RESEARCH STUDY SUBMITTED TO

Kwantlen Polytechnic University

BY

Faculty of Sustainable Agriculture and Horticulture

Title: Spent Mushroom Substrate for Water Conservation in Squash Production

Submission Date: December 17 2020

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Abstract

The effect of spent mushroom substrate (SMS) in various mulch depths (0, 5, 10, 20 cm) for the cultivation of spaghetti squash (*Cucurbita pepo*) was evaluated. Soil health indicators such as electric conductivity (EC), pH, volumetric water content (VWC) and yield, were used to measure the effect of SMS in this study. The use of SMS as mulch and soil amendment improved VWC and yield, while maintaining optimal levels of soil salinity and acidity. Therefore, the utilization of SMS as much ans organic amendment has the potential to be a productive disposal alternative in the Lower Mainland. Moreover, SMS can be suitably used for orchard site preparations at Kwantlen Polytechnic University's farm.

Introduction

Global mushroom production has increased rapidly in the past three decades, from 1.2 million metric tons in 1980 to 7.3 million metric tons in 2010 (FAO, 2012). Spent mushroom substrate (SMS) is the byproduct of mushroom production industries (Ebrahimi et al. 2019). It results after different flushes of mushrooms have been harvested (Sendi et al. 2013). For the production of each kilogram of mushrooms, 5kg of spent mushroom substrate are produced (Paredes et al., 2016). After each cycle, used spent mushroom substrate is discarded which has become a major environmental issue for the mushroom producing industries (Sendi et al. 2013). An effective disposal alternative is required, particularly because spent mushroom substrate (SMS) can be very useful. SMS can be utilized for soil and water remediation, pest control for different crops, growing media and organic amendments (Paredes et al. 2016). The effects of this substrate on crop production and soil properties have been little explored.

SMS is produced from chopped straw, poultry manure, gypsum and water (Uzun 2004). According to Uzun (2004), spent mushroom substrate alone contains a lot of salts and unstable organic material; for that reason, it must be aged for two years before using it. He proposes that the aging process will allow for leaching of organic solutes and decomposition of organic matter. SMS does not contain any pests or weed seeds because of the high temperatures associated with the composting and pasteurization processes (Uzun 2004). The electric conductivity is usually high, generally 4dS/m (Uzun 2004). In the same study, the author remarks that most fruit crops are sensitive to high levels of salinity; for example, stone fruit and pome fruits require an EC lower than 4dS/m.

Highline Mushrooms is a major producer of organic mushrooms in the BC's Fraser Valley. Its SMS is produced in three phases, carried out in enclosed facilities across BC's Lower Mainland. The substrate is made with chicken manure collected from 40 barns, wheat straw, gypsum and water. Each of these elements contributes to the quality of spent mushroom substrate, such as moisture, nitrogen content, structure and length, improved aeration and nutrient availability. The result is a substrate with 41.4-56% organic matter, 65% moisture, up to 30% ash, 14-16 C/N ratio, 51-59 meq/100g for cation exchange capacity, pH of 6.2-7.2 and EC ranging from 7.27 to 12.9 dS/m.

SMS is nontoxic and a good source of humus formation, which provides plant with micronutrients, improves water holding capacity, soil aeration and aggregation (Sendi et al. 2013). Maher (1994) observed that the addition of SMS to soil raised P, K and Mg levels. As a soil amendment, it can be used to stabilize highly disturbed soils and control plant diseases (Sendi et al. 2013). In an experiment comparing SMS with peat moss as a growing medium, Sendi et al. (2013) found that SMS had lower acidity (pH of 6.1) but higher EC (1.331 dS/m) than peat moss alone.

Composts are produced during the process of decomposition of organic matter by microorganisms in the presence of oxygen (Ebrahimi et al. 2019). They can maintain and enhance the stability and fertility of agricultural soils (Ebrahimi et al. 2019). This is done through mechanisms such as: stimulating competing organisms, reducing diseases and increasing plant resilience against them (Ebrahimi et al. 2019). As mentioned by Ebrahimi (2019), “maintaining soil fertility is one of the main factors affecting the sustainability of food production”. Approximately 30% of soils, in more than 30 countries, are suffering from deficiency of one or more micronutrients (Ebrahimi et al. 2019). Compost application is a method that can be used to combat these issues thus, maintaining soil fertility and providing long-term nutrients by gradual decomposition (Ebrahimi et al. 2019).

