ASSESSING THE IMPACT OF BIOFUMIGATION AND ANAEROBIC SOIL DISINFESTATION ON SOIL BIOLOGY, NITROGEN CYCLING, CROP ESTABLISHMENT AND YIELD IN VEGETABLE CROPPING SYSTEMS

By

Aaron J. Yoder

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ABSTRACT

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Alternative fumigation practices in horticultural production systems impact the soil in complex ways. Two practices, biofumigation (BF) and anaerobic soil disinfestation (ASD) have demonstrated success in controlling soil-borne pests, although results are often inconsistent and can have negative effects in cropping systems. Our research objectives were to: 1) investigate delayed seeding of crops as a method to reduce stand inhibition following BF, 2) monitor the impacts of BF and ASD on nitrogen availability, soil temperatures and microbial activity, and 3) evaluate the impact of BF and ASD on yields of warm season vegetable crops in southern Michigan. In one experiment, delayed seeding of muskmelon 10-15 days resulted in satisfactory emergence. Yields of melon decreased as planting date was delayed, highlighting the importance of early seeding of certain vegetable crops in Michigan. In another experiment, the 2nd and 3rd objectives were addressed. Plastic mulch treatments had substantially higher NO_3^- and NH_4^+ during and after ASD. High soil temperatures were also observed under plastic mulches in 2012 and likely have caused lower total marketable yields in tomato than bare ground treatments. This research highlights the importance of understanding how both alternative and commonly utilized cropping practices can influence environmental conditions in vegetable production, while identifying areas that must be addressed to effectively implement BF and ASD in the future for vegetable producers.

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INTRODUCTION

The need to develop alternative, environmentally benign pest management practices is one of the major challenges facing agriculture today. Many high-value horticultural cropping systems have relied on broad-spectrum fumigants as a means of mitigating a large range of biotic stresses including weeds, pathogens and insects. These high-value crops are managed intensively, often with minimal crop diversity exhibited in crop rotations. The lack of crop diversity can have major impacts on soil and subsequently plant health, and ultimately exacerbates the need for fumigation. Methyl bromide is a fumigant that has been in the process of phase-out since the 1992 Montreal Protocol after being listed as a potential ozone-depleting substance (Ware et al., 2003). Although a few exemptions currently exist for certain commodities, the availability of methyl bromide for future use is uncertain at best. There have been significant efforts to identify less persistent and more environmentally benign fumigants to replace methyl bromide; however, the broad spectrum of organisms controlled by such chemicals is at least partially responsible for their efficacy as fumigants (Yates et al., 2003). This represents a conundrum for many agricultural researchers and producers in that alternatives must be persistent and reactive enough to control targeted pests yet environmentally benign enough to ensure continued availability to growers.

The increase in demand and acreage of organically produced crops has been substantial during recent years. Organically managed systems require alternative strategies to mitigate pest pressures, as many of the synthetic chemicals available to conventional growers are not allowable under the National Organic Program guidelines

(USDA, 2014). Organic growers must utilize additional cultural, biological and physical means to sustain profitable yields.

One method with the potential to address soil-borne pest concerns in both conventional and organically managed systems is ‘biofumigation’ (BF) (Kirkegaard, 2009). Biofumigation refers to the practice of growing or utilizing biomass from specific plant species in the Brassicaceae family as a means of reducing pest (pathogens, weeds, arthropods) pressures. One of the primary mechanisms believed to be responsible for this biological suppression is through the glucosinolate-myrosinase mediated pathway. Glucosinolates (GSLs) are sulfur-containing molecules produced almost exclusively by plants in the brassica family. Upon tissue maceration, the enzyme myrosinase reacts with the normally benign glucosinolates to produce reactive isothiocyanates (ITCs) among other products (Brown and Morra, 1997; Bones et al., 1996). These ITCs and other degradation products are widely believed to contribute to observed disease suppression following BF (Lazzeri et al., 2000).

The use of the term ‘biofumigation’ has evolved in recent years to include general suppression of pest/disease organisms following incorporation of non-specific organic materials into the soil. To avoid confusion I will use the term to describe practices that utilize the GSL containing brassica family plant residues from: non-harvested green manures, partially harvested cash-crops, dried plant material, or seed meals (bi-products of oilseed extraction processing) (Kirkegaard, 2009).

Brassica cover crops can be cultivated in various cropping systems throughout the world as evidenced by their widespread use as oil crops (Leff et al., 2004; Shahidi, 1990) including the upper Midwestern U.S. (Snapp et al., 2005). Because their primary growth occurs during the cool season and they have relatively short life cycles (~45 days), brassica cover crops have the potential to fit into the short growing season of the upper Midwest; ideally, they might precede a summer vegetable crop such as tomato (*Lycopersicon lycopersicum*), muskmelon (*Cucumis melo*), or cucumber (*Cucumis sativus*).

Although many growers have embraced brassica cover crops as an additional tool for nutrient recovery, soil improvement, and disease management, some research has shown that crop establishment can be negatively affected following the incorporation of brassica residues in the spring (Ackroyd et al., 2011; Rice et al., 2007; Haramoto et al., 2005). Methods to reduce inhibition of crop emergence have not been researched extensively but must be addressed if these cover crops are to be used successfully in a cropping system. One method to address this issue is by establishing safe plant-back dates following the incorporation of brassica cover crop residues.

Although biofumigation has shown the potential to reduce disease in some cases, results are often variable and are less effective than traditional fumigation methods (Kirkegaard, 2009). Numerous factors influence the efficacy of biofumigation including cover crop biomass accumulation, GSL concentrations (Mattner et al., 2008), GSL type (as affected by cultivar selection) (Kirkegaard and Sarwar, 1998), soil type, soil moisture, soil temperature (Gimsing and Kirkegaard, 2009) and incorporation method (Morra and Kirkegaard, 2002). Many of the biologically active ITCs that are generated from BF can be lost from the soil profile after residues have been incorporated. Modifying the technique to reduce ITC losses has been cited as one of the possible ways to increase the effectiveness of BF (Matthiessen et al., 2006). A light rolling of the soil, irrigating and covering with impermeable plastic films immediately following residue incorporation are a few ways that losses might be minimized.

Anaerobic soil disinfestation (ASD) is a practice that has been recently developed and utilized in various production systems around the world including Japan (Momma, 2008), the Netherlands (Blok et al., 2000), Spain (Nunez-Zofio et al., 2011) and the U.S. (Shennan et al., 2009). This technique requires the incorporation of a readily decomposable carbon source such as rice bran, grass clippings, molasses, or ethanol. Following incorporation, the soils are covered with an impermeable film and irrigated to saturation. The fresh carbon material stimulates microbial decomposition and oxygen is quickly depleted as the impermeable film restricts re-supply of O_2 from the atmosphere. The anaerobic environment must be maintained for specified lengths of time, typically 4-6 weeks (Lamers et al., 2010) to suppress certain soil-borne diseases through changes in the microbial community, deprivation of O_2 and the development of organic acids and volatile compounds (Momma, 2008).

Because of the inherent similarities between BF and ASD, the combination of these practices seems promising and in theory stands to provide improved and more consistent pest management and crop yields. Additionally, improvements in agricultural plastic have led to the development of virtually impermeable film mulches, a technology that can be easily transferred to vegetable cropping systems and can facilitate ASD. Using brassica cover crop residues as the carbon source for ASD might allow greater suppression of soil-borne pests due to their unique biochemistry. In order to properly facilitate ASD, however, enough biomass must be incorporated into the soil. While brassica cover crops have shown the potential to accumulate significant quantities of biomass under certain conditions, germination and growth can be variable and their potential must be evaluated under various edaphic and climatic environments.

In Michigan, the window for growing summer crops is relatively short compared with states in the southern U.S. Here, low temperatures and proportionally higher precipitation during winter and spring months can delay planting of many crops into the later months of spring. Fortunately for many vegetable growers, sandy-textured soils are abundant along the southwest and central part of the state. These course-textured soils facilitate drainage of precipitation in the spring and are more easily tilled and planted than heavier clay (or fine-textured) soils.

We implemented two field experiments in 2012 and 2013 to address several objectives. The first experiment was designed to 1) evaluate delayed crop seeding as a tool for improving crop emergence following brassica cover crop incorporation, and 2) assess the impacts of delayed seeding on crop yields. The second experiment was designed to: 1) evaluate the potential of a spring-sown brassica cover crop as a carbon source for ASD under plastic mulching regimes, 2) monitor the impacts of a spring-sown brassica cover crop and plastic mulches on nitrogen availability, soil temperatures and microbial biomass following ASD treatments, and 3) evaluate the impact of biofumigation and plastic mulching on yields of warm season vegetable crops.

LITERATURE CITED

LITERATURE CITED

Ackroyd, V.J., Ngouajio, M. 2011. Brassicaceae Cover Crops Affect Seed Germination and Seedling Establishment in Cucurbit Crops. *HortTechnology*, 21: 525-532.

Blok, W.J., Lamers, J.G., Termorshuizen, A.J., Bollen, G.J. 2000. Control of Soilborne Plant Pathogens by Incorporating Fresh Organic Amendments Followed by Tarping. *Phytopathology*, 90: 253-259.

Bones, A.M., Rossiter, J.T. 1996. The myrosinase-glucosinolate system, its organization and biochemistry. *Physiologia plantarum*, 97: 194-208.

Brown P.D., Morra, M. 1997. Control of soil-borne plant pests using glucosinolate-containing plants. *Advances in Agronomy*, 61: 167-231.

Gimsing, A.L., Kirkegaard, J.A. 2009. Glucosinolates and biofumigation: fate of glucosinolates and their hydrolysis products in soil. *Phytochemistry Review*, 8: 299-310.

Haramoto, E., Gallant, E. 2005. Brassica cover cropping: I. Effects on weed and crop establishment. *Weed Science*, 53: 695-701.

Kirkegaard, J.; D. Walters, ed. 2009. Biofumigation for plant disease control- from the fundamentals to the farming system. Chapter 9, *Disease Control in Crops: Biological and Environmentally Friendly Approaches*. Blackwell Publishing Ltd. Pp. 172-195.

Lamers, J.G., Runia, W.T., Molendijk, I.P.G., Bleeker, P.O. 2010. Perspectives of Anaerobic Soil Disinfestation. *Acta Hort*., 883: 277-284.

Lazerri, L., Mancini, L. 2000. The Glucosinolate-Myrosinase System: A Natural and Practical Tool for Biofumigation. *International Symposium of Chemical Soil Substrate-Disinfestation.* 89-95.

Leff, B., Ramankutty, N., Foley, J.A. 2004. Geographic distribution of major crops across the world. Global Biogeochemical Cycles, 18: doi:10.1029/2003GB002108.

Matthiessen, J.N., Kirkegaard, J.A. 2006. Biofumigation and Enhanced Biodegradation: Opportunity and Challenge in Soilborne Pest and Disease Management. *Critical Reviews in Plant Sciences*, 25: 235-265.

Mattner, S.W., Porter, I.J., Gounder, R.K., Shanks, A.L., Wren, D.J., Allen, D. 2008. Factors that impact on the ability of biofumigants to suppress fungal pathogens and weeds of strawberry. *Crop Protection*, 27: 1165-1173.

Momma, N. 2008. Biological soil disinfestation (BSD) of soilborne pathogens and its possible mechanisms. *Japan Agricultural Research Quarterly*, 42: 7-12.

Morra, M.J., Kirkegaard, J.A. 2002. Isothiocyanate release from soil-incorporated *brassica* tissues. Soil biology and biochemistry, 34: 1683-1690.

Nunez-Zofio, M., Larregla, S., Garbisu, C. 2011. Application of organic amendments followed by soil plastic mulching reduces the incidence of Phytophthora capsici in pepper crops under temperate climate. *Crop Protection*, 30: 1563-1572.

Rice, A.R., Johnson-Maynard, J.L., Thill, D.C., Morra, M.J. 2007. Vegetable crop emergence and weed control following amendment with different Brassicaceae seed meals. *Renewable Agriculture and Food Systems*, 22: 204-212.

Shahidi, F. 1990. Rapessed and Canola: Global Production and Distribution. In Canola and Rapeseed. Springer U.S., 3-13.

Shennan, C., Muramoto, J., Koike, S.T., Daugovish, O. 2009. Optimizing anaerobic soil disinfestation for non-fumigated strawberry production in California. *Proceedings of the Annual International Research Conference on Methyl Bromide Alternatives and Emissions Reductions*. 101.

Snapp, S.S., Swinton, S.M., Labarta, R., Mutch, D., Black, R., Leep, R., Nyiraneza, J., O'Neil, K. 2005. Evaluating Cover Crops for Benefits, Costs, and Performance within Cropping System Niches. *Agronomy Journal*. 97: 322-332.

Ware, G. W., et al. 2003. "Environmental fate of methyl bromide as a soil fumigant." *Reviews of Environmental Contamination and Toxicology: Continuation of Residue Reviews*: 45-122.

Yates, S., Gan, J., Papiernik, S. 2003. Environmental Fate of Methyl Bromide as a Soil Fumigant. *Reviews of environmental contamination and toxicology*, 177: 45-122.

CHAPTER I:

DELAYED SEEDING OF MUSKMELON FOLLOWING BIOFUMIGATION IMPACTS CROP EMERGENCE, CROP QUALITY AND YIELDS

Abstract

Brassica cover crops are commonly utilized in cropping systems for their ability to scavenge residual nutrients from the soil, minimize soil erosion and reduce the incidence of soil-borne pests. Glucosinolate molecules produced by these plants are hydrolyzed to biologically reactive molecules including the isothiocyanates (ITCs) that are believed to be a primary mechanism involved in the observed suppression of pathogens and weeds. One issue that has been encountered in using these cover crops has been the reduction in stand establishment of cash crops seeded following the incorporation of cover crop residues. Because this reduction has been observed to decrease as a function of time, a field experiment was developed to evaluate delayed seeding as a strategy for minimizing stand reduction at the Southwest Michigan Research and Extension Center in Benton Harbor, MI. A previously identified susceptible muskmelon variety (*Cucurbita melo* 'Athena') was seeded at six, 5-day intervals after incorporation (DAIs) of five cover crop treatments including: Oriental mustard (*Brassica juncea* 'Forge' and *B. juncea* 'Pacific Gold'), yellow mustard (*Sinapis alba* 'Ida Gold), oilseed radish (*Raphanus sativus* 'Defender'), oats (*Avena sativa* 'Excel') and a no cover crop control. Emergence increased over time and approached control levels at 10 days for oats and 'Forge', and 15 days for 'Defender', 'Pacific Gold' and 'Ida Gold', however marketable melon yields decreased substantially after 15 DAI while un-marketable yields increased. While delayed seeding was shown to improve crop establishment, minimizing this waiting period is critical for growers to achieve higher early and cumulative marketable yields for late-maturing crops like muskmelon, particularly under the short growing season limitations imposed by the Michigan climate.

Introduction

Brassica cover crops have been utilized in many cropping systems around the world to provide numerous benefits through reduced soil erosion (De Baets et al., 2011), improved nitrogen retention (Stivers-Young, 1998), reduced nitrate leaching (Justes et al., 1999), improved soil structural qualities (Chan et al., 1996) and disease suppression (Brown et al., 1997). Biofumigation utilizes these cover crops (Kirkegaard et al., 1993) as an alternative to conventional fumigation techniques. The proliferation of this practice has increased the utilization of brassica cover crops substantially as restrictions on fumigants have become more severe in recent years and growers have demanded novel solutions to ameliorating soil-borne pest problems.

Biofumigation harnesses the unique biochemistry of many brassica (Brassicaceae) species to control certain soil-borne diseases. The primary mechanism believed to be responsible for this suppression is the glucosinolate-myrosinase pathway. Glucosinolates (GSLs) are non-reactive molecules that hydrolyze to highly reactive isothiocyanates (ITCs) in the presence of water and the enzyme myrosinase. Myrosinase is normally separated from the GSLs but comes into contact with them when plant tissues are macerated, whether mechanically or by herbivory (Brown et al., 1997). ITCs have been implicated in the suppression of numerous soil-borne pathogens including *Rhizoctonia solani* (Hansen et al., 2013), *Streptomyces scabies* (Larkin et al., 2011), *Phytophthora capsici* (Nunez-Zofio et al., 2011) and *Meloidogyne incognita* (Monfort et al., 2007). Biofumigation harnesses this phenomenon through the maceration of brassica residues (typically with a flail mower), incorporation into the soil and irrigation to facilitate the reaction and help retain the volatile ITCs in the soil profile.

Some observations have shown that crop stands can be inhibited following the incorporation of brassica cover crop residues. Two mechanisms believed to be responsible for the observed inhibition are allelopathic interactions between cover crops and crop seeds, and short-term proliferation of seed rotting pathogens facilitated by cover crop tissues. For example, muskmelon (*Cucumis melo* Group reticulatus) and cucumber (*Cucumis sativus*) emergence was greatly reduced when exposed to extracts of oilseed radish (*Raphanus sativus* var. oleiferus) in laboratory bioassays while *R. sativus, Brassica juncea* and *Sinapis alba* cover crops reduced emergence of muskmelon by 100%, 89.1% and 59.5% respectively in the field (Ackroyd et al., 2011). In another field experiment, brassica cover crops reduced emergence of a variety of crop and weed species on average by 23 to 34% and emergence was delayed by approximately 2 days, although non-brassica cover crops had comparable effects on emergence (19 to 39% reduction) and no significant differences were observed among high and low ITC brassicas (Haramoto et al., 2005). These results suggest that generalized, non-ITC related suppression by cover crop residues might be responsible for the observed inhibition. In another study, incorporation of *Brassica napus* seed meals stimulated *Pythium* spp., reducing emergence of wheat (*Triticum aestivum)* in orchard soils compared with fumigated control treatments supporting the mechanism of biologically mediated crop seed suppression (Hoagland et al., 2008). Although *S. alba* and *B. napus* amended soils dramatically inhibited lettuce emergence, no inhibition was observed in plantings 5 weeks after incorporation of seed meals, indicating that inhibitory effects can decrease with time (Rice et al., 2006).

While many studies have observed crop seed inhibition following brassica cover crop incorporation, some have also reported no effects or even germination enhancement. Fall seeded brassica cover crops improved stand establishment in spring-seeded onion while also improving crop yields (Wang et al., 2008). Fall planting of brassica cover crops has been shown to yield substantial biomass, although with biofumigation, fresh biomass, incorporated immediately prior to crop planting is often recommended due to the rapid loss of ITCs from the soil profile (Brown et al., 1997).

