg^{-1} FOB (Skinner, 2013).

Feather meal is particularly desirable because N is the only macronutrient it supplies N. It has an NPK grade of 12-0-0. This is desirable for producers dealing with years of P and K buildup. Additionally, feather meal has a relatively fast

mineralization rate for an organic fertilizer, with 75-99% of the material mineralized into plant-available N in the first year after application (Gale et al., 2006). After price, the main drawback for producers who might want to use feather meal is application difficulty. It has a bulk density around 0.6 g cm^{-3} making it light and fluffy. This can cause problems with application as the material tends to bridge in hopper bellies and drift from desired application placement. Further, it cannot be solubilized to apply as a foliar fertilizer. Several companies have produced value-added pelletized feather meal products to overcome these issues.

Many organic fertilizer products discuss stimulation of beneficial microorganisms as an overarching benefit of their product with very little specific academic research to back up the claims. Any fertilizer with substantial C will provide an energy source, this energy will stimulate microbial growth, causing the microbial population to produce more extracellular enzymes to break down soil organic matter (SOM) into plant available nutrients (PAN). An organic fertilizer does not select for beneficial or non-beneficial organisms, it simply provides various nutrients, and the energetically-favorable metabolic pathways will progress.

Studies on Algae Fertilizer Use and Benefits

In a study by Mulbry et al. (2005), algal biomass grown on anaerobically digested dairy manure was tested for N and P mineralization in a laboratory incubation and as a fertilizer on corn and cucumbers in a greenhouse trial. This

product was not the same as algal meal because the lipid fraction had not been extracted but it is the closest product to what I am testing. The algal biomass used had an NPK grade of 4.5-0.7-0.9. Approximately 3% of total N was present as mineral N at day 0. On average 26-30% of total N was mineralized in the first 21 days of a laboratory incubation held at 15^{o}C. They concluded that it would be a suitable commercial fertilizer in potting systems. Tripathi et al. (2008) provides additional evidence that photoautotrophs may be beneficial as a fertilizer source. They investigated the role of cyanobacteria in ameliorating the N demand and fly-ash stress to growth and yield of rice. They found that rice paddies treated with cyanobacteria, which are known to fix atmospheric N, lowered stress-induced thiols; reduced accumulation of cadmium, nickel, and arsenic in the rice; increased growth and yield; and reduced the N fertilizer demand.

Natural marine-grown seaweeds (macroalgae) have been used as fertilizers and soil conditioners for centuries (Blunden and Gordon, 1986; Metting et al., 1988; Temple and Bomke, 1988). Around 15 million Mgs are produced annually for use as biostimulants or biofertilizers (Khan, 2009). The concentration of mineral nutrient elements present (typically less than 1-1-1) in commercial seaweed concentrates alone cannot account for the growth responses elicited by seaweed extracts (Blunden, 1972; 1991). Many studies have been conducted to determine the modes of action for a list of benefits, including: early seed germination and establishment, improved crop performance and yield, elevated resistance to biotic and abiotic stress, and enhanced postharvest shelf-life of perishable products (Beckett and van

Staden, 1989; Hankin and Hockey, 1990; Blunden, 1991; Norrie and Keathley, 2006). Results suggest seaweed components, such as macro- and micro-nutrients, amino acids, vitamins, cytokinins, auxins, and abscisic acid-like growth substances affect cellular metabolism in treated plants leading to enhanced performance, growth, and yield (Crouch and van Staden, 1992; Crouch and van Staden, 1993; Reitz and Trumble, 1996; Durand et al., 2003; Stirk et al., 2003).

Acadian Seaplants is one of the most aggressive marketers of *Ascophyllum nodosum,* selling their dry kelp meal product above $1,600 Mg^{-1}. In order to achieve this market value, they have invested considerable resources in researching its benefits on over 80 crops, for over 25 years. Their marketing campaign touts the increased stress tolerance and stress recovery from drought, salinity, and temperature. Two major modes of action that have been investigated are the role of micronutrients and phytohormones. These are cited as reasons for improvements in nutrient uptake as the micronutrients provide essential elements for enzyme production and increase quality attributes such as firmness, color, size, and crop uniformity. Algae are known to produce essentially all of the known phytohormones of higher plants and they carry out similar physiological functions in algae as they do in plants (Tarakhovskaya, 2006). Changes in the level of exogenous cytokinins alter the regulation of physiological plant processes (Stirk and van Staden, 2010). Acadian claims that phytohormones in their products elicit natural cytokin and auxin production in plants resulting in better growth with more buds, healthier, greener leaves, and increased tolerance to environmental stresses

(AcadianSeaplants.com). It is assumed these phytohormones would also be found in algae meal (Brain, 1973); however, as the algal biofuel industry expands, more research will be needed to elucidate whether or not the benefits can be extended to crops grown with algal meal.

Conclusion

Algal meal offers an exciting opportunity for organic fertilizer production. Several algal fuel companies have completed pilot scale facilities and are demonstrating potential production at large scale, producing potentially significant quantities of algal meal. Further, by providing the, N, P, K, S, Ca, Mg and the full range of micronutrients needed for plant growth, algal meal could be a possible solution for organic producers struggling to find adequate organic nutrients to meet the ever growing demands on organic production.

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CHAPTER 2
QUANTIFICATION OF THE FERTILIZER VALUE OF ALGAL MEAL

Abstract

As the organic food industry grows, so will the need for organic nitrogen (N) fertilizers. Algal meal, the high protein co-product of algal biodiesel production, could help meet this need. The objective of this work was to quantify the fertilizer value of algal meal relative to an organic N fertilizer, feather meal, and a conventional standard, urea. A laboratory incubation was carried out to characterize the C and N mineralization of two different algal meal products. The N mineralization rate, C respiration rate, and impact on pH and EC was the same for feather meal and algal meal 2, while the mineralization rate of algal meal 1 was significantly lower. A field trial was conducted to assess the overall crop productivity of corn grown with algal meal 2, feather meal, and urea. No significant difference in corn ear yield was found between the low rate of algal meal application (101 kg N ha^{-1}) and the high rate of urea (190 kg N ha^{-1}). These results indicate that algal meal is an effective N fertilizer that should be valued relative to feather meal on a price per unit of nutrient basis.

Introduction

Nitrogen is an essential nutrient required by all organisms. In agro-ecosystems N availability is a primary limiting factor for plant growth. The United

States Department of Agriculture (USDA) Economic Research Service (ERS) reported that 11,144,000 Mg of N were applied to US fields in 2010, which was roughly three times higher than phosphorus (P) or potassium (K) inputs. In the same year the organic food industry grew 7.7% to contribute $26.7 billion in sales into our national economy (Organic Trade Association, 2011). Certified organic farmland reached more than 2,000,000 ha in 2008, growing at a 15% annual rate from 2002 to 2008 (USDA ERS, www.ers.usda.gov/data/organic). As the organic food industry grows so will the need for organic N fertilizers.

A myriad of academic and extension publications are aimed at environmentally and economically sustainable methods to help organic growers and animal producers meet the nutrient requirements of their plants with composts and manures. Timing of nutrient mineralization and immobilization (Whalen et al., 2001; Eghball et al., 2002; Norton et al., 2007), product moisture and nutrient variability (Wander, 2010), emissions of the greenhouse gasses (GHGs)(Leytem et al., 2011): nitrous oxide (N_2O), methane (CH_4), and carbon dioxide (CO_2), ammonia (NH_3) volatilization (Chantigny et al., 2004), nutrient leaching (Pang and Letey, 2000; Ball-Coelho et al., 2004), nutrient buildup (Hart et al., 1997; Zheng et al., 2004), and application methods (Moore and Hart, 1997) are all problems that have been investigated. However, even when following the best management practices, significant nutrient loss is inevitable causing negative environmental and economic consequences.

Utilizing animal manure products as a feedstock for anaerobic digestion for CH_4 biogas production, and using the subsequent liquid effluent as the nutrient broth for algae cultivation, could eliminate the current waste management difficulties facing animal producers. Extensive work at the USDA labs in Beltsville, MD has shown that algae grown on dairy effluent recovered 95% of N and 77% of P nutrient inputs (Kebede-Westhead et al., 2006). Olguin (2003) found similar results using swine effluent with 91% N removal and 87% P removal. After nutrient capture the algae can be extracted for their lipids producing a biodiesel, glycerin, and a stable high-N, organic product.

In a study by Mulbry et al. (2005), algal biomass grown on anaerobically digested dairy manure was tested for N and P mineralization in a laboratory incubation and as a fertilizer on corn and cucumbers in a greenhouse trial. The algal biomass used had an NPK grade of 4.5 -0.7-0.9. Approximately 3% of total N was present as mineral N at day 0. On average, 26-30% of total N was mineralized in the first 21 days of a laboratory incubation performed at 15°C. They concluded that it would be a suitable commercial fertilizer in potting systems. This product was not the same as algal meal because the lipid fraction had not been extracted but it is the closest product tested to those I studied.

Marine-grown seaweeds have been used as fertilizers and soil conditioners for centuries (Blunden and Gordon, 1986; Metting et al., 1988; Temple and Bomke, 1988). Around 15 million Mgs are marketed annually for use as biostimulants or

biofertilizers (Khan, 2009). The concentration of mineral nutrient elements present in commercial seaweed concentrates alone cannot account for the growth responses elicited by seaweed extracts (Blunden, 1972; 1991). Many studies have been conducted to determine the mode of action for a list of benefits, including: early seed germination and establishment, improved crop performance and yield, elevated resistance to biotic and abiotic stress, and enhanced postharvest shelf-life of perishable products (Beckett and van Staden, 1989; Hankin and Hockey, 1990; Blunden, 1991; Norrie and Keathley, 2006). Results suggest seaweed components, such as macro- and micronutrients, amino acids, vitamins, cytokinins, auxins, and abscisic acid-like growth substances, affect cellular metabolism in treated plants leading to enhanced performance, growth, and yield (Crouch and van Staden, 1992; Crouch and van Staden, 1993; Reitz and Trumble, 1996; Durand et al., 2003; Stirk et al., 2003). As the algal biofuel industry expands, more research will be needed to elucidate whether or not the benefits can be extended to crops grown with algae meal.

Many current life cycle assessments of algal fuel production do not include the value of the co-product in the overall valuation as it is assumed to be low (Pizarro et al., 2006; Chisti, 2007; Sanders and Murthy, 2009; Clarens et al., 2010), or they use animal feed values (NAS, 2012). A proper value for the algal meal, between 50-80% of the total algal biomass, could allow these operations to reach financial profitability. Many agricultural by-products have realized higher values as organic fertilizers than as feed ingredients (Skinner, 2013). Feather meal, once

primarily a high-protein cattle feedstuff has become popular and widely accepted as an effective high-N organic fertilizer increasing in price from \$350 Mg^{-1} to \$1,100 Mg^{-1} in the last 6 years (Skinner, 2013). Algal meal lends itself to use as an organic fertilizer as it has a C:N ratio of 5:1, an NPK grade around 7-3-1, and the full range of micronutrients that are essential for plant growth (A&L Labs, 2013). It holds potential as an exciting new option as a high-N organic fertilizer.

The goal of this research was to do a preliminary analysis of the viability of algal meal as a fertilizer and to determine an appropriate agronomic value. I hypothesized that algal meal is as effective as feather meal as an organic fertilizer product on a per unit of N basis. Specifically, algal meal would have a similar N mineralization rate, microbial respiration rate, extracellular enzyme production, yield of sweet corn as feather meal fertilized plots, and not have any negative impacts from the salt concentration (electrical conductivity EC). If this is true, a recalculation of the EROI of an integrated dairy-anaerobic digester-algae (DADA) production facility with the new value of algal meal priced relative to feather meal may have the potential to show an economically viable, sustainable farm-energy production system.

Materials and Methods

Organic Amendment Analyses

The algal meal products that were used in the laboratory incubation and field trial were produced as the co-products of a hexane oil extraction from algal biomass. Algal meal 1 (A1), used in the laboratory incubation, was supplied by the Center of Excellence for Hazardous Materials Management (Carlsbad, NM). Algal meal 2 (A2), used in the incubation and field trial, was made from *Nannochlosis oceanus* from a 10-ha, continuously operational facility. Feather meal 1 (F1) was a hydrolyzed, ground feather product from *Gallus gallus*, purchased from Down to Earth (Eugene, OR). The organic materials used in these trials were analyzed for nutrients and metals (Table 1 and 2 respectively) at Western A & L Agricultural Laboratory (Modesto, CA). Both algal meals used in this experiment were marine species processed from algae grown in salt water. This resulted in high EC and caused concern about potential stress. Three replicates of each amendment were tested for pH and EC prior to the laboratory incubation (Table 1). The non-nutrient metals analysis showed no analytes above the allowed levels for organic product registration (Table 2). This is consistent with work by Mulbry et al. (2006), who showed that algal biomass grown on animal manures does not contain heavy metals at concentrations that would limit its use as a fertilizer or animal feed supplement.

Field Trial

Site and Soil Information

The sweet corn field trial was conducted in 2012 at Oregon State University's Lewis-Brown Horticultural Farm in Corvallis, OR (N 44°33'10"; W 123°13'16"). The soil series is a Malabon silty clay loam (fine, mixed, superactive, mesic Pachic Ultic Argixerolls) classified by NRCS as prime, well-drained farm land. Prior to the start of the trial, the field was under a cover crop for two years. The cover crop used was the commercially available, Feed and Seed Plow Down™, which is a mix of common vetch, cereal rye, soybean, and Austrian pea. The cover crop was cut on April 28, disked May 10, spaded (Tortella Spader, Brewt Power Systems, Merced, CA) May 14, and power harrowed (Kuhn, Broadhead, WI) on May 15 and May 31. Soil samples from each experimental unit (plot) were taken April 27 to assess site uniformity. The area used was nearly level; each plot had the same gravimetric water content ($p>0.05$) and showed uniform pH and EC ($p>0.05$).

Experimental Design

A randomized block design with four blocks was used in the field trial. Each plot measured 6 m by 6 m with a 1.2-m buffer zone between plots. Soil samples taken April 27 were tested for nutrients (Table 3). Given the previous legume-cereal cover crop and these data it was determined that the soil had sufficient N for the initial 30 days of corn growth, that K was non-limiting, therefore no K was added, and that 100 kg P_2O_5 ha^{-1} were needed (Hart et al., 2010). Triple super phosphate

(TSP) was applied to every plot at the same time as N fertilizer treatments. Nitrogen rates were calculated according to the recommendation of Pre-Sidedress Nitrate Test (PSNT) (Hart et al., 2010). Using soil samples taken June 21, 2012 the NO_3^--N concentration was 10 mg kg^{-1} indicating that 140 kg N ha^{-1} was needed for maximum productivity. Six treatments were used: a control with no N addition (CTL), a low rate of algal meal at 101 kg N ha^{-1} (A Low), a medium rate of algal meal at 146 kg N ha^{-1} (A Med), a high rate of algal meal at 190 kg N ha^{-1} (A High), a high rate of feather meal at 190 kg N ha^{-1} (F High), and a high rate of urea at 190 kg N ha^{-1} (U High).

Timeline

The sweet corn (*Zea mays* L.) variety Captain™ (SE) was planted on June 6 with a double-disk planter with rows spaced 76 cm apart. Seeds were drilled approximately 20 cm apart and 4 cm deep. While planting, Lorsban™ (O,O-diethyl O-3,5,6-trichloropyridin-2-yl phosphorothioate) (Dow Agro Science, EPA registration 62719-34) was sprayed at a rate of 74 g 100 m^{-2} of row to control seed corn maggot. That evening the herbicides Outlook™ (dimethenamid-P: (S)-2-chloro-N-[91-methyl-2-methoxy)ethyl]-N-(2,4-dimethyl-thien-3-yl)-acetamide) (BASF, EPA registration 7969-156) and Atrazine™ (1-Chloro-3-ethylamino-5-isopropylamino-2,4,6-triazine) (AgroSolutions, EPA registration 1381-158) were sprayed each at a rate of 1.12 kg ha^{-1} for weed management. Fertilizers were weighed and bagged individually for each 6-m row of corn, eight per plot. In order to band the fertilizers

by hand, a pointed hoe was used to dig a small channel approximately 5 cm from the emerged corn, approximately 5 cm deep. Algal meal, feather meal, and TSP were distributed as evenly as possible in respective channels and covered on June 29. On July 4 urea was added. The difference in application timing was to account for mineralization of the organic materials, so all treatments would be optimally timed during peak N demand. Irrigation was not needed through June as there was adequate regular precipitation. Irrigation was applied once per week from July through September for one to three hours at a rate of approximately 0.84 cm hr^{-1} (Fig. 2). Using the evapotranspiration data maintained by AgriMet (Pacific Northwest Cooperative Agricultural Weather Network) the water demand for sweet corn was calculated. Four soil samples were collected and mixed from each plot every week. Soil samples were taken directly in line with the corn row, a minimum of 10 cm from any given stalk, to a depth of 15 cm.

