Effect of biochar in modifying soil salinity effects on the growth of lettuce (*Lactuca sativa* L.)

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Declaration

I, Dunya Mohi Mohsin Mohsin, declare that this is my own work and it has not been submitted in any form for another degree or diploma at any university or other institution of tertiary education. Information derived from the published or unpublished work of others has been acknowledged in the text and a list of references is given.

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Signature

Date

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Abstract

Soil salinity is widespread in Australia and globally, and with increasing pressure on maximising productivity and efficiency in food production, there is a need to identify methods of enabling farming in sub-optimal soils such as those with excess NaCl. Saline soils can managed using a range of techniques, including manipulating the plant's physiology to resist or tolerate salt uptake, and remediating the soil by removing the salt (e.g. leaching) or modifying the soil composition so that the salt does not damage the plants. In regard to modifying the soil, biochar has also been reported to ameliorate the impacts of salinity, although the specific mechanisms depend on the biochar, application rate and the soil type. This study aimed to investigate the impact of adding biochar to soils with a range of salinity levels.

Two experiments were carried out. Experiment 1 investigated the effect of biochar on lettuce growth under saline conditions, with two repeat trials conducted. Three levels of biochar (0, 5, 10 g/kg) were added to the soil prior to beginning the experiment. Two levels of soil salinity (0, 3.0 dS/m) were produced in the soil by adding 0 or 1% weight/weight of NaCl (i.e. 0.07 g/kg) to the soil. Two levels of irrigation water salinity (0, 2.4 dS/m) were applied after planting the lettuce seedlings. The saline irrigation water was prepared by adding 10.1 g NaCl to 10 L water. Lettuce was used as the test plant due to its sensitive to salinity. Seedlings were grown in a sandy loam soil for five weeks. Measurements included height, chlorophyll, shoot and root biomass, a visual health index and soil electrical conductivity (EC). Experiment 2 evaluated the effect of biochar on salt leaching using the same treatments as Experiment 1 in a soil column subjected to regularly leaching events using tap water. Leachate was collected after each event and EC and volume recorded.

Increasing biochar rates improved lettuce growth and the salinity treatments impaired lettuce growth. The interactions of biochar application and the salinity treatments indicated that adding biochar improved lettuce shoot growth with salinity irrigation or saline soil, but not both. Root dry weight was affected in the same way, but the inhibition of growth due to the two salinity treatments was more pronounced. The health index showed that the high rate of biochar resulted in the lettuce plants having less inhibition or damage in saline soil. These results are consistent with other reports about lettuce and biochar in saline soils, and may be due to changes in soil pH, microbial activity and function and organic carbon levels. Plant physiological effects may also have occurred, although some reports have found these plant processes not to have any variation in response to biochar additions in saline soils.

The leaching experiment showed that the EC did not change with increasing biochar levels, confirming that the biochar itself did not add salt. Where saline irrigation was applied (with and without soil salinity), biochar had no effect on leachate EC over time. In the saline soil treatment, biochar reduced the leachate EC initially (up to about Day 5) only, probably because of binding of the ions. Leachate volume did not vary over time in any treatment, in contrast with many reports of biochar increasing the water holding capacity of soils. This may be a genuine response (i.e. biochar is ineffective), or may be due to the sandy soil used in the trial, insufficient water volumes applied, or inadequate biochar application rates.

The experiments reported here demonstrated some benefits from biochar to plant growth in saline soils, especially at higher application rates (10 g/kg soil). The specific mechanisms were not evaluated in detail, although it was apparent that removal of salt through leaching may play a role. Further studies are needed to explore the mechanisms or processes by which biochar could influence plant growth under saline conditions.

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1 Introduction

Soil salinity is the cause of many problems related to plant growth. Salts accumulate in the soil from irrigation water, fertilizers or due to other reasons. So salinity can be either natural or induced by human beings. Efforts to manage saline soils range from those targeting the plant's physiological capacity to tolerate or resist salt uptake, to strategies aimed at improving the soil by removing the salt (e.g. leaching) or modifying the soil composition so that the salt does not damage the plants (Qadir et al. 2000, Kamaluldeen et al. 2014).

Among these techniques, biochar has also been found to manage salinity (Saifullah et al. 2018). Biochar is fine grained residue which has high carbon content. Biochar can be produced by the pyrolysis (thermo-chemical conversion) of agricultural waste materials such as wood, green waste, sewage, poultry litter, rice husks, paper pulp and other organic waste (Hu et al. 2010). This type of soil is mainly seen within the Amazon basins known as "Terra Preta" and was found to be extremely productive. The pyrolysis method used for the preparation of biochar defines the biochar alkalinity, particle size and surface area (Lehmann & Joseph 2015).

Due to its chemical and physical properties it can absorb nutrients, salts and other pollutants from the soil (Lazcano et al. 2013). It also helps in the improvement of water holding capacity (Lehmann & Joseph 2015) and soil microbial activity (Woolf et al. 2010). Biochar helps in improving soil as its highly porous structure and large surface area helps microorganisms grow well in the soil (Khadem & Raiesi 2017). It also helps in increased aeration of soil, availability of macronutrients as well as cation exchange capacity that lead to better plant growth (Tan et al. 2017). It has the ability to sequester carbon from the atmosphere due to its surface area (Brassard et al. 2016), helping reduce greenhouse gas emissions (Mohammadi et al. 2016). The

effect of biochar incorporation in soil is usually dependent upon the feed stock and pyrolysis conditions, soil type and other environmental conditions such as temperature and rainfall (Tan et al. 2017).

The action of biochar to amend the saline soil depends on the amount of the biochar applied in these soils. Also biochar and compost if used in combination have a synergistic effect on soil (Lazcano et al. 2013). It helps to increase microbial activity in saline soil that helps to retain nutrients as well as moderates soil pH also. Along with compost organic matter of the soil is increased. Also biochar has the ability to absorb carbon. Introduction of carbon activates the exchange of cations and it neutralizes the effect of electrical conduction in the soil. Biochar usage also results in the availability of phosphorous in soil whereas the nitrate salts are reduced. This is due to ability of biochar to absorb nitrogen fertilizers and inhibit process of nitrification.

Lettuce is considered a moderately sensitive to salinity (Ünlükara et al. 2008). Moderate levels of salts in the soil and water can be degrade the quality and yield of lettuce by stunting growth, reducing head formation and necrotic lesions appear on the leaves. However, lettuce is also reported to have effective mechanisms of salinity tolerance (Bartha et al. 2015). When biochar is used in soil for lettuce production it has beneficial effects on its growth and can reduce salt stress (Woolf et al. 2010). Upadhyay et al. (2014) reported an increase in pH and electrical conductivity as the rate of biochar application increased thus biochar helped in improving the growth of lettuce on saline soil. Another study showed that when biochar and cow dung in a coastal savanna soil increased lettuce yield (Frimpong et al. 2016). It increased soil pH, total organic carbon and cation exchange capacity. It temporarily increased soil respiration and microbial biomass carbon. The combined application of biochar and cow dung resulted in increased lettuce yield which was more than when each of these are used solely. The study also indicated that the source of biochar is also important for improving chemical properties of soil.

For lettuce corncob biochar has this ability of improving soil's chemical property thus leading to increased yield of lettuce. A study by (Trupiano et al. 2017) also showed the effect of biochar on lettuce plant growth and physiology. They studied its effect on the chemical and microbiological characteristics of soil. Biochar induced a positive lettuce yield whereas transpiration, stomatal conductance and assimilation rate did not show any variation.

1.1 Research aims

The broad objective of this study was to investigate the impact of adding biochar to soils with a range of salinity levels (based on salt added to soil and/or irrigation water). It was hypothesised that adding biochar can reduce the negative impacts of salinity on plant growth. A further objective was to understand the effect of biochar on salt movement in the soil. Specifically, the aims were to,

1. evaluate the effect of biochar on lettuce growth in saline soil, and

2. determine the effect of biochar on salt leaching in saline soil.