For biofumigation or brassica cover cropping in general to be adopted as a viable alternative to conventional fumigation practices, the issue of crop emergence inhibition must be addressed. One proposed method is through the determination of optimal plant back dates using a time series analysis for susceptible crops. The primary objective of this research was to determine the effects of delayed seeding of a susceptible crop (muskmelon; *Cucumis melo* Group Reticulatus var. 'Athena') on crop emergence to identify optimal plant back dates following brassica cover crop incorporation. To better assess the suitability of delayed seeding as a management tactic for growers, fruit quality and yield data were gathered to determine potential impacts on yield caused by delayed crop seeding.

Materials and Methods

In the spring of 2012, cover crop treatments were seeded using a John Deere 450 drill at the Southwest Michigan Research and Extension Center (SWMREC) in Benton Harbor, MI on an Oakville fine sand. Cover crop plot dimensions were 60'x 30' and were seeded at 18 cm between row spacing. Cover crop treatments were replicated 3 times in a randomized complete block design and included: *Brassica juncea* 'Pacific Gold' (PG), *Brassica juncea* 'Forge'(F), *Sinapis alba* 'Ida Gold'(IG), *Raphanus sativus* 'Defender'(OSR), *Avena sativa* 'Excel' (OAT) and a no cover control (C). The drill was calibrated to seed the cover crops at rates of approximately 8,8,8,11 and 134 kg/ha for PG, IG, F, OSR and OAT respectively. At flowering stage, cover crop shoot and root biomass was collected from four 25 by 50 cm quadrats in each plot, dried at 90^{o}C to a constant dry weight and then samples were weighed. Weed biomass was also collected to account for any additional biomass that might be incorporated under the various cover crop treatments (Table 1.2).

Cover crops were macerated using a flail mower (Perfect BK2-150) and incorporated into the soil using a roto-vator (Howard SM80). Immediately following cover crop incorporation, virtually impermeable film black plastic mulch was applied to eight rows running perpendicular to cover crop plots, the two outermost serving as guard rows. To minimize the impact of residue contamination during bed shaping and mulch laying a buffer zone of several feet was maintained between adjacent cover crop treatments. Following mulch application, muskmelon was seeded at 5-day intervals from 0 (day of cover crop incorporation) to 25 days. Two untreated muskmelon seeds were placed in holes set on 1' centers within each row yielding a total of 52 seeds per plot.

Crop emergence was evaluated 20 days after seeding and % emergence was calculated as the number of emerged plants divided by 52 (total seeds sown and counted per plot). Following the collection of emergence data, muskmelon plots were thinned to a minimum spacing of 1 plant per 3 feet (standard muskmelon spacing) within each row for a maximum of ten plants/plot. Muskmelon yields were collected as crops matured on weekly intervals. Fruits were graded and counted as marketable (M) or unmarketable (UM) using USDA grading standards (USDA, 2008) then counted and weighed in each plot. Statistical analysis (SAS 9.3; Cary, NC) was completed using two-way analysis of variance (ANOVA) and significance of mean differences among cover crop (main plot factor) and planting date (sub-plot factor) were evaluated using Fischer's least significant difference ($p < 0.05$) for response variables including emergence, early yields (M and UM) and total yields (M and UM). Temperature and rainfall data were gathered and summarized from an on site weather station operated through the MSU Enviro-weather network (enviroweather.msu.edu).

Results

Cover crop biomass

Dry weight biomasses accumulated by cover crops immediately prior to their incorporation are listed in Table 1.2. Though brassica cover crops have been shown to accumulate significant quantities of biomass in Michigan, these experimental plots biomass was substantially lower than previously observed. Biomass from *R. sativus, B. juncea* cultivars and *S. alba* were substantially lower than the accumulations observed in other similar studies where dry weights reached quantities of 6262, 8234, and 7092 kg/ha on muck soils (Wang et al., 2008) and 6086, 3641 and 3487 kg/ha on mineral soils (Ackroyd et al., 2011) as compared with biomass levels from this study of 1983, 1377 and 1495 kg/ha respectively. Compared with brassica cover crops, oats accumulated significantly more biomass (3269 kg/ha).

Melon emergence

There were significant interactions among cover crop and DAI treatments (Table 1.5) Within the day 0 treatment (seeded immediately following cover crop incorporation), there were no significant differences among cover crop treatments, although mean control plot emergence was substantially higher than all cover crop treatments (29% emergence vs. 2-11%) (Table 1.4). High variability among experimental blocks likely caused this lack of significance among cover crops within D0 treatments. Additionally, muskmelon seeds at later DAI treatments were exposed to higher temperatures after planting, further confounding treatment effects on emergence. Significant differences ($p < 0.05$) in emergence occurred among cover crops at 5 and 10 DAI. . These differences suggest that growers might avoid seeding muskmelon immediately following soil tillage in general, particularly following incorporation of cover crop residues. At DAI 5, however, control plots had significantly higher emergence than all the cover crop treatments, while among cover crops, numerically the *Brassica* and *Sinapis* (mustards) cover crops had the lowest, although non-significant. At DAI 10, Control plots and *B. juncea* 'Forge' and non-GSL containing oats had the highest emergence followed by the two other mustard plots and *R. sativus* 'Defender' plots.

The lack of significant differences among cover crop treatments after 10 DAI indicates that delayed seeding following cover crop incorporation can lead to improved emergence in muskmelon. The differences in melon emergence response among cover crop treatments at 5 and 10 DAI indicate that individual cover crop species and even varieties within species (as in the case of *B. juncea*) can differentially impact the duration of emergence inhibition. The lack of significant differences in emergence between non-GSL containing OAT treatments and the GSL containing brassica cover crops indicates that inhibition was not likely crop or GSL specific. These results indicate that cover crop residues (including brassicas and oats), even at low biomass can inhibit emergence of cash crops, though this suppression can be alleviated by delayed seeding. While relatively short delays improved emergence in this study, similar evaluations should be conducted following more substantial accumulations of cover crop biomass to improve recommendations under higher-biomass scenarios.

<u>Cumulative melon yields and fruit quality</u>

While cover crop effects (and interactions with DAI treatments) were not significant, for cumulative yields (M and UM), fruit numbers (M and UM) and the harvest index (marketable yield divided by the total yield), the DAI treatments were significant ($p<0.05$). One particular challenge in evaluating yield data from 0 DAI is that notably low emergence led to fewer plants in each plot to harvest yield from after thinning (Fig. 1.4). While individual plant yields were observed to be highest in DAI 0, this is likely due to decreased competition for space presented by lower plant densities, and is not necessarily a reflection of DAI treatment effects on yields. With this in mind, some important trends can be observed from these tables and graphs.

As the planting date was moved back, marketable yields decreased respectively from a mean of 25,728 kg/ha on DAI 5 to 6,210 kg/ha on DAI 25 (Table 1.6). Conversely, as DAI increased, culled (UM) fruit weight increased as well, from 15,864 kg/ha on DAI 5 to 21,134 kg/ha on DAI 25. The proportion of marketable fruit harvested from each seeding date is reflected in the harvest index, which gives the proportion of the total yield that is marketable. As DAI increased, the index decreased from a high of 62% marketable yield at 5 DAI to 24% on DAI 10. Likewise, average marketable fruit size decreased from 2.33 kg/fruit on DAI 5 to 1.68 kg/fruit on DAI 25. Melons were graded based on USDA standards for cantaloupe (AMS, 2008) where marketable fruit included both U.S. No. 1 and No. 2 grades. Much of the unmarketable fruits collected had either incomplete netting or non-uniform ripening, conditions observed to be more prevalent in later DAI plantings. This reduction in muskmelon yields at later planting dates has been observed in other field studies although the mechanisms responsible for this are poorly understood and could include the influence of day length and temperature on plant reproductive development (Baker et al., 2001). Comparison of growing degree-days among DAI treatments (Table 1.3) shows that as DAI intervals increased, growing degree days tended to decline and might help explain the concurrent observed decline in marketable crop yields.

Discussion

Although brassica cover crops have demonstrated utility in numerous growing systems, crop stand inhibition is an important consideration for growers when deciding to utilize these cover crops. Our results indicate that observing safe plant back dates could prove successful following brassica cover crop incorporation for susceptible crops such as *C. melo*, however the length of time delay required for successful crop establishment among cover crop species and varieties varies. *B. juncea* 'Forge' and oat plots required less of a delay to achieve adequate emergence than the rest of the brassicas. This demonstrates that not all cultivars within a given *Brassica* species will have the same negative impact on crop emergence and might influence their selection for use in cropping systems where shorter crop seeding delays are needed. While differences in GSL profiles are known to occur among Brassicaceae species and cultivars, it is possible that other, non-GSL related mechanisms were responsible for the differential impacts on emergence observed in our study; this notion is supported by the observed inhibition caused by the non-GSL containing oat cover crops. Changes in soil structure, fungal communities, or other allelopathic mechanisms might explain this generalized suppression following all of the cover crops in our study.

Additional strategies that might be used to reduce inhibition could be to utilize fall seeded brassicas, where living plant residues are killed by winter temperatures and decompose over the course of several months prior to planting. Wang, et al. (2008) had success using this technique in onion cropping systems where onion stand establishment was actually enhanced by fall-incorporated cover crops.

Another important consideration to be made should be the selection of the cash crop grown following the cover crops. Long-season summer crops like muskmelon can be established successfully following the growth of spring-sown brassicas, however in adhering to delayed seeding dates, precipitous declines can occur, as observed in our study after 5 and 10 days after incorporation. Selecting cash crops and varieties with short maturation or with less susceptibility to establishment inhibition could reduce the impact of delayed seeding on crop yields in narrow production windows.

Although cover crop biomass was lower than optimal in this study, the observed inhibition among cover crop treatments demonstrates that even under low biomass conditions inhibition can occur and caution should be used following their incorporation. Based on the results of our study, delaying crop seeding at least 10 to 15 days after cover crop incorporation is advisable following brassica and oat cover crops. Determination of the impact of varying levels of cover crop biomass on crop inhibition over time would be useful in as a decision making tool for growers; best planting dates might be estimated from biomass accumulation to maximize emergence and yields.

APPENDIX

Table 1.1. Cover crop dry weight biomass at incorporation

Cover Crop	Variety	Seeding rate (kg/ha)	*Mean dry weight biomass (kg/ha)
Oilseed radish	Defender	11	1983 **B**
Oriental mustard	Forge	8	1377 **B**
Oriental mustard	Pacific Gold	8	1249 **B**
Yellow mustard	Ida Gold	8	1495 **B**
Oats	Excel	134	3269 **A**
$LSD_{0.05}$			668.5
*Biomass mean values with different letters are significantly different (α=0.05) based on Fishcher's Least Significant difference.			

Table 1.2 Weed tissue dry weight biomass at incorporation

Cover Crop	Variety	*Mean dry weight biomass (kg/ha)
Control	-	425
Oilseed radish	Defender	132
Oriental mustard	Forge	215
Oriental mustard	Pacific Gold	237
Yellow mustard	Ida Gold	263
Oats	Excel	113
$LSD_{0.05}$		NS
*Values with different letters are significantly different at the α=0.05 level. NS indicates no significant difference from ANOVA (α=0.05)		

Table 1.3. Establishment dates, harvest dates, early temperatures and growing degree day (GDD) comparisons among days after incorporation (DAI) treatments

DAI Treatment	Crop planting date	Average 20 D temp. following planting	1st Harvest Date	GDD (BE, Base 50°F) from PD to 1st harvest
0	5/31/12	66.81	8/14/12	1731
5	6/5/12	71.11	8/14/12	1682
10	6/10/12	73.28	8/22/12	1711
15	6/15/12	76.16	8/22/12	1617
20	6/20/12	76.77	8/22/12	1473
25	6/25/12	77.63	8/29/12	1533

Table 1.4. Mean % emergence of *C. melo* following incorporation of cover crop residues seeded at various days after incorporation

		Oilseed radish	Oriental mustard		Yellow mustard	Oats	
DAI	**Control**	**Defender**	**Forge**	**Pacific Gold**	**Ida Gold**	**Excel**	*Pr>F*
0	29.4 **D**	3.8 **D**	2.5 **C**	3.9 **C**	6.4 **C**	10.9 **C**	**ns**
5	81.4 **BC/a**	67.9 **B/b**	47.5 **B/b**	43.6 **B/b**	59.0 **B/b**	60.9 **B/b**	**0.0164**
10	70.5 **CD/a**	41.0 **C/c**	71.8 **B/a**	47.4 **B/bc**	53.8 **B/bc**	70.5 **B/ab**	**0.0002**
15	90.4 **AB**	89.8 **A**	90.4 **A**	89.8 **A**	88.5 **A**	93.0 **A**	**ns**
20	99.4 **A**	98.7 **A**	99.4 **A**	91.0 **A**	100.0 **A**	94.2 **A**	**ns**
25	99.4 **A**	94.9 **A**	91.0 **A**	89.1 **A**	99.4 **A**	99.4 **A**	**ns**
Pr>F	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	

* Effect slices were used to determine significance of differences within treatment means across all levels of the other factor. Means were separated using Fisher's LSD, where means followed by the same letter (uppercase for comparing DAI within cover crop treatment rows, and lowercase for comparing cover crop within DAI treatment columns) are not considered significantly different (a=0.05)

Table 1.5. Significance of main effect treatments and interaction terms for % *C. melo* emergence

ANOVA table	
Effect	***Pr > F***
Cover Crop	**0.0353**
DAI	**<0.0001**
Cover Crop*DAI	**0.0421**

Table 1.6. Cumulative yields of *C.melo* following incorporation of cover crop residue

	Yield (kg/ha)		Yield (# fruit/ha)		**Harvest index
DAI	Marketable	Cull	Marketable	Cull	(mkt kg/total kg)
0	11,183 **CD**	8,120 **C**	4,651 **BC**	5,017 **C**	0.47 **C**
5	25,728 **A**	15,864 **B**	11,063 **A**	9,369 **B**	0.62 **A**
10	23,904 **A**	15,471 **B**	11,229 **A**	9,435 **B**	0.60 **AB**
15	19,552 **AB**	17,769 **AB**	9,070 **A**	10,963 **B**	0.51 **BC**
20	13,878 **BC**	17,565 **AB**	6,279 **B**	10,332 **B**	0.44 **C**
25	6,210 **D**	21,134 **A**	3,688 **C**	16,644 **A**	0.24 **D**
p > |t|	**0.0003**	**0.0057**	**0.0002**	**0.0012**	**<0.0001**

* Values followed by different letters are significantly different (α=0.05) based on Fischer's least signifcant difference.

** Harvest index was calculated by dividing the marketable yields (in kg) by the total yields

Table 1.7. Significance of main effect treatments and interaction terms on cumulative marketable and culled muskmelon yields and harvest index

p-values					
	Yield (kg/ha)		Yield (# fruit/ha)		
Effect	Marketable	Cull	Marketable	Cull	Hvst. Index
Cover Crop	0.1969	0.1264	0.2775	0.1658	0.3708
DAI	**0.0003**	**0.0057**	**0.0002**	**0.0012**	**<0.0001**
Cover Crop*DAI	0.4293	0.8565	0.3393	0.6327	0.7732

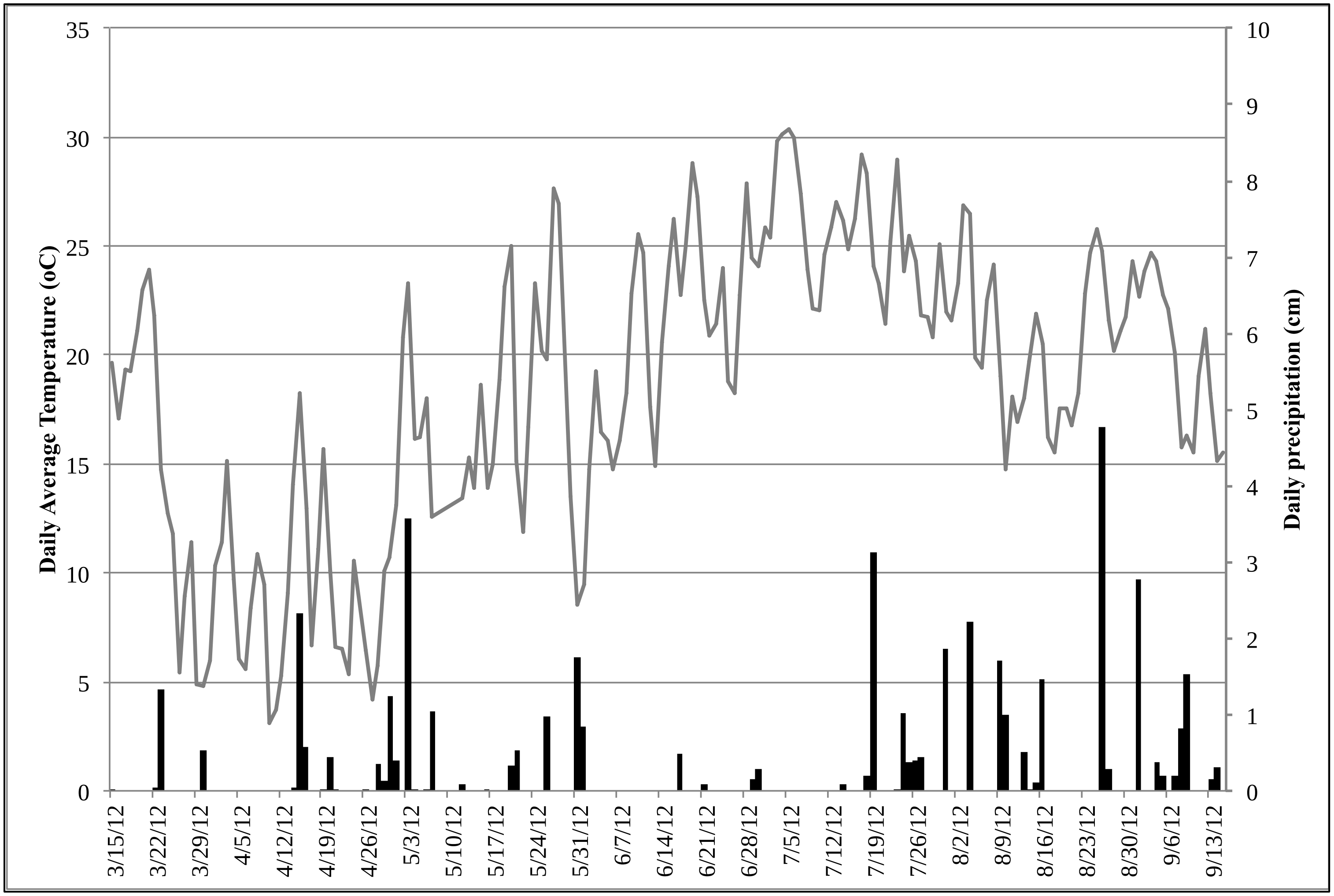

Figure 1.1. Temperature and rainfall summary from the Southwest Michigan Research and Extension Center (Benton Harbor, MI) from 3/15/12 to 9/15/12. Grey line displays average daily temperatures while black bars display daily precipitation.

