

Ceres Trust Graduate Student Grant – Final Report

Project Title: Effect of Cover and Green Manure Crops on Soil Health, Plant Health and Tuber Yield in Organic Sweet Potato Production

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Abstract

Interest in organic sweet potato production in the United States has been growing as consumers and producers are becoming increasingly concerned about how their food is cultivated. Thus, there is a growing need for information on sweet potato production under an organic production system. Organic sweet potato growers are challenged by sustainable soil management, which impacts the occurrence of soilborne disease, weeds, and other pest issues that consequently reduce the tuber yield. Being organic, their options are limited for pest control, especially fungal disease that affects the roots and/or tubers reducing their marketable quality. The research aimed to develop a strategy of using cover and/or green manure crops, and to evaluate their effects on soil and plant health, as well as disease and weed suppression. The experiment was conducted at a Certified Organic Producer's field based in central Missouri. Effects of rapeseed and cereal rye, incorporated as cover or green manure crops, were assessed in a randomized complete block design for a one-year period. Soil samples were collected for soil organic matter, pH, nutrients, and microbial analysis by using phospholipid fatty acid (PLFA) analysis three times over the course of the study. The PLFA assessed various groups of fungi and bacteria for their diversity and density. Evaluations also included weed coverage, sweet potato plant vigor, disease occurrence (*Fusarium* root rot), and sweet potato yield. Results from this experiment would increase our knowledge on how to best incorporate cover crops into organic sweet potato production by improving soil health as well as suppressing weed and disease.

Introduction

Organic farming is becoming more popular in the United States and around the world. Sweet potato, a tropical root crop that ranks 7th in terms of importance worldwide (Horton, 1987), is the fifth most valuable commodity in total sales of all organic vegetables grown in the open field. The demand for organic food production has increased in average of 24% annually in the US during the 1990s (Govindasamy and Italia, 1998). Food and environmental safety are often cited the main reason for the use of organic produce, but increasingly, economic considerations are becoming important with a rise in popularity of organically produced foods (Govindasamy and Italia, 1998; Thompson, 1998). It is estimated that a premium of 12–60% is often obtained from organic produce (Lohr, 1998), which in turn, attracts more farmers to organic production (Thompson, 1998). The U.S. Department of Agriculture ERS reports that organic foods are sold in 73% of all traditional food markets (Dimitri and Greene, 2003), and in 2005, total organic food sales were estimated at \$13.8 billion in U.S. (Organic Trade Association, OTA, 2006). Approximately 39% of those sales were fruits and vegetables, and market penetration of organic food is estimated at 2.48% of total U.S. food sales (OTA, 2006).

Despite the increased interest in organically produced sweet potato in recent years, organic sweet potato production is well below the demand due to various hurdles. Weed infestation is often cited the number one challenge for successful organic farming (Beveridge and Naylor, 1999). For sweet potato production, weed encroachment is especially an issue because the sweet potato's feeder roots are typically near the soil surface, and above-ground growth is viny in nature. These characteristics prohibit late cultivation, which leaves farmers very few options for weed control except hand removal (Treadwell et al., 2007). Without hand weeding, weed competition can lead to dramatic yield reduction up to 80% (Jackson and Harrison, Jr., 2008).

In addition to weed, sweet potatoes are also affected by soilborne diseases, especially Fusarium root and stem rot caused by soilborne fungus *Fusarium solani* (Mart.) Sacc. f. sp. *batatas* McClure. This disease has a root rot phase (also referred to as Fusarium root and stem canker, end rot, or Fusarium end rot) and a stem canker phase, and can be transmitted from infected slips, and develop on roots with wound sites, or affect the crop at any stage of production (Clark and Moyer, 1988). Without treatment, farmers could potentially face a yield reduction of 10-50% or in some cases, a 100% yield loss (Harter and Field, 1914).

Cover crops have become a viable option for sustainable agriculture because of their contribution to soil fertility and improved crop performance. Cover crops are normally grown during the off-season with an annual cash crop. When they are incorporated into the soil prior to planting the cash crop, they become a 'green manure'. Cover or green manure crops may or may not have any harvestable yield value. However, they are effective in reducing soil erosion (Creamer et al., 1997; Wall et al., 1991), increasing soil organic matter (Allison, 1973), improving soil physical and chemical characteristics (Reid and Goss, 1981), increasing microbial activity (Cook and Baker, 1983), suppressing weed populations (Shilling et al. 1985) and reducing soilborne plant diseases (Abawi and Crosier, 1992; Baker and Cook, 1974; Viaene and Abawi, 1998).

The effects of cover and green manure crops on soil health also relate to their impact on increasing soil microbial communities, which directly or indirectly reduces weeds and soilborne diseases (Abawi and Widmer, 2000; Bailey and Lazarovits, 2003; Kremer and Li, 2003). Different mechanisms have been proposed to explain the suppressive capacity including enhanced activities of antagonistic microbes (Hoitink and Boehm, 1999), release of fungicidal/herbicidal compounds during organic matter decomposition (Smolinska, 2000), or induction of systemic resistance in the host plants (Pharand et al., 2002). It is known that plants and plant products (organic amendments, crop residues, green manures) can dramatically affect soil microbial communities, and are primary drivers of soil microbial dynamics (Larkin, 2003) by increasing soil bacterial density (van Bruggen and Semenov, 2000), specifically beneficial bacteria such as fluorescent *Pseudomonas* spp. (Abawi and Widmer, 2000), and thus may also be important components in establishing and maintaining soil suppressiveness of plant pathogens (Larkin and Honeycutt, 2006) and weeds (Kremer and Li, 2003).