2 Literature review

2.1 Introduction

Plant growth is dependent on the types of soils on which the plant is expected to grow. These soils will determine the plant growth rate as well as the crop yields (Gregory & Nortclif 2012). Soil types are affected by climatic and weather conditions in different areas. However, farming and agricultural production has enhanced means of changing soil properties so that plants and crops can be grown for specific purposes especially for commercial production. Key soil constraints that affect soil fertility and function for agriculture include physical issues such as compaction and erosiveness, chemical problems including acidity and salinity, and biological limitations which can impact on nutrient availability, aggregation of soil particles, and microbial ecology (Downs 2012).

Soil salinity is a significant problem in many parts of the world (Gregory & Nortclif, 2012). The Food and Agriculture Organization reports that 397 million hectares of land are saline soils in the world (FAO 2016). Soil salinity tends to occur under two different scenarios. Dry land salinity is the type of salinuzation that is enahnced by ground water processes in non-irrigateed areas. As a result of these processes, the build up of salts in the soil occurs (Rengasamy 2006). On the other hand, irrigation salinity is another method of how soils become saline (Rengasamy 2010). As such, irrigation salinity occurs when there is an increase in salt in the soil in irrigated areas as a result of increased saline groaundwater (Podmore 2009). As such, while dryland salinity is more of a natural phenomenon, irrigation salinity is mostly considered to be as a result of human processes (Rengasamy 2006).

The aim of this literature review is to analyse the effects of organic amendments in modifying soil salinity effects on lettuce (*Lactuca sativa* L.) growth. The review will discuss salt levels and their effect on the growth of lettuce. Further discussion will also be presented on the adaptation mechanism of plants to manage salinity. The ameliorating effects of fertiliser and soil amendments such as biochar in will also be a key topic discussed.

2.2 Salinity effects on plants

2.2.1 Salt levels

Salinity in soil may be natural or human-induced. The accumulation of salts in the soil has both positive and negative effects on any plant. While some salts in the soil are important because they have nutrients that enhance plant growth, too many salts of the wrong type in the soil result in the inhibition of plant growth (Shabala & Munns 2012). Soil salinity is measured as the salt concentration in the soil, specifically through its electrical conductivity (Shainberg & Shalhevet 2012). When the electrical conductivity of a soil is 4 dS/m or more, the soil is considered to be saline. This is roughly equivalent to 40 mM NaCl (Tilbrook & Roy 2013). Highly saline soils may have concentrations of as high as 8-12 dS/m (Shainberg & Shalhevet 2012). These are concentrations that limit plant growth.

2.2.2 *Effects of salinity on plants*

There are a number of ways in which soil salinity inhibits plants growth. Salinity in soils can cause a water-deficit effect, reducing water availability to plants (Shainberg & Shalhevet 2012). In this regard, through osmosis, high soil concentrations limit the ability of the plant to take up water from the soil. As such, the plant may become adapted to growing with less water, thus limiting its growth (Shabala & Munns 2012). Additionally, salinity in soils also has the effect of affecting cell growth in the plant. Through ion-excess salinity, the plant takes up water but because of the high concentrations of ions in the water, cell growth and functioning in the plant

is constrained. It also affects the rate of transpiration in the plant resulting in stunted growth or ultimate death of the plant (Munns & Tester 2008).

Positive effects of salinity include the ability of the plant to have a higher resistance to diseases (Shabala & Munns 2012). Salinity in some plants enhances the immunity of the plant; thus making them become very resistant to diseases and as such enhancing their viability. In other plants such as cabbage, carrot, and potatoes, quality increased salinity can improve food quality characteristics, for example cabbages being more solid in high saline soils and potatoes having more starch (Grieve et al. 2012). Osmotic effects mostly enhance changes in the plant such as leaf colour changes and reduced growth rate. Osmotic effects of salinity also cause developmental issues such as the maturity rate of the plant and the root/shoot ratio (Grieve et al. 2012). However, the ionic effects of salinity commonly cause nutritional dysfunction in the plant which leads to damaging of the leaf (Shabala & Munns 2012).

Lettuce (*Lactuca sativa* L.) has a moderate sensitivity to salinity. With a petiole sap nitrate concentration of 1700-2600 during mid growth and 1300-2150 during pre harvest, the electrical conductivity of lettuce is 1.3 dS/m in its petiole sap (Ünlükara et al. 2008). In an experiment by Al-Maskri et al. (2010), salinity stress of sodium chloride on lettuce growth was conducted with different salinity levels. The study used salinity levels of 50 mM and 100 mM with a control of 0 mM. During the experiment, different elements such as the number of leaves, leaf area, shoot dry weight, and root fresh weight were highly affected by the salinity levels of the experiment (Al-Maskri et al. 2010). Another study conducted by Andriolo et al. (2008) established that increasing soil salinity levels from the between the electrical conductivity of lettuce resulted in a positive shoot fresh mass. As such, when the levels of electrical conductivity as a result of salinity stress were increased from 0.8 to 1.9 dS/m, the shoot fresh mass of the lettuce plant increased by 28.5 percent. On the other hand, further increase from 1.9 to 4.72 dS/m resulted

in a negative response on the shoot fresh mass (Andriolo et al., 2008). This increase in salinity stress resulted in a 16.5 percent decrease on the shoot fresh mass. Therefore, Andriolo et al. (2008) and Al-Maskri et al. (2010) show the moderate sensitivity of lettuce to different salinity stresses. In this regard, high salinity levels affect the growth of the lettuce plant while moderate salinity levels have a positive effect on the growth of the plant.

2.2.3 Adaptation mechanism of plants to soil salinity

Despite the fact that so many agricultural regions have becoming saline because of natural or human causes, plants are still able to grow in these conditions. This is because plants have different mechanisms that enable them deal with soil salinity and tolerate these harsh conditions. The three process that allow plants to tolerate saline soils are (A)ion exclusion, where Na+ and Cl- accumulation in the shoot is reduced by adjusting root ion transport, (B) ion tissue tolerance, where accumulated Na+ and Cl- in the leaf is compartmentalised within cells, and (C) shoot ion tolerance, where growth is maintained whilst under osmotic stress (Tilbrook & Roy 2013).

Plants use sensory mechanisms to enable salt tolerance, relying on sensory modalities that enhance their tolerance to saline conditions (Deinlein et al. 2014). First, these sensory modalities sense hyper-osmotic conditions and high sodium (Na⁺) levels in the soil solution (Shannon & Grieve 2000). High salt concentrations in soils enhance the development of hyperosmotic stress and pressure on the roots of the plants (Shrivastava & Kumar 2015). Hyperosmotic sensors in plants enable the plant to develop cytosolic responses when exposed to salt conditions such as sodium chloride (Deinlein et al. 2014).

Plants also have mechanisms that reduce the effects of osmotic stress (Deinlein et al. 2014). A common mechanism that plants use is enhancing the reduction of water loss by ensuring that

water uptake is maximised (Shrivastava & Kumar 2015). Therefore, plants ensure that the effects of salt stress such as sodium ions are reduced by ensuring that the sodium salts are excluded from the leaf tissues (Shannon & Grieve 2000). In this regard, plants ensure that salt exclusion is enhanced through increasing filtration activities on the root surface. As such, the root membrane facilitates blocking entry of sodium or other ions into the plant but allow water to get into the plant (Shabala & Munns 2012).