On September 5, the plots were harvested by hand and measured for ear count, ear yield, stalk count, and stalk yield. Three rows of the 3-m center sections of each plot were collected for ear measurements and three rows of the 3-m center sections of each plot were collected for stalk measurements. Harvesters were instructed to bypass small sections of any row that did not germinate. Only ears with a cob greater than 15 cm were collected. Only primary shoots were collected for stalk measurements. Four ears and four stalks from their respective rows were saved and coarsely chopped onsite. Samples were brought back to the laboratory, dried, ground (Plant Grinder, Thomas Scientific, Philadelphia, PA) to pass a 2-mm

sieve, and analyzed for N, P, K, S, Ca, Mg, and C by the Central Analytical Laboratory at OSU. Three weeks following harvest, soil samples were collected for residual NO_3^- in the soil at 0-15, 16-30, 31-45, and 46-60 cm depths using a soil auger.

Laboratory Incubation

The soil for the laboratory incubation came from the same field used for the field trial at Lewis-Brown Horticultural Farm. Soil samples were collected April 27 and stored at 4°C prior to incubation. The incubation consisted of eight treatments with three replications of each: two algal meals, each applied at three different rates, feather meal, and a control with no amendment. Using the bulk density of 1.21 g cm^{-3} to calculate the weight of an acre-furrow slice of Malabon soil, amendment levels were calculated to represent realistic agricultural application rates. The final letter in each treatment abbreviation indicates the N application level, L (low) represents 146 kg N ha^{-1}, M (medium) represents 190 kg N ha^{-1}, and H (high) represents 235 kg N ha^{-1}.

The soil was sieved through a 5-mm screen wet, to minimize aggregate and biological disruption while still removing rocks and large pieces of organic matter. The soil needed for each treatment type was weighed, spread on a mat, the amendment was added, mixed thoroughly, and the amended soil was distributed into 18, 50-mL centrifuge tubes. Each tube was tapped on the table 10 times to approximate natural bulk density. Each experimental unit was contained in one-quart sized canning jar that held six centrifuge tubes filled with soil and the

appropriate amendment. Jars were covered with gas permeable, water impermeable polyethylene film and incubated at 25°C for six weeks. A sample of each treatment was set aside for measurement of initial NO_3^-, NH_4^+, and H_2O contents. One tube per week was removed for measurement of NO_3^-, NH_4^+, and H_2O contents. Samples were extracted with 2N KCl for NO_3^- and NH_4^+ determination and stored at 4°C before analysis.

Analytical Methods

Nitrate was tested using the vanadium chloride (VCl_3) reduction and Griess reagent method as described by Hood-Nowotny et al. (2010) based on the technique described by Miranda et al. (2001). Ammonium was measured using the classical Berthelot reaction (Kandeler and Gerber, 1988) using sodium salicylate, sodium nitroprusside, and sodium hypochlorite. Plates for each were incubated for 60 min at 37°C and measured colorimetrically on a microplate reader (BioTek, Synergy 2.0 Gen5™ multimode microplate reader. Winooski, WI). Nitrate was read at 540 nm; NH_4^+ was read at 650 nm. Standards were prepared at each measurement using dilutions of stock 1M NO_3^- and NH_4 solutions.

The mineralization rate for each fertilizer treatment was calculated as the difference between the total inorganic N at the end and beginning of the incubation, divided by the days of incubation:

[Final (NO_3^--N +NH_4^+-N) – Beginning (NO_3^--N +NH_4^+-N)]/incubation days (1)

For each amendment, the inorganic N mineralized through the incubation minus the inorganic N mineralized in the control was divided by the total N added to the soil by the respective amendment to calculate the % N mineralized:

[Final (NO_3^--N +NH_4^+-N) - Control (NO_3^--N +NH_4^+-N)]/Total N added (2)

All CO_2 measurements were made using a Picarro™ Isotopic CO_2 Analyzer model A0311 (Sunnyvale, CA). Each sample was sealed and CO_2–C was measured for two minutes at two different time points. In the laboratory incubation CO_2-C was measured weekly using all remaining tubes in the jar with one hour between beginning and final measurements. For the field trial, a 30-g sample of field moist soil was put in a 125-mL Erlenmeyer flask, with two hours between measurements. The difference between second reading (sum of $^{13}CO_2$-C + $^{12}CO_2$-C) and the first reading (sum of $^{13}CO_2$-C + $^{12}CO_2$-C) was used to calculate the total CO_2-C evolution rate.

For each amendment, the CO_2-C respired through the incubation minus the CO_2-C respired from the control was divided by the total C added to the soil by the respective amendment to calculate the % amendment C respired.

[Final ($^{13}CO_2$-C + $^{12}CO_2$-C) - Control ($^{13}CO_2$-C + $^{12}CO_2$-C)]/Total C added (3)

I estimated the amount of C remaining in the soil of the field trial from the amendments using the total C added to the soil by each amendment minus the % amendment C respired from the laboratory incubation.

Extracellular enzyme activity was measured at four time points for each experiment. Alpha-glucosidase and beta-glucosidase were selected to assess carbohydrate catabolism and leucine-aminopeptidase (LAP) was a proxy to assess protein catabolism. Fluorescent methylumbelliferone was used for alpha- and beta-glucosidase enzyme assays and fluorescent methylcoumarin substrate was used for LAP enzyme assays according to German et al. (2011).

Wet soil was weighed, dried at 60°C for a minimum of 48 hr, and reweighed to obtain the gravimetric moisture content of all soil samples. EC and pH were measured using a 1:2 soil:water slurry. Samples were put on the shaker for 30 minutes, allowed to return to atmospheric conditions (Gavlak et al., 1997) and measured with a Hanna™ GroCheck meter (Smithfield, RI).

Statistical Methods

Statistical results were computed using SAS 9.2 (SAS Institute Inc., Cary, NC). Repeated measure tests for data with equal time between measurements were run as multivariate with autoregression because compound symmetry could not be assumed and autoregression was the best-fit model of correlation. Repeated measure tests of the CO_2 -C respiration from the laboratory incubation were run as multivariate with unstructured regression because unequal spacing of measurement times eliminated autoregression as an option. Data from a single time were analyzed with a randomized block design ANOVA and LSD test for multiple comparisons as treatment structures were not well defined. All harvest data was

analyzed as a randomized block design with subsamples. Results with a p-value < 0.05 are considered significant. Graphical representations were produced with KaliedaGraph Synergy Software version 4.1.3 (Reading, PA).

Results

Laboratory Incubation

Ammonium-N concentrations were elevated for two weeks and subsequently declined (Fig. 3). Nitrate-N accumulated throughout the incubation (Fig. 4). Multivariate repeated measures tests of all the NO_3^--N and the NH_4^+-N concentrations showed that there was a significant treatment effect, there was a significant time effect, and there was a significant interaction. When testing NO_3^--N accumulation there was no significant difference in the regression line of F1M against A2M or A2H treatments, all other treatments were significantly lower.

Nitrogen mineralization of the organic amendments, measured as the accumulation of inorganic N minus the N mineralized by the control, proceeded more or less linearly throughout the incubation (Fig. 5). The N mineralization rate (Table 4) was the highest in F1M with no statistical differences among F1M and A2M and A2H. A1 at all rates was lower than A2 and F1M. The fraction of N mineralized of each amendment (Eqn. 2) was highest in F1M and A2L (Table 4). For both A1 and A2, the efficiency was higher at the lower N application rates, though not statistically different among A1 rates.

Respiration rates peaked early during the incubation with a reduced production rate thereafter (Fig. 6). Cumulative CO_2-C respiration (Fig. 7) showed significant treatment, time, and interaction effects. When comparing A2M against F1M, there was no significant treatment effect; however, there was significantly more CO_2-C released from A1H and A2H than A1M, A2M, or F1M. There was no significant difference in CO_2-C evolved between A1H and A2H.

Extracellular enzyme activity in the laboratory incubation showed a clear temporal trend in both the alpha- and beta-glucosidase extracellular enzyme production (Fig. 8) but no treatment differences were found. There was a significant treatment difference in levels of leucine-aminopeptidase activity; however, there were no consistent trends among treatment performance.

The pH of the control did not change during the laboratory incubation but pH in all amended soils decreased about 0.5 units (Table 5). There was no significant difference in the pH reduction among treatments. The EC change varied by amendment. Algal meal products had higher inherent EC and the mean increase in the EC of the soil at the end of the incubation was 0.37 mS cm^{-1} (SE=0.01), that was significantly higher than the EC increase in the F1 and CTL treatments. The feather meal increased EC by 0.19 mS cm^{-1} (SE=0.02), significantly more than the control. The EC of the control did not change significantly through the incubation.

Field Trial

Nitrate concentrations decreased throughout the field trial until August 1, or 56 days after planting (Fig. 9). At this time, the corn N uptake rate was significantly reduced. The increased levels of NO_3^--N in the soil after August 8 indicate that the microbial N mineralization rate did not slow as significantly as the corn N uptake. Comparing the regression through time of NO_3^--N concentrations showed that there were significant treatment and time effects, but no significant interaction. The NO_3^--N values were not statistically different among treatments during the first 7 weeks of the trial. August 15, 22, 29 and September 5th there were differences in the NO_3^--N remaining in the soil (Table 6), with F High and U High often higher than other treatments.

Ammonium levels were low prior to fertilization, peaked after fertilization, declined, and increased again once the rate of corn N uptake declined (Fig. 10). There were no significant differences between treatments in soil NH_4^+-N.

In order to determine if algal meal fertilizer would be more or less prone to leaching N than feather meal and urea, soil samples were taken 3 weeks after harvest to test residual NO_3^--N in soil to determine if N mineralization from all amendments continued at the same rate after crop removal. In the top 15 cm, the CTL and A Low plots had statistically lower NO_3^--N concentrations than F High plots. Although all algal plots trended lower than U High and F High plots in the top 45 cm, there were no statistical differences in A High, F High or U High at any depth (Fig. 11).

The CO_2-C respiration rate of the treatments ranged from 8 to 26 μg CO_2-C g^{-1} dry soil day^{-1}, with a mean of 14.6 μg CO_2-C g^{-1} dry soil day^{-1}. There were no significant differences among treatments or through time in the field trial (Fig. 12).

There were no significant differences among treatments or with time in any of the enzyme assays from the field trial (Fig. 8). The soil pH in the field trial increased by about 0.5 (SE=0.03) from 5.8 to 6.3. There was a significant difference through time but no difference among treatments. This is within the typical range of soil pH seasonal increase due to differences in moisture (Hart et al., 2010). The EC in the field trial decreased by 0.03 mS cm^{-1} (SE=0.004); from 0.08 to 0.05 mS cm^{-1} there were no treatment differences.

Harvest Data

The mean stalk count for each 3-m row was 13 with no significant difference among any of the treatments; this extrapolates to 53,600 corn plants ha^{-1} (Fig. 13). There was a significant difference in stalk yield among treatments. The urea treatment had the highest stalk yield, medium and high rate organic fertilizer treatments were in the next group, the low rate of algal meal was significantly lower than urea, and the control was significantly lower than any of the treatments. The control plots had fewer ears than the other treatment plots, averaging 42,700 ha^{-1}; there was no significant difference in the total number of ears from any of the fertilized plots, which had a mean of 56,800 ha^{-1}. Although the fertilized plots had higher ear yield than the control, the fertilized plots were not significantly different

from each other. The ear harvest yield for the control was 60% of the yield from the highest yielding plot, F High. The ear mean yield for fertilized plots was 26,200 kg ha^{-1} (12 tons $acre^{-1}$) (Fig. 13).

Plant Tissue Analysis

The total N concentration in the stalk was lowest in the control, significantly higher in feather meal and urea treatments, and intermediate for the algal treatments. The N concentration in the ear was lowest in the control and significantly higher in the A Med, A High and F High (Table 7). The P concentration in the stalk was significantly higher in the low rate of algal meal application than in the high rate of urea with all other treatments being in between. There were no significant differences in the percentage of K, S, Ca, or Mg in the ears or stalk tissue analysis among fertilized plots.

Discussion

Algal Characteristics

Each algal meal product varied in composition presumably depending on nutrient input, salinity of growing medium, algae strain, and post harvesting procedures. The fertilizer analysis of A2, the *Nannochloropsis oceanus* algal meal used in the laboratory incubation and the field trial, showed a desirable NPK grade of 7.2-3-1.2 and C:N ratio of 5.2:1 (Table 1). The A1 treatment had a similar C:N ratio (5.7:1) but a lower concentration of NPK nutrients with a 5.8-0.2-0.1 (Table 1). In

the laboratory incubation % C respired as CO_2-C and the total N added were the same but the % N mineralized through the lab incubation was significantly lower with the A1 than the A2 treatment (Table 4).

There were several measured differences between the two algal meals that could help to explain the discrepancy in N mineralization. A1 had a substantially lower pH (2.47) than the other organic amendments (Table 1). However, the direct pH of fertilizers is not typically measured. Rather, the potential acidity or basicity, describing the pH reaction of the soil solution after the product is applied, is reported as calculated by the Pierre equation in units of equivalent $CaCO_3$ displacement or quantity of $CaCO_3$ needed to neutralize (Pierre, 1928; Argo and Fisher, 2008). Using the Pierre equation, A1 and A2 would have the same potential acidity because they had the same influence on the pH of the soil. From this I would not expect that the low pH of the amendment retarded the N mineralization.

The high concentration of sulfur (S) in A1 (7.8%) stands out as another major nutritive difference but synthetic fertilizers such as ammonium sulfate (24% S) are regularly used without observed adverse N mineralization effects (Havlin et al., 1999). Amendment A1 had 5.5% Ca whereas each of the other organic amendments had about 1% Ca. It has been shown that, in neutral to basic soils, Ca^{2+} can form cation bridges between mineral surfaces and OM, leading to aggregation of clay particles with OM stabilizing the soil structure and potentially decreasing the availability for microbial use (Oades, 1988). In a field trial using isotopically labeled C inputs the presence of Ca^{2+} increased the residual ^{14}C in the soil for up to 6 months

(Oades, 1988). The soil used in this trial started with a pH of 5.7 and I did not measure a difference in the CO_2 evolution from equivalent rates of amendment application; however, the high Ca in A1 may have formed cation bridges impacting microbial availability and reducing the rate of N mineralization.

Extraction chemicals and processes, drying temperatures, and algal anatomy and physiology have an impact on the molecular structure of algal meal (Van Der Meulen, 2013). Structural differences impact the affinity of organic material to adsorb to mineral soil and therefore their availability for microbial degradation (Kleber and Johnson, 2010). For instance, diatomaceous algae species have silica dioxide in their cell walls, whereas most others rely on cellulose for cellular structural stability (Shelef and Soeder, 1980). These differences impart different bonding structures and a different consortium of soil biota would be responsible for the decomposition of the cellular components. The preliminary data from the incubation give reason to believe that different algal production methods may yield co-products of varying value. Each production facility may be able to market an algal meal that meets the needs of soils that are deficient in specific nutrients. Further testing is necessary to determine the resulting chemical composition of algal meals from different technologies.

Nitrogen

It has been recorded for more than 2,000 years that OM is an important aspect of soil fertility (Long, 1842). In recent years, it has become generally accepted

that the balance between N mineralization and immobilization is a function of the relative availability of C and N in a substrate and the metabolic needs of the microbial biomass (Myrold and Bottomley, 2008), and is driven by the depolymerization of N-containing OM by microbial extracellular enzymes (Chapin et al., 2002; Schimel and Bennett, 2004). Bacterial cells have a generally accepted C:N ratio of 5:1 (Myrold and Bottomley, 2008). Since the fungal community can have C:N ratios up to 15:1, a general C:N ratio of 8:1 for total microbial biomass is typically used (Myrold and Bottomley, 2008). With C:N ratios ranging from 4:1 to 5.7:1 I am confident that none of the organic amendments used in these trials induced net N immobilization.