Organic matter is rich in essential elements for plant growth, such as nitrogen, phosphorous and potassium in organic forms (Ebrahimi et al. 2019). Its use depends on the quantitative and qualitative characteristics, climate conditions, plant type, soil organisms and management method (Ebrahimi et al. 2019). Organic manures enhance the different properties of soil: physical, chemical and biological (Ali et al. 2018). They can increase the water holding capacity of the soil, which in studies has resulted in a stable crop production and quality (Ali et al. 2018). In a study comparing different organic manures and organic manure combinations for bulb production, plants receiving poultry manure had the tallest plant height, in contrast with plants that were not treated with this manure type (Ali et al. 2018). In the same study, spent mushroom substrate ranked as one of the manures with the highest plant growth, bulb weight and total yield (Ali et al. 2018).

In an experiment using soil-based potting media with different rates of SMS, Ribas et al (2009) observed that applying smaller rates resulted in a greater aboveground biomass on dry weight than with larger rates. In the same experiment, no significant changes in the salt content of the soil solution were noticed due to the variation of SMS rates (Ribas et al. 2009).

Spent mushroom substrate represents a promising renewable agricultural organic resource that can improve soil properties (Herrero-Hernandez et al. 2012). In a study, spent mushroom substrate was found to be high in organic matter content and nutrients (Alvarez-Martin et al. 2016). On this basis, SMS was assessed as a tool for pesticide biodegradation and leachates control. Alvarez-Martin et al. (2016), found that the dissipation rate of the fungicides cymoxanil and tebuconazole was higher in soils with this amendment, this was due to the increased absorption. SMS is also associated with retention, decreased mobility and reduced bioavailability of trace metals (Herrero-Hernandez et al. 2012). Despite the studies proposing the addition of organic amendments to accelerate the natural remediation of metal sorption, precipitation and complexation reaction; Herrero-Hernandez et al. (2012) found that moderate doses of spent mushroom substrate did not cause a significant accumulation of metals in a 3-year period.

Repeated applications of SMS can increase organic matter remarkably when used at composted state (Herrero-Hernandez et al. 2012). The use of this amendment can be advantageous to increase the pH of soil because of its alkaline nature and chalk content (Ali et al. 2018). Additional studies by Ebrahimi et al. (2018), found that high rates of spent mushroom substrate produced less tomato in row application, which might be related to high salinity around the root area. However, treatments with SMS at medium rate with broadcast application produced the maximum amount of tomato in the same study (Ebrahimi et al. 2019).

A way to counteract high salinity problems would be by using salt resistant rootstocks, which take up less chloride (Uzun 2004). Related problems are said to arise when too much SMS is applied on the soil, or when plants are grown directly on it (Uzun 2004). Depending on rainfall, conductivity will gradually decrease through leaching during the weathering process (Uzun 2004). During the first year of weathering, most of the salts originally present will be released (Uzun 2004).

Squash (*Cucurbita pepo)* are a warm season annual crop (Newenhouse 2012). *Cucurbita pepo* is the species with the greatest monetary value of the genus Cucurbita L. (Gong et al., 2012). They need heat, long growing seasons and well-drained soils that warm up quickly like, light sandy soils (Macdonald 2014). Most of the plants in the Cucurbitaceae family form tendrils, which wrap around stems as the plant grows (Newenhouse 2012). They grow flat on the ground and develop vines (Newenhouse 2012). Squash have shallow roots that grow in a wide area; consequently, spacing margins must be taken into consideration for when they are planted (Newenhouse 2012). As recommended, plants must be spaced at a minimum of 90-120 cm (35-47 in) apart in rows 120-180 cm (47-70 in) apart (Macdonald, 2014). Soils with high levels of organic matter may turn the fruit soft and not as sweet (Newenhouse 2012). Squash is able to grow in a wide pH range of 5.5-

7.5 (Newenhouse 2012). Plants from the Cucurbitaceae family need to be irrigated regularly, approximately 1 inch of water every week, because they use a lot of it to grow big leaves (Newenhouse 2012).