Gene regulation in roots is also another mechanism of salt tolerance in plants (Shannon & Grieve 2000). Gene regulation involves the formation of transcription factor family genes. The transcription factor is expressed differently as it reacts to different levels of salinity. Gene regulation is only possible with the action of different plant hormones such as abscisic acid, gibberelic acid, and brassinosteroids, among others. As such, gene regulation minimises lateral root elongation towards very high salt concentrations (Deinlein et al., 2014).

Additionally, chromatin modifications are also a means of plants tolerating with salinity stress (Shannon & Grieve 2000). Chromatin modification creates salt resistance for the plant in the same way the salt stress is generated (Downs 2012). Chromatin modification enhances the change of structure in histone modification and DNA methylation (Kouzarides 2007). As such, these modifications enable the regulation of gene networks that are stress responsive, especially in saline environments (Kim et al., 2015).For lettuce growth, salinity stress tolerance is achieved by the application of plant hormones such as brassinosteroids. These compounds enhance the physiological processes of the lettuce plant by enabling xylem differentiation, ethylene biosynthesis, and abiotic stress mechanisms (Downs 2012). This study by Ekcinci et al. (2012) showed that the application of brassinosteroids such as 24-EBL treatments results in the reduction of salinity effects on lettuce growth and development. This, therefore, shows the importance of such hormones in the plant. In the case of high salinity stress, the plant produces

these hormones in high amounts so that the normal processes in the plant can continue to be done (Downs 2012). These hormones also facilitate high yields in lettuce farming.

2.2.4 Salinity sensitivity of lettuce

Lettuce is one of those plants that are known to have effective mechanisms of salinity tolerance. In lettuce salinity tolerance is unlike other plants because of the lack of exclusion of sodium ions but rather enhances the inclusion of the ions in the shoot system (Bartha et al. 2015). The ability of the root and the leaves of lettuce to react differently to salt enhance the survival of this crop in saline areas. In the experiment by Bartha et al. (2015) five cultivars of lettuce of different types were subjected to different conditions of salt stress. The results of the study showed that exposure to salt stress resulted in physiological and biochemical modifications in the plant. The experiment also showed that the shoot fresh weight growth rate was reduced in all the cultivars because of high amounts of sodium chloride. Stomatal conductance was also reduced as a result of the enhanced salt stress in the plant. However, it was noted that in the experiment, the growth rate of the root was affected minimally as compared to the growth rate of the leaf. This is also in accordance with the ability of the root to withstand high salt levels as compared to the leaf (Gregory & Nortclif 2012).

The results of this study are in agreement with the results of the study by Unlukara et al. (2008) where results showed that minimum yield in lettuce plants was obtained from samples that had undergone salt treatments. On the other hand, plants in the control experiment had maximum yield. In lettuce, high salinity levels reduce the number of leaves in each plant as well as the plant height of the crop; resulting in the ultimate decrease or plant weight (Ünlükara et al. 2008). However, the number of leaves in the lettuce plant is unaffected by low salinity levels. High salinity levels of over 4.17 dS m⁻¹ reduce the number of leaves in the plant. Salinity levels have no effect on the taste of the lettuce plant (Ünlükara et al. 2008). Both studies by Bartha et al.

(2015) and Unkulara et al. (2008) agree that salinity stress on the lettuce plant affects root hydraulic conductance. This is because the toxicity of the sodium and chloride ions in root plasma reduce any activity in this area (Shainberg & Shalhevet 2012).

Herbaceous crops like lettuce have more sensitivity as compared to woody crops (FAO 2009). This is attributed to the fact that herbaceous woody crops have more complex physiology when it comes to ion-specific elements than herbaceous crops. Lettuce is considered moderately sensitive with a ECe threshold of 1.3 dS/m (Ayars et al., 1951; Bernstein et al., 1974; Osawa, 1965).

2.3 Farm management

Managing saline soils in farms is a major problem for many farmers in the world, including soil salinity. Various techniques can be applied to enhance farm management under saline conditions, depending on the cause of the salinity (Uphoff et al. 2006). In the case of irrigation-related salinity, it is important to remember that the type of irrigation that is chosen for different soils has a direct impact on the nature, rate and type of salt accumulation in the soil (Shainberg & Shalhevet 2012).

2.3.1 Irrigation as a farm management technique for salinity management

The type of irrigation applied and the amount or volume of water used in the irrigation process directly influence the nature of salt accumulation in soils (Shainberg & Shalhevet 2012). It also affects the nature of salt distribution in these salts (Downs 2012). For this reason, in many areas, the use of flood irrigation as part of the leaching process has been used as a farm management technique in enhancing. Flood irrigation is preferred as a farm management technique (Rietz & Haynes 2003). Through flood irrigation, infiltration into the soils is enhanced and the removal of salts is done. This method has been preferred by many farmers because of its ability to

remove salts below the root zone of the plant (Sharma & Minhas 2005). As such, it ensures that all the salts in the soil are removed from very deep. Other irrigation methods such as sprinkler irrigation systems also achieve the same results as the flood irrigation (Sharma & Minhas 2005).

On the other hand, other types of irrigation such as furrow irrigation have also been used as farm management practices. For furrow irrigation, the soluble salts that are present in the soil are moved through the entire wetting front (Rietz & Haynes 2003). As such soluble salts are concentrated at the point of termination and removed through constant wetting of the furrow irrigation system (Devkota et al. 2015). As such, the movement of the water through the soil is through capillary action resulting in the concentration of the salts in the intervening beds between the furrows (Sharma & Minhas 2005). Drip irrigation is also a common practice managing salinity in soils. In this type of irrigation, the soil (Sharma & Minhas 2005).

The water quality that is used for irrigation matters in enhancing the effects of the irrigation practice in the management of saline soils (Krenkel 2012). In this regard, the water that is used for irrigation should be of good quality that is considerably fresh. This means that salty water should not be used for irrigation because it increases the amount of salts in the soil instead of reducing this effect (Krenkel 2012). Irrigation water quality is enhanced by the nature of salt concentration in the water. The use of high-salt concentrated waters results in the reduction of plant moisture (Shainberg & Shalhevet 2012). This has an immediate effect on crop growth and an ultimate effect on crop yield (Rietz & Haynes 2003, Krenkel 2012). It is, therefore, important to enhance effective water management practices through irrigation to enable the effective farm management of saline soils.

2.3.2 Tillage

Tillage is another farm management technique that has been applied over years as means of enhancing crop yield. The cultivation of crops through land preparation enhances the ability of the farmers to determine different practices that are essential in ensuring that crops are grown in the best way possible and yields are enhanced (Gregory & Nortclif 2012). Some researchers such as Devkota et al. (2015) argue that tillage is not the appropriate method for managing saline salts. They explain that no-till lands have a greater chance of being management effectively for salinity reduction than tilled lands and that tillage can bring sub-surface salts to the soil surface (Devkota et al. 2015) and closer to the root zone, affecting the growth of crops by limiting water uptake (Shabala & Munns 2012). Tillage can also enable water to rise through capillary action in the soil profile and transfer soluble salts from the deep layers, and also enhances the process of drying out the soil and concentrating salts in the soil further (Qureshi et al., 2008).

For this reasons, no-till is seen as a better strategy for managing saline soils (Devkota et al. 2015). It is recommended that for lettuce growth, the seedbed preparation should be the only tilling to be done (Munns & Tester 2008). This will enable the plant to enhance its mechanism of survival from an early point and reduce soil salinity stress on the growth of the lettuce plant. The reduction in tillage is less likely to be a problem for weed infestation in a short-season crop such as lettuce (Kristiansen et al. 2008).