In a study on potential methods for estimating the N fertilizer value of organic residues, Delin et al. (2012) used 15 common agricultural by-products and a mineral N source (ammonium nitrate) in a ryegrass greenhouse trial. The above-ground N uptake for each treatment was plotted against the mineral N fertilizer rate. From this regression, the mineral N fertilizer rate corresponding to the above-ground plant N uptake for each treatment with organic fertilizer was derived to determine the mineral fertilizer equivalent (MFE), expressed as the percentage of total N applied. Considering several possible predictors, they concluded that the C:N ratio was the best predictor of a product's MFE. An aerobic incubation of the organic amendments also correlated well ($r^2 = 0.78$).

$$MFE = 0.87 - 0.05*(\text{C:N ratio of product}) \qquad (r^2 = 0.83) \qquad (4)$$

Using Eqn. 4, the calculated MFE value of A1 was 59%, A2 was 61%, and F1

was 67%. This over-predicts net N mineralization for A1 but is a reasonable estimate for A2 and F1 in the laboratory incubation (Table 4). The accuracy of an MFE value should be assessed based on plant N uptake; as it was determined in the Delin et al. (2012) study.

The predicted MFE does not agree with the plant tissue analysis or the yield results from the field trial (Table 4). Using the MFE, one would expect the N uptake from A High to be 61% that of U High; however, total N uptake with A High was 92% of uptake from U High and there was no significant difference. Meaning that A High was as effective at delivering N to the corn as U High. The difference could be a result of the range of products tested by Delin et al. (2012), as products with higher C:N ratios impacted the slope of the C:N regression, possibly under-predicting the N mineralization from these low C:N products.

The N use efficiency (NUE) (treatment N uptake-control N uptake divided by total N applied) was relatively high in this trial, ranging from 61% to 88% of total N applied (Table 4). The more typical range is from 30% to 70% (Legg and Meisinger, 1982) depending on N loss from denitrification, NH_3–N volatilization, NO_3^--N leaching, and sampling and analysis errors with variability ranges dependent on the mineral soil constituents (Hargrove, 1988; Nannipieri et al., 1990; Nannipieri et al., 1999). However, it has been shown that NUE can be in this range (70-80%) if the N is applied below the soil surface and in phase with crop demand (Meisinger et al., 2008). It has been shown that applying the correct rate of N is the single most important factor in improved NUE (Power and Scheper, 1989; Magdoff, 1991;

Freeman et al., 2008), leading to a possible conclusion that 100 kg N ha^{-1} (equivalent to the A Low treatment) was the optimal rate in this field trial.

As every effort was made to reduce N losses, I am confident in these NUE calculations. I assume minimal loss from NH_3 volatilization as all N fertilizers were banded at a depth of 5 cm, irrigated that evening, and had a pH of 5.7 (Bouwmeester et al., 1985; Hargrove, 1988; Meisinger et al., 2008). Denitrification can also be assumed to be minimal, because the soils were well drained and the soil was not saturated, even when sampled shortly after a heavy irrigation event. Urea hydrolysis occurs rapidly, with peak NH_4^+-N concentrations measured after 2 days (Nannipieri et al., 1990). The trend toward lower NUE in urea treatments may have been a result of the initial NH_4^+-N release rate being greater than the corn N uptake rate, causing more N to be lost to the environment. I did see elevated NH_4^+-N concentrations after fertilization but U High was not different than other fertilizer treatments one week after application. Nitrate-N was not measured at depth throughout this trial but the reduction of surface soil NO_3^--N levels through the growing season, coupled with the results that all post-harvest NO_3^--N concentration values were below 10 μg g^{-1} dry soil indicating low leaching potential (Marx et al., 1999), led me to conclude that little N was lost by leaching. A portion of the additional N could also have gone into microbial biomass; however, the literature would suggest that soils fertilized with synthetic N may decrease microbial biomass N, as well as microbial respiration and potential enzymatic activity (Treseder, 2008; Ramirez et al., 2012). Any N not lost

from the system is considered sorbed to the mineral surface with typical ranges from 20% to 60% (Hargrove, 1988; Francis et al., 2008).

One assumption pervasive throughout N mineralization efficiency calculations (Eqn. 2) is that the microbial population responsible for mineralization of organic matter to inorganic N mineralizes the same amount of N from the soil organic matter (SOM) regardless of the rate at which N is added. However, Zaman et al. (1999) and Ramirez et al. (2012) have shown that during periods of luxury N availability, less of the SOM N is mineralized. Without isotopic labeling of amendments, it is impossible to calculate how the N mineralization dynamics of the SOM changed with increased N application; however, looking at the NUE of plots fertilized with different rates, one can see that in plots with lower application rates more of the "amendment N" was taken up, so it would appear that more was mineralized (Table 4). It is possible that at lower rates of N addition more N was mineralized from the SOM in order to meet the total N demand. Whereas at high application rates the microbial community did not need to invest as much energy in extracellular enzyme production to acquire the same amount of N. Further, reports showing net N immobilization during the entire growing season (Nadelhoffer et al. 1984; Giblin et al., 1991; Polglase et al., 1992), and N mineralization rates well below estimates of plant uptake based on N accumulation in plants (Dyck et al., 1987; Chapin et al., 1988), suggest that, at least, in low-N ecosystems the core assumption underlying N mineralization assays may be invalid. Further testing with

isotopic labeling is needed to quantify the difference in SOM N mineralization dynamics under varying levels of C and N additions.

There was no significant difference between the ear yield, ear count, or ear N concentration (Fig. 13, Table 7) of the amended treatments. This raises questions about the N application rates, which were based on PSNT values. The PSNT, done with samples taken on June 22, when the plants had five leaves, indicated that 140 kg N ha^{-1} were needed for optimal crop performance. The PSNT protocol recommends taking a 30 cm soil sample for NO_3^- analysis in between rows, away from the fertilizer band (Hart et al., 2010); however, soil was sampled to a depth of 15 cm, as it was during all soil samples collected during the growing season. This difference in sampling depth probably did not affect the PSNT because no statistical difference was found post-harvest in soil NO_3^- -N between the 0-15 cm and 16-30 cm depths. The A High, F High, and U High treatments were applied above the recommendation for optimal yield. This was done because I estimated that 75% of the N applied (Gilmour, 1998; Gale et al., 2006; Sullivan et al., 2010) would be mineralized from the organic amendments, resulting in expected PAN of 76 kg N ha^{-1}, 117 kg N ha^{-1}, and 143 kg N ha^{-1} for the low, medium, and high rates, respectively. The urea was applied at the same N rate as the high rate for organic amendments to capture a maximum productivity value and for statistical representation. In future work multiple rates of urea should be used for comparison.

One of the most significant economic concerns for organic producers is the timing of N mineralization of organic amendments (Pang and Letey, 2000). Compost

and manures have a relatively slow N mineralization rate, with means around 0.6-1 mg N kg^{-1} day^{-1} with immobilization periods of one month being typical (Sullivan et al., 1999). For this reason, it is often recommended that composts and manures are applied substantially before peak crop N demand in order to build up the available N. However, if too much mineralization occurs before peak crop N demand the potential for NO_3^--N leaching increases. In order to reduce N inputs to the whole system and increase NUE, N should be applied so as to be available only when the crop needs it (Raun and Johnson, 1999). The N uptake curve for corn follows a sigmoidal pattern with a very slow uptake and growth for the first month after emergence until it reaches a critical mass for leaf area and photosynthetic activity (Freeman, 2008; Hart et al., 2010). The next 4-week period is marked by rapid N uptake and rapid biomass growth (Richie and Hanway, 1982; Hart et al., 2010). This is the most critical period for N nutrition; if adequate N is not available during this period it will result in a significant loss of yield (Richie and Hanway, 1982). After silking, the rate of N uptake slows to a more moderate pace and the N in the corn is reallocated from the leaves into the grain. In order to assess the timing of N mineralization relative to corn N uptake demands, the % N mineralized during the laboratory incubation (26^o C) was plotted relative to growing degree-days (GDDs) (data from Hyslop weather station, 3455 NE Granger Rd, Corvallis, OR, 97330), such that one month of GDDs in the field was equivalent to 21 GDDs in the laboratory. During this period the A2L mineralized the equivalent of 100 kg N ha^{-1}, A2M mineralized 107 kg N ha^{-1}, A2H mineralized 105 kg N ha^{-1}, and F1M mineralized 81

kg N ha^{-1} from the fertilizers alone. The estimated crop N uptake during this month is 146 kg N ha^{-1} (Freeman, 2008). Charting the N release from the laboratory incubation, including the N mineralized from SOM over the growing season, the N mineralized in the lab incubation from treatment A2M and F1M align closely with a corn N uptake curve modeled by Richie et al. (2005) while the N mineralized from the control soil and the N mineralized from A1 would not meet the sweet corn N demand (Fig. 14).

In a paper on the mechanisms and controls on stabilization and destabilization of soil organic matter, Sollins et al. (1996) reported the percent of degradation of many different polymers, including proteins from *Chlorella*, a green alga. In the non-allophanic soils (which caused increased OM sorption and decreased mineralization), the proteins were 58-67% degraded after 12 weeks of laboratory incubation as measured by ^{14}C-labeled substrates (Zunino et al., 1982). This is similar to our results from the A2 and F1 treatments, which showed 51-73% of the applied N was mineralized (Table 4). Our results are also similar to rates reported in Delin et al. (2012), who found up to 5% N mineralization per day during the first few days and about 14% per week for the first month. Gale et al. (2006) also found similar weekly mineralization of the feather meal tested through a 70-day laboratory incubation where 65% and 74% of the total N added was mineralized.

The organic amendments tested here, with C:N ratios lower than that of the total microbial population, mineralized about 25 μg inorganic N g^{-1} dry soil in the first week after application. Though a large quantity of inorganic N was not

immediately available the mineralization rate kept pace with the corn N uptake, this allows growers to time the application of these products similarly to how they would use a conventional fertilizer, possibly at lower than conventionally-recommended rates. Further testing with multiple rates of feather meal, urea, and algal meal is necessary to determine if the low rate of algal meal can sustainably induce N uptake equal to the higher rates that experience N abundance scenarios.

Carbon

Soils can be a source or a sink for C as they represent one of the largest reservoirs of organic C on the global scale (Schlesinger, 1995). The amount of soil C storage is dependent on inputs by net primary production and organic amendments, and their decomposition rates (Lutzow et al., 2006). There is uncertainty about how OM will respond to climate change (Heimann and Reichstein, 2008); however, it is expected that as global average temperatures increase, the rate of microbial respiration will increase, further tipping the C balance from the soil to the atmosphere (Schlesinger, 1995; IPCC, 2007). Long-term agricultural soil microbial populations tend to be dominated by heterotrophic bacteria that obtain C, energy, and reducing equivalents from SOM (Sylvia et al., 2005). The rate at which these organisms reproduce, produce enzymes, and turn over nutrients is dependent upon the quantity and quality of organic matter available (Schneider et al., 2012; Booth et al., 2005). Since nutrient mineralization is dependent on the activity of microbial

communities, adding organic matter to the soil is considered necessary to sustain soil fertility (Stewart et al., 2007; Freeman et al., 2008). Organic materials with low C:N ratios are typically considered high quality, leading to rapid decomposition and mineralization (Manzoni and Porporato, 2009; Booth et al., 2005).

To measure the rate of organic matter decomposition and calculate the residual C remaining in the soil, CO_2-C respiration was measured in both the laboratory and field trials. The method used to measure the CO_2-C in the field was not frequent enough and did not provide enough precision to detect differences among treatments; however, the lab incubation had lower variability and was not confounded by plant interactions. The control soil respired roughly 75% as much CO_2–C as the amended soils. The total CO_2-C respiration from each treatment was 69%, 71%, 63%, and 91% of the total C added in the A2L, A2M, A2H, and F1M treatments respectively (data not shown). When the respiration from the control soil was subtracted from that of the amended soils, about 20% of the added C was calculated as lost as CO_2, leaving about 80% of the amendment C in the soil. Applying these percentages of added C remaining in the soil from the laboratory incubation to the field scale I estimated the amount of the net C input: A Low (441 kg C ha^{-1}), A Med (673 kg C ha^{-1}), A High (810 kg C ha^{-1}), and F High (635 kg C ha^{-1}). These additions represent 0.2-1.3% of the total C in the soil, a marginal increase that must be tested further before credits for C sequestration can be determined.

The high percentage of N mineralized from the amendment relative to the C respired from the amendment would suggest that C was limiting to the microbial

decomposers. It is generally thought that N limits plant growth and C limits microbial growth (Schimel and Bennett, 2004; Myrold and Bottomley, 2008). This is consistent with the high corn NUE and optimized corn yield found in this trial. Because a low percentage of the C was respired, one would expect that C would accumulate in the soil, becoming less limiting to microorganisms in the future (Kleber and Johnson, 2010). However, the C could also become sorbed to the mineral soil surfaces becoming protected from future microbial degradation (Kleber and Johnson, 2010).

Potential Additional Benefits

Although the focus of my study was primarily on the potential of algal meal to supply plant available N, and to a lesser extent, microbially available C, it is important to remember that there may be additional benefits of algal meal additions. Research on agricultural use of organic amendments has consistently shown soil bulk density and penetration resistance decreasing with increasing amendment rate, while aggregate stability, porosity, and infiltration rate increase with amendment rate (Cogger et al., 2004). In a field trial with corn, Paul and Beauchamp (1996) showed that a single application of dairy cattle slurry increased the microbial biomass C, resulting in greater N mineralization, and greater corn N uptake than treatments amended with urea. In an extensive review on the effects of organic amendments on soil physical properties Khaleel et al. (1981) found that organic C content increased and bulk density decreased as organic amendment

increased. They also found a significant correlation between increased organic C percentage and the increase in water holding capacity (Khaleel et al., 1981). However, these benefits are largely seen when adding manures, composts, and biosolids at high rates resulting in large C additions and may not be applicable to algal and feather meal used at agronomic rates.

Studies on seaweed components, such as macro- and micronutrients, amino acids, vitamins, cytokinins, auxins and abscisic acid-like growth substances, show that these components affect cellular metabolism in treated plants leading to enhanced performance, growth, and yield (Beckett and van Staden, 1989; Blunden, 1991; Crouch and van Staden, 1992; Crouch and van Staden, 1993; Reitz and Trumble, 1996; Durand et al., 2003; Stirk et al., 2003). The current study shows equivalent corn yield from a plot fertilized with 101 kg N ha^{-1} of algal meal and plots fertilized with 190 kg N ha^{-1} of urea. The evidence would suggest that the value of algal meal is greater than the sum of the macronutrient content. Further testing will be required to determine if the benefits of low-N seaweed amendments, composts, and manures can also be extended to algal meal.

Market Valuation

Algal co-products from each different kind of proprietary production method have their own inherent C:N ratio, NPK content, concentration of micronutrients, and electrical conductivity, each lending to the overall nutritive value. By showing that algal meal can be as effective as both the conventional and organic standard at

delivering N to a crop, I have validated assigning a price equivalent to the current market value of conventional and organic fertilizers on a price per unit of nutrient basis (Table 8). Urea delivers a 45% N product to the grower for \$611 Mg^{-1} (USDA ERS, 2010), this equates to \$1.36 kg^{-1} of N. Therefore algal meal is worth \$85 Mg^{-1} in the conventional market based on the value of N alone. However, organic high N products average \$10.41 kg^{-1} N (Skinner, 2013) making algal meal worth \$729 Mg^{-1} based on the value of organic N alone. Assuming P and K are as available from algal meal as they are from other organic sources, if the value from the P and K is included the conventional price reaches \$140 Mg^{-1} and the organic price reaches \$1054 Mg^{-1}. Under current market conditions, including marketing costs and transportation expenses, an algae facility could expect a sale price of \$800 Mg^{-1} for dry algal meal with an NPK of 7-3-1. This valuation does not include any potential benefits of the product beyond the macro nutrient values. Higher values should be included after quantification of the benefits in C sequestration, aggregate stability, micronutrient additions, phytohormone interactions, and energy consumption offsets if any.