Permanent crops such as fruit trees are the result of decisions that may take years to manifest, consequently these need to be considered carefully (Pissonnier 2017). Orchards play an important role in promoting the development of the fruit industry, regional economies, the environment and global warming mitigation (Yang et al. 2020). Orchards require mulching management strategies to increase soil organic matter, yields and quality (Yang et al. 2020). Mulching can significantly change the soil organic carbon content and dissolved organic matter (Yang et al. 2020). Consequently, the input of organic matter and pH will have an effect on the soil microbial community, which will reflect on soil quality (Yang et al. 2020). As recommended by a report from the Pennsylvania State University (2016), orchard soil is preferred to be deep, well drained and well aerated sandy loams or loams.

SMS should be analyzed for nutrient content and applied at the rate determined by local soil conditions and the choice of the cultivar (Uzun 2004). According to Uzun (2004), the benefits of this compost in orchards are modest during the first year but will increase considerably in the years after. Although SMS tends to be applied in a 2-inch (5 cm) thick layer when used as mulch, Tsaoir and Mansfield (2000) propose 4-inches (10 cm) to provide adequate weed control. Mulching with SMS provides reasonable weed control but it may have a negative effect in the yield of apple orchards, due to toxic levels of plant nutrients (Uzun 2004).

Objectives

1. Compare four mulch depths of spent mushroom substrate, on the basis of various soil quality indicators, in preparation for the establishment of an orchard site at Garden City Lands in Richmond, BC.
2. Evaluate whether the use of SMS is an ecologically sound disposal alternative of spent mushroom waste.
3. Explore the interaction of SMS with the mineral soil layer over the native peat at Garden City Lands and highlight its abilities as mulch and soil amendment. Ideally, the results will encourage local farmers to use it more readily.
4. Encourage mushroom companies like Highline Mushrooms to introduce SMS to the local and national market.
5. Further the understanding of the effects of SMS for the cultivation of squash and orchard trees, with an emphasis in water conservation.

The hypothesis in question is defined by whether different mulch depths of spent mushroom substrate have an effect on various soil quality indicators (electrical conductivity, pH, volumetric water content and yield).

Materials and methods

Study site

The study was carried out at Kwantlen Polytechnic University's farm on Richmond's Garden City Lands, located between Westminster Highway, Alderbridge Way, Garden City Way and No. 4 Road (49°10'26.9"N 123°07'09.7"W). It covers approximately 2.7 ha with variable soil composition, due to the fact that the upper 70 cm is imported sandy-clay loam with varying proportions of sand and clay (City of Richmond 2020). Average high and low temperatures during the growing season were 22 and 12 °C, respectively. Total precipitation was 48 mm between June 17 and August 28 (Environment and Natural Resources 2020, Farmwest 2020).

Experimental Treatments

The experimental treatments were four depths of spent mushroom substrate (0, 5, 10, 20 cm; Highline Mushrooms, Langley BC), applied as mulch on June 17th, 2020. The material had been used as a substrate for organic certified mushroom production. The no mulch control was used to standardize the study and allow comparisons to untreated soil. Bare soil is susceptible to erosion, competition with weeds and, reduced levels of organic matter.

The study was arranged in a randomized complete block design with four replicate blocks of four plots each, for a total of 16 plots (Fig. 1). Blocks were separated by a one-meter path. Each block measured 2 x 8 m, while plots were 2 x 2 m. Plots were subdivided into four subplots, measuring 1 x 1 m. Each of the 64 subplots were randomly assigned to a different crop; this report concerns the plots containing squash.