2.4 Effects of fertilisers and soil amendments on soil salinity

The process of changing soil properties for enhanced soil growth is referred to as soil amendment (Uphoff et al. 2006). Soil amendments can either be organic or inorganic (Pardo et al. 2014). Organic soil amendments are considered to be the most effective because they change a number of physical properties in the soil (Kristiansen et al., 2008). Organic amendments

include the use of compost, manure, humus, sphagnum peat moss, sewerage sludge, among others, to change the physical nature of the soil for better properties for plant growth (Tejada et al. 2006, Schulz & Glaser 2012). On the other hand, inorganic amendments include the use of lime, perlite, vermiculite, and sulphur, among others as means of changing the pH of the soil (Pardo et al. 2014). The use of these organic and inorganic soil amendments can be applied on different types of soil to modify soil properties and functions by changing their physical, biological and chemical properties (Lehmann & Joseph 2015).

2.4.1 Organic soil amendments

Organic soil amendments are important for limiting or rectifying soil salinity. In this regard, these soil amendments may be preferred because of their ability to improve soil properties without causing significant longer-term side effects to the soil (Pardo et al., 2014). Organic soil amendments reduce soil compaction (Uphoff et al. 2006), enable soil to become more aerated (Downs 2012), improve soil moisture retention thus diluting salt levels and electrical conductivity (Lakhdar et al. 2010, Gregory & Nortclif 2012) and increase water filtration (Tejada et al., 2006). These largely physical changes increase NaCl leaching (Lakhdar et al. 2010).Organic amendments also enhance the stimulation of dehydrogenase activity in the soil, increasing microbial activity (Lakhdar et al. 2010). Lakhdar et al. (2010) used municipal solid waste and sewage sludge as a means of reducing soil salinity. The study showed that these organic amendments reduced soil pH and enhanced enzymatic activities within the soil lower soil salt levels.

2.4.2 Gypsum and its modifying effects on soil salinity

Fertiliser management requires caution in saline soils because of the high concentration of soluble salts that mineral fertilisers contain (Gregory & Nortclif 2012). The high concentration of these salts change soil structure (Deinlein et al. 2014) and reduce soil biological activity

(Hasbullah & Marschner 2015). Gypsum (calcium sulphate dehydrate)has been found as an effective fertiliser for modifying saline affected soils (Kordlaghari & Rowell 2006), having been used by farmers for years to manage soil and increase crop yield (Abdel-Fattah 2012).Gypsum enhances soil infiltration (Kordlaghari & Rowell 2006) and improves porosity and drainage (Gregory & Nortclif 2012). This is especially important in saline soils as infiltration increases the leaching of salts irrigation or rain water (Abdel-Fattah 2012).

Gypsum can be applied in soils that are highly affected by sodium salts. Sodium ions in the soils react with Ca⁺⁺ ions in the gypsum by replacing Na⁺ in the soils (Abdel-Fattah 2012). Gypsum is a source of calcium and sulphur which are important nutrients that can increase plant growth (Abdel-Fattah 2012), including fruit development (Locascio & Hochmuth 2002) and roots of crops such as lettuce (Chutichudet et al. 2009). The application of gypsum in lettuce production is done before sowing so that the effects of gypsum can occur and the soluble salts released do not affect subsequent lettuce growth (Chutichudet et al., 2009).

2.4.3 Biochar as a soil amendment strategy for managing salinity effects

Biochar is made from waste biomass under a limited supply of oxygen (Lazcano et al. 2013). It is carbon-rich and extremely porous (Woolf et al. 2010). Biochar has been used in different parts of the world as a soil amendment strategy for various functions, including managing soil salinity (Lehmann & Joseph 2015). Biochar's chemical and physical characteristics enable it to absorb nutrients, salts and pollutants from the soils (Lazcano et al. 2013) and improves water holding capacity (Lehmann & Joseph 2015) and soil microbial activity (Woolf et al. 2010).

As a soil amendment technique, the application of biochar has been effective in enhancing the reduction of leaching processes especially in sandy soils (Schulz & Glaser 2012). This is because biochar is effective in enhancing soil moisture content and improving soil texture for

plant growth in sandy soils. Through this, water quality application is enhanced in the soils (Woolf et al. 2010). The reduction of soil acidity is an important element in the application of biochar. This is because it ensures that the soil can withstand different plant systemic responses that enhance the immunity of the plant especially to diseases that are caused and enhanced by soil salinity (Lehmann & Joseph 2015). The implementation of biochar in amending saline soils is dependent on the amounts of biochar applied in these soils (Woolf et al. 2010).

Biochar may be spread on the land according to the required levels and percentages for the land (Woolf et al. 2010). It can also be applied on the soil surface and the soil covered with other organic materials to enhance the properties and functions of biochar (Schulz & Glaser 2012). On saline soils it can be mixed with compost on the land. In other places, it can be applied when it is made as liquid slurry specifically in large scale biochar application. Compost and biochar have a synergistic relationship in soil (Lazcano et al. 2013). The application of compost with biochar results in the increased ability of biochar to enhance microbial activity in saline soils enhancing nutrient retention (Schulz & Glaser 2012), and moderates soil pH (Woolf et al. 2010).

Biochar enhances the soil amendment of saline soils by increasing soil organic matter as discussed above especially through compost application. Soil organic matter in increased in the soil due to the ability of biochar to ensure that the amounts of carbon in the biochar have been introduced into the soil (Woolf et al. 2010). The introduction of carbon into the soils activates the exchange of cations that enhance and neutralise the effect of electrical conduction in the soil. Studies have revealed that the application of biochar results in a high availability of phosphorus in the soil. This is attributed to the ability of the biochar to enhance a high retention of nutrients (Zheng et al. 2010). Additionally, the increased amounts of phosphorous means that a reduction of nitrate salts are enhanced. The reduction of these nitrate salts in the soil is

attributed to the ability of the biochar to absorb nitrogen fertilisers and inhibit nitrification (Woolf et al. 2010).

The beneficial effect of biochar on the growth of lettuce is also evident in the literature. Woolf et al. (2010) tested whether biochar reduced the stress of salt in the soil and whether the biochar was efficient in interacting with mycorrhizal fungi. The study showed that biochar reduced salt stress in vegetable crops in saline soils. Biochar interacted with mycorrhizal fungi to reduce the negative impacts of soil salinity and increased the crop growth. Upadhyay et al. (2014) also reported that biochar improved the growth of lettuce in saline soils. The experiment found an increase in pH and electrical conductivity as the rate of biochar application increased (Upadhay et al., 2014).

2.4.4 Leaching as a soil amendment strategy

Leaching is an important element in the management of saline soils. This is attributed to its ability of removing salts in the soils from the root zones (Barnard et al. 2010). This is the process where water applied to the soil surface in a ponding process to allow it to infiltrate into the soil slowly. The main condition of leaching saline soils is that heavily saline soils are treated through washing out (Sharma & Minhas 2005). This means that for soils that are heavily affected by salinity, flood irrigation is proposed (Barnard et al. 2010). It must be noted, however, that leaching may only be effective when there are drainage channels in the farm to discharge the salty water away from the farm (Krenkel 2012). Without discharging this water, the salt remains in the soil resulting in the contamination of groundwater and further damage to the soil. However, where there is adequate natural drainage, there is no need for surface discharging of the water.

Leaching is done to soils whose soil moisture content is very low. When the soil moisture content of land is high, the water will be unable to infiltrate and carry of salts from the soils (Podmore 2009). It is also preferred in porous soils that enhance effective water infiltration from the soils. The process of leaching should also be done in soils where there is a deep water table (Shainberg & Shalhevet 2012) as the deep water table will ensure that the leaching process is done without raising the water table and the leachate moves well beyond the root zone of crops.

For saline soils, leaching is done at times when temperatures are relatively low like in the late autumn, when evaporation rates are minimal, and, in areas with low rainfall in summer, it is also the time when the groundwater level is also low (Shainberg & Shalhevet 2012). Such times provide the best conditions for the leaching process so that maximum effects can be obtained. The main condition of leaching saline soils is that heavily saline soils are treated through washing out (Sharma & Minhas 2005). This means that for soils that are heavily affected by salinity, flood irrigation is proposed (Barnard et al. 2010) as it can ensure that salts are dissolved and the water containing these salts is drained to lower soil horizons and to the ground water.