Conclusions

The results of this one-year preliminary trial support the original hypothesis: algal meal can be as effective as feather meal as an organic fertilizer product on a per unit of N basis. Specifically, algal meal had statistically the same N

mineralization rate, though A2 had a faster initial release while F1 peaked later. Additions of algal meal and feather meal resulted in statistically similar microbial respiration rates, though the algal treatments added more C per unit of N and respired a smaller portion of what was added resulting in a greater net addition of C to the soil. I did not detect a treatment difference in extracellular enzyme production. The yield of sweet corn showed that algal meal was at least as effective as feather meal and could be just as effective at lower application rates. In this irrigated, silty clay loam soil the high salts did not result in any visible signs of osmotic stress (such as leaf curling, leaf-tip browning, or cob nubbins).

In calculating the EROI of an algae biofuel production facility, the value of all products must be estimated in both the near and long term. Biodiesel prices and government tax credits are difficult to predict and subject to changes in political will, making it difficult for investors to have confidence providing a 10-year loan to cover the capital expenditure of the start-up costs. The algal industry currently undervalues the co-product, using values from 0 to \$170 Mg^{-1} (Chisti, 2007; Cantrell et al., 2008; Sanders and Murthy, 2009; Sakthivel et al., 2011; NAS, 2012). Utilizing the fertilizer value of algal meal at \$800 Mg^{-1} could help algae fuel start-up companies secure financing and economic profits.

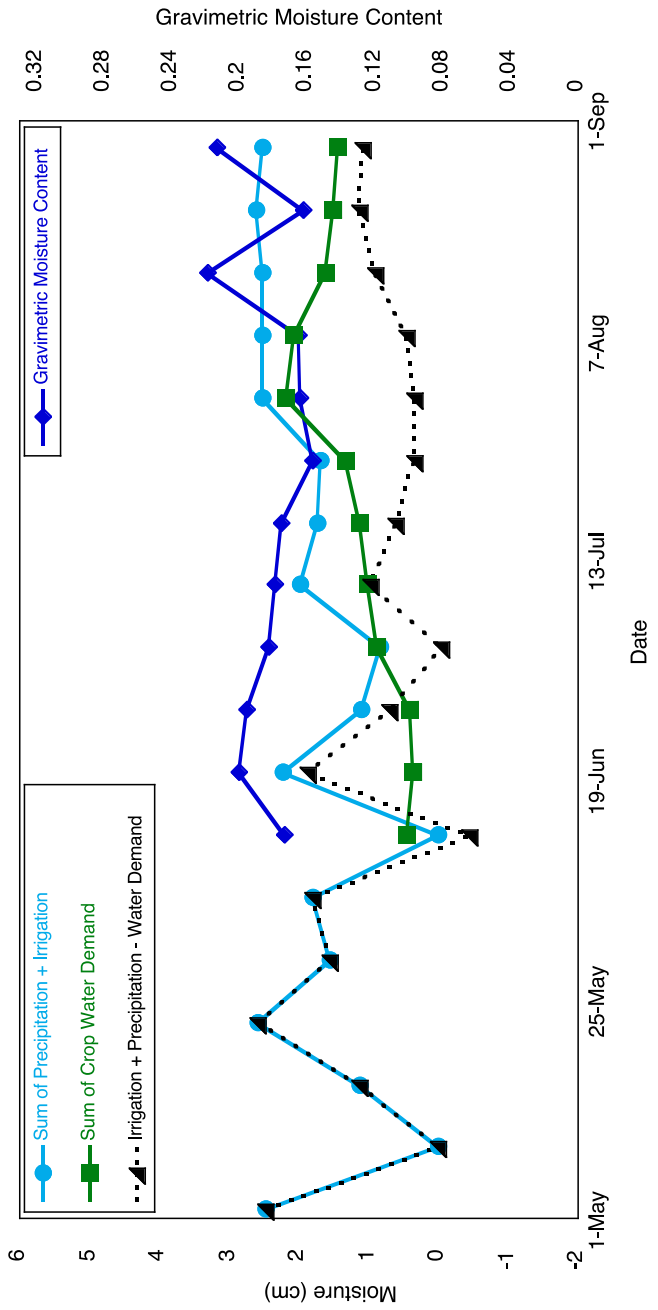

Fig. 2. Field Trial Soil Moisture Status
Weather data from Corvallis, OR Hyslop Farm and AgriMet. Data points are the sum of the respective moisture measurement for the previous week. Crop water demand was calculated using Agrimet evapotranspiration reference for alfalfa multiplied by a coefficient for sweet corn. The precipitation + irrigation – crop water demand shows that the crop was not under moisture stress.

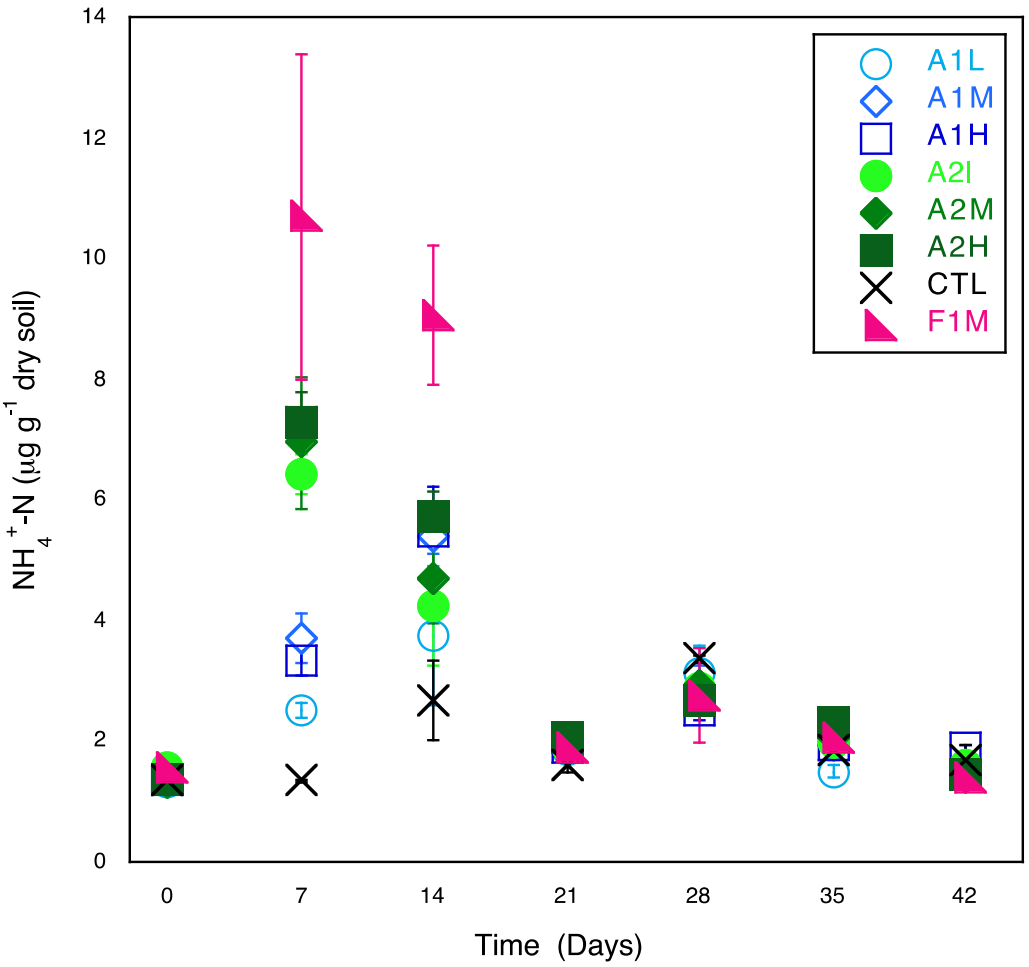

Fig. 3. Ammonium Production through the Laboratory Incubation
Treatments are: A1 = algae meal 1, A2 = algae meal 2, CTL = no amendment added, F1 = feather meal, with the last letter indicating the rate (L= low, M=medium, or H=high). Data are means (n=3). Error bars represent standard error.

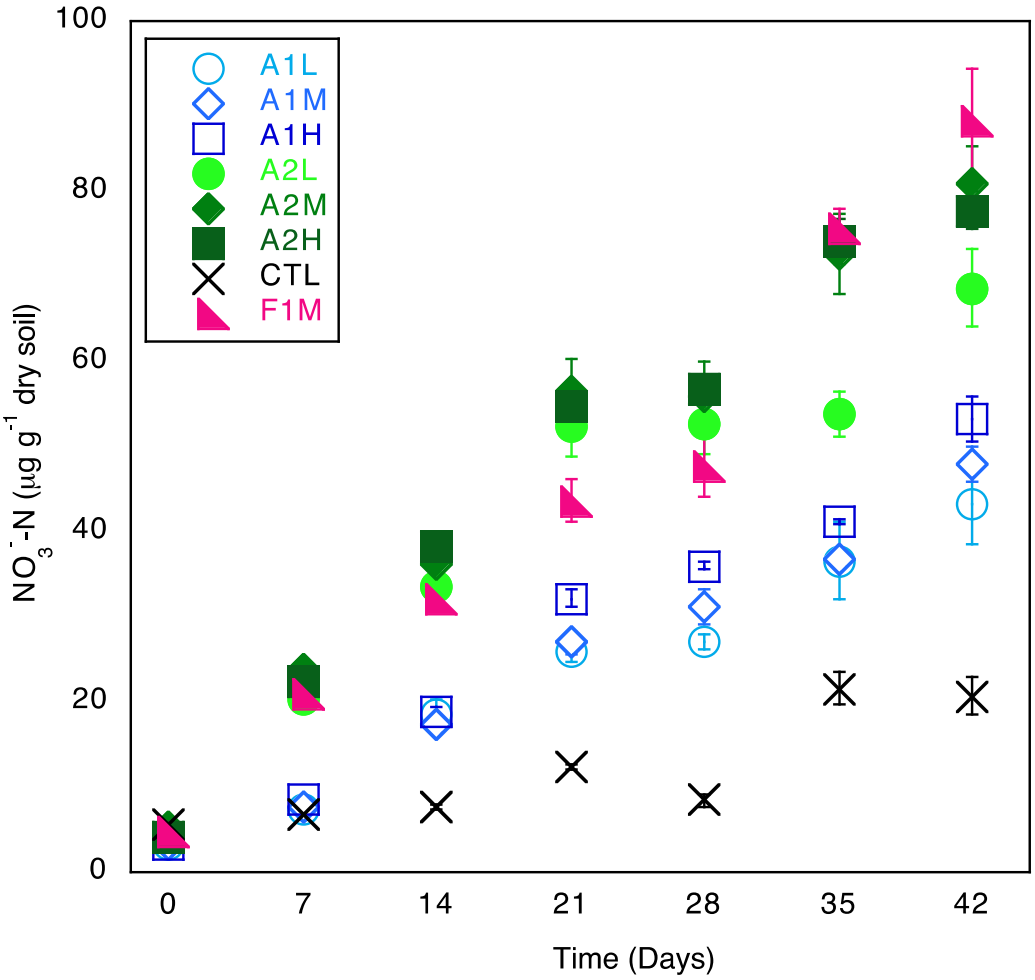

Fig. 4. Nitrate Accumulation through Laboratory Incubation
Treatments are: A1 = algae meal 1, A2 = algae meal 2, CTL = no amendment added, F1 = feather meal, with the last letter indicating the rate (L= low, M=medium, or H=high). Data are means (n=3). Error bars represent standard error.

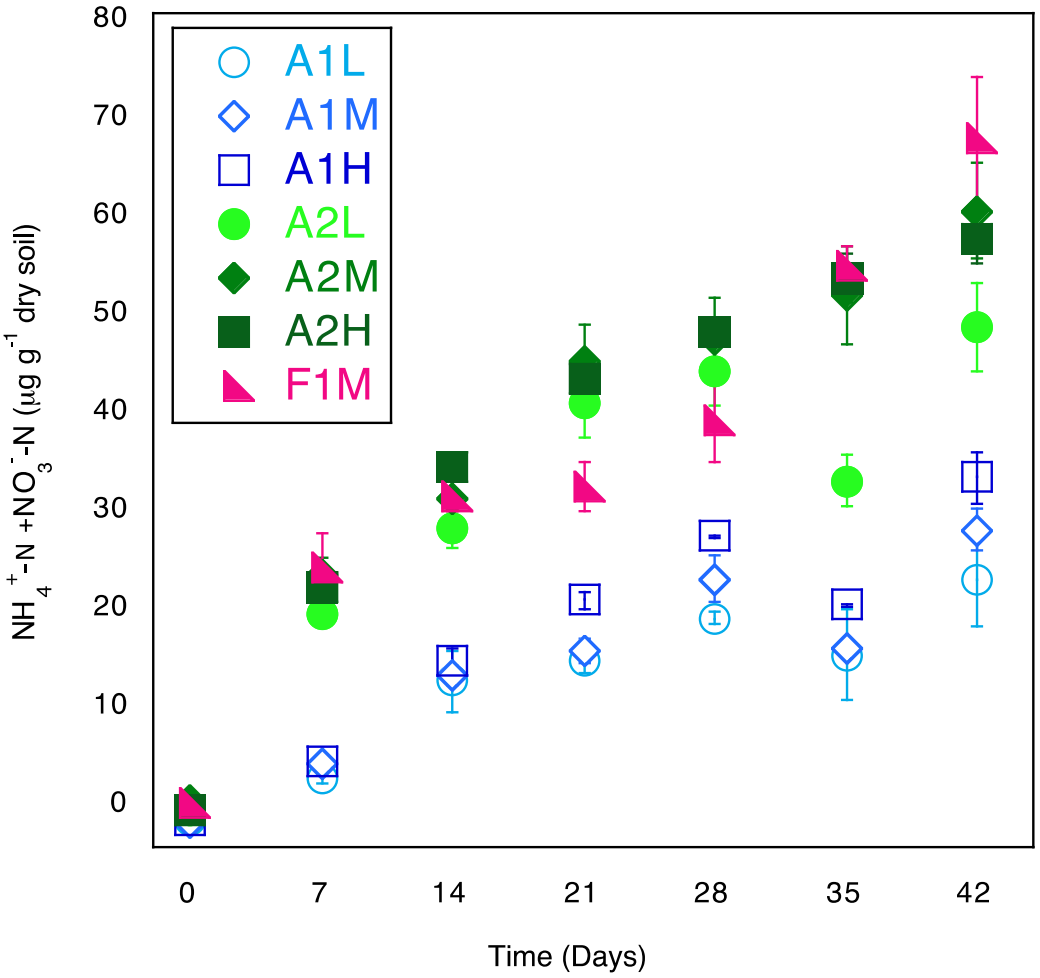

Fig. 5. Amendment N Mineralization
(Treatment NH_4^+-N +NO_3—N [final-beginning])-. (Control NH_4^+-N +NO_3—N [final-beginning]). Treatments are: A1 = algae meal 1, A2 = algae meal 2, CTL = no amendment added, F1 = feather meal, with the last letter indicating the rate; L= low, M=medium, or H=high. Data are means (n=3). Error bars represent standard error.