A few studies conducted on the use of cover crops in organic sweet potato production systems showed improved plant performance when cover crops were used (Treadwell et al., 2003; Jett and Talbot, 1998). No studies have been conducted on the use of cover crops relating to soil health and plant health, hence the study which was conducted with the goal of evaluating the effects of cover/green manure crops on soil health, soil microbial activity and sweet potato yield in organic sweet potato production.

Materials and Methods

Experimental Site

Field plots were established at the Share-Life Farms (Longitude: -93.2225, Latitude: 39.1341) located in Central Missouri, 6.5 miles north of I-70 in Saline County. Share-Life farms is a central Missouri Community-Supported Agriculture (CSA) farm that has been Certified Organic continuously since 2004. The soils are silt loam, topography is upland loess and the elevation is 764 ft (233 m).

Experimental Design

Treatments were arranged in a split-plot with the whole plot variable being the use of cover crop species as cover or green manure, in addition to control. The sub-plot variable was the cover crop species cereal rye (*Secale cereale* L.) or rapeseed (*Brassica napus* L.). Individual whole-plot measured 6 m × 6 m, subplot measured 3 m × 6 m, and the total plot area measured 18 m x 34 m. Whole plot variables were arranged in a randomized complete block design with 3 replications, with a 7.6 m-wide alleyway between blocks to provide room for equipment.

Plant Material and Planting

Organic rapeseed and cereal rye seeds were obtained from Johnny's selected seeds (955 Benton Ave, Winslow, ME 04901) and GrowOrganic (Peaceful Valley Farm & Garden Supply, 125 Cydesale Court, Grass Valley, CA 95945), respectively. The sweet potato slips "Beauregard" were purchased from Southern Exposure seed exchange (P.O Box 460, Mineral, VA 23117). The rapeseed and cereal rye were established on the 3rd of October 2016 at rates of 7 lbs and 100 lbs/acre respectively, using a mechanical hand planter (Figure 1). The sweet potato slips were planted on the 22nd of June 2017 using a planter (Figure 2), at an intra and inter row spacing of 30 cm and 150 cm, respectively.

Termination of cover crops

The cereal rye and rapeseed were first terminated on the 24th April, 2017 using a mower (Figure 3) followed by a second time on the 6th of June, 2017 using a roller crimper (Figure 4), due to the regrowth of the cover crops. A tiller (Figure 5) was used to incorporate the cover crops into the soil for plots that were designated to be green manure plots, on both the 24th of April and 6th June following termination.

Measurements

Data were collected for visual assessments of cover crop plant vigor and coverage once monthly in February, March and April 2017. Plant vigor was estimated based on a 1-5 scale where 5 were highest, 3 and 1 were average or lowest, respectively. The coverage was estimated as the percent area of the sub plot covered by the cover crops. The percent weed cover was estimated once monthly from February to October, using a 61 cm x 61 cm frame (Figure 6) placed 3 times randomly in each subplot.

Soil samples were collected three times: after planting cover crops (10th October, 2016), after terminating cover crops but prior to planting sweet potato (22nd June, 2017), and before sweet potato harvesting (6th November, 2017). From each sub-plot, ten random soils cores were collected by a steel probe with diameter of 10 centimeters to a depth of 15 centimeters. The cores were separated from rocks by passing through a 2 mm sieve. Soil samples from each sub plot were then mixed thoroughly, and stored at room temperature for further analysis.

Soil samples were analyzed for Cation Exchange Capacity (CEC), Neutralizable Acidity (NA), Organic Matter (OM), pH, and plant available nutrients including P, K, Fe, Ca, S, Mn, and NH₄-N and NO₃-N. Soil microbial community structure and biomass were determined by analyzing soil phospholipid fatty acid (PLFA) following the procedure described by Bligh and Dyer (1959), and later modified by Petersen and Klug (1994). Briefly, soil samples between 2 and 2.5 g were added to Teflon-lined screw cap cultures tubes and freeze-dried before extracting the total lipids. The total lipid extract was then fractionated into glycol-, neutral, and polar lipid fractions, and the polar lipid fraction was trans-esterified with mild alkali to recover PLFA as methyl esters (Ibekwe *et al.* 2002). A gas chromatograph equipped with flame ionizer detector was used to separate, quantify and identify the methyl esters against an internal standard. The peaks of the samples were then compared with the database of known microbial fingerprints to determine the molar responses of taxonomic microbial groups of bacteria, fungi, gram-positive bacteria, gram-negative bacteria, actinomycetes, and mycorrhizae, and the ratios of fungi to bacteria (F/B) ratio, cyclopropyl 17 to monoenoic precursor (Cy17/pre) ratio, and saturated to monounsaturated fatty acid (Sat/mono) ratio. Total PLFA concentration (p mole g⁻¹ soil) was also determined as the total microbial biomass.

During growth, the sweet potatoes were visually assessed for plant vigor and insect damage on a monthly basis from the time of transplanting in June to right before harvesting in October 2017. The sweet potatoes were harvested over several days from 6th of November to 13th November, using a potato digger (Figure 7). A week after harvest, the numbers of tubers from each sub plot were counted and graded based on the USDA market grade standards, and marketable (at least 3.81 cm in diameter and free of blemish, spot and disease – Figure 8) and culls (roots of any size that exhibited wounds, breakage, or severe cracking that would reduce storage life – Figure 9) (USDA, 1981) were separated. The tubers were also assessed and recorded for disease symptoms.