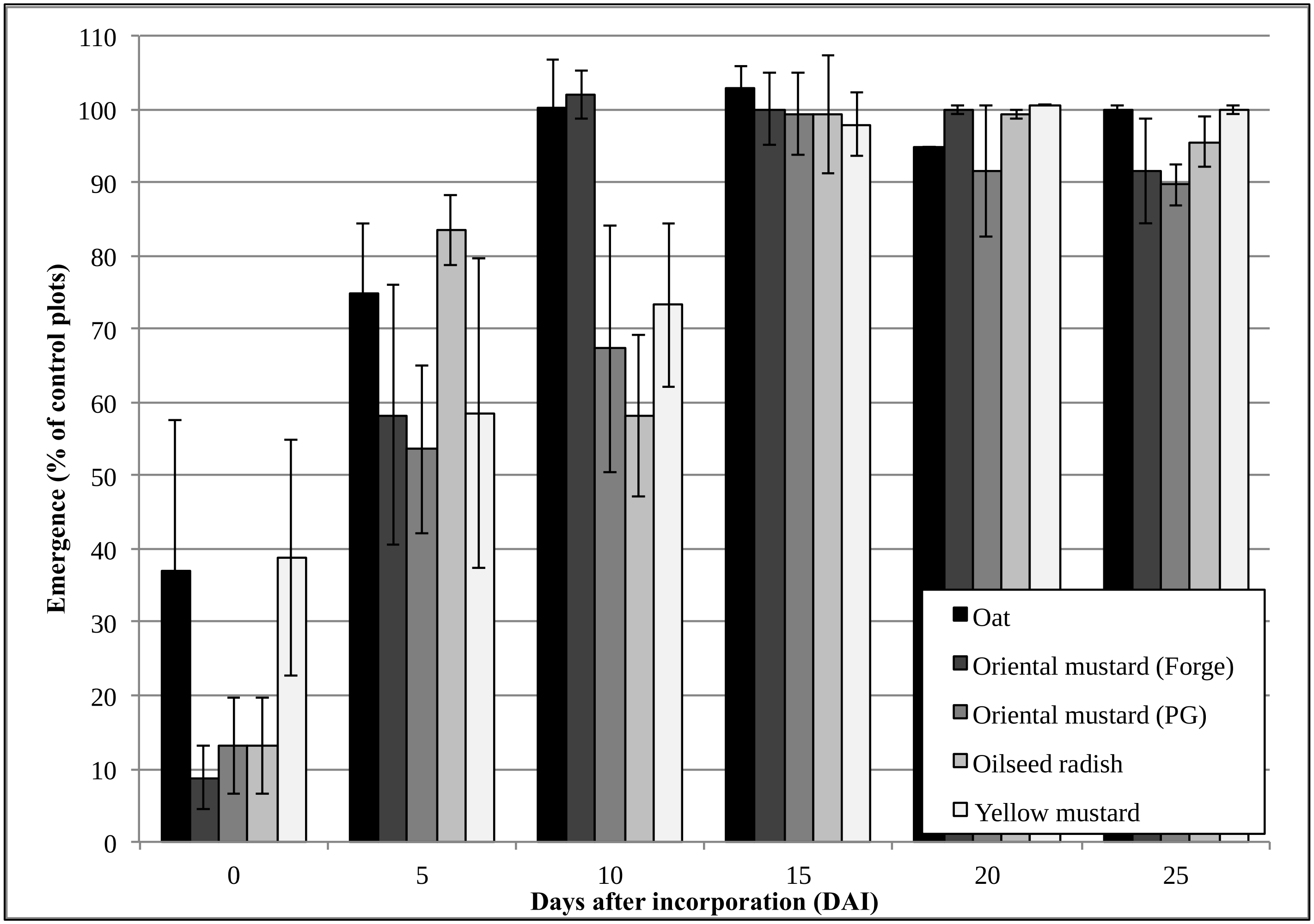

Figure 1.2. Mean and standard errors for the emergence of *C. melo* ‘Athena’ following incorporation of five cover crops at six delayed seeding dates. Emergence here is expressed as a percentage of control plots.

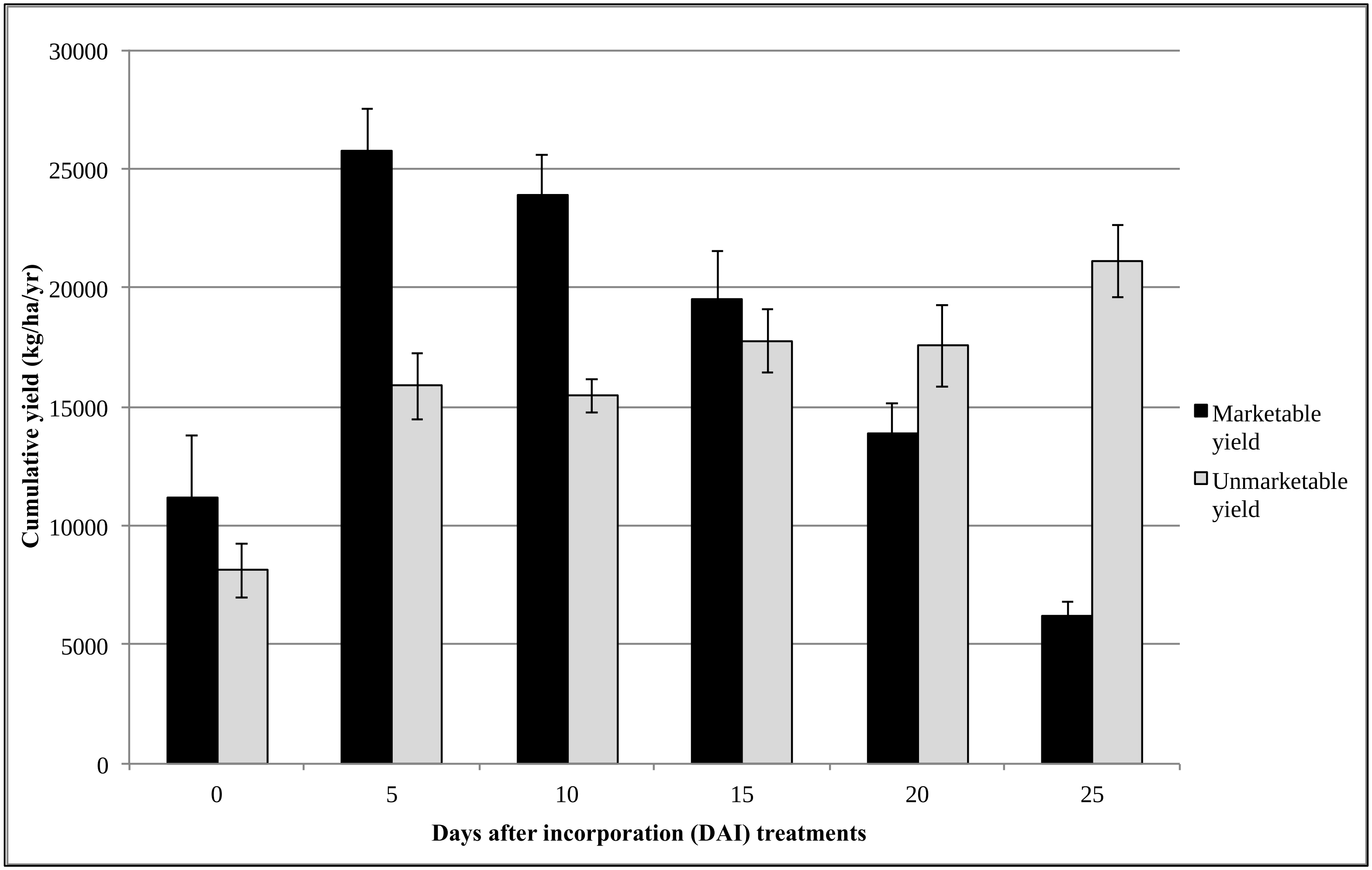

Figure 1.3. Mean and standard errors for cumulative marketable and non-marketable (cull) yields of *C. melo* 'Athena' seeded at 6 dates after incorporation of cover crops (DAI treatments). Yields were graded based on USDA standards

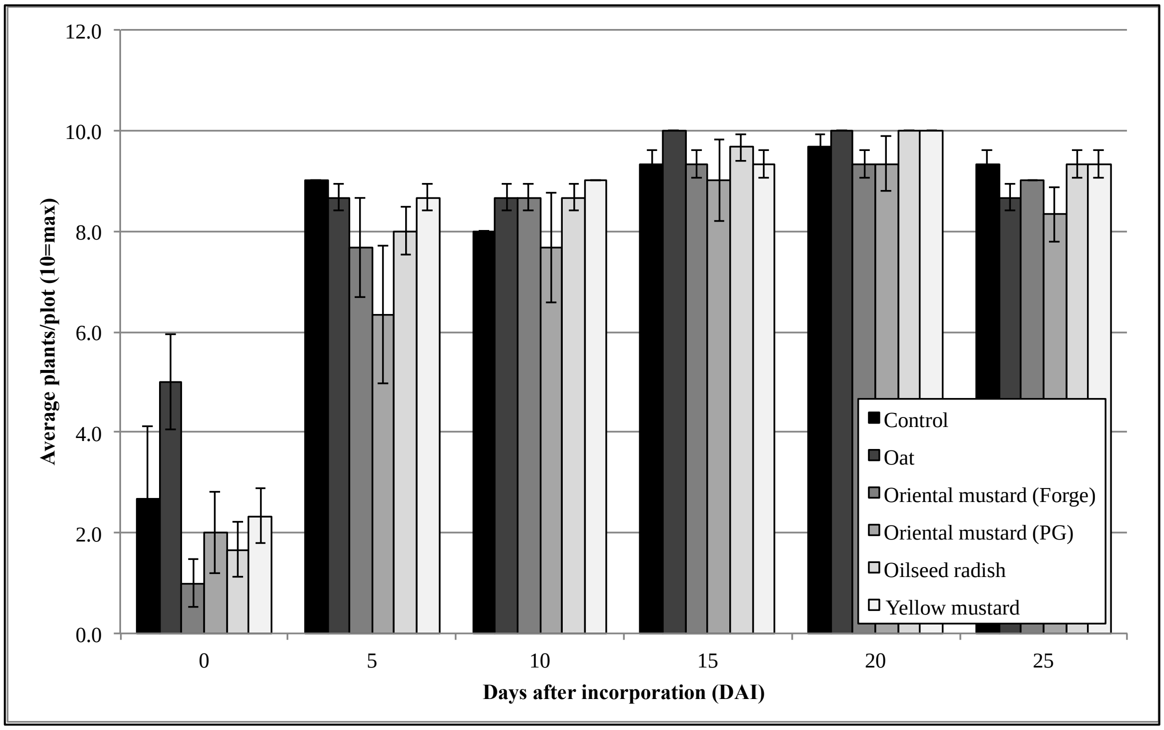

Figure 1.4. Plant density (*C. melo* 'Athena') following crop thinning. Plants were thinned to a spacing of at least 60 cm between plants. Early treatments had lower densities due to reduced crop emergence.

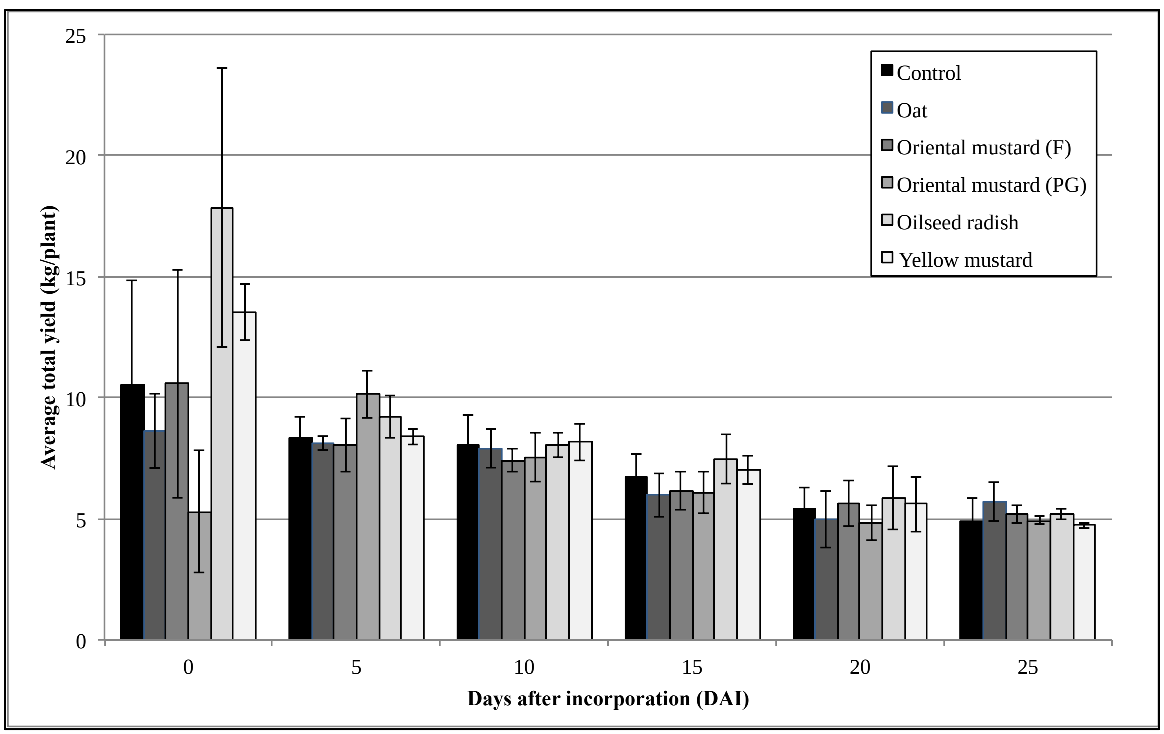

Figure 1.5. Mean and standard errors for average plant yields (yearly) from individual *C. melo* 'Athena' plants. High means for early DAI treatments reflect lower plant densities from reduced emergence.

LITERATURE CITED

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Agricultural Marketing Service. 2008. United States Standards for Grades of Cantaloups. United States Department of Agriculture.

Ackroyd, V.J., Ngouajio, M. 2011. Brassicaceae Cover Crops Affect Seed Germination and Seedling Establishment in Cucurbit Crops. *HortTechnology*, 21: 525-532.

Baker, J.T. Reddy, V.R. 2001. Temperature Effects on Phenological Development and Yield of Muskmelon. *Annals of Botany*, 87: 605-613.

Brown P.D., Morra, M. 1997. Control of soil-borne plant pests using glucosinolate-containing plants. *Advances in Agronomy*, 61: 167-231.

Chan, K.Y., Heenan, D.P. 1996. The influence of crop rotation on soil structure and soil physical properties under conventional tillage. *Soil and Tillage Research* 37: 113-125.

De Baets, S., Poesen, J., Meersmans, J., Serlet, L. 2011. Cover crops and their erosion-reducing effects during concentrated flow erosion. *Catena*, 85: 237-244.

Hansen, Z.R, Keinath, A.P. 2013. Increased pepper yields following incorporation of biofumigation cover crops and the effects on soilborne pathogen populations and pepper diseases. *Applied Soil Ecology*, 63: 67-77.

Haramoto, E., Gallant, E. 2005. Brassica cover cropping: I. Effects on weed and crop establishment. *Weed Science*, 53: 695-701.

Hoagland, L., Carpenter-Boggs, L., Reganold, J.P., Mazzola, M. 2008. Role of native soil biology in Brassicaceous seed meal-induced weed suppression. *Soil Biology and Biochemistry*, 40: 1689-1697.

Justes, E., Bruno M., Nicolardot B. 1999. Comparing the effectiveness of radish cover crop, oilseed rape volunteers and oilseed rape residues incorporation for reducing nitrate leaching. *Nutrient cycling in Agroecosystems*, 55: 207-220.

Kirkegaard, J.A., Gardner, P.A., Desmarchelier, J.M., Angus, J.F. 1993. Biofumigation-using *Brassica* species to control pests and diseases in horticulture and agriculture. *Proceedings 9th Australian Research Assembly on Brassicas*. 77-82.

Larkin, R.P, Honeycutt, C.W., Griffin, T.S., Olanya, O.M., Halloran, J.M, He, Z. 2011. Effects of Different Potato Cropping System Approaches and Water Management on Soilborne Diseases and Soil Microbial Communities. *Phytopathology*, 101: 58-67.

Monfort, W.S., Csinos, A.S., Desaeger, J., Seebold, K., Webster, T.M., Diaz-Perez, J.C. 2007. Evaluating *Brassica* species as an alternative control measure for root-knot nematode (*M. incognita*) in Georgia vegetable plasticulture. *Crop Protection*, 26: 1359-1368.

Nunez-Zofio, M., Larregla, S., Garbisu, C. 2011. Application of organic amendments followed by soil plastic mulching reduces the incidence of Phytophthora capsici in pepper crops under temperate climate. *Crop Protection*, 30: 1563-1572.

Rice, A.R., Johnson-Maynard, J.L., Thill, D.C., Morra, M.J. 2007. Vegetable crop emergence and weed control following amendment with different Brassicaceae seed meals. *Renewable Agriculture and Food Systems*, 22: 204-212

Stivers-Young, L. 1998. Growth, Nitrogen Accumulation, and Weed Suppression by Fall Cover Crops Following Early Harvest of Vegetables. *HortScience*. 33: 60-63.

United States Department of Agriculture, Fresh Products Branch. 2008. United States Standards for Grades of Cantaloups. 1-6.

Wang, G., Ngouajio, M., Warncke, D.D. 2008. Nutrient Cycling, Weed Suppression, and Onion Yield Following Brassica and Sorghum Sudangrass Cover Crops. *HortTechnology*, 18: 63-74.