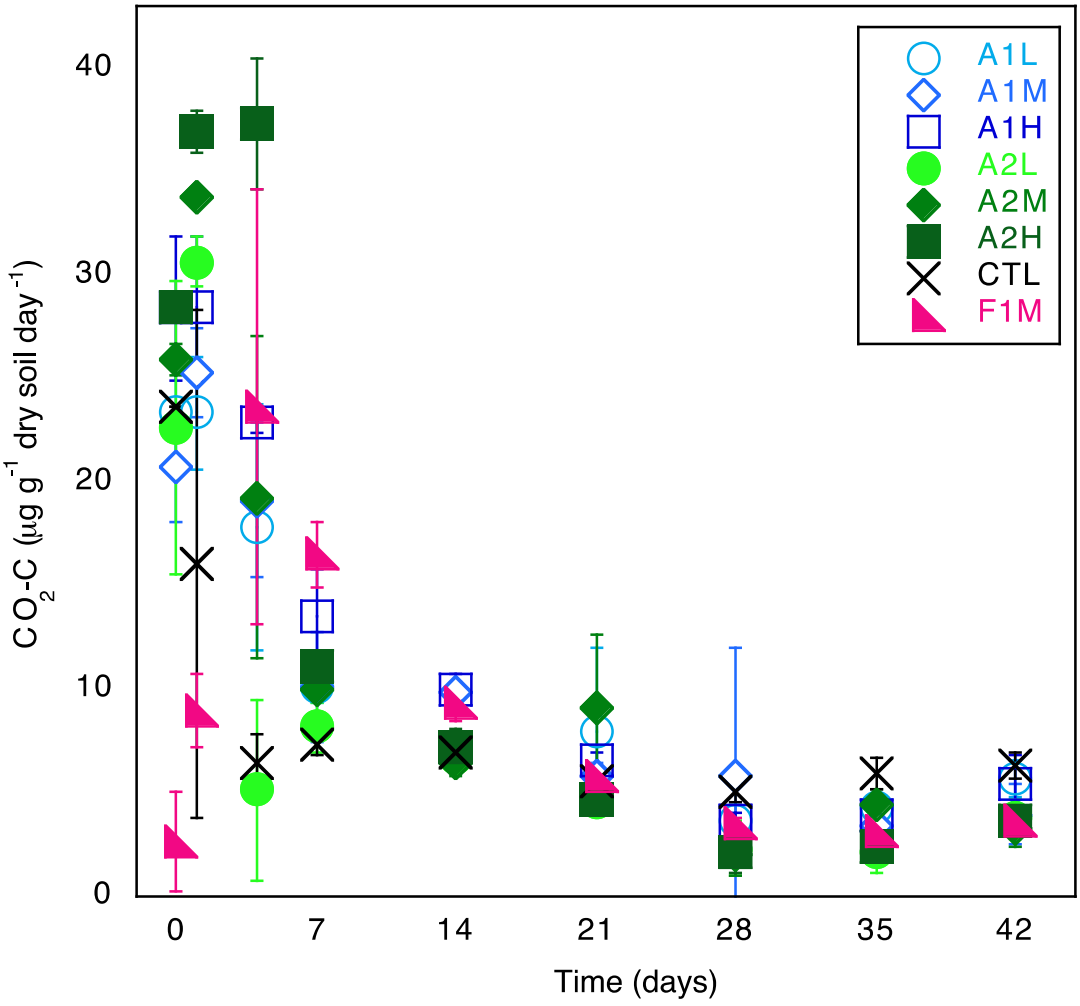

Fig. 6. Carbon Dioxide–C Production through Laboratory Incubation
Treatments are: A1 = algae meal 1, A2 = algae meal 2, CTL = no amendment added, F1 = feather meal, with the last letter indicating the rate; L= low, M=medium, or H=high. Data are means (n=3). Error bars represent standard error.

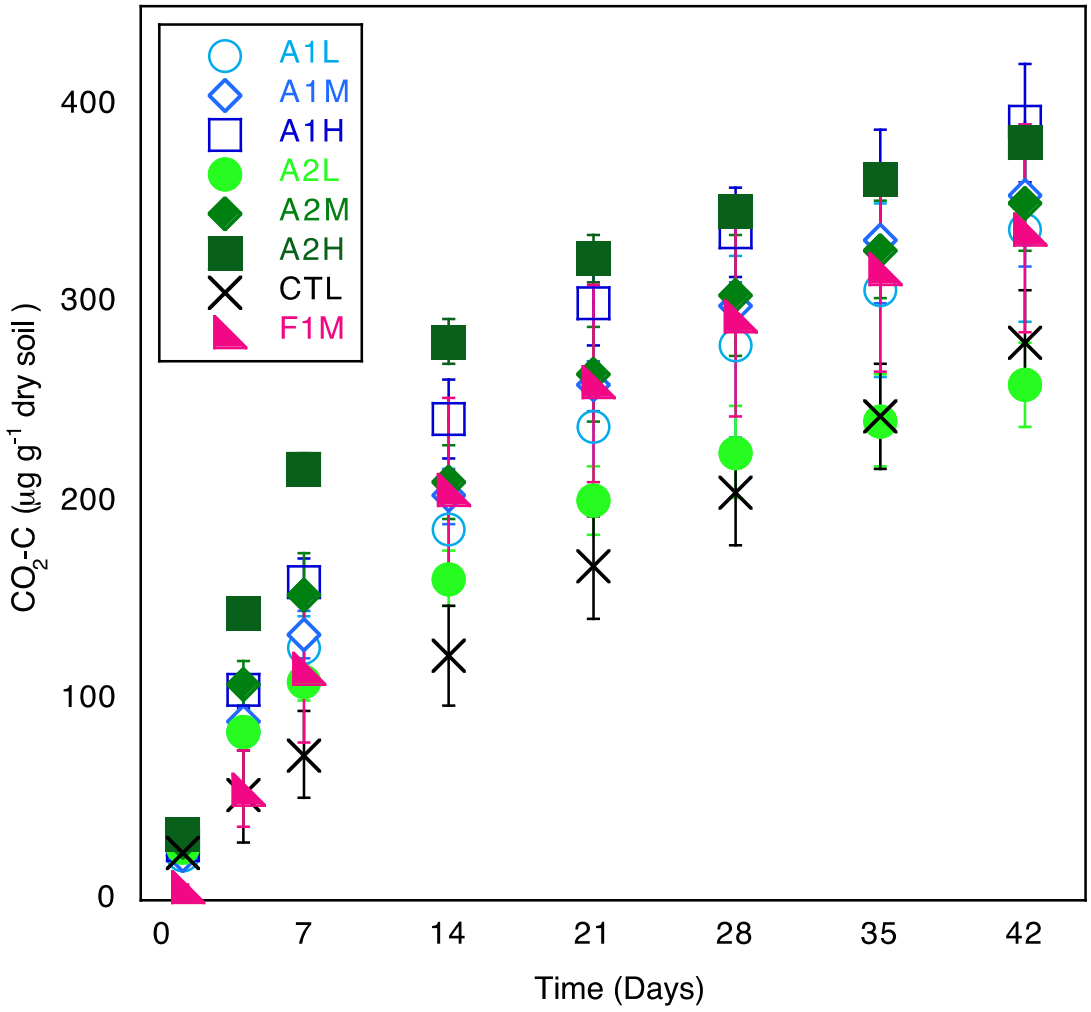

Fig. 7. Cumulative CO_2-C Respiration through Laboratory Incubation
Treatments are: A1 = algae meal 1, A2 = algae meal 2, CTL = no amendment added, F1 = feather meal, with the last letter indicating the rate; L= low, M=medium, or H=high. Data are means (n=3). Error bars represent standard error.

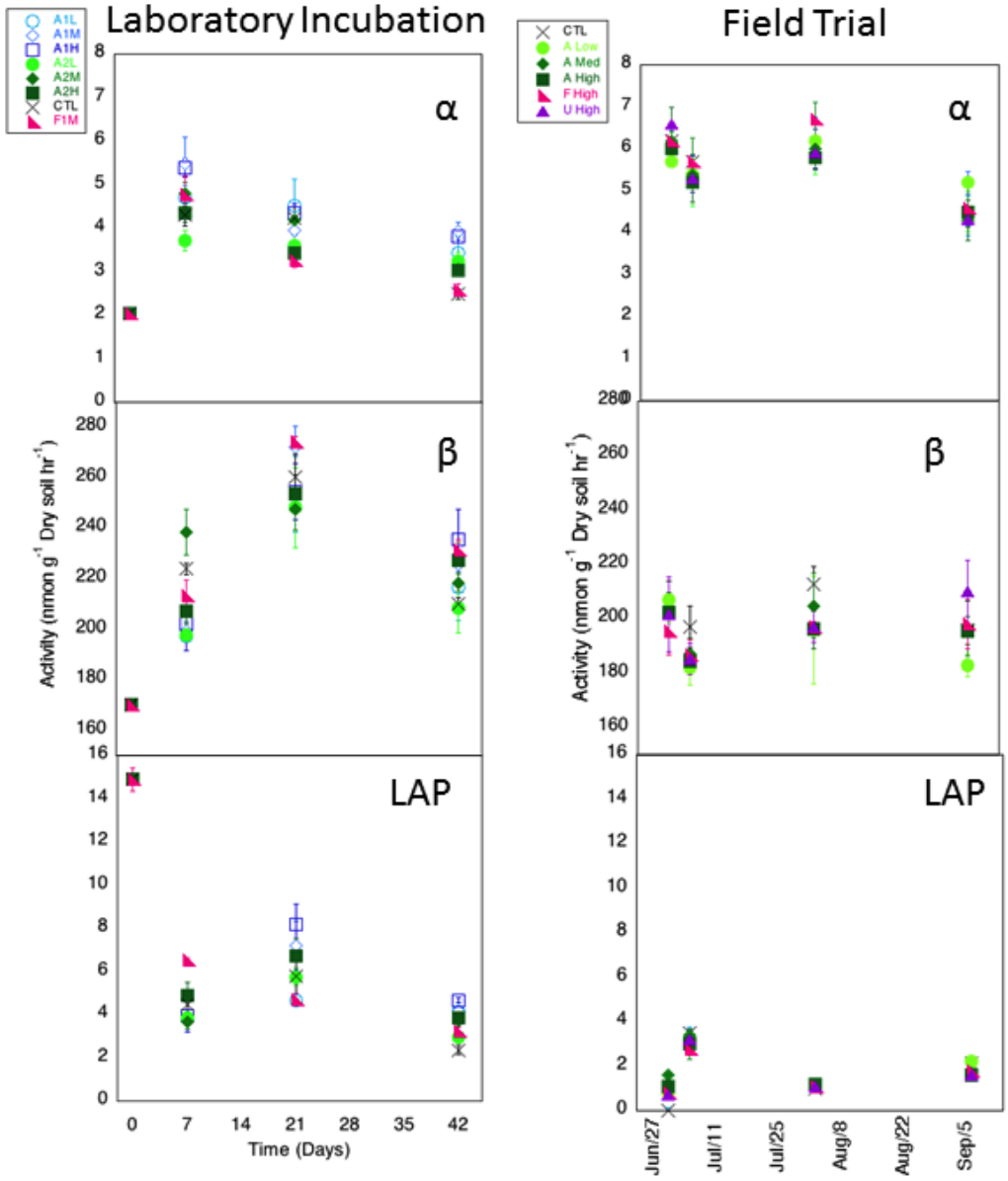

Fig. 8. Extracellular Enzyme Activity in Laboratory Incubation and Field Trial Laboratory incubation treatments are: A1 = algae meal 1, A2 = algae meal 2, CTL = no amendment added, F1 = feather meal. The last letter indicates the rate; L= low, M=medium, or H=high. Field trial treatments are: CTL= no N added, A Low = A2 101 kg N ha^{-1}, A Med = A2 146 kg N ha^{-1}, A High = A2 190 kg N ha^{-1}, F High = F1 190 kg N ha^{-1}, U High = Urea 190 kg N ha^{-1}. α = alpha-glucosidase β = beta-glucosidase LAP = leucine-aminopeptidase. Data are means (laboratory n=3, field n=4). Error bars represent standard error.

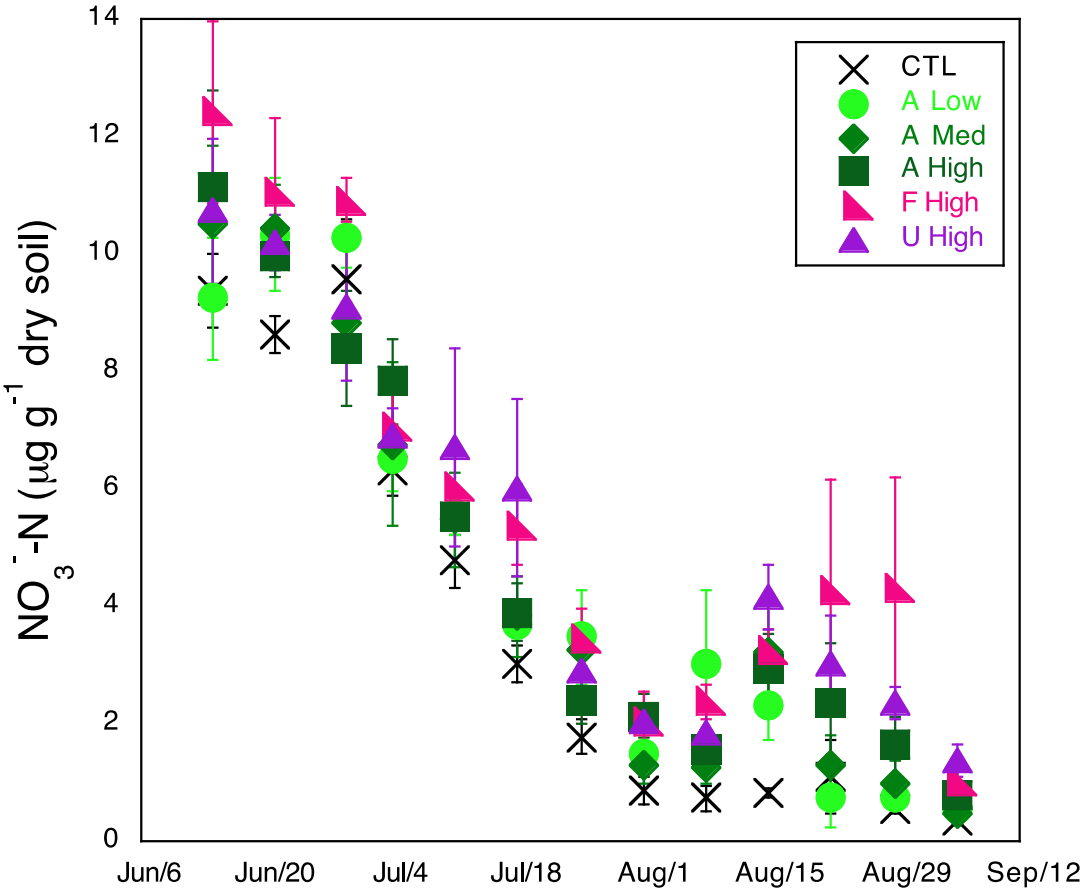

Fig. 9. Nitrate Concentrations through Field Trial
Treatments are: CTL= no N added, A Low = A2 101 kg N ha^{-1}, A Med = A2 146 kg N ha^{-1}, A High =A2 190 kg N ha^{-1}, F High =F1 190 kg N ha^{-1}, U High =Urea 190 kg N ha^{-1}. Organic fertilizers applied June 29th, urea fertilizer applied July 4, before sampling. Data are means (n=4). Error bars represent standard error.

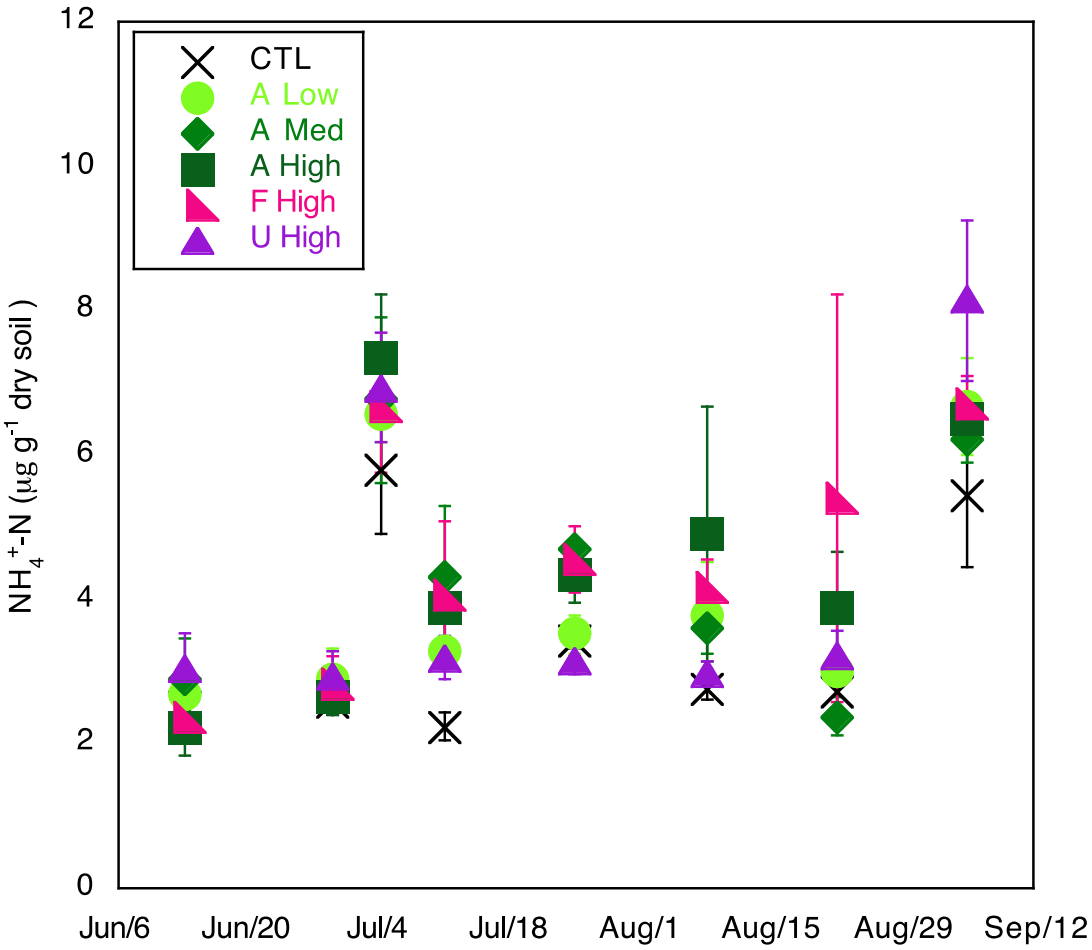

Fig. 10. Ammonium Concentration through Field Trial
Treatments are: CTL= no N added, A Low = A2 101 kg N ha^{-1}, A Med = A2 146 kg N ha^{-1}, A High =A2 190 kg N ha^{-1}, F High =F1 190 kg N ha^{-1}, U High =Urea 190 kg N ha^{-1}. Organic fertilizers applied June 29th, urea fertilizer applied July 4, before sampling. Data are means (n=4). Error bars represent standard error.