Statistics

All data collected were subjected to ANOVA using SAS (9.4), and significant means were separated by Fisher's Protected LSD at $P < 0.05$.

Results and Discussion

Cover crops

The cover crops generally performed well, with cereal rye showing greater initial plant vigor than rapeseed. The cereal rye maintained good plant vigor, while the rapeseed improved over time with the highest plant vigor before termination (Figure 10). The plant cover on the other hand showed no significant differences between treatments, except for the rapeseed used as cover crop which was lower during the evaluation in March (Figure 11). When it was time for termination, both cereal rye and rapeseed were flowering and covered the entire plots. Figures 12 through 15 illustrate this.

Weeds

There were a number of weed species established in the plot area during the course of the study, including grasses like yellow foxtail, and broadleaf weeds like Amaranth, clover, dandelions, morning glory, henbit, purslane, Queen arms lace, crownvetch and milkweed. The weeds were significantly suppressed when the cover crops were still in the field from October, 2016 to 24th April, 2017 and no significant differences were observed between cover crop species used (Figure 16). There is wide agreement that a vigorous living cover crop will suppress weeds growing at the same time as the cover crop (Akobundu et al., 2000; Blackshaw et al. 2001; Brennan and Smith, 2005; Creamer and Baldwin, 2000; Favero et al., 2001; Grimmer and Masiunas, 2004; Peachey et al., 2004; Stivers-Young, 1998). It does so by absorbing red light and thereby reduce the red: far red ratio sufficiently to inhibit phytochrome mediated seed germination, whereas cover crop residue has a minimal effect on this ratio (Teasdale and Daughtry, 1993). Furthermore, a living cover crop competes with emerging and growing weeds for essential resources and inhibits emergence and growth more than cover crop residue does (Reddy and Koger, 2004; Teasdale and Daughtry, 1993).

The cover crop plots showed significant increase of weed cover in May, 2017 (Figure 16) since these plots were not tilled. In comparison, minimal weed cover was found in green manure plots and control plots since they were tilled first on 24th April and a second time on 6th June. Once the cover crop was terminated, the weeds increased significantly later in June through October, with the highest coverage found in the control plots (Fig 17). Dead cover crop residue does not suppress weeds as consistently as live cover crops do (Reddy and Koger, 2004; Teasdale and Daughtry, 1993). Furthermore, the magnitude of weed suppression by residue is usually higher for weed emergence measured early in the season than for weed density or biomass measured later in the season. Once seedlings become established, cover crop residue usually has a negligible impact on

weed growth and seed production or may even stimulate these processes through conservation of soil moisture and release of nutrients (Haramoto and Gallandt, 2005; Teasdale and Daughtry, 1993).

Regardless of species, plots with cover crops tilled into the soil as green manure showed significantly less weed cover from July to October (Figure 17). There were no differences in weed suppression between cover crop species for the most part except for June, just after termination of the cover crops. Figure 18 to 20 illustrate the weeds in all plots in August 2017, one and half months after the sweet potato slips were transplanted and established.

Soil microbes (Phospholipid Fatty Acid Analysis)

The total microbial biomass was significantly influenced by sampling date, with the highest numbers being observed at the 1st sampling on 10th October 2016 (Figure 21). The numbers decreased significantly during the second sampling on 22nd June, 2017, possibly due to the tillage done in preparation for planting of sweet potatoes. It has been shown that tillage reduces soil macroaggregate content which provides an important microhabitat for microbial density, diversity, and activity (Ranjard and Richaume 2001; Six *et al.*, 2002). The last sampling carried out on 6th November was performed on undisturbed soil right before harvesting; this may explain the 9% increase of microbial biomass since the second sampling (Figure 21). There was however, no significant effect of the treatments on the total microbial biomass, which was different from early reports where increase in microbial biomass was observed following utilization of cover crops (Finney *et al.*, 2017; Mendes *et al.*, 1999; Wang *et al.*, 2007). Bossio and Scow (1998) determined that seasonal effects on microbial community composition in tomato fields were greater in magnitude than factors associated with management practices such as manure, cover crop, and mineral fertilizer inputs. It is likely that several seasons of cropping are necessary before such a change can be found.

Sampling date also significantly affected the fungi, mycorrhizae, gram positive, gram negative, anaerobes and eukaryotes groups (Figure 22). The fungi and gram positive bacteria decreased overtime, while the AM Fungi and gram negative increased overtime. The fungal and bacterial densities may have been influenced by tillage which has been demonstrated to affect soil temperature and humidity, thus influencing strongly soil microbial abundance and in particular fungal development (Frey *et al.*, 1999; Spedding *et al.*, 2004). Additionally, tillage causes direct tissue damage to the fungi leading to a reduction in their abundance at soil surface (Balesdent *et al.*, 2000; Six *et al.*, 2002). No till systems have been demonstrated to have a positive impact on soil physical and chemical properties (Six *et al.*, 2002), and on soil microbial biomass and activity (Sapkota *et al.*, 2011).