CHAPTER II:

EVALUATING THE POTENTIAL FOR BIOFUMIGATION AND ANAEROBIC SOIL DISINFESTATION IN MICHIGAN VEGETABLE PRODUCTION SYSTEMS: IMPACTS ON SOIL NITROGEN, MICROBIAL BIOMASS AND YIELDS OF FRESH-MARKET TOMATO AND SLICING CUCUMBER

Abstract

A field experiment was implemented in 2012 and 2013 at the Michigan State University Horticulture Teaching and Research Center (HTRC) in Holt, MI to investigate three objectives: 1) evaluate the potential of a spring-sown brassica cover crop as a carbon source for anaerobic soil disinfestation (ASD) under different plastic mulching regimes, 2) monitor the impacts of a spring-sown brassica cover crop and plastic mulches on nitrogen availability, soil temperatures and soil microbial biomass following ASD treatments and 3) evaluate the impact of biofumigation and plastic mulching practices on yields of the long season fresh-market tomato (*Lycopericon lycopersicum* 'Big Beef') and short season slicing cucumber (*Cucumis sativus* 'Cortez'). Soil redox potential was measured using a HYPNOS III continuous logging system. No significant differences were found among cover crop treatments regarding nitrogen availability and tomato yields, although responses in mulch treatments varied considerably. The addition of molasses to virtually impermeable film (VIF) treatments (to stimulate ASD) dramatically decreased plant available nitrogen during the early part of the growing season and led to substantially lower yields than other mulch treatments. Bare ground treatments (with no plastic mulch) had significantly higher marketable yields in 2012 and 2013 (non-significant in 2013). Soil temperatures are believed to have caused these declines in yields, where root-zone temperatures routinely exceeded 100^{o}F in 2012. The results of our study indicate that nitrogen dynamics and soil temperatures are affected considerably by ASD mulching practices and molasses additions and should be considered in future work on ASD in Michigan attempting to optimize this practice.

Introduction

The need to develop sustainable management practices is one of the greatest challenges for agriculture today. In the horticultural sector, restrictions on the use of fumigants like methyl bromide have required innovation in conventionally managed crops for pest management, while in organically managed systems even greater restrictions create even greater challenges. In both conventional and organic production systems, Integrated Pest Management (IPM) has been promoted and largely embraced by growers. With IPM, decisions for managing pests are weighed in a cost/benefit analysis that takes into consideration the impacts and interests of growers, society and the environment (Kogan, 1998). Although implementation of IPM tactics varies considerably, in general, it involves understanding pest biology/life cycles, preventive measures to manage pest outbreaks (cultural, biological and monitoring based approaches) and often rely on chemical controls as a last resort.

Although the importance of preventative measures for pest management should not be understated, situations involving heavy pest pressure may necessitate the use of response-oriented strategies to achieve desirable pest suppression. Restrictions on the use of broad-spectrum fumigants have limited the ability of growers to respond to pest outbreaks when they occur. Alternative, control-based strategies have recently been developed that seek to minimize negative environmental impacts, while concurrently providing effective control of soil-borne pests.

<u>Biofumigation</u>

Biofumigation (BF) utilizes the unique biochemistry of certain plant families (most notably, the Brassicaceae or mustard family) to suppress primarily soil-borne pathogens and weeds. The suppressive effects of brassicas have been observed for decades, but mechanisms have been poorly understood until recent years. Glucosinolates (GSLs) are plant secondary metabolites produced by mustards that, upon contact with the enzyme myrosinase in the presence of water, hydrolyze to form an assortment of biologically reactive products including nitriles, thiocyanates, and volatile isothiocyanates (ITCs). ITCs exhibit broad biocidal activity against numerous pests (Kirkegaard, 2009). The GSL profiles of species within the Brassicaceae vary considerably, although they tend to remain consistent within a given species (Kirkegaard and Sarwar, 1998). The ability to suppress pests through biofumigation largely hinges on the ability to accumulate sufficient quantities of biomass (directly related to GSL quantity), effectively convert GSLs to ITCs, and maintain ITCs in the soil profile where they can react with pest organisms. In practice, mustard residues are incorporated either as cover crops, inter-crops, or as seed meals (bi-product of the oil extraction process). Macerating plant residues is crucial to the release of GSLs and can be achieved through flail mowing or, less effectively, through disking-in plant residues without mowing. Following incorporation, the soil is irrigated to facilitate hydrolysis and to help 'seal' the volatile ITCs into the soil profile, reducing their loss to the atmosphere.

Anaerobic Soil Disinfestation

Another practice developed recently is anaerobic soil disinfestation (ASD), utilized to varying degrees around the world but most notably in Japan (Momma, 2008), the Netherlands (Blok et al., 2000), Spain (Nunez-Zofio et al., 2011) and the United States. Within the U.S., specific pathogens have been targeted with ASD including, perhaps most notably *Verticillium dahliae* in California strawberry production (Shennan et al., 2009) and *Fusarium oxysporum* and *Meloidogyne incognita* in Florida bell pepper and eggplant production (Butler et al., 2009; Butler et al., 2012).

To induce an anaerobic state, labile carbon sources such as molasses (Butler et al., 2012), rice bran (Shennan et al., 2009), wheat bran (Momma, 2008) or other forms of plant biomass (Mehissa et al, 2007) are incorporated into the soil. Fields, or targeted areas within the field are irrigated and covered with impermeable films for a period of time, typically 4 to 6 weeks (Lamers, 2010). During ASD, soil oxygen is rapidly depleted and the redox potential of the soil decreases dramatically. Under prolonged, sufficiently low reducing conditions in the soil, organic acids are produced by fermentative decomposition of residues. The prolonged anoxic conditions and organic acid production are believed to be the mechanisms largely responsible for disease suppression, although changes in the microbial communities (Mehissa et al., 2007; Nunez-Zofio et al., 2011; Momma, 2008) and long-term suppressiveness (Goud et al., 2004) following ASD implicate microbially mediated mechanisms of disease suppression as well.

Incorporating sufficient quantities of biomass into the soil is critical for both of these practices to be effective in suppressing disease. Generating sufficient quantities of ITCs for biofumigation is directly related to biomass accumulation by brassica species. Likewise, sufficient biomass is needed to facilitate the anaerobic conditions required for ASD. Utilizing cover crop residue as a carbon source for ASD is attractive due to the numerous agronomic and ecological benefits that cover crops impart. Warm-season cover crops can effectively generate anaerobic conditions in Florida where summer-fallow periods provide windows for cover crops (Butler et al., 2011). In temperate regions where the growing season is shorter, warm-season cover crops can be challenging to fit into the production cycles with warm-season cash crops and requires the substitution of income generating cash crops with non-harvested cover crops. Although many long-term benefits can be attained through the use of cover crops, using summer cover crops requires that fields be moved out of production temporarily, negatively impacting producers' profitability. However, opportunities for utilizing cool-season cover crops as carbon sources for ASD exist in these regions; particularly attractive options include brassica family cover crops that thrive in cooler temperatures and can accumulate significant, albeit variable quantities of biomass (Snapp et al., 2005).

Plastic mulching practices

Recent developments in agricultural plastics have led to the widespread availability of impermeable films. Compared with the traditional low-density polyethylene mulches, virtually impermeable films (VIFs) have been shown to better retain commercially available fumigants in the soil profile (Austerweil et al., 2006). While VIFs have been used primarily for improving the efficacy of chemical, manufactured fumigants, they might also be used to enhance the efficacy of biologically based fumigation practices such as BF and ASD.

Black plastic is the standard mulch in horticultural production worldwide, although other types of colored mulches have shown promise and even widespread adoption in different regions (Tarara, 2000). Black plastic is used extensively in fresh-market tomato (*Lycopersicon lycopersicum*) production, particularly in cooler production regions due to its ability to increase root-zone soil temperature (Teasdale et al., 1995; Decoteau et al. 1989), increase early harvests (Abdul-Baki et al., 1992; Teasdale et al., 1995) and total yields (Abdul-Baki et al., 1992), and to control weeds.

Although studies have identified optimal conventionally applied nitrogen fertilizer rates for crops grown with black plastic (Abdul-Baki et al., 1997), little information exists on nitrogen dynamics under plastic mulch, particularly in organic production systems that rely extensively on cover crops for fertility. Nitrogen mineralization has been shown to increase as a function of temperature (Macdonald et al., 1995) but is also influenced by oxygen availability (Parr et al., 1959; Moore et al., 1992). Management practices such as soil flooding or those that lead to soil compaction (e.g., heavy equipment usage) can dramatically influence the oxygen availability within soils, and can influence nitrogen mineralization (Jensen et al., 1996). Nitrogen availability in soils is further complicated by nitrogen transformations (denitrification, ammonia volatilization) and microbially mediated immobilization (Robertson et al., 2007). Anaerobic soils are characterized by high rates of ammonium accumulation, denitrification, and low biological immobilization (Ponnamperuma et al., 1984). Although plastic mulching influences certain soil qualities such as temperature, its influence on nitrogen dynamics has yet to be studied in systems utilizing VIF mulches for ASD or fumigation enhancement purposes.

<u>Project objectives</u>

The objectives of this study are to 1) evaluate the potential of a spring-sown brassica cover crop as a carbon source for ASD under various plastic mulching regimes, 2) monitor the impacts of a spring-sown brassica cover crop and plastic mulches on nitrogen availability, soil temperatures and microbial biomass following ASD treatments and 3) evaluate the impact of biofumigation and plastic mulching practices on yields of a long season (fresh-market tomato (*Lycopericon lycopersicum* ‘Big Beef’)) and short season (slicing cucumber (*Cucumis sativus* ‘Cortez)) vegetable crop.

Materials and Methods

Field design and treatment implementation

A two-year field experiment was conducted at the Michigan State University Horticulture Teaching and Research Center (HTRC) in Holt, MI (42°40'34"N, 84°29'5"W) from 2012 to 2013. The fields used in the experiment had been cropped from 2009 to 2011 with organically managed bell peppers and cucumbers using hairy vetch/rye cover crop mixtures. To prepare for the 2012 experiment, one field was seeded with a sorghum sudangrass cover crop in 2011, with residue removed from the field to reduce variability within the field caused by previous experimental treatments. The same procedure was followed in 2012 in the adjacent field to prepare the 2013 study.

In 2012, four cover crops were seeded in 8.5 x 13.4 m plots within a randomized complete block design. These treatments were replicated 4 times across the field and each block included a no cover crop control plot. Cover crops included: oriental mustard (*Brassica juncea* ‘Pacific Gold’) (PG), yellow mustard (*Sinapis alba* ‘Ida Gold’) (IG), oilseed radish (*Raphanus sativus* ‘Defender’) (OSR) and oats (*Avena sativa* ‘Excel’) (OAT). Standard seeding rates of 8,8,11 and 134 kg/ha were used for PG, IG, OSR and OAT respectively. Prior to cover crop seeding, the field was fertilized at the rate of 112 kg N/ ha (Mcgeary’s Organic Fertilizer, 8-2-2) using an oscillating spreader. Cover crops were then evenly broadcast by hand in each plot and incorporated using a rolling-basket implement. Due to poor stand establishment in 2012, seeding methods were modified in 2013 by using a multi-row push seeder to better distribute the seeds over the surface and improve seed depth placement. Poor stand establishment among IG plots in 2012 also required altering the seeding rate to 18 kg/ha for 2013.

At approximately 50% flowering, cover crops were sampled for above and below ground biomass using four 25 x 50cm quadrats from each plot. Plant residue was then dried at 90^{o}C for two weeks and weighed. Following biomass sampling, cover crops were mowed using a flail mower and immediately incorporated to the soil using a roto-vator. Following incorporation, sub-plot mulch treatments were immediately applied using a mechanical plastic mulch layer, including bare ground (BG), standard low-density black polyurethane (BP) and black virtually impermeable film (VIF). In 2013, an additional mulching treatment (VIF+M) was applied to all main plot treatments; this included the application of molasses at the within bed rate of 19.9 Mg/ha as a standard ASD treatment comparison (Butler et al., 2012). After observing a two-week ASD period, fresh-market tomato ‘Big beef’ and slicing cucumber ‘Cortez’ (Osborne International Seed co., Mt. Vernon, WA) transplants were planted at 61 cm centers within plots. Guard plants were established at the plot ends and included a roma-type tomato, *L. lycopericon* ‘Mariana’ and *C. sativus* ‘Lemon Cucumber’. Only two mulching treatments were evaluated in cucumber (bare ground and VIF) for both years, while in tomatoes all mulch treatments were applied (three in 2012, four in 2013). All crops were managed using organic production methods. Weeding was accomplished by hand (cultivation and hand weeding) as needed. Cucumber insect pests were managed using OMRI approved pyrethrin formulations (Pyganic**®,** Mclaughlin Gomley King Company**)** for control of spotted *(Diabrotica undecimpuncta*) and striped (*Acalymma vittatum*) cucumber beetles.

Objective 1 methods

In 2013, two cover crop treatments (IG and no cover control) and four mulching sub-plot treatments were evaluated to determine suitability for ASD. Following incorporation of cover crops and plastic mulch application, treatments were irrigated using drip tape. To assess anaerobic conditions, two methods were used. Throughout ASD, gas samples were collected from plots using hypodermic needles/syringes and transferred to Exetainer® vials (Labco Ltd., Ceredigion, UK). Vials were first flushed by venting with sample gas and then filled to an over pressurized state to prevent sample loss. Samples were analyzed at the Kellogg Biological Station (KBS) using gas chromatography to assess concentrations of CO_2 (IRGA detector) and N_2O (ECD detector).

Soil redox potential (Eh) was also measured using the HYPNOS III continuous Eh logging system (Vorenhout et al., 2004) using Pt electrodes and an Ag/AgCl reference electrode. Three probes (each measuring at 10cm and 25cm depths) were placed in six plots of varying treatment combinations (Table 2.3). Redox measurements were set to be logged at 15 minute intervals during the duration of the ASD treatment. Measured redox potential (Em) was adjusted to standardized (Eh) redox potential (to relate to the standard hydrogen electrode). In determination of critical redox potential values (CEh), aggregated pH values were used from each main plot. The following equation was used to determine the CEh (Butler et al., 2011):

$$\text{CEh} = 595\text{mV} - (60\text{mV} * \text{soil pH})$$

Cumulative soil anaerobicity (mVh beneath the CEh) was assessed for each Pt electrode by dividing each measurement by 4 (to generate hourly units from 15 minute logging intervals) and summing each electrode dataset.

Objective 2 methods

Two main plot treatments (IG and no cover) and four sub-plot mulching treatments (BP, VIF, VIF+M and NM) were sampled in 2013 to evaluate nitrogen availability throughout the growing season in tomato plots. Soil composite samples were collected (at 6” depth) from twelve individual cores at several dates throughout the growing season. Samples were mixed and later extracted with 1 M KCl. Solutions were then analyzed for NO_3^- and NH_4^+ using flow through analysis (Lachat QuikChem ® 8500 series, Lachat Instruments, Loveland, CO) at the MSU soil testing lab. To track nitrogen availability during ASD while maintaining the integrity of the plastic, ion exchange resin strips were also used. Anion and cation resin sheets (GE osmonics Inc., Minnetonka, MN) were cut into 2.5x10 cm strips. Three pairs of anion and cation strips were placed in each plot from 0-10 cm depth starting at the beginning of ASD and were extracted and replaced every two weeks during the growing season. Resin strips were collected and then extracted using a 2 M KCl solution and analyzed at the MSU soil testing lab for NO_3^- and NH_4^+. After resin strip placement in the soil, mulched treatments were covered and sealed using black-colored duct tape. In the same plots, soil temperature was monitored by burying HOBO® temperature data loggers (Onset Computer Corp., Bourne, MA) 10 cm beneath the soil surface. Loggers were set to collect temperature data every 30 minutes and were retrieved after the final crop had been harvested.

Microbial biomass was determined through the chloroform fumigation-incubation method (Jenkinson et al., 1976). Soil samples collected immediately after ASD were stored at 4^{o}C until analysis. Field samples were separated into three fumigated and three unfumigated lab reps following sieving with 4 mm mesh screen. Soil moisture content for each sample was determined and all samples were adjusted to 50% gravimetric water holding capacity. Samples were placed in desiccators and fumigated for 24 hours with chloroform. Following fumigation, samples were incubated in air-tight quart-sized mason jars with butyl septa inserted into the lids. After 10 days, CO_2 concentrations were determined by using an infrared gas analyzer (Qubit S151 CO_2 Analyzer, Qubit System Inc., Kingston, Ontario, Canada). Soil microbial biomass was calculated using the following equation: $1.73*F_C-0.56*UF_C$, where F_C and UF_C are the mineralized carbon from fumigated and unfumigated soil samples (Horwath et al., 1996).

Objective 3 methods

As crops matured, tomato and cucumber yields were collected weekly from all cover crop x mulching treatment plots . Yields were graded and classified as marketable or unmarketable based on USDA grading standards (USDA, 2008). The sorted yields were then counted (fruit number) and weighed. Cumulative yield data was analyzed (SAS 9.3, Cary NC) using ANOVA and mean differences were evaluated using Fischer's LSD (a=.05). The same procedure was used for evaluating early yields on individual harvest dates.

Results

Cover crop biomass

Although seeding methods seemed to improve cover crop stand establishment in 2013, in both years dry weight biomass of cover crops was substantially lower than anticipated (Table 2.2). Previous studies using spring seeded brassica cover crops in southern Michigan have demonstrated biomass yields of 6068, 3641 and 3487 kg/ha for oilseed radish, Oriental, and yellow mustard respectively (Ackroyd et al., 2011), a notable difference from the 2291, 1235 and 1133 kg/ha generated in this study (Table 2.2). Cultivars like 'Pacific Gold' are noted to be quite sensitive to day-length and can begin to flower before substantial biomass has accumulated (Snapp et al., 2006). Oilseed radish accumulated the greatest quantity of biomass for the brassicas (Table 2.2) while yellow and oriental mustards had the lowest in both years. Among all of the cover crops seeded, oats accumulated the most biomass in 2012, and had substantially lower mean biomass in 2013, although biomass was quite variable from plot to plot (Table 2.2).

Redox potential and soil gas monitoring during ASD

Establishing anaerobic conditions proved to be more challenging than anticipated (Table 2.3). Of the 32 sensors installed in the field, only three reported Eh values below the CEh (182-198mV): two were in VIF+M plots (35-10,513 mVh beneath CEh), and another was under VIF (268 mVh beneath CEh). Also, it is worth noting that these sensors were also all placed within the cover crop treatments which may have contributed to the lower Eh from the added biomass. Although biomass estimates from the yellow mustard plot were lower than anticipated (1063 kg/ha), in a greenhouse study, Butler et al. (2011) showed that similar rates of cover crop biomass produced high cumulative anaerobicity, although the authors noted that anaerobic conditions are often more challenging to establish under field conditions than in greenhouse pot studies (Butler et al., 2011). Other studies have demonstrated that anaerobic conditions can be maintained under field conditions, where mVh beneath the CEh can exceed 50,000 within two weeks depending on soil type, irrigation and plastic characteristics (Shennan et al., 2010). To attain cumulative anaerobicity closer to those values needed for successful ASD (as determined by previous research) more work could be focused on manipulating irrigation techniques, timing, and evaluating different carbon sources suitable for use in the Michigan climate.