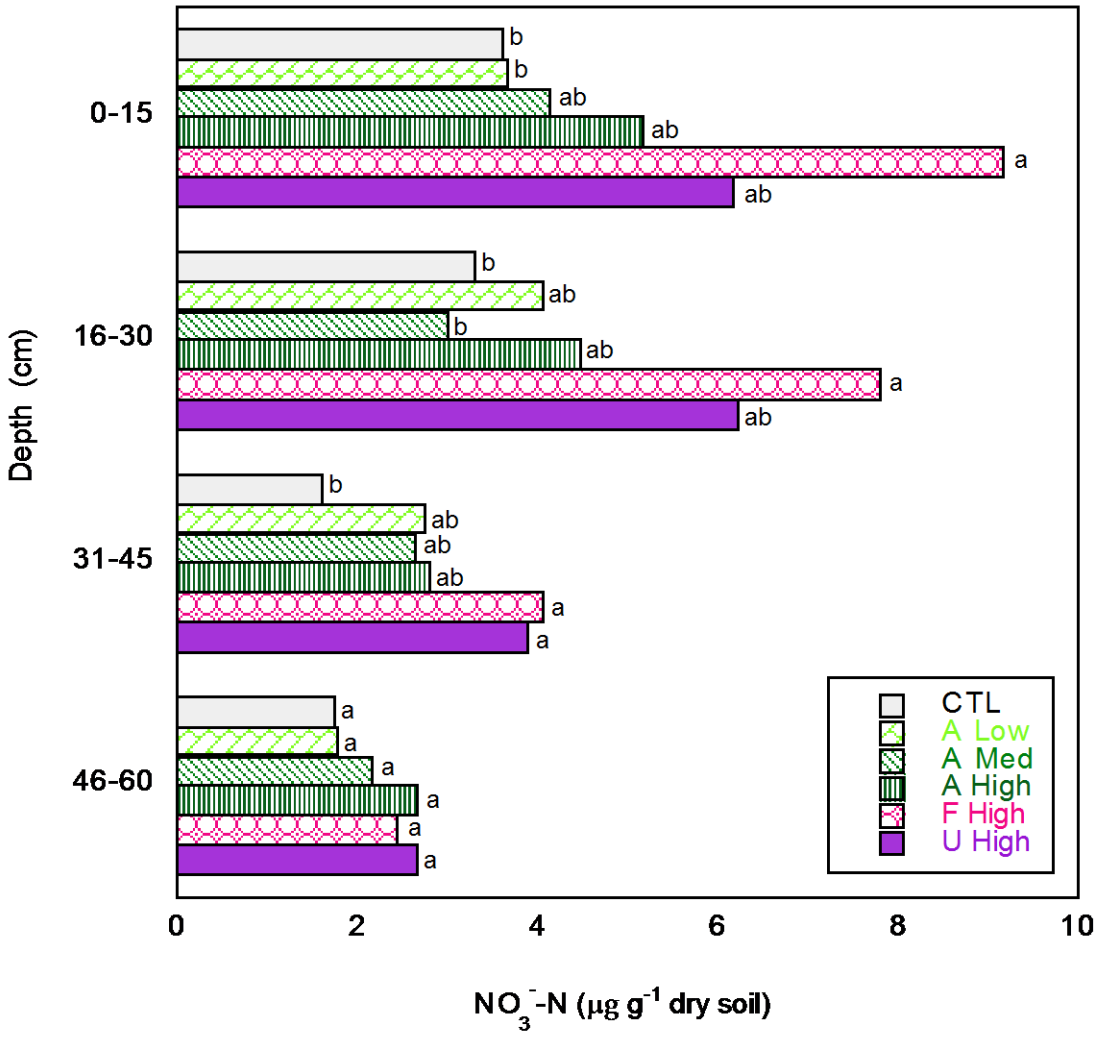

Fig. 11. Nitrate Concentration in Soil 3 Weeks After Harvest
Treatments are: CTL= no N added, A Low = A2 101 kg N ha^{-1}, A Med = A2 146 kg N ha^{-1}, A High =A2 190 kg N ha^{-1}, F High =F1 190 kg N ha^{-1}, U High =Urea 190 kg N ha^{-1}. Data are means (n=4). Error bars represent standard error.

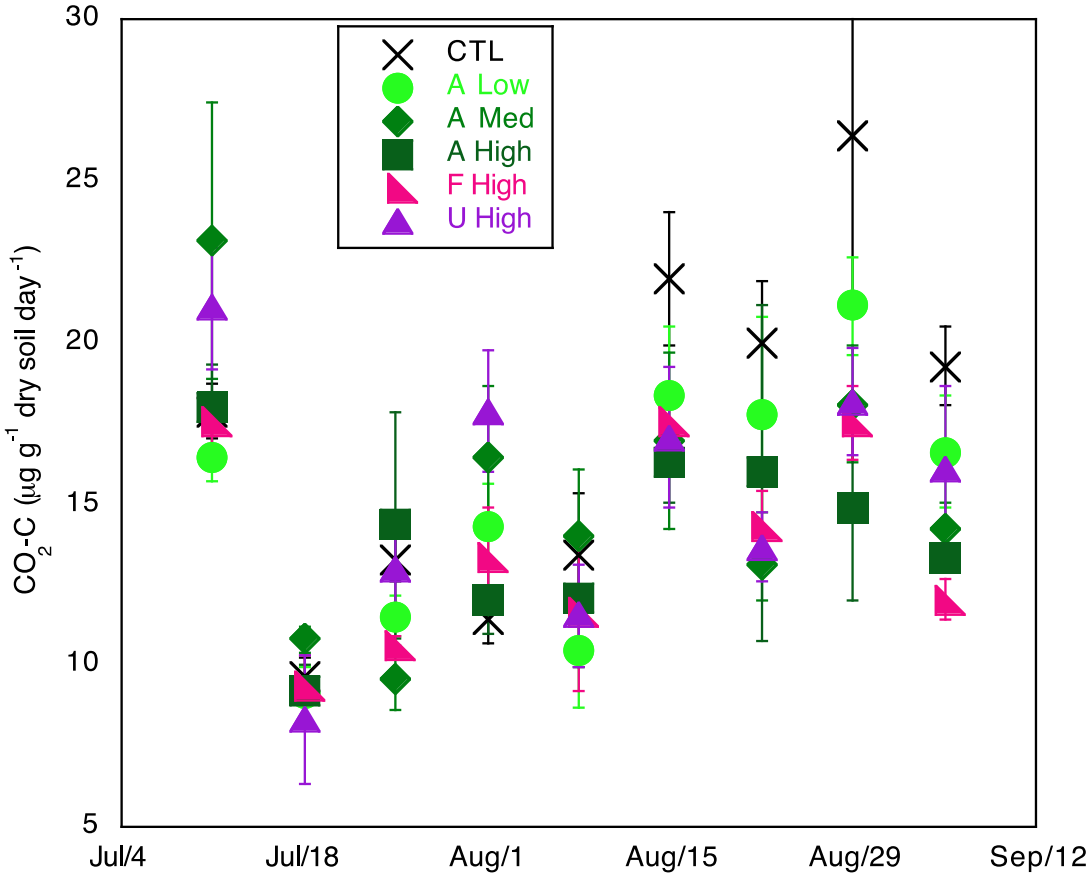

Fig. 12. Carbon Dioxide Concentration through Field Trial
Treatments are: CTL= no N added, A Low = A2 101 kg N ha^{-1}, A Med = A2 146 kg N ha^{-1}, A High = A2 190 kg N ha^{-1}, F High = F1 190 kg N ha^{-1}, U High = Urea 190 kg N ha^{-1}. Data are means (n=4). Error bars represent standard error.

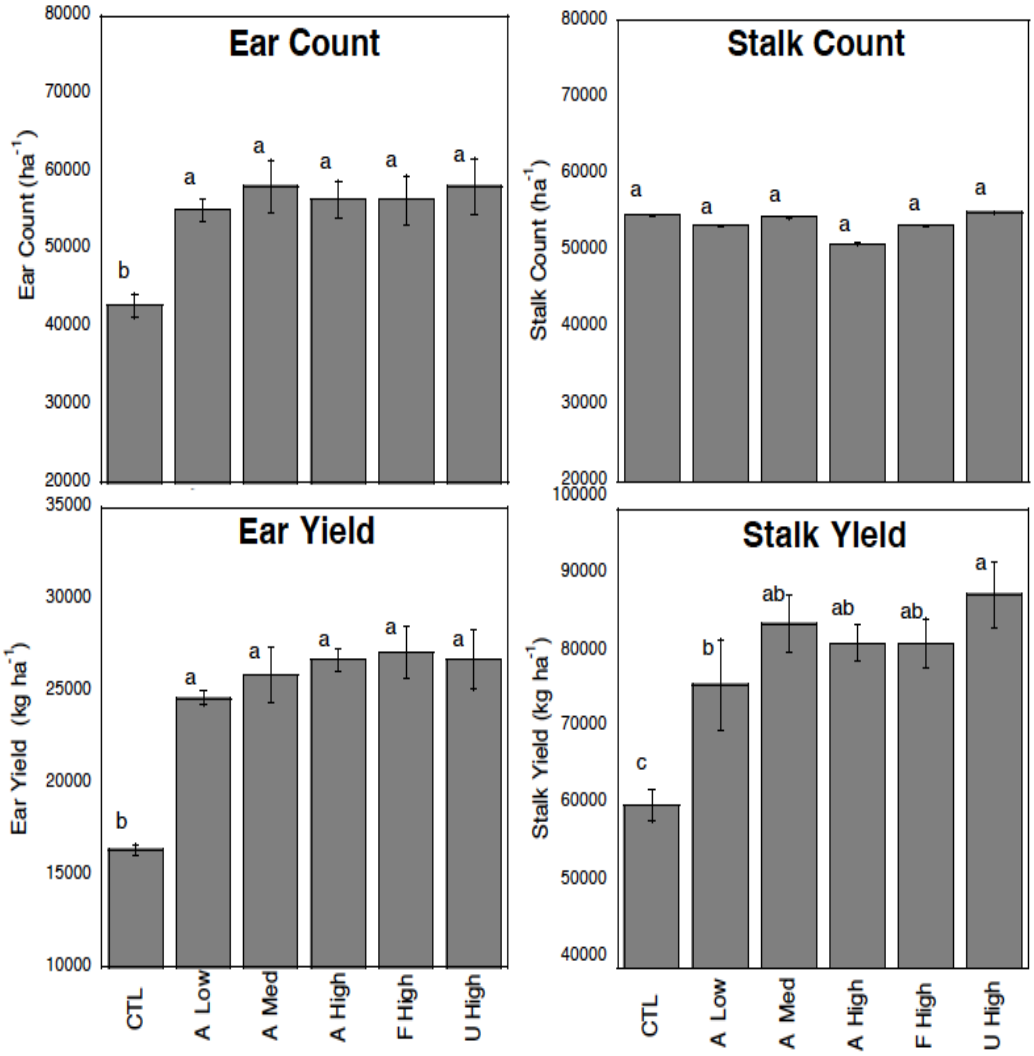

Fig. 13. Harvest Yield of Captain Sweet Corn 2013
Three meters of 3 rows of each experimental unit for each parameter were measured. Ha yields are extrapolations. Treatments are: CTL= no N added, A Low = A2 101 kg N ha^{-1}, A Med = A2 146 kg N ha^{-1}, A High = A2 190 kg N ha^{-1}, F High = F1 190 kg N ha^{-1}, U High = Urea 190 kg N ha^{-1}. Data are means of subsamples (n=12). Error bars represent standard error.

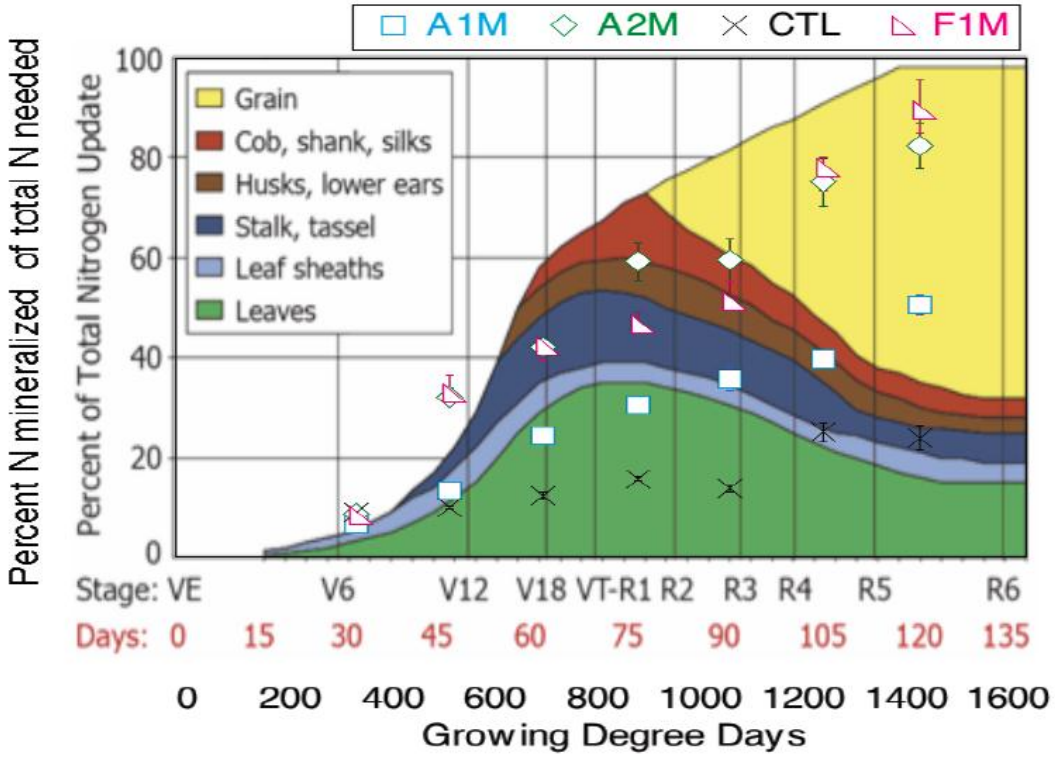

Fig. 14. Nitrogen Mineralization and Corn Uptake Timing
Treatments are: A1M = algae meal 1 med rate, A2M = algae meal 2 med rate, F1M = feather meal 1 med rate. Medium rate used in laboratory incubation was calculated to be the same as the high rate used in the field trial. Days are based off of field data from Richie et al. (1982). The lab incubation data was plotted on degree days, rather than incubation days. N uptake of sweet corn adapted from Richie et al., 1982. Howe a corn plant develops. Error bars represent standard error.