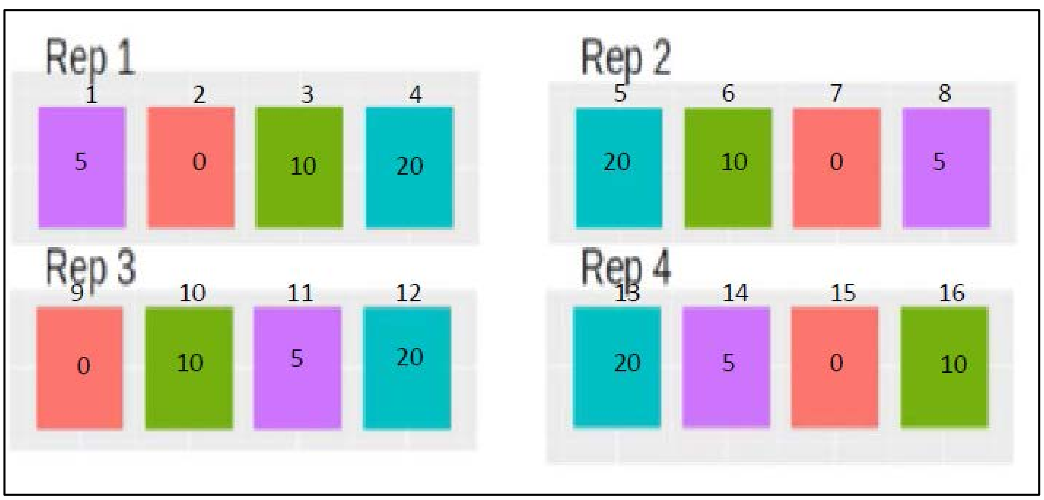

Figure 1. Plot map of randomized complete block design for spaghetti squash (*C. pepo*) cultivation with various mulch depths of spent mushroom substrate, n=4.

Research Subject

Squash (*Cucurbita pepo)* was the test crop in this study, specifically vegetable Spaghetti Squash. This crop is able to reach maturity within 100 days (West Coat Seeds 2014). The seeds used were Orangeti F1 Spaghetti Squash (Certified Organic, High Mowing Organic Seed Batch 08040).

The spaghetti squash was grown from transplant using a standard potting mix, instead of direct seeding to prevent premature damage to the germinates. Germinates are very delicate and can be harmed due to climatic conditions and/or animal predation (e.g. heavy rains or crow feeding).

The seeds were started in a solar dome greenhouse at the KPU Sustainable Agriculture Farm on May 28, 2020. A single plant was transplanted into the center of each subplot on June 19, 2020. Plants were transplanted into the soil below the mulch by digging a well through the mulch layer. Plants were irrigated occasionally with a single drip tape running through the plots, to ensure that all plots received the same amount of water.

Data Collection

EC was measured every two weeks from July 3 to August 28 of 2020 with a Soil EC Meter (FieldScout Direct EC Meter, Spectrum Technologies). Soil was saturated by spot watering before the probe tip was inserted 5 cm below the soil surface at the base of one plant in each subplot. Volumetric Water Content (VWC) was monitored every two weeks from July 17 to August 28 using a Time Delay Reflectometry Sensor (FieldScout TDR 350, Spectrum Technologies) with 20 cm probes. Probes were inserted into the soil below the mulch at the base of one plant in each subplot before irrigation. A soil pH meter (ExStick 110, Extech Instruments) was used to measure pH on October 1, after harvest. Marketable squash weight was measured for each plot on August 28, using a field scale.

Statistical Analysis

Statistical analysis was conducted using the jamovi (jamovi Project 2020, v. 1.2.9) interface to the R statistical computing environment, (R Core Team 2019, v.3.6). Independent variables were various mulching depths (0, 5, 10, 20 cm) and monitoring date. pH and yield were analyzed by ANOVA; EC and VWC were analyzed by Repeated Measures ANOVA. Means were separated by Tukey’s Honestly Significance Difference when significant treatment effects were found. A critical threshold of $\alpha = 0.05$ was maintained throughout the study.

Results and discussion

Electric conductivity. Compost depth had a significant effect on soil EC ($p \leq 0.001$) (Fig. 2). Plots treated with 20 cm of SMS mulch had the highest EC (1.34 dS/m) and control plots had the lowest (0.1 dS/m). No significant effect of monitoring date on EC was detected ($p \leq 0.181$), despite an apparent tendency for EC to decline through July and August. No interactions were observed between date and mulch depth ($p \leq 0.847$) or date and replicate ($p \leq 0.248$). The highest EC value noted was 5.17 dS/m, and it appeared on a plot treated with 20 cm of SMS. Uzun (2004) mentions a gradual decrease in electric conductivity in a study discussing the advantages and disadvantages of using SMC in fruit growing. This was probably due to leaching during the weathering process. Paredes et al. (2016) obtained similar results in a study which evaluated the addition of two spent mushroom substrates on the characteristics of calcareous clayey-loam soil and the yield and nutritional status of lettuce. Given the relatively low soil EC detected in mulched treatments in this study, mulching squash and orchard trees with SMS appears to be feasible.