Community change and stress indicator ratios to depict shifts in populations and stress respectively, were determined and significant sampling timing effect were observed (Figure 23). Stress indicator ratios including the saturated to monounsaturated fatty acids (Sat/mono) and

cyclopropyl 17/precursor (Cycl/pre) were developed to detect soil microbial shifts in response to unfavorable conditions, such as extreme temperatures and pH, suboptimal substrates and water and toxin accumulations (Bank *et al.*, 2014). The stress factors may be enhanced or weakened by other factors such as soil tillage or incorporation of organic amendments (Van Bruggen, & Semenov, 2000). The ratio of saturated to monounsaturated fatty acids decreased over time while the cyclopropyl 17/precursor increased at the second sampling date and then decreased. The cyclopropyl 17/precursor also showed significant differences among treatments for the sampling on 10th October 2016 (Figure 24). It appeared that rapeseed used as cover crop showed reduced cyclopropyl 17/precursor ratio than other treated plots. This effect, however, was only observed in one sampling date shortly after establishment of cover crops on the 3rd of October 2016. Collectively, these stress indicators indicate an overall decline in stress conditions for the bacteria overtime, thus indirectly suppress fungal activities.

The ratio of gram positive to gram negative bacteria showed a decrease over time (Figure 23), indicating a shift to a dominance of gram negative bacteria. This increase in gram-negative bacteria with concurrent decreases in gram-positive bacteria may indicate an increase in labile carbon availability throughout the course of the experiment. In various studies, gram-negative bacteria have been considered indicative of higher substrate availability (Bossio and Scow 1998; Fierer *et al.*, 2003). The gram positive versus gram negative bacteria likely reflects the dominant metabolic strategy used by each of these bio marker groups. Specifically, gram positive bacteria tend to utilize more recalcitrant carbon sources, while gram negative bacteria tend to exploit more labile carbon sources (Fierer *et al.*, 2007).

Soil chemical property analysis

Soil chemical property analysis was performed to determine the pH, Cation Exchange Capacity (CEC), Neutralizable Acidity (NA), nitrate, ammonium, phosphorus, potassium, magnesium, calcium, sulphur, manganese, copper and iron. The calcium, magnesium, copper, manganese and iron had significant increments over time (Figure 25). The CEC was also significantly influenced by the sampling date, with a significant increase over time (Figure 26). The rest of the measured soil parameters were not significantly affected by either the sampling date or treatments used. Collectively, these results indicated that cover crops, regardless of species or method of termination, did not significantly affect soil chemical properties compared to control.

Sweet potato yield

After transplanting, the sweet potato slips showed adequate growth in the plot area, with significantly higher plant vigor in the green manure plots regardless of crop species (Figure 27). The cover crop plots showed reduced plant vigor, likely due to the weed presence since these plots were not tilled prior to planting. The plant vigor was correlated to the sweet potato yield, in that the green manure plots and the control plots, both received tillage prior to planting, produced marketable tubers, while the cover crop plots had no yield at all (Figure 28). Compared to the control plots, plots that received green manure, regardless of species, produced 48% or more total

tubers. Plots that received cereal rye as green manure resulted in 2.4× of marketable yield compared the control. The weight of the marketable tubers was positively correlated to the marketable yield, with the green manure plots having higher yield compared to control which had lower marketable yield. At harvest, all tubers were evaluated for possible disease or mice damages. The damages caused by disease or mice all followed the same trend of tuber yield which was influenced by treatment, with higher damage in the green manure plots.

Outreach

Some of the results from the study were shared at a field day that took place at Share Life Farms on August 16 2017. Invited speakers from the University of Missouri and Lincoln University, included Dr. Robert Kremer, Dr. Jaime Pinero, Dr. Reid Smeda, Mr. Tim Reinbott and Ms. Kathi Mecham. Topics included soil health, insect control and pollinator plants, weed control and fertility in organic production systems. Despite heavy rainfall, the half day event attracted more than 40 people representing farmers, industry and government representatives, community gardeners, and students and extension specialists from 17 Missouri towns (Figure 29). The majority of participants expressed satisfaction in attending the field day, and 96% of them stated their willingness to change their current practices based on what they learned. Brochures were prepared to share with field day attendees, addressing issues related to the research as well as other aspects of organic production. Lastly, an oral presentation of some of this work was presented at the Agronomy Society of America-Crop Science Society of America-Soil Science Society of America annual meeting in Tampa Florida during a Soil Health Symposium, with about 30 in attendance. The results will also make a part of my graduate thesis.

Figures



Figure 1. Mr. Jim Thomas illustrating how to use the mechanical planter



Figure 2. Transplanting sweet potatoes using planter.



Figure 3. Mower used for 1st termination of cover crops.



Figure 4. Roller crimper used for 2nd termination.



Figure 5. Tiller used for incorporation of cover crop in green manure plots.



Figure 6. Frame used for weed evaluation.



Figure 7: Digger used to harvest sweet potato tubers.



Figure 8: Marketable tubers.



Figure 9: Blemished tubers.