Table 1: Organic Fertilizer Nutrient Report for Amendments Used in Laboratory and Field Trial

	C	N	P_2O_5	K_2O	S	Mg	Ca	Na	Fe	Al	Mn	Cu	Zn	B	pH	EC	C:N
	----------%----------								----------mg kg^{-1}----------						-log[H+]	mS/cm	
A1	32.59	5.76	0.16	0.12	7.82	1.24	5.54	0.99	520	1849	24	57	22	31	2.47	9.02	5.7
A2	37.29	7.22	3.02	1.19	1.21	1.47	1	4.7	4583	2963	133	12	127	148	6.4	10.57	5.2
F1	50.1	12.5	1.05	0.13	1.5	0.05	1.02	0.11	304	76	11	20	98	25	5.63	2.47	4.0

Analysis done by A & L Western Agricultural Laboratories, Inc. Modesto, CA

Table 2: Metals Analysis of Organic Amendments Used in Laboratory and Field Trial

Analyte	A1	A2	F1	Detection Limit	Method Reference
	----------mg kg^{-1}----------				
Arsenic	0.63	1.68	BDL	0.25	EPA SW846-6016
Cadmium	0.05	BDL	0.03	0.03	EPA SW846-6016
Lead	1.1	1.7	BDL	0.5	EPA SW846-6016
Nickel	12.9	3.8	0.7	0.1	EPA SW846-6016
Mercury	BDL	BDL	BDL	0.05	EPA SW846-7471A

Analysis done by A & L Western Agricultural Laboratories, Inc. Modesto, CA

Sample Prepartation Method: EPA SW846-3050B

Table 3: Initial Soil Test Results

LOI	C	N	P	K	Ca	Mg	S	Na	Cu	Mn	Zn	Fe	B	NO_3-N	pH	EC
----------	%----------		----------						mg kg^{-1}	----------					-log[H+]	mS/cm
3.25	1.49	0.11	31	332	2430	656	100	60	2.8	40.9	0.2	76.7	0.3	0.67	5.8	0.08

Composite Sample - Taken 2012 Apr 27

Table 4: Nitrogen Mineralization, Uptake, and Efficiency

Fertilizer	Laboratory Incubation			Field Trial		
	Mineralization Rate	Fraction of N mineralized	Fraction of N mineralized less control N	Gross N Uptake	Net N Uptake	N Use Efficiency
	µg inoragnic N g^{-1} dry soil day^{-1}	µg inorganic N g^{-1} dry soil µg N^{-1} added	µg inorganic N g^{-1} dry soil µg N^{-1} added	kg N ha^{-1}	kg N ha^{-1}	treatment-ctl kg N uptake ha^{-1} N^{-1} added
A1L	0.95c	0.65c	.35c	no	no	no
A1M	1.07c	0.57c	.32c	no	no	no
A1H	1.20c	0.50c	.31c	no	no	no
A2L / A Low	1.54b	1.02a	.70a	206ab	89a	0.88a
A2M / A Med	1.80ab	0.89ab	.65ab	222ab	104ab	0.71a
A2H / A High	1.75ab	0.71b	.51b	234ab	117ab	0.61a
CTL	0.37d	na	na	118a	na	na
FIM / F High	1.98a	0.96a	.73a	239ab	121ab	0.64a
U High	no	no	no	253b	135b	0.71a

Means follwed by the same letter are not significantly different. Fied trial n=4; laboratory incubation n=3 α=0.05

na = not applicable, divided by 0

no = treatment not included in experiment

Table 5. pH and EC of Laboratory Incubation

Fertilizer	pH		EC mS cm^{-1}	
	week 1	week 6	week 1	week 6
A1L	5.67	5.17b	0.05	0.38c
A1M	5.7	5.17b	0.05	0.45ab
A1H	5.73	5.17b	0.05	0.46a
A2L	5.73	5.13b	0.05	0.38bc
A2M	5.73	5.17b	0.07	0.44abc
A2H	5.63	5.13b	0.07	0.44abc
CTL	5.67	5.60a	0.07	0.09e
F1M	5.63	5.13b	0.07	0.26d

Means follwed by the same letter are not significantly different n=3 α=0.05

Table 6. Nitrate Concentration at End of Growing Season

Treatment	----------μg NO_3^--N g^{-1} dry soil----------			
	8/15/12	8/22/12	8/29/12	9/5/12
CTL	0.80c	1.10c	0.55b	0.38c
A Low	2.33bc	0.75bc	0.78b	0.53bc
A Med	3.20ab	1.30bc	0.98b	0.45c
A High	2.98ab	2.35abc	1.65b	0.75bc
F High	3.25ab	4.30a	4.33a	1.03ab
U High	4.18a	3.03ab	2.38ab	1.38a

Means follwed by the same letter are not significantly different. n=4 α=0.05

Table 7. Plant Tissue Analysis for Macronutrients

Treatment	Nitrogen %		Phosphorus %		Potassium %	
	Stalk	Ear	Stalk	Ear	Stalk	Ear
Control	0.58b	1.00b	0.23ab	0.30a	1.63a	1.08a
A Low	0.84ab	1.23ab	0.24a	0.30a	1.71a	1.01a
A Med	0.79ab	1.32a	0.23ab	0.31a	1.64a	1.01a
A High	0.89ab	1.34a	.024ab	0.30a	1.68a	1.06a
F High	0.90a	1.42a	.023ab	0.30a	1.72a	1.09a
U High	0.99a	1.22ab	.022b	0.29a	1.59a	1.03a

Means follwed by the same letter are not significantly different. n=4 α=0.05

Table 8. Fertilizer Prices

PRODUCT	Conventional or organic	Nutrient delivered	Valued Nutrient of total product	Valued nutrient of total nutrients	Farm Delivered Price	Product Price	Nutrient Price
	c or o	NPK	%	%	per Mg	per kg	per kg
Urea	c	N	45	100	$ 610.68	$ 0.61	$ 1.36
Anhydrous ammonium	c	N	82	100	$ 863.11	$ 0.86	$ 1.05
Feather meal	o	N	12	100	$ 1,212.54	$ 1.21	$ 10.10
Blood meal	o	N	13	100	$ 1,433.01	$ 1.43	$ 11.01
Bat guano*	o	N	9	69	$ 912.71	$ 0.91	$ 10.13
Bat guano	o	P	3	23	$ 304.24	$ 0.30	$ 10.13
Bat guano	o	K	1	8	$ 101.85	$ 0.10	$ 10.18
Diammonium Phosphate	c	N	18	28	$ 224.08	$ 0.22	$ 1.24
Diammonium Phosphate	c	P	46	72	$ 576.20	$ 0.58	$ 1.25
Triple super phosphate	c	P	45	100	$ 733.04	$ 0.73	$ 1.63
Rock Phosphate	o	P	3	100	$ 496.04	$ 0.50	$ 16.52
Bone Meal	o	P	18	100	$ 661.39	$ 0.66	$ 3.67
Muriate of Potash (KCl)	c	K	60	100	$ 713.20	$ 0.71	$ 1.19
Sulphate of Potash	o	K	52	100	$ 1,102.31	$ 1.10	$ 2.12
AVERAGES OF							
Nitrogen Products	c	N	48		$ 565.96	$ 0.57	$ 1.22
Phosphate Products	c	P	46		$ 654.62	$ 0.65	$ 1.44
Potassium Products	c	K	60		$ 713.20	$ 0.71	$ 1.19
Nitrogen Products	o	N	11		$ 1,186.09	$ 1.19	$ 10.41
Phosphate Products	o	P	8		$ 487.22	$ 0.49	$ 10.11
Potassium Products	o	K	52		$ 1,102.31	$ 1.10	$ 2.12

From this we can calculate the conventional market value and the organic market value of algae

PRODUCT	Nutrient delivered	Nutrient %	Conventional Nutrient Price	Organic Nutrient Price	Conventional Farm Delivered Price	Organic Farm Delivered Price
	NPK	%	per kg	per kg	per Mg	per Mg
Algal Meal	N	7	$ 1.22	$ 10.41	$ 85.27	$ 729.61
Algal Meal	P	3	$ 1.44	$ 10.11	$ 43.22	$ 303.50
Algal Meal	K	1	$ 1.19	$ 2.12	$ 11.89	$ 21.20
Sum Nutrient Value					**$ 140.38**	**$ 1,054.31**

* For products that deliver multiple nutrients the price of the total product is multiplied by the percent of valued nutrient delivered

Conventional fertilizer prices from: Agricultural Prices, National Agricultural Statistics Service, USDA. 2012

Organic fertilizer deliverd bulk price averages from 201: Advanced Marketing Group, LLC Aurora, OR

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CHAPTER 3
COMPARATIVE ANALYSIS OF AN INTEGRATED SYSTEM UTILIZING ALGAL MEAL AS AN ORGANIC FERTILIZER

Abstract

Algae have shown great potential as a source for renewable fuels; however, current production schemes have not been able to prove a sustainable energy return on investment due to the high costs of nutrient addition and the energy required for drying the biomass. Integrated algae-dairy production systems have been posited as a potential solution for algal production barriers as well as a way to capture environmentally problematic nutrients excreted by animals in concentrated animal feeding operations. I hypothesized that, when considering environmental benefits and current policy incentives, utilizing the fertilizer value of algal meal at \$800 Mg^{-1} will show the integrated production facility to be an economically viable manure management system. Utilizing data from similar operations, I calculated the potential net present value, showing that the fertilizer value of algal meal adds enough income to make the system profitable.

Introduction

The impetus for quantifying the fertilizer value of algal meal was that the algal fuel industry is leaving money on the table by undervaluing 50-80% of their product. The protein-rich biomass, algal meal, has received little attention in the algal energy research world. Economic assessments (Chisti, 2007; Cantrell et al.,

2008; Sanders and Murthy, 2009; Sakthivel et al., 2011) have expressly stated that the value of the co-product has not been included in the overall valuation. Algal by-products from each different kind of proprietary production method have their own inherent C:N ratio, NPK content, concentration of micronutrients, and electrical conductivity, each lending to the overall nutritive value. By showing that algal meal can be as effective as both the conventional and organic standard at delivering N to a crop (Andrews, 2013) I have validated assigning a price equivalent to the current market value of conventional and organic fertilizers on a price per unit of nutrient basis (Table 8). Under current market conditions the conventional nutrient equivalent delivered to the grower was \$140 Mg^{-1} and the value to an organic grower was \$1054 Mg^{-1}. Including marketing costs and transportation expenses, an algae facility could expect a sale price of \$800 Mg^{-1} of dry algal meal with an NPK of 7-3-1 (Andrews, 2013). This valuation does not include any potential benefits of the product beyond the macro nutrient values.

Though increasing the assigned market value of 50-80% of the total biomass produced (algal meal) may go a long way toward economic feasibility, there are bigger issues facing the algal fuel industry that need to be addressed. The production of nutrients for algal biodiesel production has been demonstrated to be the major contributor to the total greenhouse gas emissions of the system (Clarens et al., 2010; NAS, 2012). In order to meet the US Department of Energy goal of 136 billion L (17% of total fuel consumption) of biofuel production by 2022, with only 57 billion L from corn ethanol, the current methods of algae oil production would

use an estimated 14-36 million Mg of N and 2-5.5 million Mg of P (NAS, 2012). The largest energy input of algae production, 89% of total energy requirements, is in the drying of the algal cake in preparation for extraction (Sander and Murthy, 2009). Further, evaporative losses during algae growth are the largest quantity of water consumption. The NAS (2012) estimated that the mean US evaporation rate from a raceway pond is 0.9 m^3 m^{-2} yr^{-1}.

Integrated algae-dairy models have been designed in order to overcome nutrient capture and greenhouse gas (GHG) management issues faced by industrial animal agriculture and in so doing, overcome the algae production barrier of nutrient costs (Lincoln et al., 1996; Craggs et al., 2004; Mulbry et al., 2005; Woertz et al., 2009; Wang et al., 2010; Zhang et al., 2013). I have proposed a dairy-anaerobic digestion-algae system (DADA) in which the dairy-housing manure is sent to an anaerobic digester (AD) where a succession of microbes transform the organic matter into a methane-rich biogas, heat, nutrient-rich liquid effluent, and solid compost. The liquid effluent is diluted and used as the nutrient broth for the algae production. The heat is incorporated into drying of the algal biomass. The composted solids are used as bedding in the free stalls or sold into the horticultural markets. The algal biomass is extracted for fatty acid methyl esters (FAME biodiesel). From this process glycerin and "biomass" are also produced. The biomass co-product, algal meal, is used on farm or sold as an organic fertilizer. Logistically, the actual biodiesel refinery should not be co-located on a dairy. Instead the dairy

will produce the total dry biomass and sell it to a biorefinery for extraction, at a price that accounts for the total value of each of the co-products.

The methane biogas can be used to power algae, dairy, and AD operations on farm with any remainder being sold back to the energy grid. The biodiesel is used to operate large farm equipment with the remainder sold as transportation fuel. The glycerin can be used to optimize the AD performance. Amon et al. (2006) demonstrated a three-fold increase in biogas yield when glycerol (at 6% of total biomass) was added to pig manure slurry. It has been shown to be a suitable replacement for corn as a feed energy source (at least up to 15% of the ration) without adverse effects on milk production or milk composition (Donkin et al., 2009). The solid effluent from the AD is ready-made compost and is reused as dairy bedding, as is commonly the practice of dairymen utilizing an AD and solids separator systems (Meyer et al., 2011). The size of the algal raceway can be scaled based on the optimal concentration of P addition and the characteristics of the liquid effluent from the AD. Several studies have shown P removal from dairy waste by algae is nearly 100%, commonly greater than removal of N (70-90%), indicating P is the limiting algal growth factor when using dairy effluent as the nutrient media (Lincoln et al., 1996; Mulbry et al., 2008; Woertz et al., 2009).

In a non-integrated production models the cost of drying the algal biomass for lipid extraction has been one of the cost prohibitive steps in the life cycle analysis. However, the heat and energy produced in the AD has been calculated to be more than enough to cover the energy requirements of drying the algal cake

(Pizarro et al., 2006). If we assume we can dry algal biomass to 20% solids through a screw press or solar drying (Kadam, 2001; Pizarro et al., 2006; Mulbry et al., 2008) and 220kg of dried algal biomass is produced on each hectare, the energy required to dry the biomass is 3900 MJ ha^{-1} d^{-1} to 90% DM. Using the 11 ha treatment area needed for 1000 cows, producing algae 270 days a year the total energy needed is 1.17 x 10^7 MJ yr^{-1} (Pizarro et al., 2006). Assuming each cow produces 5.7 kg of manure VS d^{-1} (Van Horn et al., 1994) and anaerobic digestion yields 350 L of biogas kg^{-1} VS (Amon et al., 2006; Homan et al., 2013), biogas production should be 2000 L cow^{-1} d^{-1}. Methane has an energy content of 0.037 MJ L^{-1}, the biogas is composed of 60% methane. Therefore, the total energy provided by the AD would be 1.63 x 10^7 MJ yr^{-1} (Pizarro et al., 2006). The energy requirement for drying is met as well as the estimated 30% of total heat production that is needed to keep the AD at 35°C. Seventy-five percent of the energy produced by the AD is heat (Wright, 2001). Further biogas is difficult to store, compress, or liquefy, requiring low temperatures (-83°C) and high pressure (5000psi) (Homan et al., 2013). Therefore in order to utilize the energy efficiently it would be very difficult to use the biogas for anything but continuous on site consumption.

The primary difference between this project and others who have evaluated integrated algae-dairy operations is that I use the algae, not just to capture nutrients, but also to extract the biomass for biodiesel, yielding high value co-products. It has been posited that unless algal lipid content is at least 40% of the biomass, the total biomass will yield more energy as a feedstock for AD than from

biodiesel extraction (Sialve et al., 2009). Farm-scale work by the USDA ARS lab in Beltsville, MD has only realized 12% lipid content using algal turf scrubber (ATS) technology to capture nutrients from dairy wastes (Mulbry, 2007). These works are cited as the driving reason that Zhang et al., (2013) evaluated their integrated system utilizing the total algal biomass as AD feedstock for their life cycle analysis and life cycle costing. However, in outdoor bench-scale cultures of algae grown on dairy wastewater, Woertz et al., (2009) was able to reach a maximum lipid productivity of 29% (not at peak biomass production). In the Mulbry system, the primary goal was dairy waste nutrient capture, therefore they allowed an indigenous algal consortia, dominated by *Rhizoclonium hieroglyphicum,* to naturally colonize the ATS in early spring (Mulbry et al., 2008) instead of selecting for species with high lipid productivity. The use of controlled seeding with benthic algae, such as *Amphora*, which have demonstrated 40% lipid content, (Griffiths and Harrison, 2009) is a possible way to increase the total value of the ATS system.