The manner in which saline soils are remediated by leaching depends on the chemical and physical properties of the soil, and the extent of the salinity in the soil (Barnard et al., 2010). The ratio of soluble salts of calcium and sodium ions that are present in the soil determines the degree of salinity (Shainberg & Shalhevet 2012). Soils that are highly affected by calcium are easily amended by leaching. On the other hand, soils that are highly affected by chloride salts are more complex in that the physical properties of the soil are altered in order for successful leaching to be done (Devkota et al. 2015).

Chapter 2. Literature Review

2.5 Conclusions

In summary, soil salinity has affected the growth and yields of crops over many years. Salinity in soils can occur naturally or through human-made processes such as irrigation practices. Soil salinity limits the growth of crops by reducing the amount of nutrients and water that the plant can take up and enhancing the development of diseases in these crops. In lettuce, soil salinity limits the ability of the plant to develop in terms of shoot and root growth. Under particularly adverse conditions, the effects of salinity can lead to plant death. There is a need to enhance effective farm management techniques to limit the negative impacts of salinity on the growth and development of the crops. For this reason, irrigation practices such as flood irrigation have been enhanced in areas where soil salinity severe. In other areas, tillage practices have been improved to address this issue, though there is some evidence that minimal tillage practices lead to lower salinity impacts. Soil amendment strategies such as leaching, organic fertiliser application, adding gypsum, and biochar application have been reported to reduce the effects of soil salinity on crops.

In terms of knowledge gaps, there are various issues that need further research when it comes to information about soil salinity, its effects on the growth of different plants and options for management. A key information gap is related to the effects of soil amendments and their application in different conditions to reduce salinity impacts. Currently, the information that exists identifies the types of soil amendments that are present in the market. However, there is a gap in the provision of information about modifying different soil properties using different types of soil amendment strategies. Further research is also needed regarding the types of amendments that are suited to different environmental conditions. Such information will enable recommendations for soil amendments to become more reliable and applicable to farmers who continue to lose crop yields because of saline soils.

Based on these information gaps, it is evident that there needs to be further research done on this topic to enhance soil salinity management practices. In this regard, further research needs to be done on the effects of saline soils on the growth of lettuce on amended soils. Some studies have suggested that the processes of soil amendment may be temporary and salinity may appear again over time. As such, research needs to establish the conditions in which lettuce will grow effectively once the soil amendments have been done on saline soils. Further research also needs to be done on the debate about the impacts of no-till practices on soil salinity. There is also need to carry out research on other farm management practices apart from tillage and irrigation that can be demonstrated to farmers so that they can address the issue of soil salinity and have better crop yields. Research on these issues will enable farmers, students, organisations, and governments to have more information about the practicalities of saline soils management and their effects on plant growth.

3 Methods

3.1 Experiment 1a: Effect of biochar on lettuce growth under saline conditions

This study was conducted at the School of Environmental and Rural Sciences, University of New England (UNE) from 26/10/2016 to 1/12/2016, under glasshouse conditions to study the effect of biochar in modifying soil and irrigation salinity effects on lettuce (*Lactuca sativa* L.) growth. Three treatments were combined in a full three-way multifactorial trial with (a) biochar, (b) soil salinity and (c) irrigation salinity used at various rates. The levels selected were based on common rates reported in the literature for biochar (Lehmann & Joseph 2015), and soil and irrigation salinity toxicity in lettuce (Unlükara, et al., 2008).

A sandy loam soil was collected from a nearby UNE farm (30.44 S, 151.67 E, 1,013 m elevation) and prepared by sieving through a 2 mm sieve and air-drying until the weigh was stable. The initial salinity of soil was measured by the electrical resistance of a 1:5 soil: water suspension. Ten grams of dry soil was weighed, 50 ml of deionised water added and the mixture was mechanically shaken (end over end) in a closed system for 1 hour at 25 °C. The mixture was allowed to settle for 30 minutes and the electrical conductivity (EC) was measured using EC meter (TPS Pty Ltd, Model labCHEM cond.TDS and Temp). The pH of the mixture was also measured using a pH meter (TPS Pty Ltd Model 901-CP). Texture components had previously been measured by the UNE laboratory staff (Table 1). Tap water (EC of 0.307 dS/m) was used throughout the experiment

Table 1. Characterisation of son used in Experiment 1a and 1b.				
Soil characteristic	Value			
Sand	83.5%			
Clay	10.5%			
Loam	6.0%			
Electrical conductivity	0.034 ds/m			
рН	7.93			

Table 1. Characterisation of soil used in Experiment 1a and 1b.

3.1.1 Experimental design and preparation of the treatments

There were five replicates of each treatment and the 60 pots (3 biochar levels \times 2 soil salinity levels \times 2 irrigation salinity levels \times 5 replicates) were laid out in a fully randomised design. Three levels of biochar (0, 5, 10 g/kg) were added to the soil prior to beginning the experiment. The biochar was made from coconut carbon and supplied by Filchem Australia Pty Limited. The appropriate weight of biochar was added to separate containers of soil and all containers (including the zero biochar control) equally stirred to fully incorporate the biochar.

Two levels of soil salinity (0, 3.0 dS/m) were produced in the soil by adding 0 or 1% weight/weight of NaCl (i.e. 0.07 g/kg) to the soil. All soil containers (including the control) equally stirred to fully incorporate the NaCl.

Two levels of irrigation water salinity (0, 2.4 dS/m) were applied after planting the lettuce seedlings. The saline irrigation water was prepared by adding 10.1 g NaCl to 10 L water. This solution and the control solution were kept in sealed containers during the experiment and the both containers were fully stirred during to each irrigation event.

3.1.2 Experimental conditions

The experiment was conducted in a glasshouse set to a diurnal temperature range of 15 - 25 °C. Plastic pots (130 mm diameter and 10.5 mm depth) were lined with paper towel to prevent soil loss and filled with 500 g of soil treated with biochar and/or NaCl and labelled. Half of the pots were subsequently watered with plain water and half with saline water. Small trays were placed under each pot and leached water was returned to the pots.

Lettuce seedlings (about 4 weeks old) were bought from a commercial supplier and planted individually (Figure 1). Each pot was irrigated at a rate of 100 mL of plain or saline water every two days for the first week, then daily after that. Once a week, all pots were fertilised using a solution of Aquasol (23.0% nitrogen, 23.95% phosphorus, 14.0% potassium) added at a rate of 0.1 g / 100 ml to the plain and saline irrigation water.

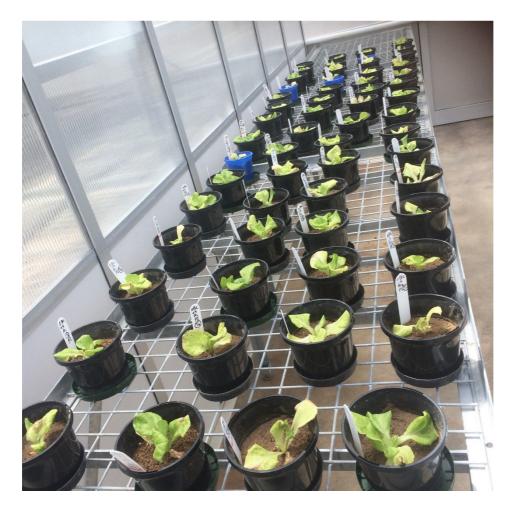


Figure 1. Experimental set-up in the glasshouse.