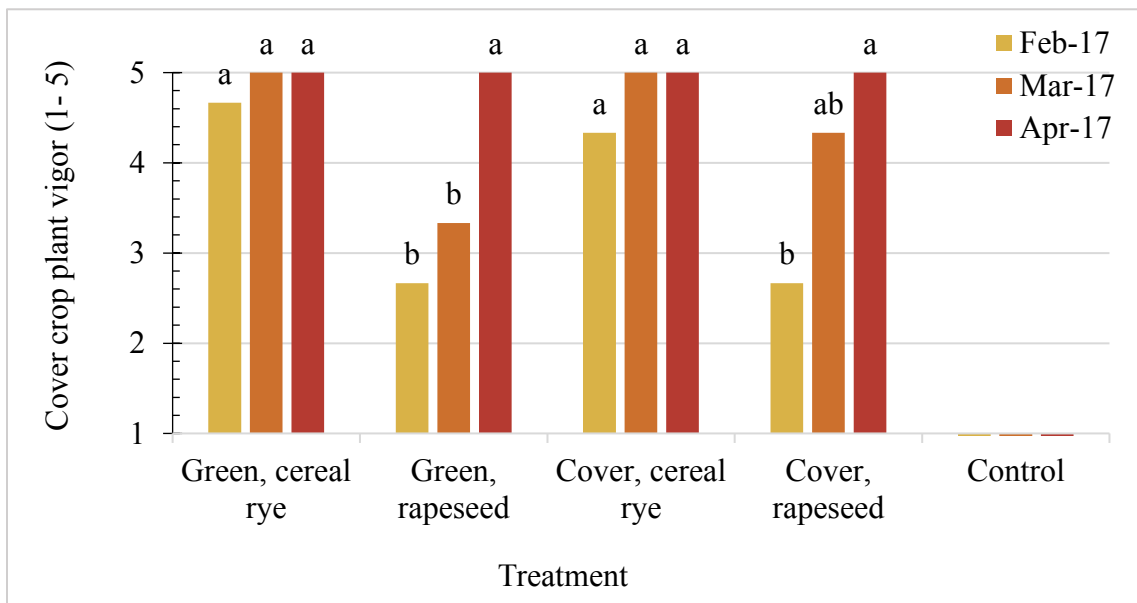


Figure 10. Cover crop plant vigor (1-5) influenced by treatment applied. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

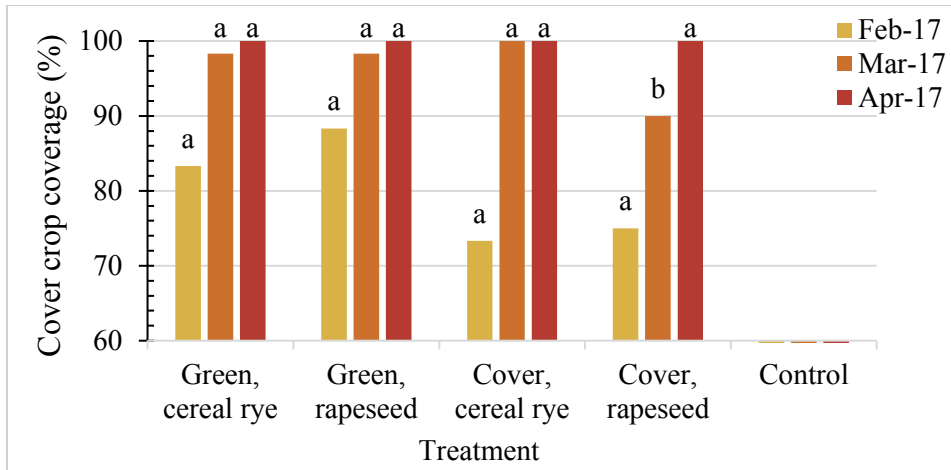


Figure 11. Cover crop cover (%) influenced by treatment applied. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.



Figure 12. Cover crops on 28th October 2016.



Figure 13. Cover crops on 3rd February 2017.



Figure 14. Cover crops on 7th April 2017.



Figure 15. Plot view on 24th April 2017.

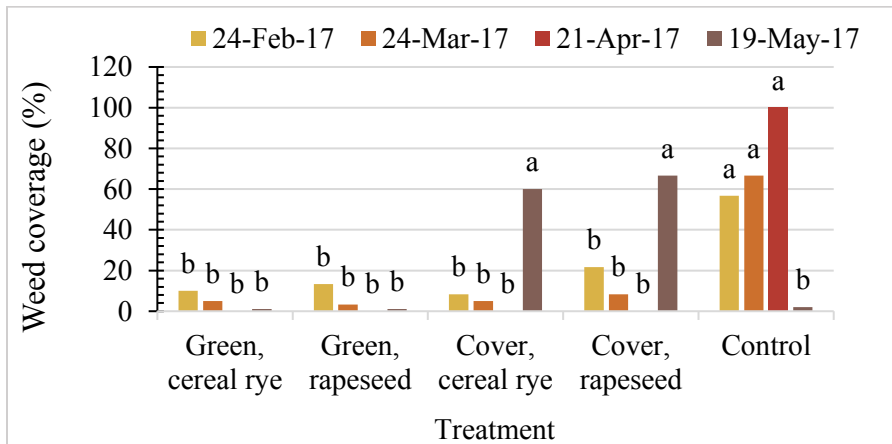


Fig 16. Weed cover (%) from February to May influenced by treatment applied. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