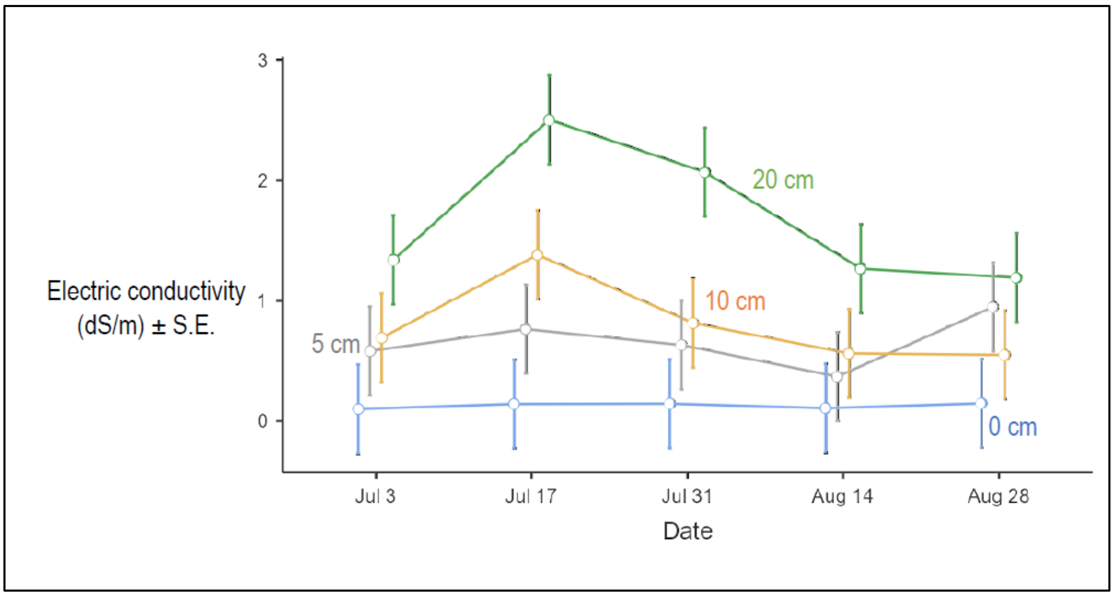

Figure 2. Relationship between electric conductivity (dS/m) and collection dates for spaghetti squash (*Cucurbita pepo*) mulched with various depths of spent mushroom substrate (0, 5, 10, 20). Bars show standard error around each mean.

pH. No significant mulch depth effect on pH was found ($p \leq 0.519$) (Fig. 3), but a slight tendency toward higher pH appeared with more mulch (Figure. 4). pH seems to be moving in the direction of neutrality therefore, almost all replicates measured within the ideal range, 5.5-7.5 (Newenhouse 2012). Sendi et al. (2013) found in a study aiming to confirm the feasibility of replacing peat moss with spent mushroom substrate that the SMS treatment had the lowest acidity level among other treatments. It is difficult to predict whether pH would become more basic with the addition of more mulch. In the same fashion, through repeated pH readings, it would be possible to notice an increase in pH in the same mulch depths over time. Lou et al. (2017) reports similar findings in a study developing a nutrient retention strategy during SMS application using mushroom substrate biochars. They observed from the groups without biochars that the pH values were distributed within the range of 7.0 and 8.5. pH levels are a direct indictor of nutrient availability and so, it is imperative to deduce if a cumulative effect will produce a beneficial or toxic environment for plants. This is particularly important hence toxic levels of plant nutrients can have a negative effect in the yield of apple orchards, according to Uzun (2004).