3.1.3 Measurements

A 'health index' was measured every week for five weeks. The visual health index was a ranking from 1 to 4, based on the colour (green, yellow, greenish yellow and dead) of the foliage. At five weeks after transplanting, the lettuces were harvest by removing them from the soil, washing thoroughly and cutting them into shoot and root portions. The samples were dried in an oven at 40 °C for 48 hours and the shoot and root dry weight was recorded.

3.2 Experiment 1b: Effect of biochar on lettuce growth under saline conditions

3.2.1 Experimental treatments and conditions

The results for Experiment 1a were possibly not reliable as the control treatment grew poorly, limiting the ability of the trial to detect further growth suppression due to (soil and/or irrigation) salinity. The pots may have lacked sufficient watering towards the latter part of the experiment. Therefore the experiment was repeated, using the same treatments and under the same growing conditions, with more careful attention to plant husbandry. Experiment 1b was carried out from 10/3/2017 to 5/5/2017. Three treatments were combined in a full three-way multifactorial trial with (a) biochar, (b) soil salinity and (c) irrigation salinity. The levels selected were based on those used in Experiment 1a.

3.2.2 Measurements

Lettuce height, leaf chlorophyll concentration (Minolta SPAD-502) was measured at harvest (five weeks after transplanting). The lettuces were then cut at ground level and the shoot dry weight determined after drying the samples in an oven at 40 °C for 48 hours. A 50 ml sample of soil was also collected and the EC measured using the method described for Experiment 1a.

3.3 Experiment 2: Effect of biochar on salt leaching

This study was conducted at the School of Environmental and Rural Sciences, University of New England (UNE) from 15/12/2016 to 12/1/2017, under glasshouse conditions to study the effect of biochar on salt leaching. Three treatments were combined in a complete three-way multifactorial trial with (a) biochar, (b) soil salinity and (c) irrigation salinity used at various rates. The levels selected were based on those used in Experiment 1.

A sandy loam soil was collected from a nearby UNE farm (30.44 S, 151.67 E, 1,013 m elevation) and the EC (as for Experiment 1b) and pH measured (pH meter – Model 901-CP). Texture components had previously been measured by the UNE laboratory (Table 1). Tap water (EC of 0.307 ds/m) was used throughout the experiment.

3.3.1 Preparation of the treatments

Three levels of biochar (0, 5, 10 g/kg soil) were added to the soil prior to beginning the experiment. The biochar was made from coconut carbon and supplied by (Filchem Australia Pty Limited). The appropriate weight of biochar was added to separate containers of soil and all containers (including the zero biochar control) equally stirred to fully incorporate the biochar. Two levels of soil salinity (0, 3.0 dS/m) were produced in the soil by adding 0 or 1% weight/weight of NaCl (i.e. 0.03 g/kg) to the soil. All soil containers (including the control) equally stirred to fully incorporate the NaCl.

Two levels of irrigation water salinity (0, 2.4 dS/m) were applied after put the soil in the pot. The saline irrigation water was prepared by adding 22 g NaCl to 22 L water. This solution and the control solution were kept in sealed containers during the experiment and the both containers were fully stirred during to each irrigation event.

3.3.2 Experimental design

There were four replicates of each treatment and the 48 pots (3 biochar levels \times 2 soil salinity levels \times 2 irrigation salinity levels \times 4 replicates) were laid out in a fully randomised design in a glasshouse set to a diurnal temperature range of 15 – 25 °C. Plastic pots (8 mm diameter and 13 mm depth) were lined with paper towel to prevent soil loss and filled with 2.56 g of soil treated with biochar and/or NaCl and labelled. Half of the pots were subsequently watered with plain water and half with saline water. Small trays were placed under each pot to collect leaching water. Each pot was irrigated at a rate of 100 mL of plain or saline water every two days for the first week, then daily after that. EC and volume was measured daily in the lab. After 24 days, each pot was divided for three levels, top, middle and bottom.

3.4 Statistical analyses

For all experiments, statistical analyses were carried out with an analysis of variance (ANOVA) using the R program version 3.2.2 (R Core Team 2017). Assumptions of heterogeneous variances and normal distributions were confirmed or log transformations were carried out to stabilise the data. Data were tested for significant differences at P = 0.05. Significantly different means were separated using 95% confidence limits (1.96 × standard errors) (Afshartous & Preston 2010). Linear regression was also used to assess the relationship between soil electrical conductivity (EC) and plant height, chlorophyll and shoot biomass. In Experiment 2, the variation of EC over time was summarised using the loess function in R to produce a non-parametric, locally smoothed curve.

4 Results

4.1 Experiment 1a: Effect of biochar on lettuce growth under saline conditions

4.1.1 Shoot dry weight

Lettuce shoot dry weight was significantly affected by irrigation salinity, soil salinity and biochar (P < 0.024), and interaction of soil salinity and biochar (P < 0.001) (Figure 2). Adding BC improved lettuce growth under non-saline conditions (SS 0 + IS 0), and with only saline irrigation water (SS 0 + IS 2.4) or only saline soil (SS 3 + IS 0). With saline soil and irrigation water (SS 3 + IS 2.4), lettuce growth was reduced for the BC 0 and BC 10, but not different for the BC 5 treatment.

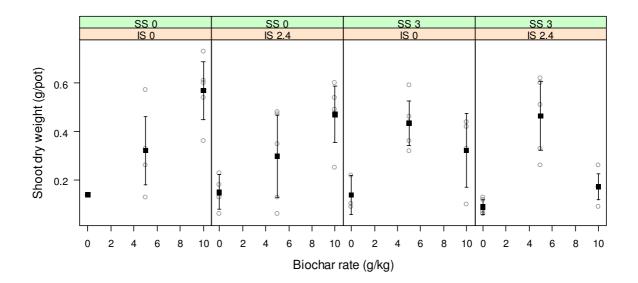


Figure 2. Effect of biochar, soil salinity (SS: 0 or 3 dS/m) and irrigation salinity (IS: 0 or 2.4 dS/m) lettuce shoot dry weight. Circles show the raw data, the black squares are the treatment means and the error bars are the 95% confidence intervals.

4.1.2 Root dry weight

Lettuce root dry weight was significantly affected by irrigation salinity, soil salinity and biochar (P < 0.042), and interaction of soil salinity and biochar (P = 0.047) (Figure 3). The response

patterns were similar to that of shoot dry weight, except that the inhibition of growth by the two salinity treatments was stronger.

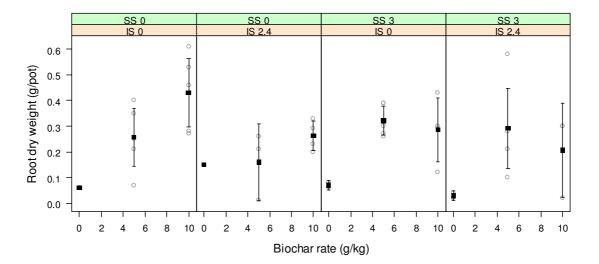


Figure 3. Effect of biochar, soil salinity (SS: 0 or 3 dS/m) and irrigation salinity (IS: 0 or 2.4 dS/m) lettuce root dry weight. Circles show the raw data, the black squares are the treatment means and the error bars are the 95% confidence intervals.

4.1.3 Health index

The health index (HI) decreased over time for most treatments (P < 0.001), including the control. While the soil and irrigation salinity treatments were not significant (P > 0.236), increasing the application rate of biochar improved the HI (P < 0.001). Biochar and soil salinity interacted (P < 0.001), with the BC0 and BC5 having no difference in control or saline soils, but the high rate of biochar having a lower HI in saline soil.