CO_2 concentrations were observed to be much higher under VIF mulch treatments than under standard black plastic during the entirety of ASD confirming that VIF mulch is less permeable than the standard black plastic mulch (Figure 2.2). The addition of molasses also created substantially higher concentrations of CO_2 under the mulch. N_2O concentrations followed similar patterns where by VIF mulch with molasses generated the highest concentration of N_2O throughout the ASD period. Methods used to determine gas concentrations do not permit quantification of the actual generation of gases over time among plastic mulch treatments (fluxes), but the data suggest that the molasses amendment generated greater quantities of N_2O under VIF mulch (Figure 2.3). Interestingly, cover crop treatments all diverged from no cover treatments on the sampling date prior to ASD termination (June 17).

<u>Nitrogen dynamics and microbial biomass</u>

Differences in NO_3^- and NH_4^+ concentrations were observed at various times throughout the 2012 and 2013 growing seasons among mulch treatments, while cover crop treatment differences were not significant within each year (α=0.05). Two mulch treatments were monitored in 2012 (NM and VIF), while all 4 were monitored in 2013 (NM, BP, VIF, VIF+M). NO_3^- and NH_4^+ were significantly higher during ASD and the first four (for NO_3^-) and two (for NH_4^+) weeks after transplanting (Figure 2.5). For the last four weeks, this trend was reversed where NM treatments sustained significantly higher levels of NO_3^- and NH_4^+, although the magnitude of these differences was less substantial than at earlier sampling dates. Differences in NH_4^+ concentrations were not significant after the first four weeks in 2012 (Figure 2.4). In 2013, NO_3^- was significantly higher in VIF and BP treatments than NM and VIF+M during ASD and the first two weeks after transplanting (Figure 2.5). This trend was reversed from 7/18-7/31 where NM and VIF+M soils had significantly higher NO_3^- than under BP, and again from 8/14-8/28 NM plots had higher NO_3^- than all other plots. NH_4^+ concentrations were highest in VIF+M treatments for the first two weeks following ASD and the last two weeks of the season. NH_4^+ data proved to be quite variable, particularly under mulched treatments although these differences were significant (α=0.05) on the second and last sampling date in 2013 (Figure 2.5), where VIF+M treatments were the highest. Mean microbial biomass carbon and soil respiration were the highest in the VIF+molasses treatments, although differences were not significant (α=0.05) (Figure 2.7).

Soil temperatures

2012 proved to be an exceptionally warm growing season. Historical heat data at this site shows that 2012 had substantially higher days with temperatures exceeding 32.2^{o}C (90^{o}F) than in the previous 6 years or in 2013 (Figure 2.10). Likewise in 2012 soil temperatures reached exceedingly high levels under plastic mulch treatments, when at mid-day in July, average temperatures of 40^{o}C (104^{o}F) were recorded following air temperature readings of 37.7^{o}C (100^{o}F) (Figure 2.10). Plots without plastic mulch displayed substantially lower soil temperatures during these heat events, reaching highs of 32.7^{o}C (91^{o}F) on the same date (Figure 2.11). Although tomatoes are known as summer, heat-loving crops, previous work has indicated that optimal root-zone temperatures for tomato growth and yields are around 26^{o}C (78.8^{o}F) and maximum temperatures (the point at which growth ceases) at 29.3^{o}C (84.7^{o}F) (Diaz-Perez et al., 2002). While aerial heat stress in tomato has been shown to reduce pollen release, pollen viability and fruit set (Firon et al., 2006), less is known about heat stress in tomato roots. Monthly mean root-zone temperatures from 2012 indicate that in July and August, plots without plastic mulch maintained mean root-zone temperatures closer to the cited optimum of 26^{o}C (78.8^{o}F) while under BP and VIF, mean temperatures (28.3^{o}C (83^{o}F) in June, 29.4^{o}C (85^{o}F) in July) were above optimal (figure 2.12). In 2013, mean temperatures under all mulch treatments were substantially lower during the summer months, by at least 5^{o}F. Because temperature plays an integral role in plant development, growth and reproduction, differences in root-zone temperature caused by mulch treatments were a likely contributor to differences in tomato yields.

<u>Tomato yields and quality</u>

In 2012 and 2013, cover crop treatments did not have significant effects on tomato yields, while mulching treatments did (table 2.4). In 2012, early marketable yields (from first four harvests) were highest in plastic mulch treatments compared with no mulch, while late marketable yields (last four harvests) were significantly higher in no mulch plots than in both black plastic and VIF treatments. While early marketable yields were substantially greater under plastic, the fraction of the total marketable yield accounted for by early yields was substantially less than that of later yields in no mulch (7% vs. 75%), black plastic (15% vs. 55%) and VIF (10% vs. 67%). This demonstrates that the majority of tomato yields are harvested later in the season, a potential trade-off faced by producers wanting to maximize early yields while also attaining high cumulative yields. 2013 marketable yields followed similar trends based on these three mulching treatment, although total marketable yield differences were not significant (α=0.05).

VIF+M treatments yielded significantly less total marketable yields than all other mulch treatments where moderate early and late yields did not compensate overall for the differences in high early (BP, VIF) and high late (NM) yields of the other mulching treatments. Interestingly, total unmarketable yields were greater in VIF treatments than in black plastic in 2012 while in 2013 no significant differences in total unmarketable yields were observed among plastic mulch treatments (although they were significantly greater in NM plots). Several studies have noted that extreme heat under black plastic mulch in causes reductions in total yields of tomato when compared with other plastic (Ngouajio et al., 2005) and organic-residue mulches (Tindall et al., 1991; Teasdale et al., 1995). While black plastic is currently the standard mulch for tomato production in Michigan, increases in early yields may be offset by lower late yields, particularly during warm years as observed in this study. Using plastic or organic mulches that transfer less heat to the root-zone than black plastic mulches might be a management strategy worth adopting by growers in this region.

Cucumber yields

Overall cucumber yields were substantially higher in 2012 than in 2013 across all treatments. In 2012, there was no significant difference among cover crop or mulch treatments on marketable or unmarketable yields (Table 2.5). However, in 2013, plants mulched with VIF had significantly higher marketable and unmarketable yields. Unlike tomato yields, cucumbers yields were not signifcantly affected by VIF mulch in the unusually warm year of 2012 although mean yields under no mulch treatments were higher than under VIF. Although optimal root-zone temperatures have been cited as being relatively similar to tomatoes at 24-30^{o}C (75-86^{o}F) (Gosselin et al., 1985), it is possible that cucumber leaves shade soil more effectively than tomatoes to reduce soil heat accumulation, reducing the impact of excessive temperatures on crop yields grown on black plastic mulch. Many of the harvested cucumber fruits in 2012 from the VIF plots had symptoms of heat exposure (white, bleached areas) that were not common in NM plots which contributed to the higher unmarketable yields under VIF. While nitrogen was not monitored under cucumber plots, it seems likely that NO_3^- availability would be higher under cucumber plots as increases in soil temperature under black plastic would increase mineralization and subsequently plant available nitrogen early in the season as was seen in tomato plots; this could help to explain greater yields under VIF in the cooler 2013 season.

Discussion

Brassica cover crops are known to exhibit a variable affinity for scavenging nutrients, accumulating biomass, and reducing disease incidence for subsequent crops. In our study, spring-seeded brassica cover crops did not demonstrate differences in affecting nitrogen availability or yields of fresh-market tomato. While no statistically significant differences were observed among cover crop treatments, low cover crop biomass accumulation could be masking potential differences that might occur under environments more favorable for cover crop growth. In addition, the use of these cover crops as a carbon source for ASD would likely be improved if greater biomass accumulation occured. The lack of anaerobic conditions generated under nearly all of the cover crop and mulch treatment combinations indicate that methods for attaining sufficiently low Eh need to be developed for this region prior to subsequent investigations regarding ASD. Researchers in California determined that 50,000 mVh under the CEh were needed to effectively reduce *Verticillium dahliae* microsclerotia in strawberry production systems and would provide a logical initial benchmark for anaerobicity here. While anaerobic conditions were documented from a few locations in this experiment, they were not maintained for a sufficient length of time to be considered effective for ASD. Optimizing irrigation levels and delivery systems could likely improve the establishment of anaerobic conditions as irrigation has been shown to be critical in ASD establishment in other studies. Under VIF, higher concentrations of CO_2 and N_2O were maintained during ASD, which reinforces its utility in maintaining a localized anaerobic environment. Amendment with molasses led to substantially higher CO_2 and N_2O concentrations under VIF but caused dramatic reductions in plant available N throughout the rest of the growing season. Total marketable yields from molasses amended plots were significantly lower than all other mulch treatments. Plants in these plots were visibly stunted for a period of time following ASD, likely due to severe N deficiency. Under plastic mulch treatments, NO_3^- and NH_4^+ were significantly higher during the first month following ASD termination and exhibited higher early yields. During the mid-season of 2012, an unusually warm year for mid-Michigan, soil temperatures under all plastic mulch treatments reached extremes of over 100^{o}F and could explain differences in total marketable tomato yields. Bare ground treatments had the highest late and cumulative yields in 2012 and 2013, although 2013 yield differences were not statistically significant (with the exception of molasses amended plots). For tomatoes, utilizing black plastic mulches can afford benefits in the way of weed control and warming the soil in the early spring when temperatures are still cool; however, in later plantings it could be advantageous to utilize mulches that transfer less heat to the soil such as white or reflective mulch for better maintenance of soil temperatures and ultimately crop yields. Neutral (2012) and positive (2013) effects of black VIF mulch on cucumber yields observed in this study demonstrate its utility as a model crop for subsequent research that seeks to utilize these black plastic mulches in south-central Michigan.

APPENDIX

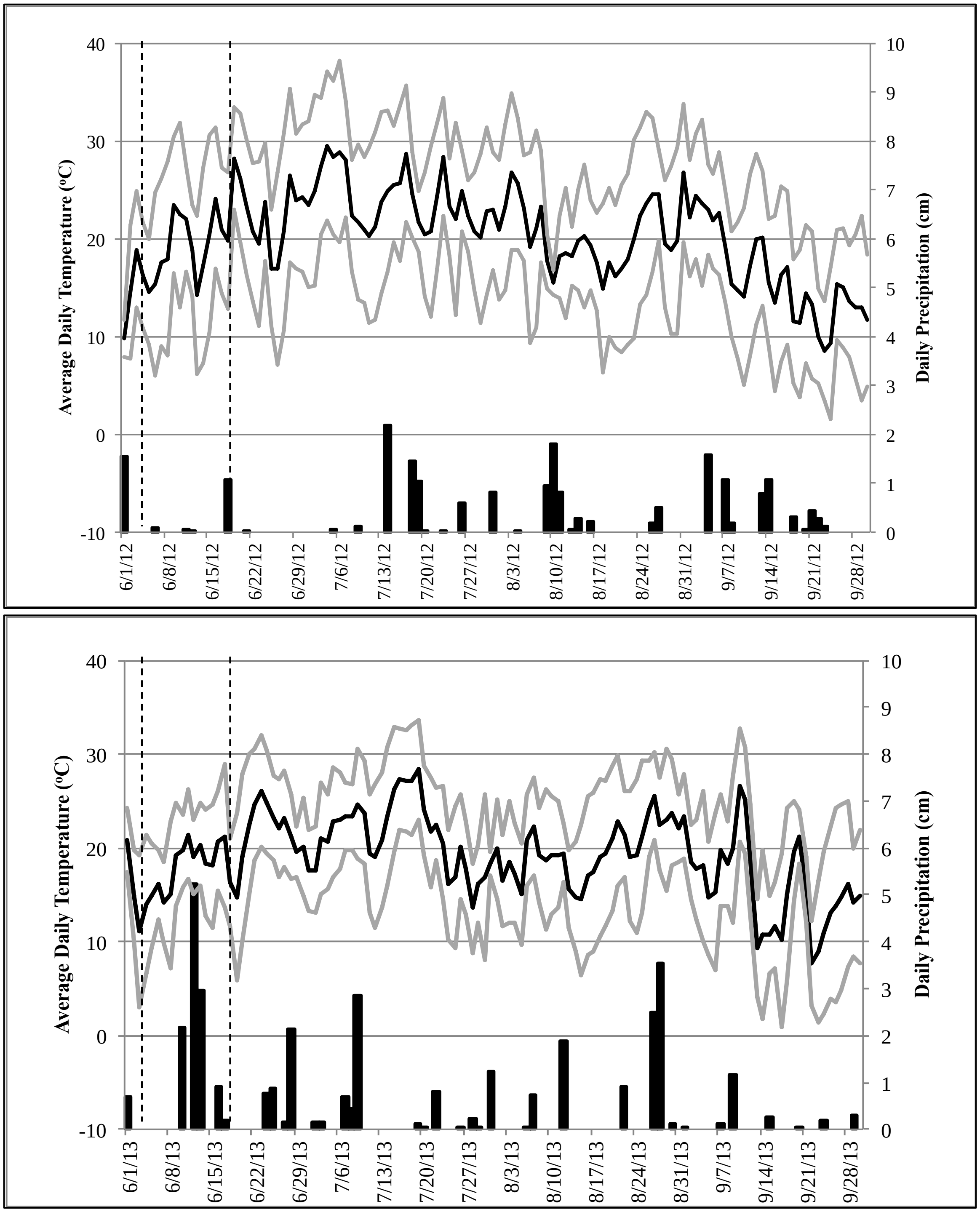


Figure 2.1. Average daily (black solid line) minimum and maximum (grey solid lines) and daily precipitation (bars) at the HTRC (Holt, MI) for the 2012 (above) and 2013 (below) growing season. The first and second vertical dotted line indicate the initiation and termination of ASD respectively.

Table 2.1. Growing degree day, heat stress and precipitation at HTRC weather station from June 1- Oct. 1[1]

Year	Degree Days (F, Base 40)[2]	Temp. greater than 90 F (days)	Rainfall (inches)
2008	3400	1	43.2
2009	3179	2	32.3
2010	3464	2	30.5
2011	3414	7	35.3
2012	**3491**	**16**	**20.1**
2013	**3199**	**6**	**34.0**

[1]Dates analyzed based on growing season for southern MI.
Years in bold type denote years when experiment was conducted.
[2]Calculated according to Baskerville-Emin method using a base temperature of 40^{o}F.

Table 2.2. 2012 and 2013 Cover crop seeding rates, mean dry weight cover crop and weed biomass at incorporation, accumulated total N, and residual soil N prior to ASD[1]

Cover Crop/ variety	Seeding rate (lbs/ac)	Cover crop biomass (kg/ha)	Weed biomass (kg/ha)	[2]Accumulated N (kg/ha) from cover crop biomass	[3]Accumulated N (kg/ha) from weed biomass	Soil NO_3^- (mg/kg) at incorporation	Soil NH_4^+ (mg/kg) at incorporation
2012							
Control (no cover)	-	-	819 ± 99	0	-	-	-
Oilseed radish/ 'Defender'	10	2291 ± 301	148 ± 27	44.3 ± 7.2	-	-	-
Oat /'Excel'	120	2418 ± 431	136 ± 24	36.9 ± 4.2	-	-	-
Yellow mustard / 'Ida gold'	7	1133 ± 165	388 ± 103	28.9 ± 4.5	-	-	-
Oriental mustard / 'Pacific gold'	7	1235 ± 216	311 ± 19	26.8 ± 4.6	-	-	-
2013							
Control (no cover)	-	-	732 ± 114	0	13.8 ± 4.5	3.43 ± 0.44	1.53 ± 0.22
Oilseed radish/ 'Defender'	10	2097 ± 408	133 ± 31	51.3 ± 8.2	3.9 ± 0.8	2.91 ± 0.23	1.36 ± 0.13
Oat /'Excel'	120	1459 ± 968	122 ± 28	27.3 ± 5.8	3.2 ± 0.5	2.99 ± 0.19	2.19 ± 0.62
Yellow mustard / 'Ida gold'	10	1002 ± 183	347 ± 119	24.6 ± 2.8	11.3 ± 3.2	3.26 ± 0.34	2.05 ± 0.34
Oriental mustard / 'Pacific gold'	7	1246 ± 348	278 ± 22	28.3 ± 7.4	8.3 ± 0.9	3.32 ± 0.33	1.35 ± 0.10

[1] Cell values (except for seeding rates) represent mean values followed by standard errors.
[2] Accumulated N was calculated by multiplying the dryweight biomass estimates by plant %N data derived from subsamples of dried biomass.
[3] N accumulated from weed biomass was estimated in 2013 only.

Table 2.3. Cumulative mVh beneath critical redox threshold (CEh) at two depths under cover crop and plastic mulching treatments

Cover Crop	Mulch type	Probe	mVh below CEh (7.5cm depth)[1]	mVh below CEh (25.5 cm depth)	pH (before ASD)[2]	pH (after ASD)
Yellow mustard	No mulch	1	0	0	6.6	6.8
		2	0	0		
		3	0	0		
	Black plastic	1	0	0		7.0
		2	0	0		
		3	0	0		
	VIF	1	268	0		6.8
		2	0	0		
		3	0	0		
	VIF+ Molasses	1	0	35		7.2
		2	10,513	0		
		3	0	0		
No cover	No mulch	1	0	0	6.9	6.7
		2	0	0		
		3	0	0		
	VIF+ Molasses	1	0	0		7.0
		2	0	0		
		3	0	0		

[1] Cumulative mVh (millivolt hours) below CEh was calculated by adding 220 mV (reference value for H electrode) to measured values (E_m), subtracting these standardized E_h values from CEh thresholds, dividing cell values by 4 (to obtain hour units) and summing these values for each electrode data set.
[2] Soil pH values from main plots (aggregates of 12 cores) were used for adjustments to CEh calculations.