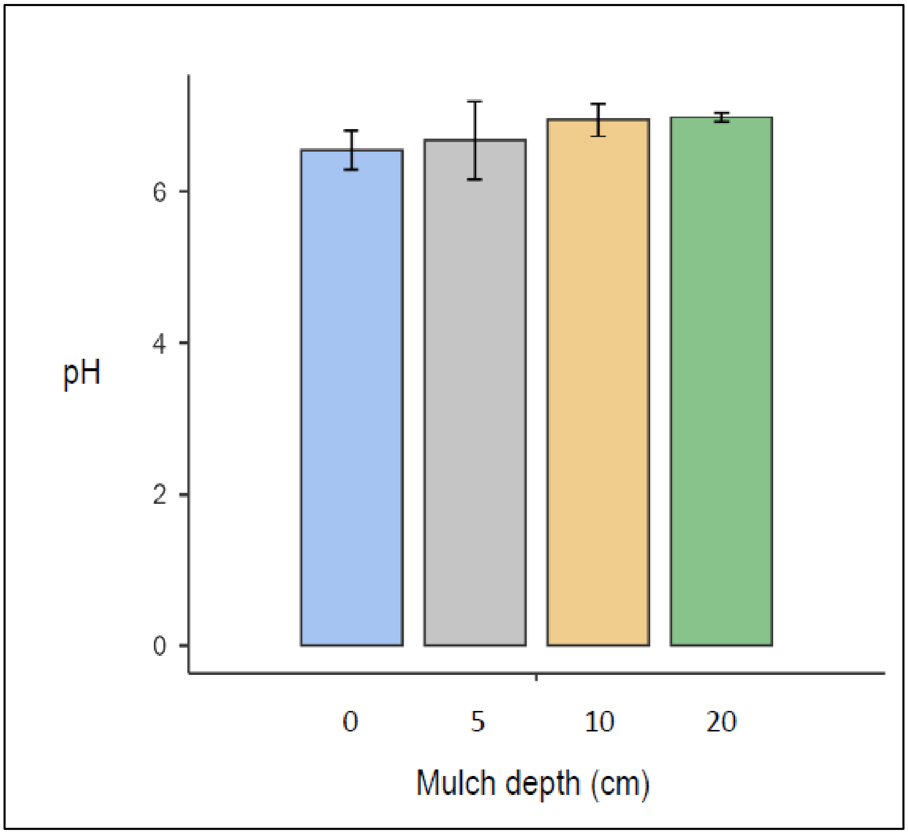

Figure 3. Plot bars show the pH of spaghetti squash (*Cucurbita pepo*) mulched with various depths of spent mushroom substrate (0, 5, 10, 20 cm).

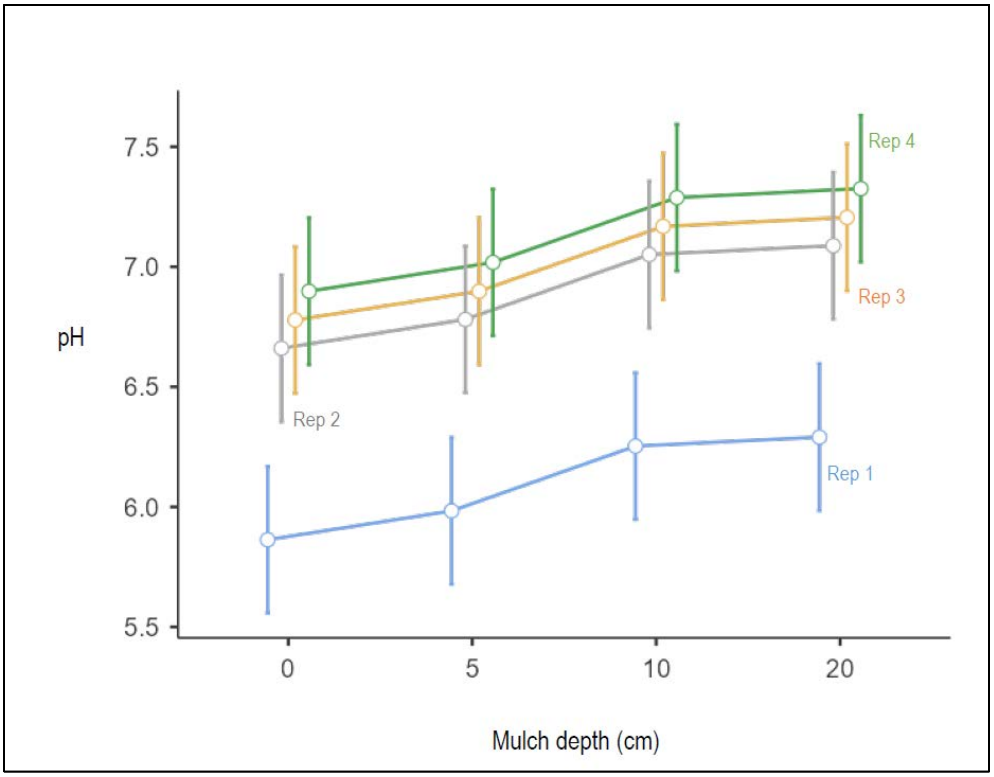

Figure 4. Relationship between spent mushroom substrate mulch depths and replicate. Bars show the standard error around each mean.