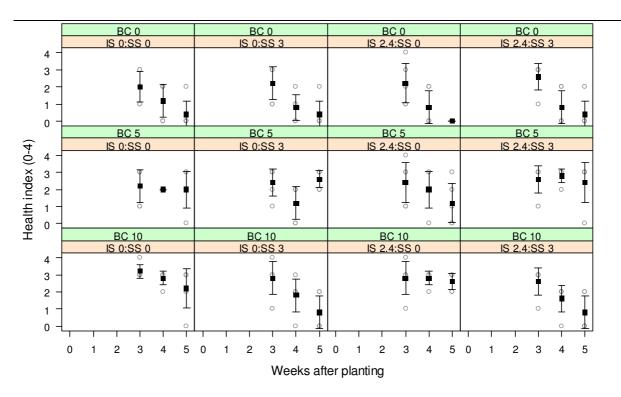


Figure 4. Effect of biochar (BC: 0, 5 or 10 g biochar per kg soil), irrigation salinity (IS: 0 or 2.4 dS/m) and soil salinity (SS: 0 or 3 dS/m) on lettuce leaf health over time. Circles show the raw data, the black squares are the treatment means and the error bars are the 95% confidence intervals.

4.2 Experiment 1b: Effect of biochar on lettuce growth under saline conditions

4.2.1 Plant height

The main terms (biochar, irrigation salinity and soil salinity) and their three-way interaction were all significant ($P \le 0.027$) for plant height (Figure 5). The effect of BC was no difference by using levels of biochar with no salt irrigation and no salt soil and no difference in SS 0 with SS 0 and IS 0, but BC 10 was better than BC0, BC5 in the SS 3. IS and Combined there is difference and BC 10 helps, but BC 5 did not.

Chapter 4. Results

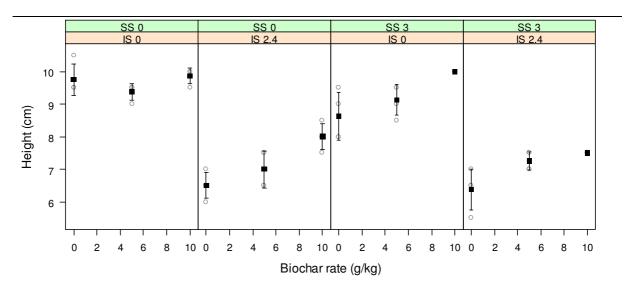


Figure 5. Effect of biochar (BC: 0, 5 and 10 g/kg), soil salinity (SS: 0 and 3 dS/m) and irrigation water salinity (IS: 0 and 2.4 dS/m) on plant height. Circles show the raw data, the black square is the mean, the error bars show the 95% confidence limits.

4.2.2 Chlorophyll

Figure 6 shows there was no significant effect of biochar (P = 0.766) on leaf chlorophyll leaves, but irrigation salinity (P < 0.001) and soil salinity (P < 0.001) IS reduced chlorophyll by about 40%, SS by about 10%, and the combined salinity treatment by about 50%.

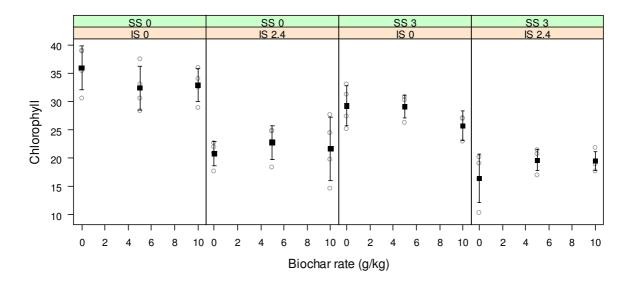


Figure 6. Effect of biochar (BC: 0, 5 and 10 g/kg), soil salinity (SS: 0 and 3 dS/m) and irrigation water salinity (IS: 0 and 2.4 dS/m) on leaf chlorophyll. Circles show the raw data, the black square is the mean, the error bars show the 95% confidence limits.

4.2.3 Shoot dry weight

The main terms (biochar, irrigation salinity and soil salinity) and their three-way interaction were all significant ($P \le 0.027$) for shoot dry weight (Figure 7). The response patterns were similar to those reported for plant height.

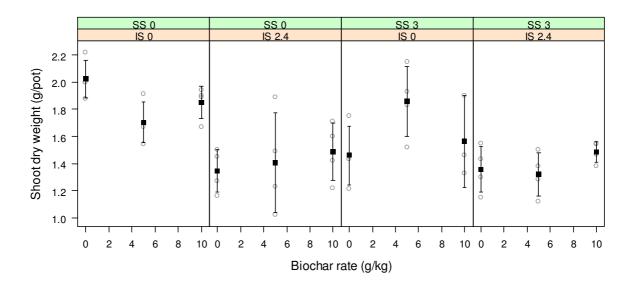


Figure 7. Effect of biochar (BC: 0, 5 and 10 g/kg), soil salinity (SS: 0 and 3 dS/m) and irrigation water salinity (IS: 0 and 2.4 dS/m) on shoot dry weight. Circles show the raw data, the black square is the mean, the error bars show the 95% confidence limits.

4.2.4 Soil electrical conductivity

All terms were significant ($P \le 0.047$), except the IS×BC interaction (P = 0.179) (Figure 8). Soil EC was lower at higher rates of biochar in the non-saline treatment (SS 0 + IS 0) and the high salinity treatment (SS 3 + IS 2.4).

Chapter 4. Results

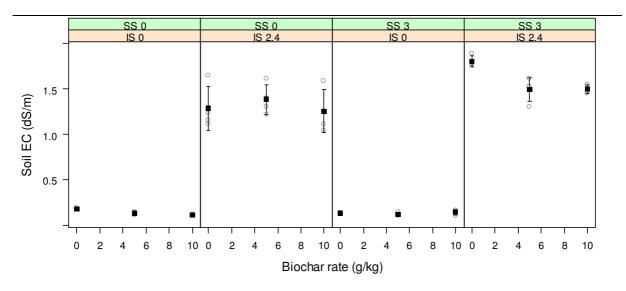


Figure 8. Effect of biochar (BC: 0, 5 and 10 g/kg), soil salinity (SS: 0 and 3 dS/m) and irrigation water salinity (IS: 0 and 2.4 dS/m) on soil electrical conductivity (EC). Circles show the raw data, the black square is the mean, the error bars show the 95% confidence limits.

4.3 Experiment 2: Effect of biochar on salt leaching

4.3.1 Leachate EC over time

The top row of plots in Figure 9 show that, with no salinity added in salt or irrigation, the EC did not change with increasing biochar levels. This indicates that there were no significant inputs of salt from biochar itself. The responses for the irrigation salinity treatment were different, and were plotted separately so that the changes in leachate EC are easier to visualise (see Figure 11 and Figure 10).

Chapter 4. Results

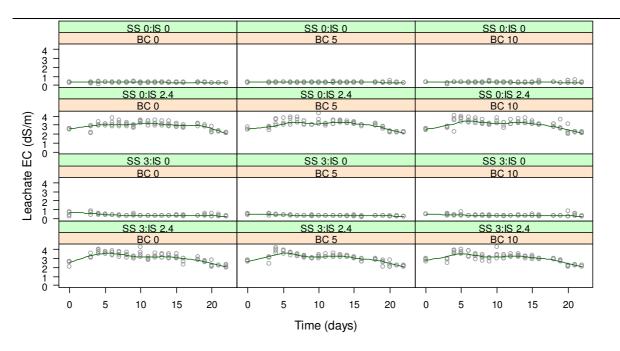


Figure 9. Effect of biochar (BC: 0, 5 and 10 g/kg), soil salinity (SS: 0 and 3 dS/m) and irrigation water salinity (IS: 0 and 2.4 dS/m) on leachate electrical conductivity (EC) over time. Circles show the raw data and the line is a local smoothing curve (loess).