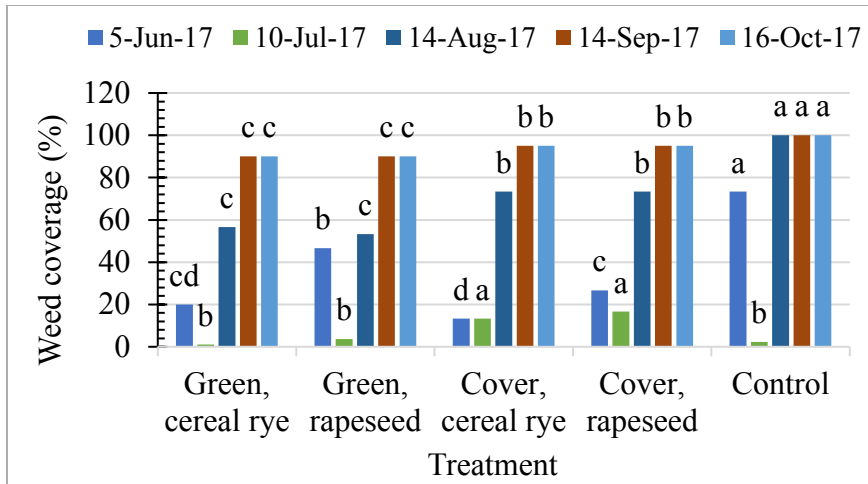


Fig 17. Weed cover (%) from June to October influenced by treatment applied. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

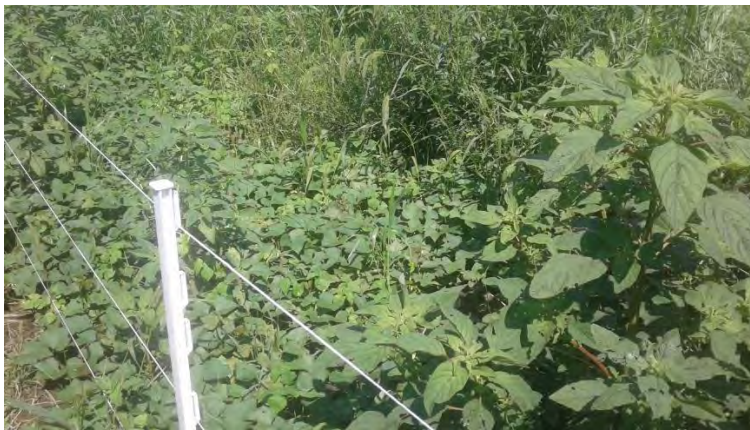


Figure 18. Weeds in green manure plots on 8th August 2017.



Figure 19. Weeds in cover crop plots on 8th August 2017.



Figure 20. Weeds in control plots on 8th August 2017.

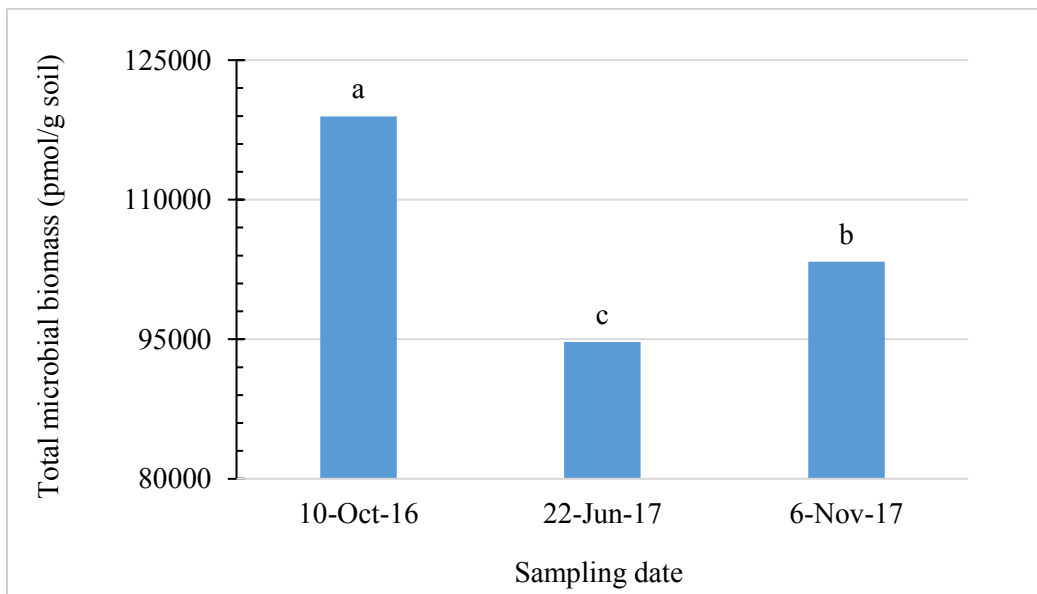


Figure 21. Total Microbial biomass (pmol/gram soil) influenced by date. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