Volumetric water content. Collection date and mulch depth both had a significant effect on VWC ($p \leq 0.001$) and a marginally significant interaction between collection date and mulch depths was also observed ($p \leq 0.057$) (Fig. 5). VWC tended to decline throughout August, reflecting a regional drying pattern due to low precipitation in the summer. Therefore, the graph reflects the weather behavior and the water retention abilities of the spent mushroom substrate. Plots with 20 cm of mulch had the highest volumetric water content throughout the growing season, despite the decrease at the end of August. The other three mulch depths displayed a similar trend, until reaching a similar VWC. Arthur et al. (2012) reported a similar effect, SMS increased the water content of sandy loam soil. On a different study which explored the use of new mulches as an alternative to conventional ones, Farzi et al. (2017) found that mulch application has a great efficiency on soil water conservation in semi-arid regions. Our findings for VCW are consistent with those of Pi et al. (2017) who showed that a variety of mulches can improve water storage. Consequently, mulching with spent mushroom substrate can reduce the reliance on irrigation, by retaining soil moisture during the growing season.

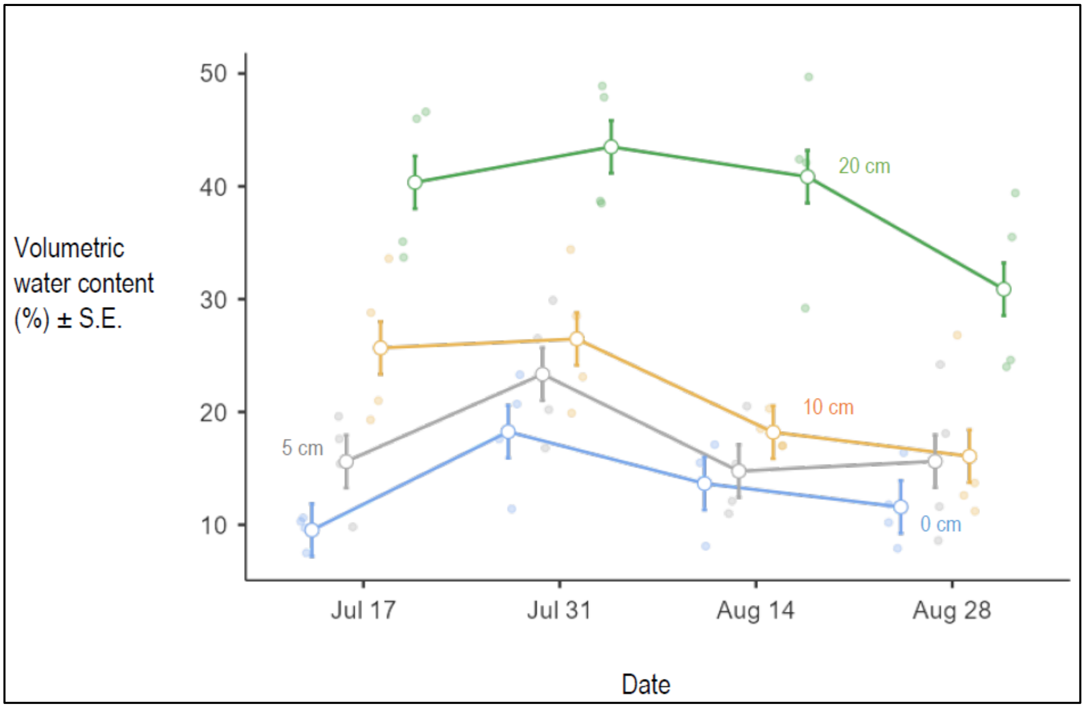

Figure 5. Relationship between volumetric water content (%) and collection dates for spaghetti squash (*Cucurbita pepo*) mulched with various depths of spent mushroom substrate (0, 5, 10, 20 cm). Bars show the standard error around each mean.