The recent paper by Zhang et al. (2013) reported a complete "cradle to gate" life cycle analysis (LCA) and life cycle costing (LCC) of four different dairy manure management practices calculating the net present value (NPV) of each. A reference land application scenario (REF), an AD with land-application of liquid digestate operation (ADO), an AD with recycling of liquid digestate to an open-pond algae cultivation system (OPS), and an AD with recycling of liquid digestate to an algae turf scrubber system (ATS) were evaluated for net energy output, reductions in net eutrophication potential, and reductions in global warming potential. They found

that all three "improved" systems were environmentally favorable. Further, if robust nutrient credits, valued at \$20 kg^{-1} N in Chesapeake Bay Watershed are available, the algae systems are much more financially attractive considering: initial outlay, annual operating costs, and annual revenue (Table 9).

I hypothesize that when considering environmental benefits (nutrient capture, GHG emissions reduction, and energy displacement), and current policy incentives of integrating algal biodiesel and AD into dairy production, utilizing the fertilizer value of algal meal at \$800 Mg^{-1} (Andrews, 2013) to the producer will show the integrated DADA production facility to be an economically viable manure management system. More field trials using algal meal as a fertilizer are needed before this value is firmly established. Following price validation, a complete life cycle analysis must be performed in order to assess this hypothesis. In order to evaluate the potential viability of DADA, the detailed, supplemental production and accounting data by Zhang et al. (2013), will be used for estimates and comparison.

Materials and Methods

Utilizing the supplemental data from the LCC of Zhang et al. (2013), I estimated the NPV of the DADA system (Table 9). To calculate the capital expenditure, I used the cost of the anaerobic digester from the ADO scenario as well as the cost of the algae cultivation. I subtracted the cost of the pretreatment of algae for the AD and added \$50,000 as an estimated cost for a dryer being built into the system. To estimate the operational cost, I used reported values from the ADO AD

operation and the ATS algae operation. For the total revenues, I excluded the corn productivity from the area that the algal facility occupied and any profit from the sale of bio-electricty as the AD energy generation will likely be completely used on farm for the algae drying process. The value of the digestate is the same as from the ADO. Using Zhang's published ATS algal productivity of 1.53 Mg cow^{-1} yr^{-1}, estimating 25% of the total algal biomass is FAME, using 0.883 g cm^{-3} as the density (www.biofuelsb2b.com), and using a sale price of \$3.00 gal^{-1} (Chisti, 2007), I calculated the profit from the sale of biodiesel for 100 $cows^{-1}$ yr^{-1}. The algal meal value was calculated as 70% of the total biomass at \$800 Mg^{-1}. Additional revenue from glycerin and omega-3 oils were not included in this preliminary analysis but offer additional potential revenue sources. The total capital cost was amortized over 20 years without accounting for a discount rate and subtracted from the annual profit to achieve the NPV (Table 9).

Results and Discussion

Utilizing robust nutrient trading programs, all of the algae systems returned a net profit to the dairyman (Table 9). However, nutrient trading programs are only established in the highly-sensitive Chesapeake Bay Watershed states of MD, PA, VA, and WV. Without the nutrient credits for the OPS and ATS systems, an operator would spend roughly 40 times as much for their annual manure management system as the potential operator of a DADA system. The additional monetary gain

for the energy provided by the algal biomass in the AD system was $9,954 and $12,439 for OPS and ATS. This is less than the calculated economic benefit of the biodiesel product and considerably less when including the $82,600 100 cow^{-1} yr^{-1} generated by the sale of organic fertilizer.

As reported in Zhang et al. (2013), the environmental benefits of each of the systems was far greater than from the reference scenario for both global warming potential and eutrophication potential. These benefits will also apply to the DADA system. Economic benefits of accounting for positive environmental externalities are currently possible through government tax credits, infrastructure grants, and C trading markets. The production tax credit reduces the federal income tax of qualified owners of renewable energy projects, paying $22 MW h^{-1} based on total electrical output for the first 10 years of operation (Goodward and Gonzalez, 2010). The Investment Tax Credit (ITC) allows tax payers to deduct 10-30% (depending on facility type) of their capital investment from their federal income tax when the equipment is placed into service (Goodward and Gonzalez, 2010). California has established a C trading market with current values for short-term trades at $14 Mg^{-1} CO_2e (e=equivalence) reduction (Weisberg, 2013). If the fuel is sold to transportation fuel refiners, biodiesel from the algal production is also eligible for Renewable Identification Numbers (RINs). One caveat is that the RIN (1 RIN=77,000 BTUs) is owned by the fueling station and should be included in the contracted sale price of the fuel. The current RIN price average is $0.52 RIN^{-1} (Weisberg, 2013). Biogas and biodiesel both have lower C intensity than crude oil, qualifying both for

credits in the California markets under the Low Carbon Fuel Standard (Weisberg, 2013). Regardless of the fact that there are many potential policy incentives for the DADA system, most cannot be presented to a loan board because they are too volatile in price and the expiration dates for each program tend to have moving targets (Weisberg, 2013). Additionally the frequency of fraudulent claims and the lack of set metrics for assessing improvement parameters have infused doubt into the market, all but eliminating long-term contracts or anything other than spot market prices (Weisberg, 2013).

The original analysis by Zhang, et al. (2013), was done on a 100-cow dairy that is typical in the Chesapeake watershed. The theory of economies of scale is based on the cost advantages that businesses obtain due to increased size. The cost per unit of output generally decreases with increasing scale as fixed costs are spread out over more units of output, to a point (McConnell and Brue, 2002). Often operational efficiency is also greater with increasing scale, leading to lower variable cost as well (McConnell and Brue, 2002). Goodrich (2005) calculated that for a 100-cow herd the capital expenditure for an AD setup is over $60,000. Increasing the herd size to 200 or 300 would increase the capital and operational costs by 1.4 and 1.6 times respectively (Goodrich, 2005) reducing the total cost per animal. Dairies with small herds do not have a large appetite for high capital investment for manure management, while larger operators may see greater incentive and lower marginal risk. The US EPA Climate Change Division of the Office of Atmospheric Programs mandates that facilities produce aggregate GHG emissions greater than 27,500 Mgs

of CO_2e per year monitor and report emissions, whereas a facility with less than 3,200 mature dairy cows are not currently required to report emissions (US EPA, 2009). For these reasons future work should be modeled with more than 3200 cows in order provide data to those who will face nutrient and GHG management challenges caused by changing regulations.

Conclusions

This rough accounting, relying heavily upon the work of Zhang, et al. (2013), shows that including the fertilizer value of algal meal into the overall economic assessment pushes an environmentally favorable practice toward economic viability. In a recent phone interview with an algae industry professional I was told that these results will have a significant impact on the industry, potentially changing the single-minded lipid focus (Van Der Meulen, 2013). This gives me confidence that an integrated dairy-anaerobic digester-algae production facility has the potential to address a variety of management, environmental, and economic issues faced by large-scale animal agriculture while addressing the issues facing an industry that has been shown to have the greatest potential for bioenergy production efficiency. In addition to diversified production opportunities; the sale of biodiesel, electrical energy, and high value fertilizers, this system would create high-tech jobs on farms, potentially enhancing rural economic development.

Table 9. Net Present Value Comparison with Zhang et al., 2013 for 100 Cow Dairy

Cash Flow	REF	ADO	OPS	ATS	DADA
Capital costs (sum)	**$ 102,000**	**$ 334,349**	**$ 574,987**	**$ 595,832**	**$ 571,455**
Capital Cost After ITC		$ 257,192	$ 442,298	$ 458,332	$ 439,581
Manure Land Application	$ 102,000	$ 95,696			
Anaerobic digestion		$ 238,653	$ 283,430	$ 288,530	$ 238,653
Algae cultivation			$ 291,557	$ 307,302	$ 307,302
Annual operational costs (sum)	**$ 9,200**	**$ 40,741**	**$ 94,835**	**$ 106,391**	**$ 92,793**
Manure on-farm land application	$ 9,200	$ 8,631			
Anaerobic digestion		$ 32,109	$ 43,851	$ 45,507	$ 32,109
Algae cultivation			$ 50,984	$ 60,684	$ 60,684
Annual revenues (sum)	**$ 12,637**	**$ 57,267**	**$ 311,544**	**$ 315,737**	**$ 398,225**
Corn	$ 12,637	$ 5,321			
Bio-electricty		$ 16,827	$ 26,781	$ 29,266	
Digestate		$ 1,844	$ 2,748	$ 3,034	$ 1,844
Biodiesel					$ 34,254
Algal meal					$ 82,600
N credit sale		$ 26,662	$ 271,742	$ 271,742	$ 271,742
P credit sale		$ 61	$ 123	$ 123	$ 123
Carbon credit sale		$ 1,264	$ 1,733	$ 2,374	$ 2,374
Production Tax Credit		$ 5,288	$ 8,417	$ 9,198	$ 5,288
Totals					
Amortization of Capital Cost (20yr)	$ 5,100	$ 16,717	$ 28,749	$ 29,792	$ 28,573
Amortization of Capital Cost ITC (20yr)	$ 510	$ 12,860	$ 22,115	$ 22,917	$ 21,979
Gross Income	**$ 12,637**	**$ 23,992**	**$ 29,529**	**$ 32,300**	**$ 118,698**
Net Present Value with no alt markets	**$ (1,663)**	**$ (33,466)**	**$ (94,055)**	**$ (103,883)**	**$ (2,668)**
Net Present Value only using ITC	**$ 2,927**	**$ (29,609)**	**$ (87,421)**	**$ (97,008)**	**$ 3,926**
Net Present Value using all listed income	**$ 2,927**	**$ 3,667**	**$ 194,594**	**$ 186,429**	**$ 283,453**

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SUMMARY

Algae have shown great potential as a source for renewable fuels. The 2012 report by the National Academy of Sciences recommends further research into the use of wastewater and fertilizer to meet algal production sustainability. My objective was to quantify the fertilizer value of the co-product, algal meal, for the economic evaluation of an integrated dairy-anaerobic digester-algae production facility. The algal meal performed similarly to the feather meal in the laboratory incubation mineralization tests. In the field trial the algal meal treatments, 101 kg N ha^{-1}, 146 kg N ha^{-1}, and 190 kg N ha^{-1} produced the same corn yield and ear N concentration as both feather meal treatments and urea treatments fertilized at the highest rate. This gave me confidence to quantify the fertilizer value of this algal meal relative to feather meal on a price per unit of nutrient basis. This 7-3-1 NPK algal meal is worth \$800 Mg^{-1} to the producer at current market prices. This price only includes the value of the macronutrients N, P, and K. Each algal biomass will vary depending on processing, extraction, algal strain, and nutrient media. After further testing to fully quantify the value of the micronutrients, phytohormones, C sequestration, and required application rates, a higher market value is expected.

Integrated algae-dairy production systems have been posited as a potential solution for algal production barriers as well as a way to capture environmentally problematic nutrients excreted by animals in concentrated animal feeding

operations. In a preliminary accounting, the inclusion of the fertilizer value of algal meal for an integrated dairy-anaerobic digester-algae production facility takes it from a system that shows great environmental impact reduction that may work with the right policy incentives, to a system that is profitable even with few government incentives. Future work should include a life cycle analysis and life cycle costing of the proposed farm facility to confirm that an integrated dairy-anaerobic digester-algae system can be an economically viable, manure-management, farm-energy production system.

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APPENDIX

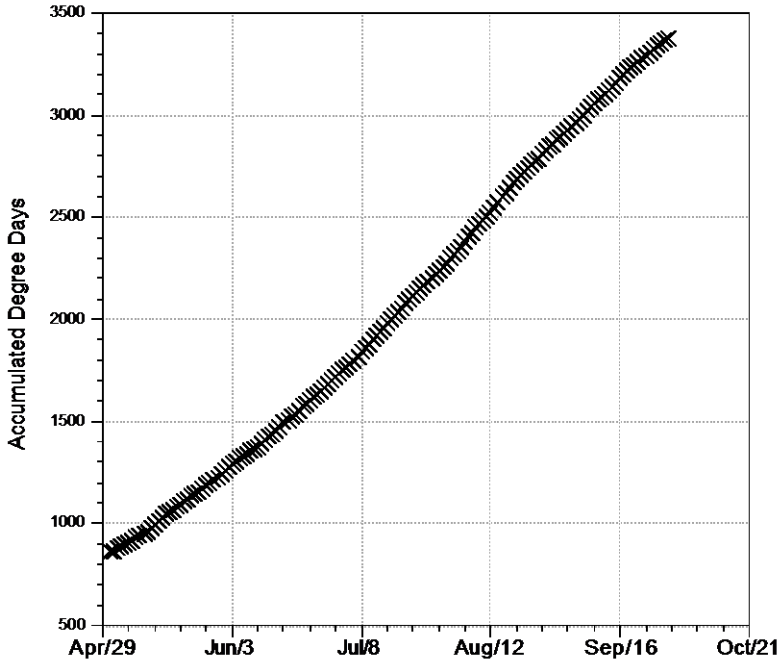

Fig. A1. Accumulated Degree Days through 2012 Growing Season
OSU College of Agricultural Sciences Hyslop Weather Station.

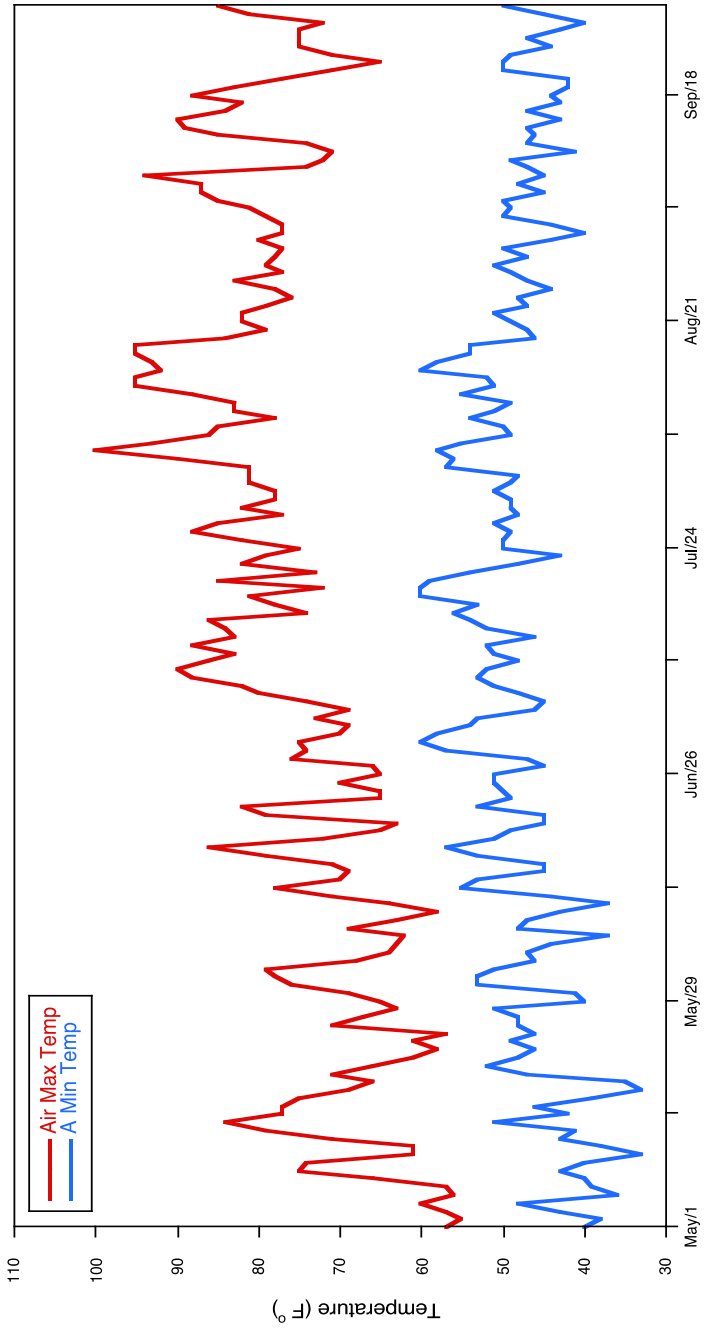

Fig. A2. Field Trial Atmospheric Temperature
Reported as daily maximums and daily minimums. OSU College of Agricultural Sciences Hyslop Weather Station.