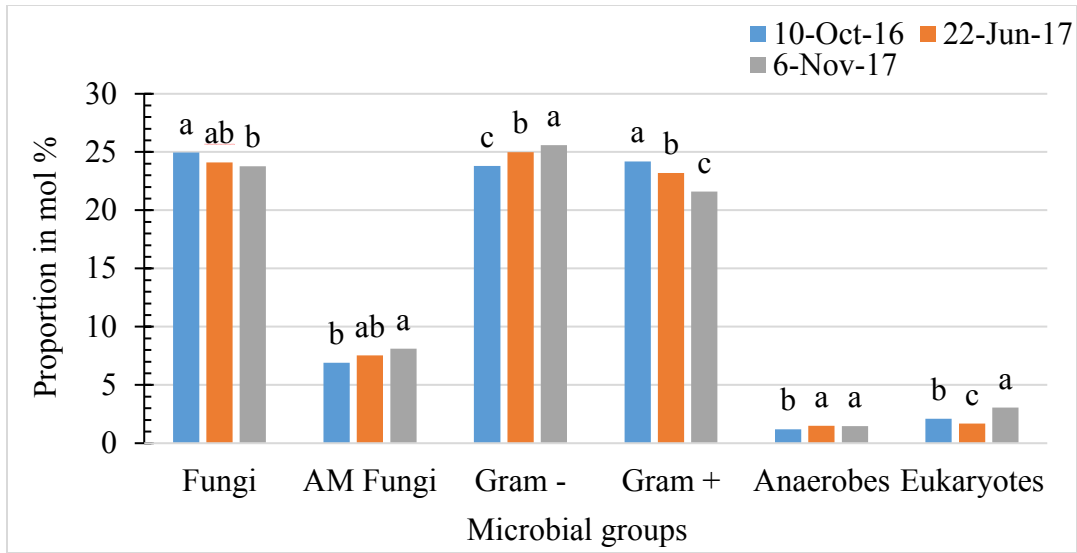


Figure 22. Microbial groups (mol %) influenced by date. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

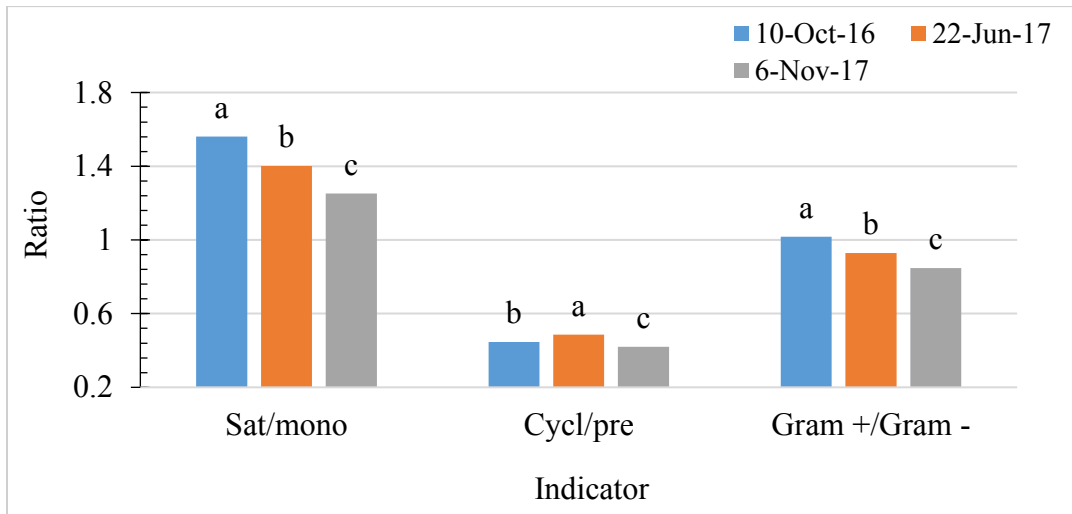


Figure 23. Indicator ratios influenced by date. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

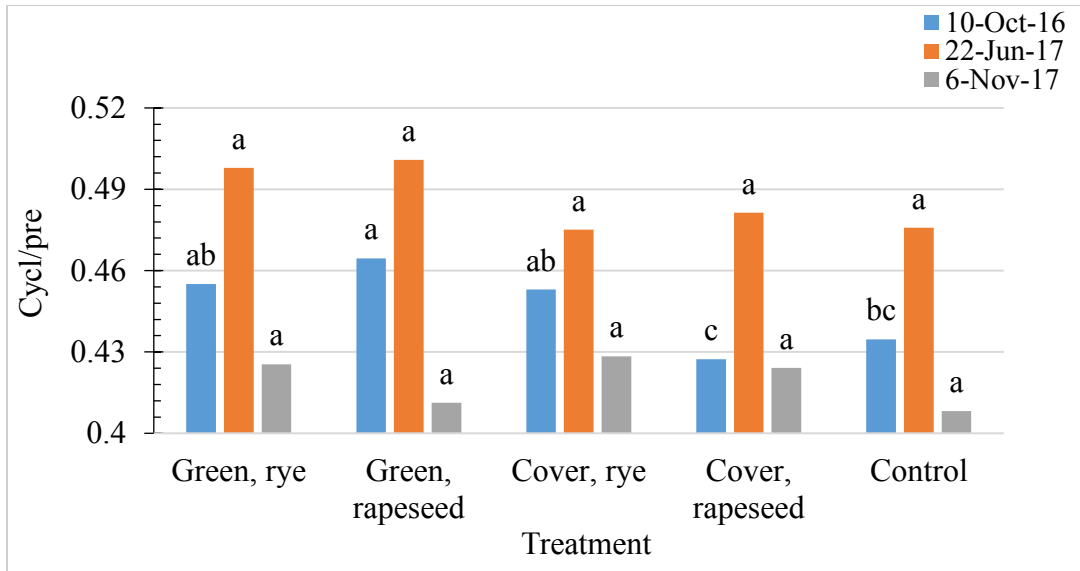


Figure 24. Cyclopropyl 17/precursor ratio influenced by treatment applied. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