Yield. Mulch depth had no significant effect on yield ($p \leq 0.37$) (Fig. 7). The average squash yield was 178 ± 12 t/ha which compares favorably to a typical squash yield of 56 t/ha. Although mulch depth did not have a significant effect on yield, the 20 cm depth displayed the highest yield mean of all treatments (Table 1, Fig. 7). Likewise, Ali et al. (2018) observed in an experiment carried out to investigate the organic manures effect of bulb production of onion cultivars under semiarid condition, that the total yield was significantly affected by organic manures, including spent mushroom substrate. In the same experiment, it was mentioned that the increase in yield by organic manures was related to their effect on water holding capacity and ability to provide nutrients for a long duration, due to less leaching of nutrients (Ali et al, 2018). Moreover, Ebrahimi et al. (2018) assessed four different types of compost in two methods of application for their potential to support organic tomato yield. In this study, they found that treatments with SMS at medium rate with broadcast application produced the maximum amount of tomato.

Table 1. Estimated marginal means of spaghetti squash (*Cucurbita pepo*) yield separated by mulch depth (cm).

			95% Confidence Interval	
Compost Depth (cm)	Mean	SE	Lower	Upper
0	16.7	2.45	11.2	22.3
5	16.3	2.45	10.7	21.8
10	16.4	2.45	10.9	21.9
20	21.8	2.45	16.2	27.3

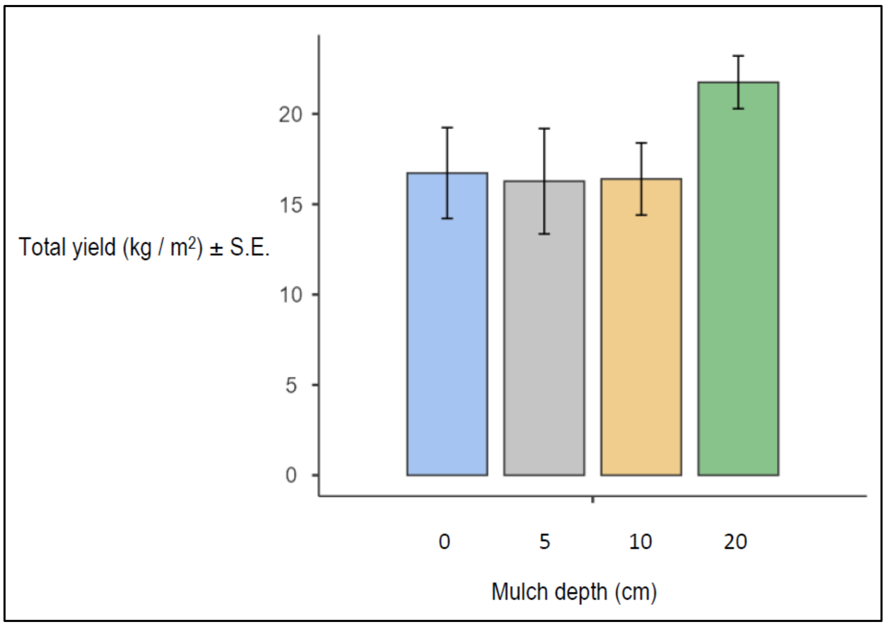

Figure 7. Yield of spaghetti squash mulched with various depths of spent mushroom substrate. Bars show the standard deviation around each mean.

Conclusion

Mulching spaghetti squash (*Cucurbita pepo*) with SMS improved water holding capacity without compromising yield. The improvement was greatest with 20 cm of SMS mulch, the highest rate tested. Mulching reduces reliance on irrigation in our local climate. Spaghetti squash yield was almost three times the expected amount. Mulching with SMS did not affect negatively the electric conductivity or the pH. No toxic effects due to high salinity or accumulation of nutrients were observed. The increase in yield could be the consequence of improved water holding capacity and prolonged nutrient availability before the natural regional drying pattern, due to low precipitation.

Acknowledgements

Thank you Mike Bomford for all your patience and advice, Lindsay Dodds for the support, as well as Andy Smith and fellow students, Sarah-Jane Copeland and James Oswald, who assisted the experimental set up and maintenance.

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