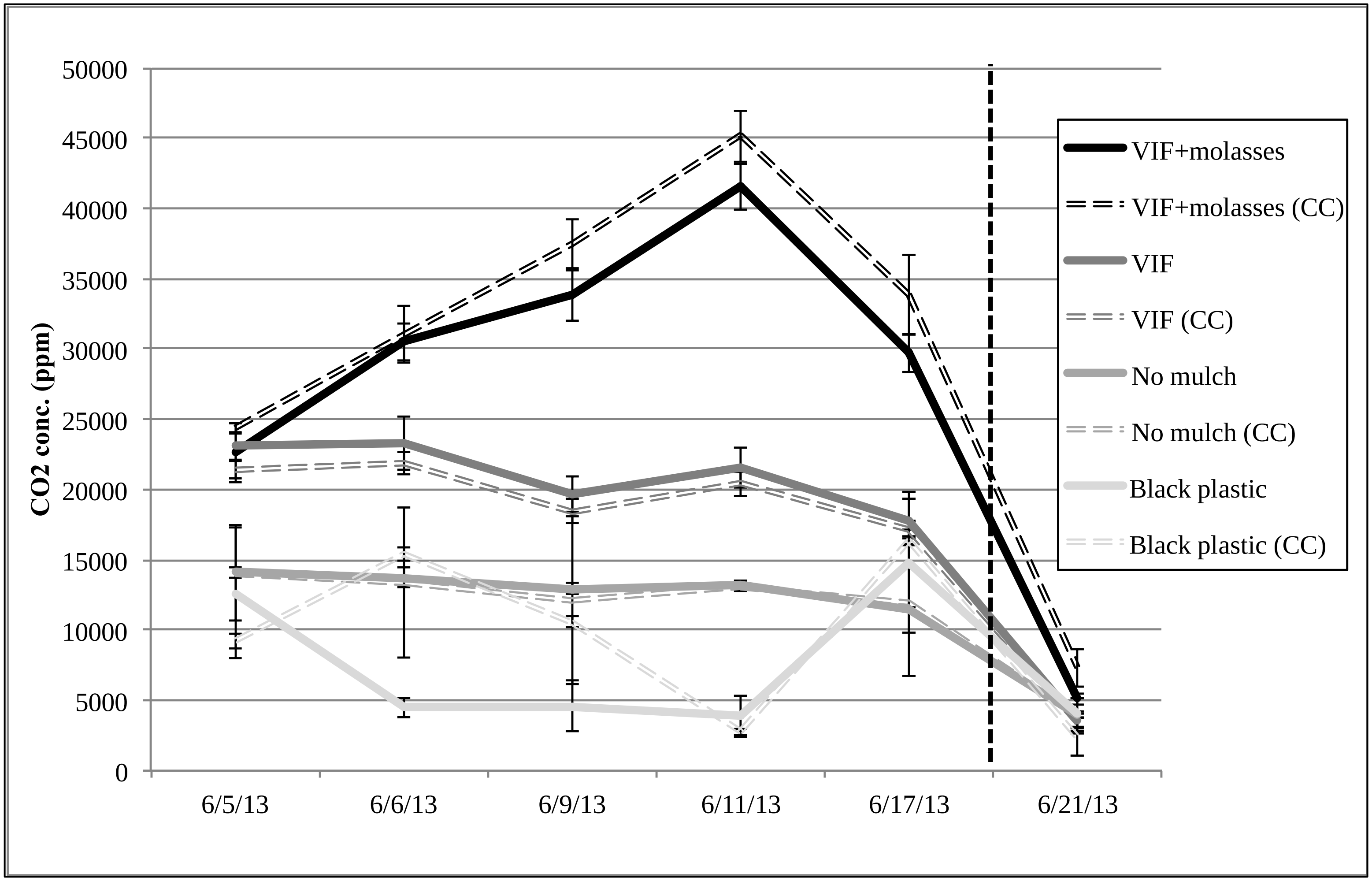


Figure 2.2. Concentrations of CO_2 collected from beds with various mulch and cover crop treatments. Samples were collected immediately following the initiation of ASD (6/5) and sampled intermittently until transplants were set (6/19; indicated by the dashed line. Error bars indicate standard errors from 4 replications of each treatment combination sampled.

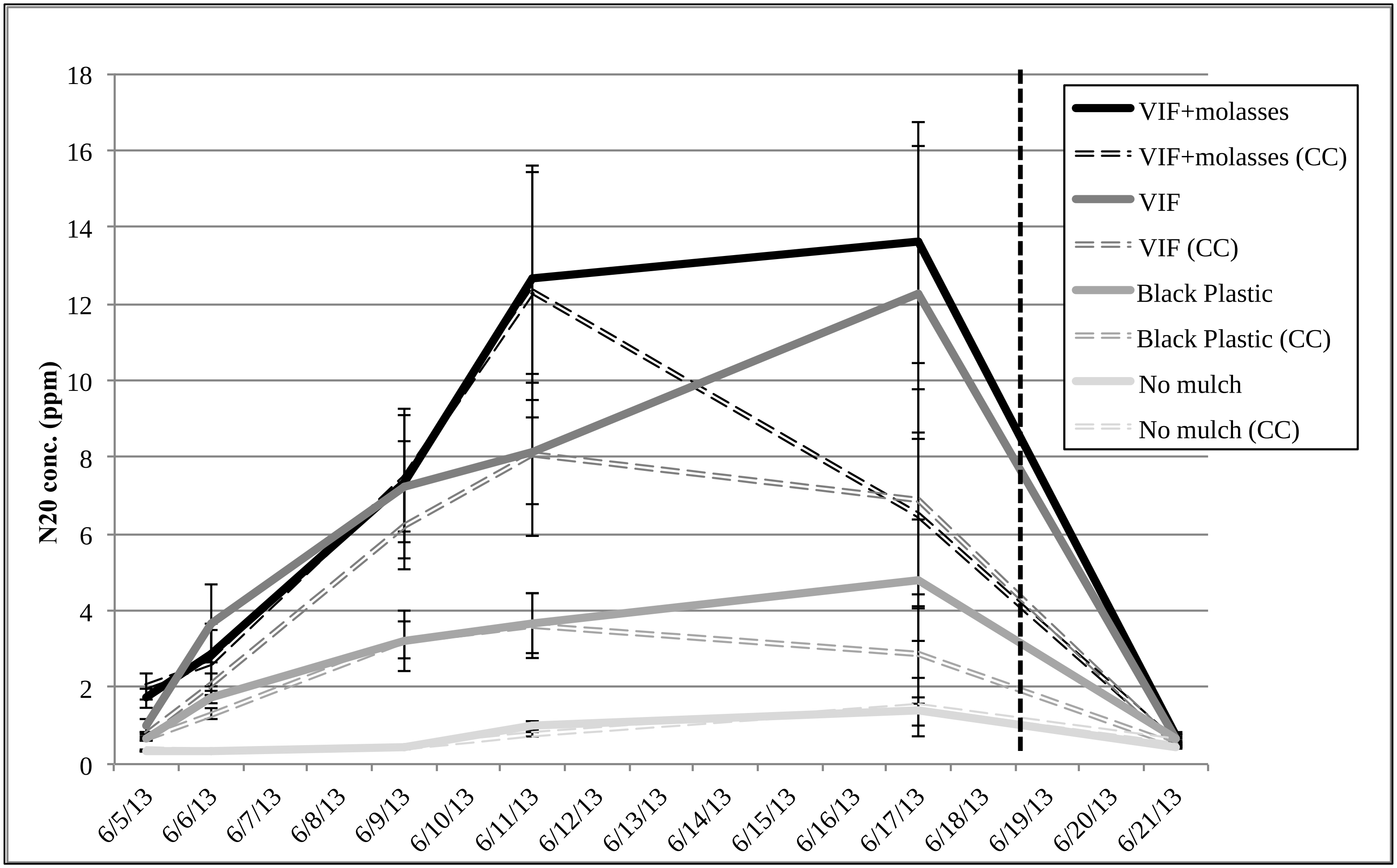


Figure 2.3. Concentrations of N_2O collected from beds with various mulch and cover crop treatments. Samples were collected immediately following the initiation of ASD (6/5) and sampled intermittently until transplants were set (6/19; indicated by the dashed line. Error bars indicate standard errors from 4 replications of each treatment combination sampled.

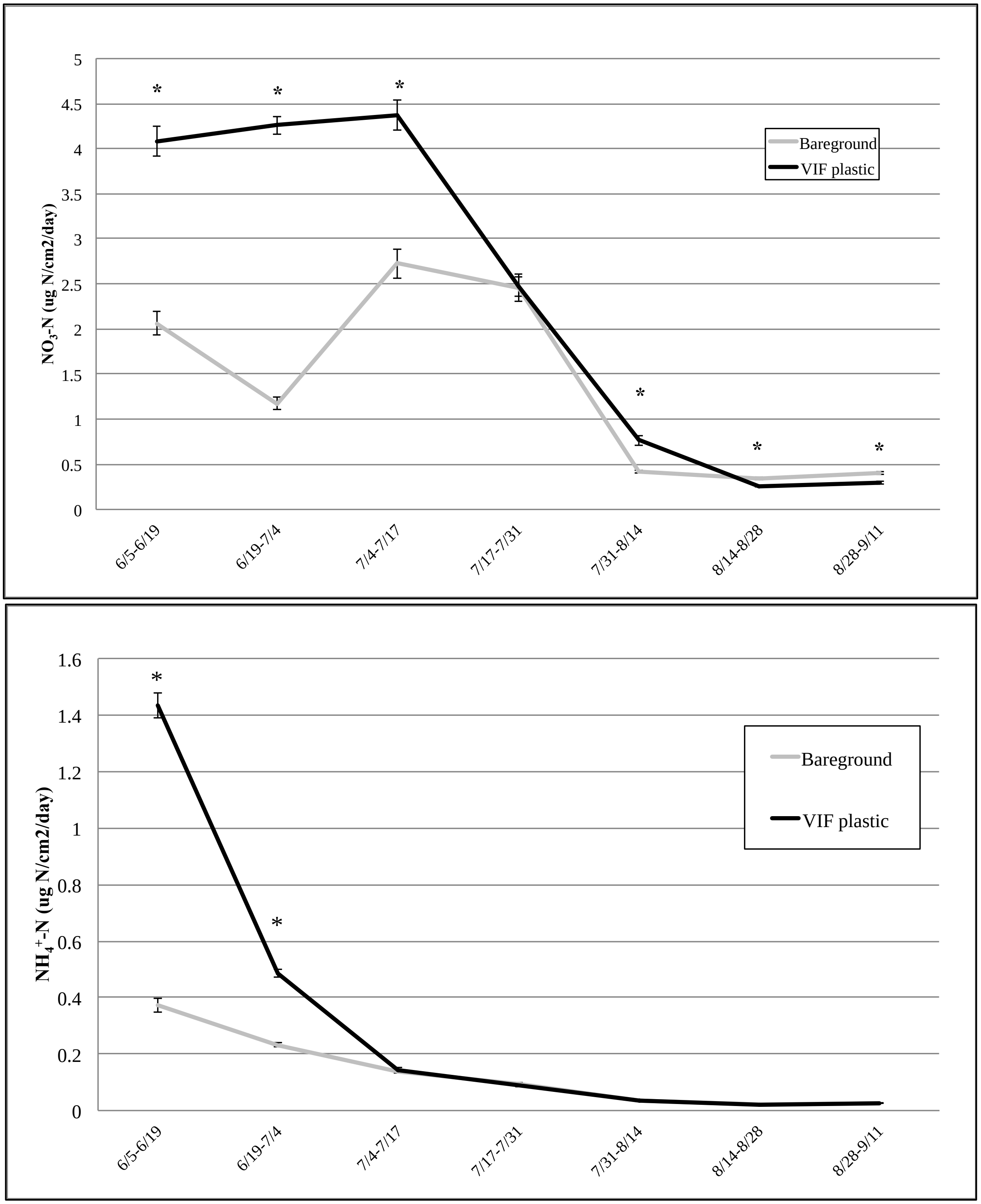


Figure 2.4. NO_3^- (above) and NH_4^+ (below) extracted from ion exchange resin strips in 2012. Main effects of mulch treatment were analyzed for each sampling date after determining lack of significance among cover crop treatments analyzed. Note different scales between NO_3^- and NH_4^+ graphs.
*Indicates significant difference detected (α=0.05)

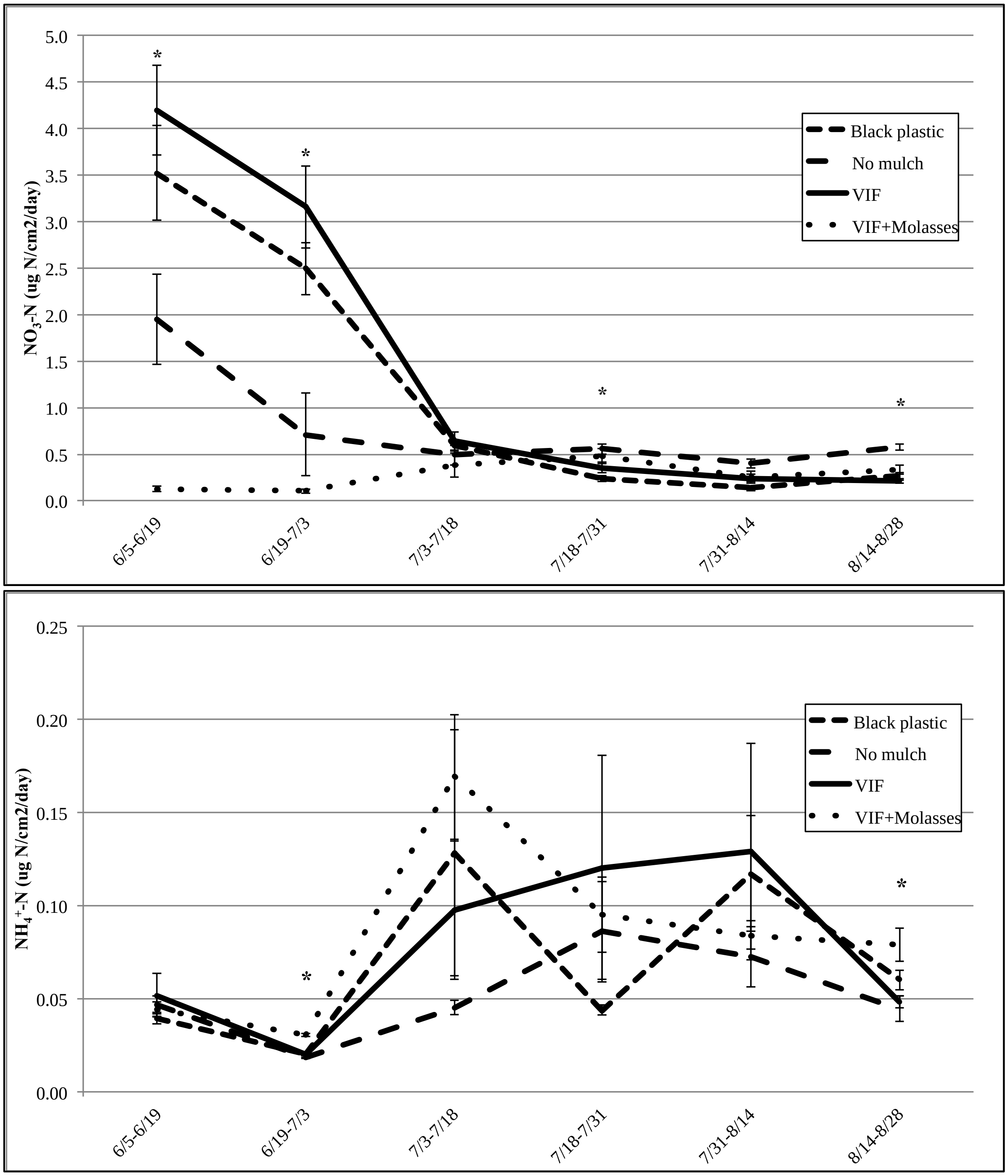


Figure 2.5. NO_3^- (above) and NH_4^+ (below) from ion exchange resin strips in 2013. Main effects of mulch treatment were analyzed for each sampling date after determining lack of significance among cover crop treatments analyzed. Note different scales between NO_3^- and NH_4^+ graphs.
*Indicates significant difference detected (α=0.05)