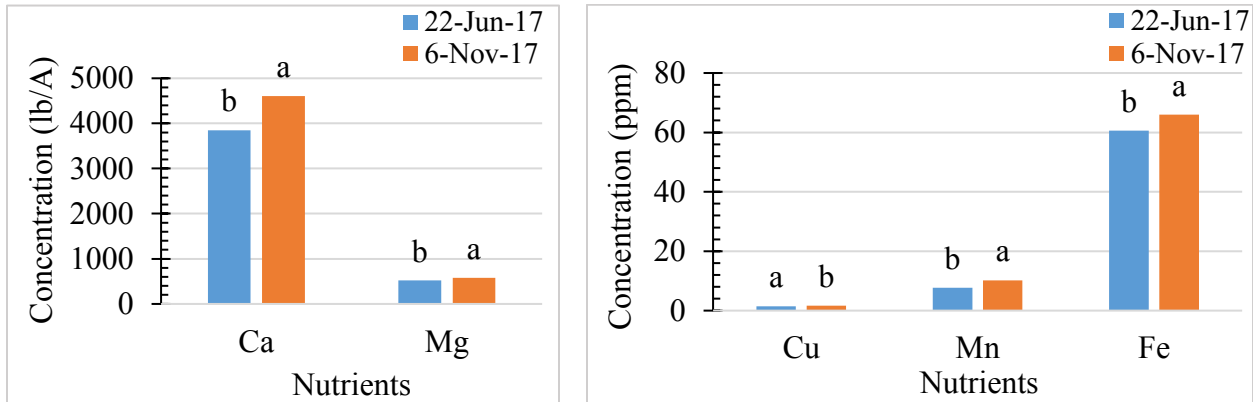


Figure 25. Soil nutrients influenced by date in lb/A (left) and ppm (right). Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

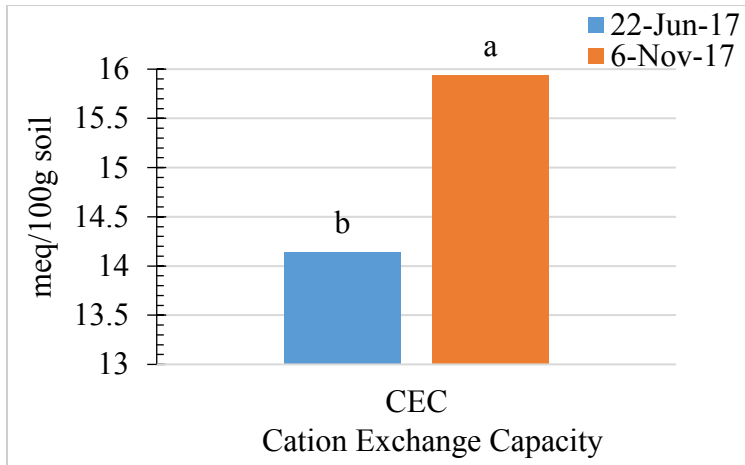


Figure 26. Cation Exchange Capacity (meq/100g) influenced by date. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

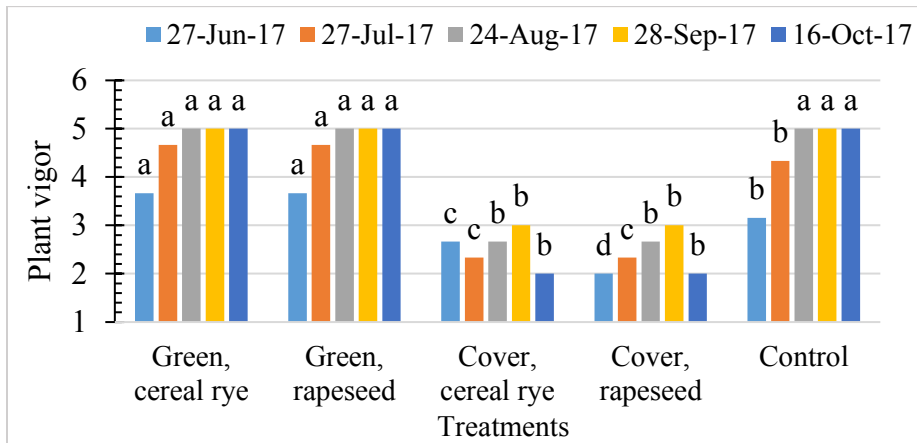


Figure 27. Sweet potato plant vigor (1-5) influenced by treatment applied. Bars labeled with the same letters at each evaluation timing are not significantly different based on Fishers' Protected LSD at $P < 0.05$.

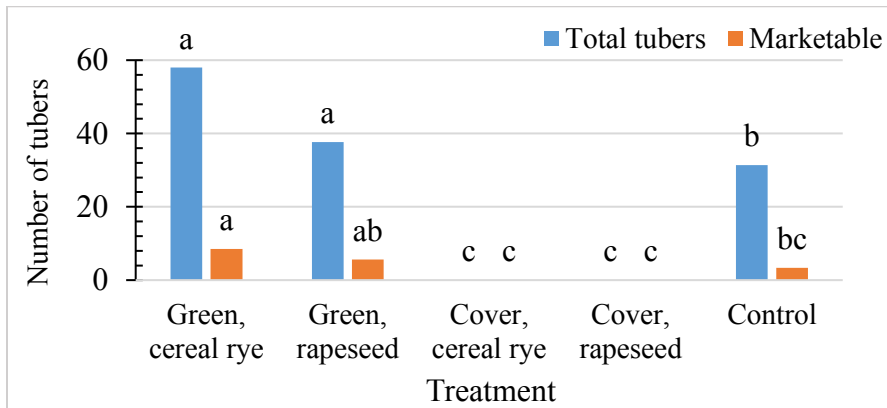


Figure 28. Sweet potato total and marketable yield influenced by treatment applied.



Figure 29. Field day at Share Life Farm on 16th August 2017.

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