Figure 10 shows there was no effect on leachate EC over time at different BC levels with saline irrigation.

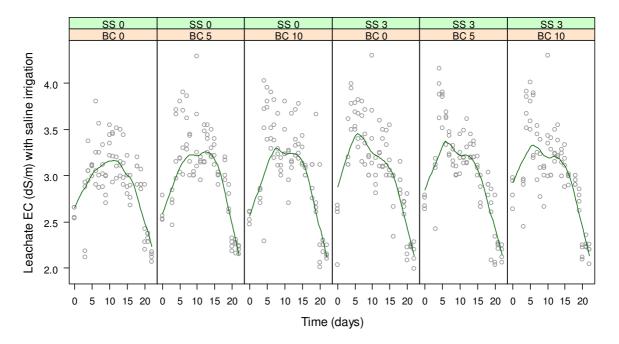


Figure 10. Effect of biochar (BC: 0, 5 and 10 g/kg) and soil salinity (SS: 0 and 3 dS/m) on leachate electrical conductivity (EC) over time using saline irrigation water (2.4 dS/m). Circles show the raw data and the line is a local smoothing curve (loess).

Without saline irrigation, leachate EC was considerably lower, and it declined over time. In the non-saline soil treatment (SS 0), this decline did not differ at increasing rates of biochar. In the saline soil (SS 3) treatment, BC 5 and BC 10 reduced the initial EC levels up to about Day 5, after which they were similar.

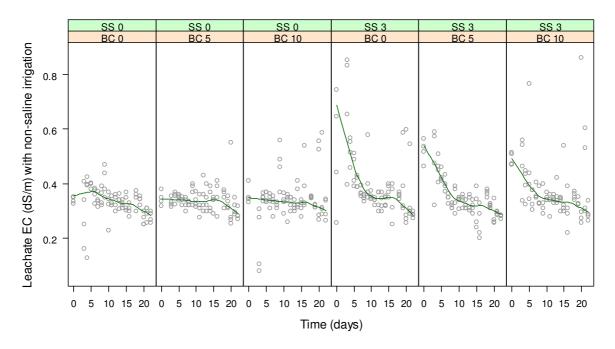


Figure 11. Effect of biochar (BC: 0, 5 and 10 g/kg) and soil salinity (SS: 0 and 3 dS/m) on leachate electrical conductivity (EC) over time using non-saline irrigation water (0 dS/m).

4.3.2 Leachate volume over time

The variation in leachate volume over time was unaffected by all the treatment combinations

(Figure 12).

Chapter 4. Results

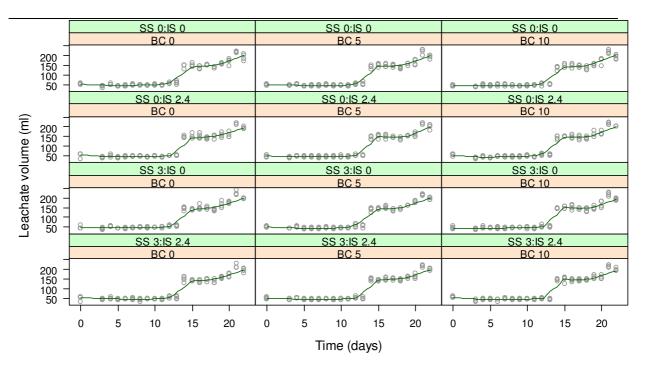


Figure 12. Effect of biochar (BC: 0, 5 and 10 g/kg), soil salinity (SS: 0 and 3 dS/m) and irrigation water salinity (IS: 0 and 2.4 dS/m) on leachate volume over time.

5 Discussion

Given the widespread occurrence of soil salinity in Australia and globally, and the increasing pressure on maximising productivity and efficiency per unit of land, there is a need to identify methods of enabling plant production in sub-optimal soils such as those with excess NaCl. Saline soils can managed using a range of techniques, including manipulating the plant's physiology to resist or tolerate salt uptake, and remediating the soil by removing the salt (e.g. leaching) or modifying the soil composition so that the salt does not damage the plants (Qadir et al. 2000, Kamaluldeen et al. 2014). In regard to modifying the soil, biochar has also been reported to ameliorate the impacts of salinity, although the specific mechanisms depend on the biochar, application rate and the soil type (Woolf et al. 2010, Saifullah et al. 2018).

This study aimed to investigate the impact of adding biochar to soils with a range of salinity levels. The salinity levels were imposed by adding salt to soil directly (soil salinity) and in the irrigation water (irrigation salinity). Lettuce was used as the test plant as it is known to be moderately sensitive to salinity, with easily recognised symptoms including stunted growth and necrotic lesions (Ünlükara et al. 2008).

5.1 Effect of biochar on lettuce growth under saline conditions

The results for Experiment 1 were not considered not reliable as the control treatment (nonsaline soil and irrigation water) grew poorly. Therefore, the ability of the experiments to demonstrate the suppression of growth due to salinity may have been limited. It was assumed that the pots have lacked sufficient watering towards the latter part of the experiment. Nonetheless, the results from Experiment 1 showed that increasing biochar rates improved lettuce growth and that the salinity treatments impaired lettuce growth, especially soil salinity. The beneficial effect of biochar is widely reported (Nieto et al. 2016, Trupiano et al. 2017), as are the negative effect of salinity. However the interactions indicated that adding biochar improved lettuce shoot growth with salinity irrigation or saline soil, but not both. Root dry weight was affected in the same way, but the inhibition of growth due to the two salinity treatments was more pronounced. The health index showed that the high rate of biochar resulted in the lettuce plants having less inhibition or damage in saline soil.

Experiment 2 was conducted as a repetition of Experiment 1. It showed similar results as those observed in Experiment 1, with a clearly demonstration of the beneficial effects of biochar, especially for plant height and leaf chlorophyll.

These results are consistent with other reports about lettuce and biochar in saline soils, and may be due to changes in soil pH, microbial activity and function and organic carbon levels (Woolf et al. 2010, Frimpong et al. 2016). Effects on plant physiology (e.g. transpiration, stomatal conductance) may also have occurred (Bartha et al. 2015), but Trupiano et al. (2017) reported that these plant processes did not show any variation in response to biochar additions in saline soils.

5.2 Effect of biochar on salt leaching

The leaching experiment showed that the EC did not change with increasing biochar levels, confirming that the biochar itself did not add significant inputs of salt to the soil column. Where saline irrigation was applied (with and without soil salinity), biochar had no effect on leachate EC over time. In the saline soil treatment, biochar reduced the leachate EC initially (up to about Day 5) only, probably because of binding of the ions (Zheng et al. 2013). In practice, these EC

levels are biologically relatively low (Beltrão et al. 1997). However, although they may become important over time, with regular biochar additions and where salt inputs to the soil are of concern (Qadir et al. 2000). The upper soil layer had about two times higher salinity than the middle and lower layers.

Leachate volume did not vary over time in any of the treatment combinations. This contrasts with many reports of biochar increasing the water holding capacity of soils (Nieto et al. 2016, Haider et al. 2017). This result may be a genuine response showing an inherent lack of effectiveness by biochar. However, other explanations for the lack of effect on leachate volumes include the very sandy soil used in the trial, insufficient water volumes applied to the soil column, or inadequate biochar application rates.

5.3 Conclusion

The experiments reported here demonstrated some benefits from biochar to plant growth in saline soils, especially at higher application rates (10 g/kg soil). The specific mechanisms were not evaluated in detail, although it was apparent that removal of salt through leaching may play a role. A recent review on the application of biochar for remediating of salt-affected soils, Saifullah et al. (2018) note that uncertainty remains about the effect of biochar on the properties of salt-affected soils. They recommend that further intensive studies are needed to explore the mechanisms or processes by which biochar could influence plant growth under saline conditions.

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