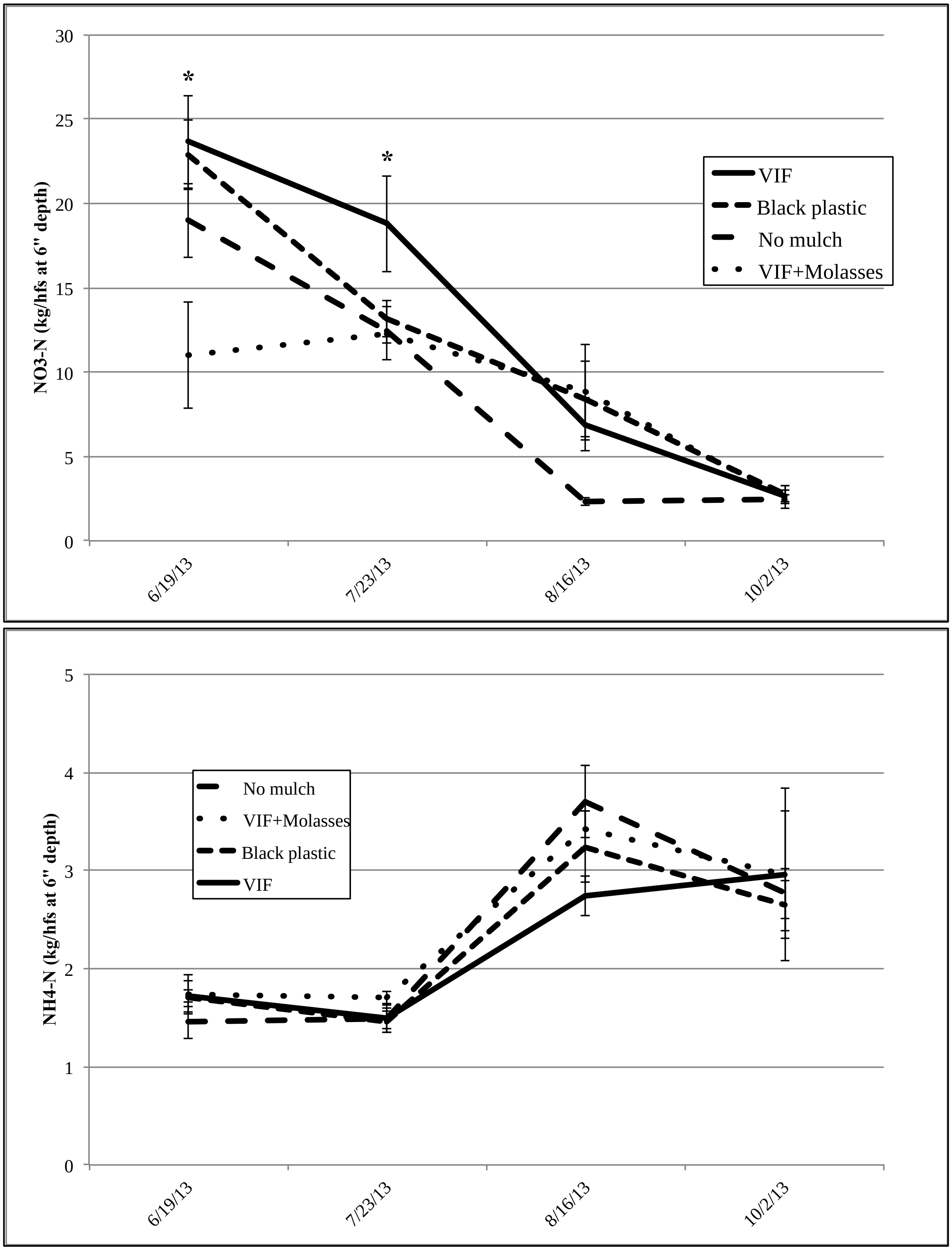
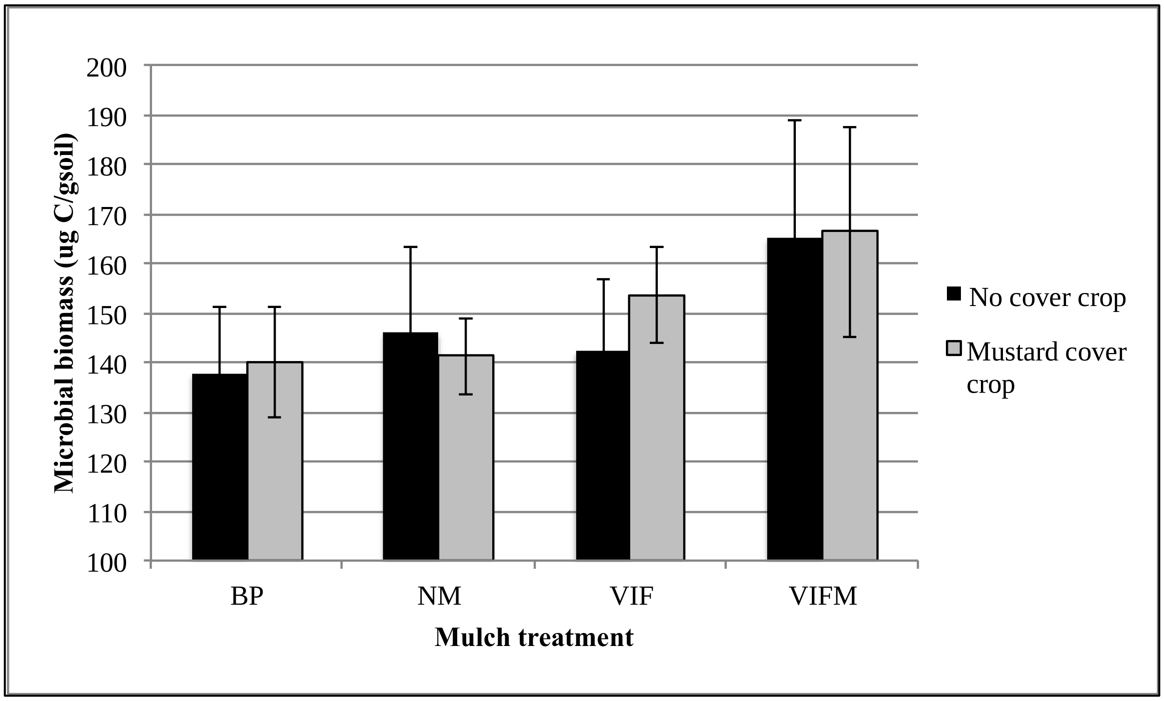


Figure 2.6. Soil NO_3^- (above) and NH_4^+ (below) collected from soil cores during the 2013 growing season. Main effects of mulch treatment were analyzed for each sampling date after determining lack of significance among cover crop treatments analyzed. Note difference in scale between NO_3^- and NH_4^+ graphs.
*Indicates significant difference detected (α=0.05)



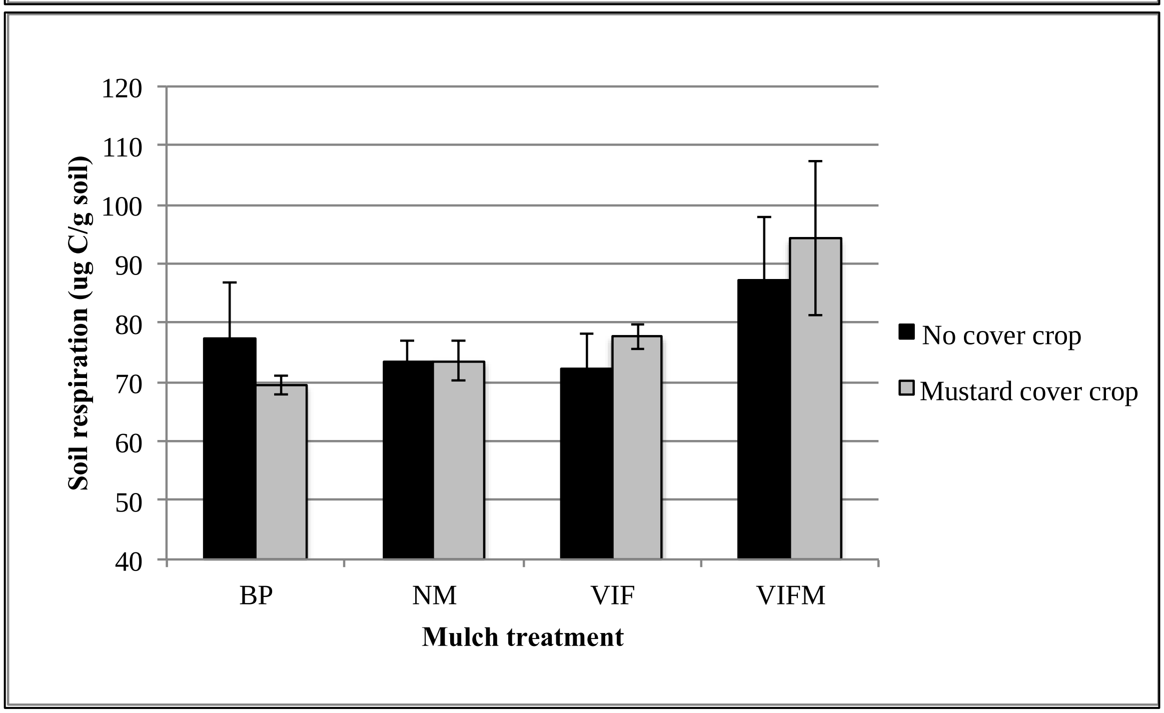


Figure 2.7. Microbial biomass carbon (above) and soil respiration (below) collected from soil samples immediately after ASD treatment. No significant differences were detected among mulching x cover crop treatment combinations (α=0.05) likely due to the high variability among field replicates.

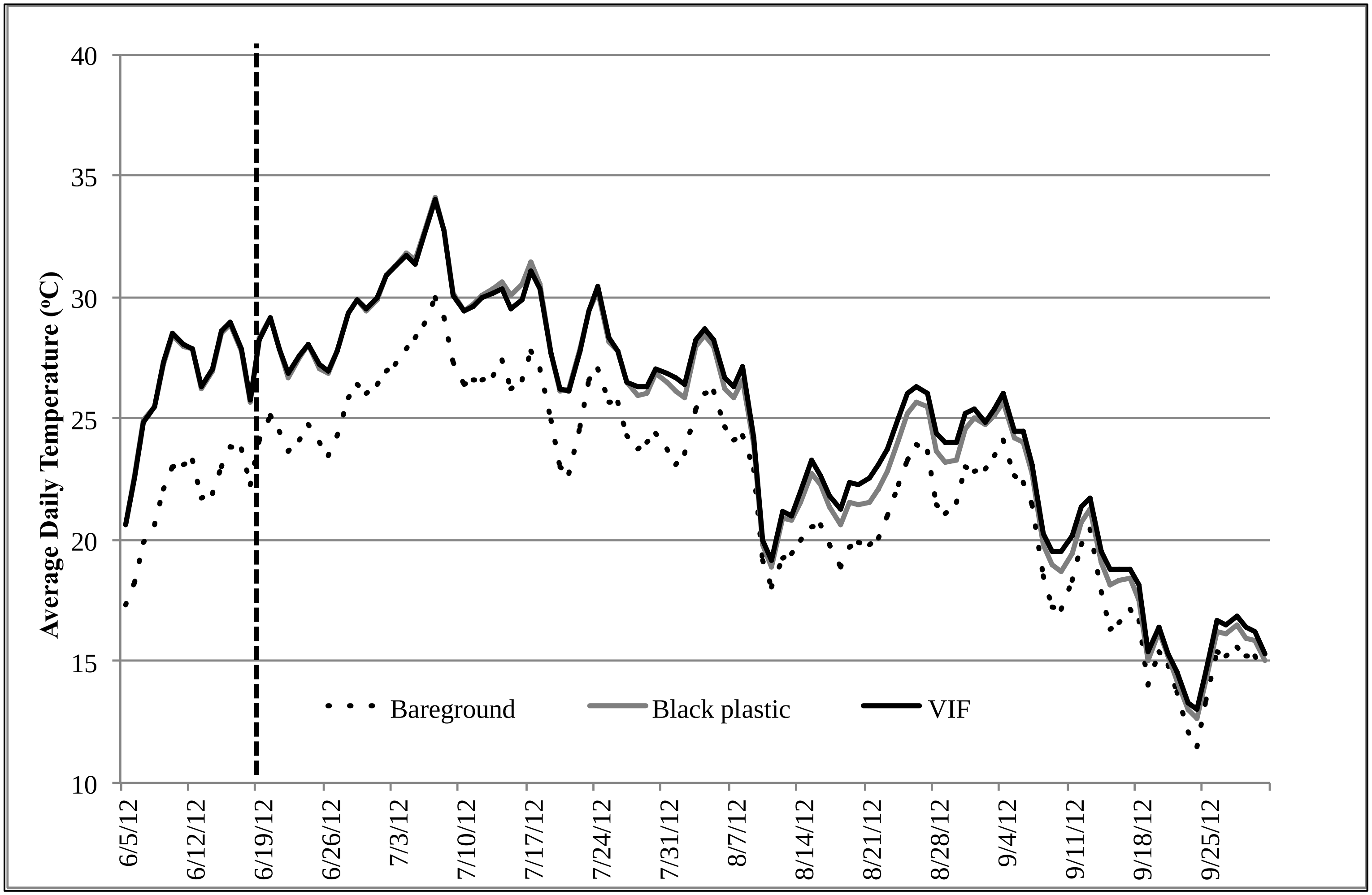


Figure 2.8. Average daily soil temperatures (recorded at 10 cm depth) under various mulch treaments at the HTRC (Holt, MI) during the 2012 growing season. The dotted vertical black line indicates the end of ASD treaments and when transplants were set in the field (June 19th).

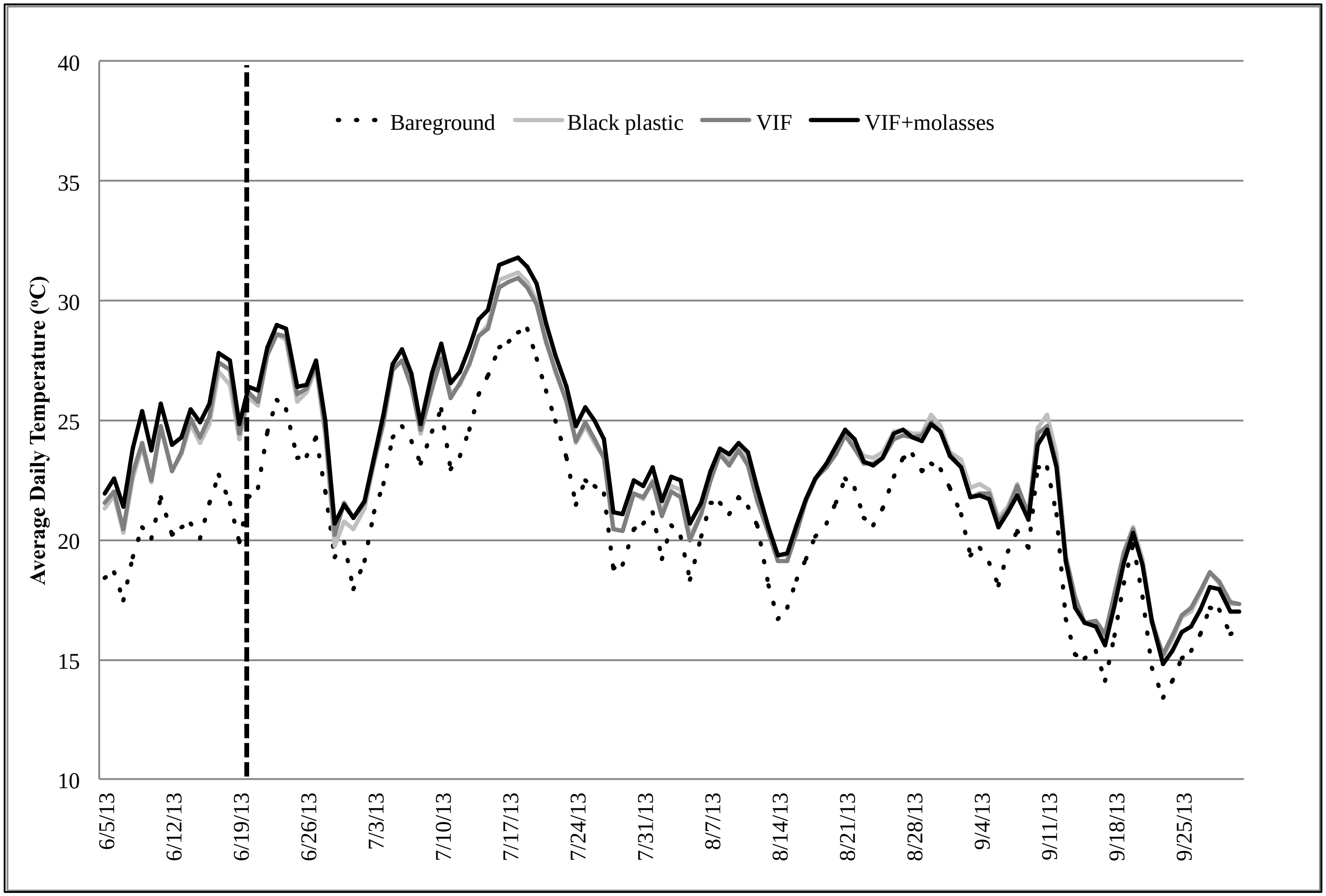


Figure 2.9. Average daily soil temperatures (recorded at 10 cm depth) under various mulch treaments at the HTRC (Holt, MI) during the 2013 growing season. The dotted vertical black line indicates the end of ASD treaments and when transplants were set in the field (June 19th).

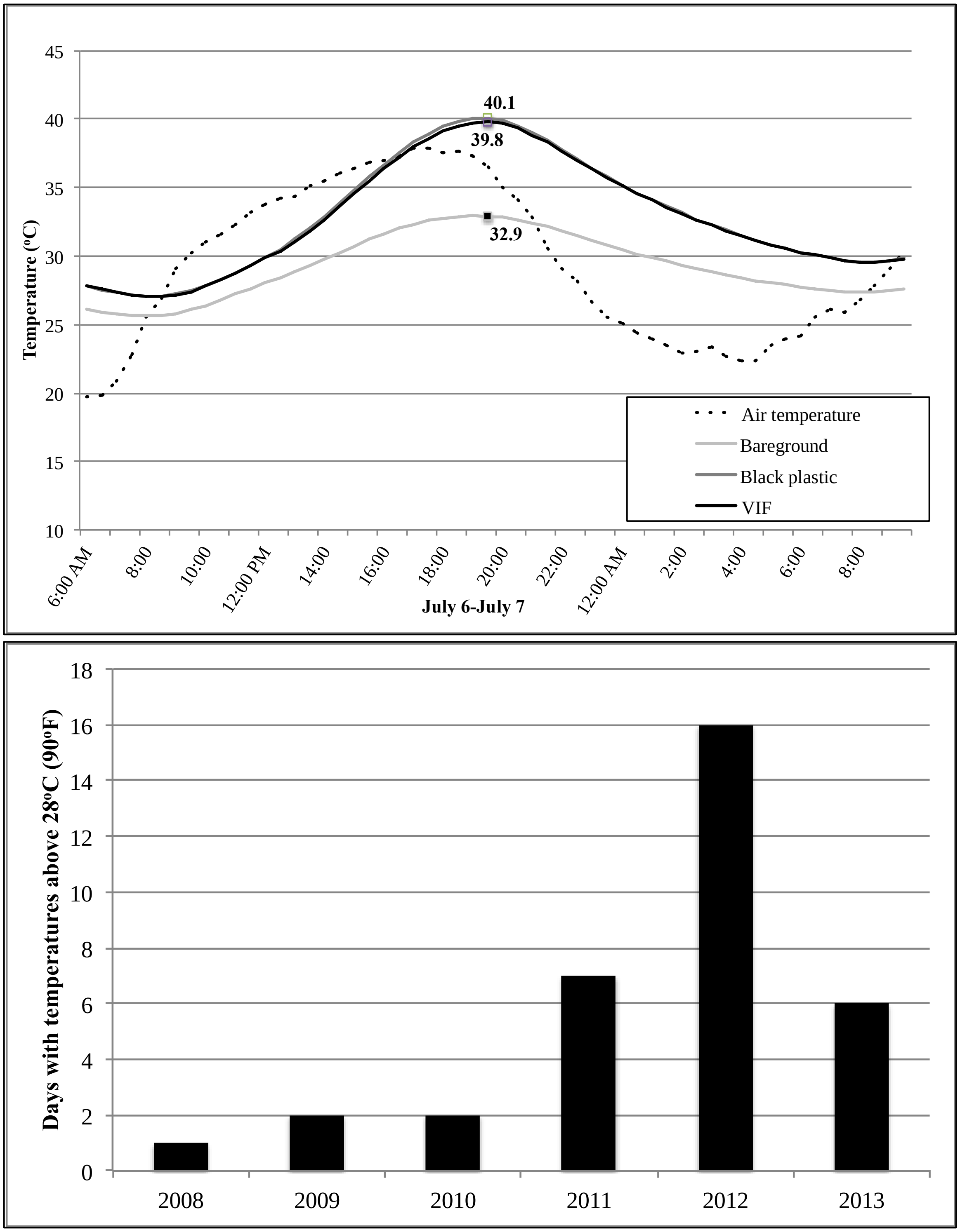


Figure 2.10. Figures demonstrating extreme heat stress during summer of 2012 including a snapshot of diurnal fluctuation of soil and air temperatures in early July at the field site (above) and historical record of the number of days with temperatures exceeding 28^{o}C (90^{o}F) (below) at the weather station located at the HTRC (Holt, MI).

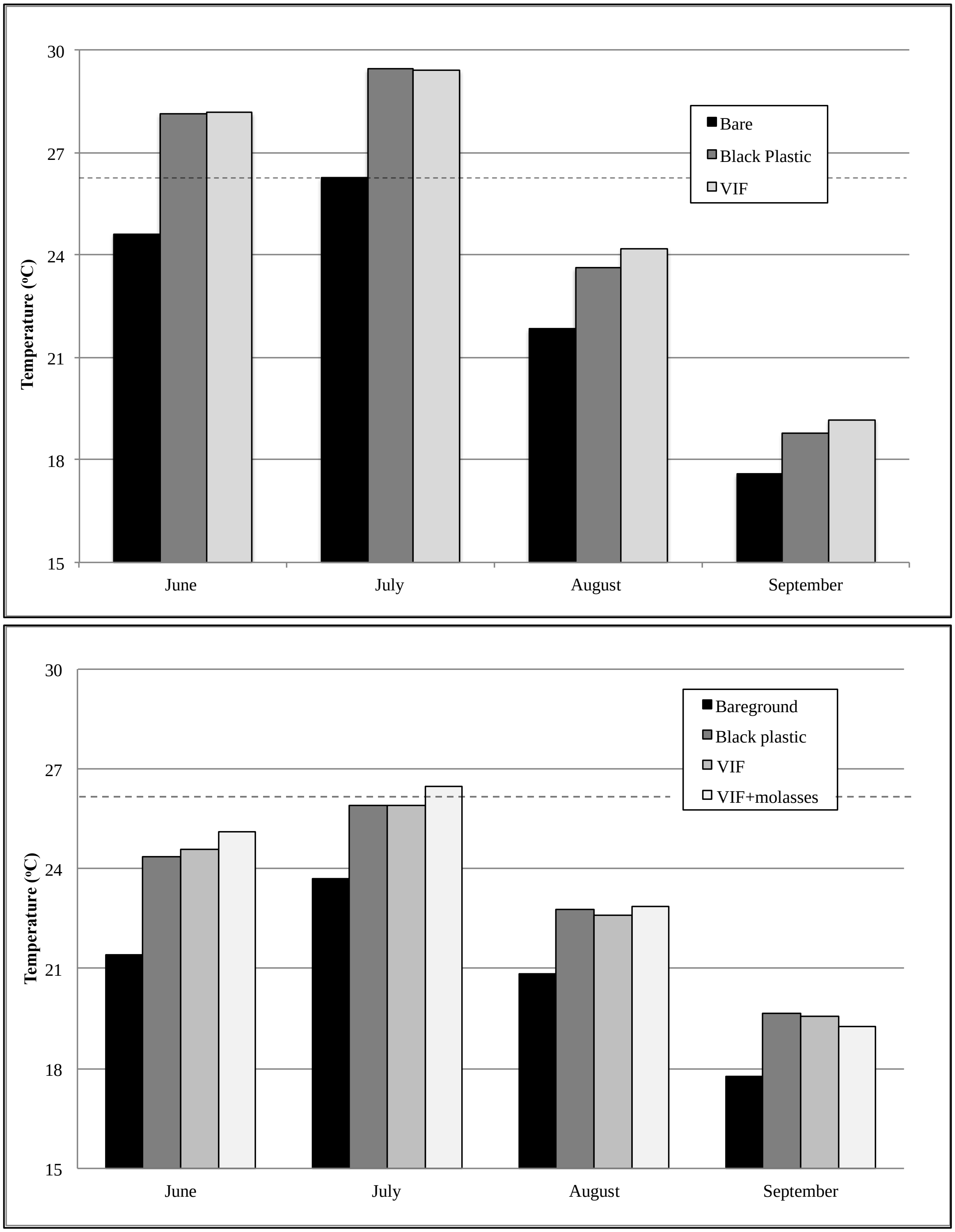


Figure 2.11. Monthly mean root-zone temperatures collected from HOBO™ data loggers buried 10cm under the soil surface. Individual bars represent mean values from 15 (2012) and 8 loggers (2013). Horizontal dotted lines indicate optimal root-zone temperatures for tomato growth (Diaz-Perez, 2002)

Table 2.4. Fresh market tomato yields under various mulch treatments in 2012 and 2013

Mulch Treatment	[1]Early yields (Mg/ha)		[2]Late yields (Mg/ha)		[3]Cumulative yields (Mg/ha)	
	Marketable	Cull	Marketable	Cull	Marketable	Cull
2012						
No Mulch	[4]3.7 **C**	1.6 **B**	41.9 **A**	29.5 **A**	55.5 **A**	31.9 **A**
Black Plastic	6.9 **A**	4.4 **A**	31.5 **B**	18.0 **B**	47.5 **B**	23.7 **C**
VIF	4.3 **B**	3.7 **A**	30.9 **B**	22.7 **C**	45.9 **B**	28.0 **B**
Pr > F	*<0.0001*	*<0.0001*	*<0.0001*	*0.0003*	*0.0014*	*0.0041*
2013						
No Mulch	9.4 **B**	1.6 **B**	51.1 **A**	27.8 **A**	60.5 **A**	29.4 **A**
Black Plastic	21.9 **A**	3.5 **A**	33.1 **B**	15.2 **B**	55.0 **A**	18.8 **B**
VIF	18.8 **A**	2.8 **A**	39.6 **B**	17.6 **B**	58.5 **A**	20.4 **B**
VIF+molasses	10.6 **B**	1.6 **B**	33.5 **B**	18.7 **B**	44.2 **B**	20.3 **B**
Pr > F	*<0.0001*	*0.0002*	*<0.0001*	*<0.0001*	*0.0002*	*<0.0001*

[1]Early yields included the first four harvests in 2012 and the first three harvests in 2013
[2]Late yields included the last four harvests from 2012 and the last three harvests from 2013
[3]Yields were collected weekly for a total of 9 harvests in 2012 (Aug. 13-Oct. 3) and 6 harvests in 2013 (Sept. 4
[4]Means followed by different letters within columns are significantly different (α=0.05)

Table 2.5. Slicing cucumber yields under mulch treatments in 2012 and 2013

Mulch Treatment	Cumulative yields (Mg/ha)	
	Marketable	Cull
2012		
No Mulch	22.3	12.4
VIF	20.5	13.8
Pr > F	*0.1781*	*0.1522*
2013		
No Mulch	[1]5.2 **B**	1.5 **B**
VIF	10.1 **A**	3.3 **A**
Pr > F	*<0.0001*	*<0.0001*

[1]Means followed by different letters within columns are significantly different (α=0.05)

LITERATURE CITED

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Abdul-baki, A., Spence, C. 1992. Black Polyethylene Mulch Doubled Yield of Fresh-market Field Tomatoes. *HortScience*, 27: 787-789.

Abdul-Baki, A.A., Teasdale, J.R., Korcak., R.F. 1997. Nitrogen Requirements of Fresh-market Tomatoes on Hairy Vetch and Black Polyethylene Mulch. *HortScience*, 32: 217-221.

Austerweil, M.,Steiner, B., Gamliel, A. 2006. Permeation of soil fumigants through agricultural plastic films. *Phytoparasitica,* 34: 491-501.

Blok, W.J., Lamers, J.G., Termorshuizen, A.J., Bollen, G.J. 2000. Control of Soilborne Plant Pathogens by Incorporating Fresh Organic Amendments Followed by Tarping. *Phytopathology*, 90: 253-259.

Butler, D.M., Rosskopf, E.N., Kokalis-Burelle, N., Muramoto, J., Shennan, C. 2009. Field evaluation of anaerobic soil disinfestations in a bell pepper-eggplant double crop. *Proc. Annual Int. Res. Conference on Methyl Bromide Alternatives and Emissions Reductions.*

Butler, D.M., Rosskopf, E.N., Kokalis-Burelle, N., Albano, J.P.., Muramoto, J., Shennan, C. 2011. Exploring warm-season cover crops as carbon sources for anaerobic soil disinfestation (ASD). *Plant and Soil*, 355: 149-165.

Butler, D.M., Kokalis-Burelle, N., Muramoto, J., Shennan, C., McCollum G.T., Rosskopf, E.N. 2012. Impact of anaerobic soil disinfestation combined with soil solarization on plant-parasitic nematodes and introduced inoculum of soilborne plant pathogens in raised-bed vegetable production. *Crop protection*, 39: 33-40.

Decoteau, D.R., Kasperbauer, D., Daniels, D., Hunt, P.G. 1989. Plastic mulch color effects on reflected light and tomato plant growth. *Scientia Hort*, 34: 169-175.

Diaz-Perez, J.C., Batal, K.D. 2002. Colored Plastic Film Mulches Affect Tomato Growth and Yield Via Changes in Root-zone Temperature. *J. Amer. Soc. Hort. Sci.*, 127: 127-136.

Firon, N., Shaked, R., Peet, M.M., Pharr, D.M., Zamski, E., Rosenfeld, K., Althan, L., Pressman, E. 2006. Pollen grains of heat tolerant tomato cultivars retain higher carbohydrate concentration under heat stress. *Scientia Horticulturae*, 109: 212-217.

Goud, J.C., Termorshuizen, A.J., Blok, W.J., van Bruggen, A.H.C. 2004. Long-Term Effect of Biological Soil Disinfestation on Verticillium Wilt. *Plant Disease*, 88: 688-694.

Gosselin, A., Trudel, M.J. 1985. Influence of root-zone temperature on growth, development and yield of cucumber plants cv. Toska. *Plant and Soil*, 85: 327-336.

Horwath, W.R., E.A. Paul, D. Harris, J. Norton, L. Jagger, and K.A. Horton. 1996. Defining a realistic control for the chloroform fumigation-incubation method using microscopic counting and ^{14}C-substrates. *Canadian Journal Of Soil Science,* 76:459-467.

Jenkinson, D., Powlson, D.S. 1976. The effects of biocidal treatments on metabolism in soil: V. A method for measuring soil microbial biomass. *Soil Biology and Biochemistry*, 8: 209-213.

Jensen, L.S., McQueen, D.J., Shepherd, T.G. Effects of soil compaction on N-mineralization and microbial-C and –N. I. Field Measurements. *Soil Tillage and Research*, 38: 175-188.

Kirkegaard, J.A., Sarwar, M. 1998. Biofumigation potential of brassicas I. Variation in glucosinolate profiles of diverse-field grown brassicas. *Plant and Soil*, 201: 71-89.

Kirkegaard, J.; D. Walters, ed. 2009. Biofumigation for plant disease control- from the fundamentals to the farming system. Chapter 9, *Disease Control in Crops: Biological and Environmentally Friendly Approaches*. Blackwell Publishing Ltd. Pp. 172-195.

Kogan, M. 1998. Integrated pest management: historical perspectives and contemporary developments." *Annual review of entomology* 43: 243-270.

Lamers, J.G., Runia, W.T., Molendijk, I.P.G., Bleeker, P.O. 2010. Perspectives of Anaerobic Soil Disinfestation. *Acta Hort*., 883: 277-284.

MacDonald, N.W., Zak, D.R., Pregitzer, K.S. 1995. Temperature Effects on Kinetics of Microbial Respiration and Net Nitrogen and Sulfur Mineralization. *Soil Science Society of America Journal*, 59: 233-240.

Mehissa, A.S., van Diepenigen, A.D., Wenneker, M., van Beuningen, A.R., Janse, J.D., Trudie, G., Coenen, C., Termorshuizen, A.J., van Bruggen, A.H.C., Blok, W.J. 2007. Biological Soil Disinfestation (BSD), a new control method for potato brown rot, caused by R*alstonia solanacearum* race 3 biovar 2. *Eur J Plant Pathol*, 117: 403-415.

Momma, N. 2008. Biological soil disinfestation (BSD) of soilborne pathogens and its possible mechanisms. *Japan Agricultural Research Quarterly*, 42: 7-12.

Moore, P.A., Reddy, K.R., Graetz, D.A. 1992 Nutrient Transformations in Sediments as Influenced by Oxygen Supply. *Journal of Environmental Quality,* 21: 387-393.

Ngouajio, M., Ernest, J. Changes in the Physical, Optical, and Thermal Properties of Polyethylene Mulches during Double Cropping. 2005. *HortScience*, 401: 94-97.

Nunez-Zofio, M., Larregla, S., Garbisu, C. 2011. Application of organic amendments followed by soil plastic mulching reduces the incidence of Phytophthora capsici in pepper crops under temperate climate. *Crop Protection*, 30: 1563-1572.

Parr, J.F., Reuszer, H.W. Organic Matter Decomposition as Influenced by Oxygen Level and Method of Application to Soil. *Soil Science Society of America Journal*, 23: 214-216.

Ponnamperuma, F.N. Ed. Kozlowski, T.T. 1984. Chapter II: Effects of Flooding on Soils, in *Flooding and Plant Growth*. Academic Press, Inc. Orlando, Florida. 10-42.
Shennan, C., Muramoto, J., Koike, S.T., Daugovish, O. 2009. Optimizing anaerobic soil disinfestation for non-fumigated strawberry production in California. *Proceedings of the Annual International Research Conference on Methyl Bromide Alternatives and Emissions Reductions*. 101.

Robertson, G.P., Groffman, P.M. Ed. Paul, E.A. 2007. Nitrogen transformation. Soil Microbiology, Biochemistry, and Ecology. Springer, New York, NY. 341-364.

Shennan, C., Muramoto, J., Koike, T., Daugovish, O. 2010. Optimizing Anaerobic Soil Disinfestation for Non-fumigated Strawberry Production in California. *California Strawberry Commission Annual Production Research Report.* 149-161.

Snapp, S.S., Swinton, S.M., Labarta, R., Mutch, D., Black, R., Leep, R., Nyiraneza, J., O'Neil, K. 2005. Evaluating Cover Crops for Benefits, Costs, and Performance within Cropping System Niches. *Agronomy Journal*. 97: 322-332.

Snapp, S. S., Date, K., Cichy, K., O'Neil, K. 2006. Mustards- A Brassica Cover Crop for Michigan. Extension Bulletin E-2956. Michigan State University.

Tarara, J.M. 2000. Microclimate Modification with Plastic Mulch. HortScience, 35: 169-180.

Teasdale, J.R., Abdul-Baki, A.A. 1995. Soil Temperature and Tomato Growth Associated with Black Polyethylene and Hairy Vetch Mulches. *Journal of the American Society for Horticulture Science*, 120: 848-853.

Tindall, J.A., Beverly, R.B., Radcliffe, D.E. 1991. Mulch Effect on Soil Properties and Tomato Growth Using Micro-Irrigation. *Agronomy Journal*, 83: 1028-1034.

Vorenhout, M., van der Geest, H.G., van Marum, D., Wattel., K., Eijsackers, J.P. 2004. Automated and Continuous Redox Potential Measurements in Soil. *Journal of Environmental Quality*, 33: 1562-1567.

United States Department of Agriculture, Fresh Products Branch. 1997. United States Standards for Grades of Cucumbers. 1-7.

United States Department of Agriculture, Fresh Products Branch. 1997. United States Standards for Grades of Fresh Tomatoes. 1-13.

CONCLUSIONS AND FUTURE RESEARCH

Biologically based management practices such as biofumigation and ASD represent novel approaches for managing pests. While our understanding of the mechanisms governing BF and ASD have improved dramatically in the recent past, more research is needed to further optimize these practices if they are to be adopted by growers. Tailoring these practices will require regionally based research, which can adapt these practices to regional climatic, edaphic and pathogenic conditions that exist.

Using delayed seeding to improve crop stand establishment was successful for muskmelon in southwest Michigan. Seeding crops at least ten days following the incorporation of brassica cover crops for biofumigation can reduce detrimental crop stand inhibition that often accompanies these cover crops. While these results are encouraging and can generally improve management of brassica cover crops, recommendations should be made to growers with the understanding that these results might not be appropriate under different circumstances (higher cover crop biomass, soil type, crop type, etc.). Additionally, delaying of seeding can have adverse effects on crop yields. In our study, marketable crop yields began to decline when seeded 15 days after incorporation. While this observation warrants a cautionary approach for many long-season vegetable crops such as muskmelon, shorter season crops might not be as adversely affected by delayed seeding and might make a more appropriate fit for this type of management tactic. This is particularly important in areas where the summer growing season is short and planting windows are narrow.

While biologically based pest management practices are logically focused on disease control, the utilization of cover crop residues, carbon amendments and plastic mulch can have important impacts on nutrient availability and crop yields. While anaerobic soil disinfestation has been shown to be an effective method for controlling soil-borne diseases, these methods have not been established for vegetable production in northern areas of the U.S. Due to the narrow growing season in Michigan, methods that seek to create rapidly reducing conditions in the soil would be beneficial to avoid lengthy periods of 'idle' field space. Yellow mustard residues did not prove to be effective at achieving sufficiently reduced conditions required for successful ASD. Due to the lack of reduction in nearly all of the plots measured (including molasses controls) more research is needed to appropriately establish anaerobic conditions in our region (irrigation methods/levels, other carbon sources, timing, etc.). Using winter-hardy, fall-seeded cover crops that regrow in the spring might be better suited as a carbon source than the spring-seeded brassicas due to their ability to accumulate large quantities of biomass and ease of establishment.

Modifying nitrogen fertilization might be necessary following the incorporation of a highly carbonaceous material for ASD. We observed substantial declines in nitrogen availability following molasses application with visible and quantifiable decreases in plant growth and crop yields. Using black plastic mulches can increase nitrogen availability for crops following ASD, particularly early in the growing season although these differences are diminished later. Understanding how plastic mulches (different colors & permeabilities) impact nitrogen dynamics could improve nutrient management, particularly in systems that utilize frequent additions of residues. Despite the higher early season nitrogen availability, total marketable tomato yields were significantly lower in plastic mulched beds than under no mulch treatments. High summer temperatures led to extreme high soil temperatures under mulch and are believed to have been responsible for this decline in crop yields. Improving our understanding of root-zone temperature effects on crop performance and their manipulation through mulching practices would be of practical significance for growers, particularly in northern production areas where use of black plastic is a standard practice and warmer summer temperatures are expected due to a changing climate.