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Thesis

Submitted in Fulfillment of the Requirement for the Degree of

Doctor of Philosophy in Soil Science

(Soil Fertility and Plant Nutrition)

Nutrient Availability and Wheat Growth as Affected by Plant Residues and Inorganic Fertilizers in Saline Soils

Soil and Land Systems Earth and Environmental Sciences The University of Adelaide

2008

IN THE NAME OF GOD

GRACIOUS AND MERCIFUL

DEDICATION

The thesis is dedicated to my father, who has been a great source of motivation and inspiration. It is also dedicated to my mother, who gave love and support.

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Declaration

This thesis contains no material, which has been accepted for the reward of any other degree or diploma in any other university, and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where references have been made in the text.

I give contest to copy of thesis, when deposited in the University Library, being available for loan and photocopying.

Ahmed ELGHARABLY

Acknowledgement

I would like to extend my thanks for my supervisors Dr Petra Marschner and Dr Pichu Rengasamy, for their guidance and suggestions through out the course of my study. I am quite grateful for Dr Ed Barrett-Lennard for critical examination of the material and the useful suggestions that helped in the interpretation of the results.

I also would like to acknowledge the Egyptian Government for the scholarship given to me during my studies.

Last, but not least, I express my gratitude for my family for their encouragement and support.

Abstract

Over 10% of the world's land is salt affected. Salt accumulation is a major soil constraint for agricultural sustainability in arable or newly cultivated soils. As a result of salinity, soil chemical, physical and biological properties deteriorate, plant uptake of water and nutrients, particularly P, decreases and plant growth declines. Application of plant residues can enhance the activity of soil microorganisms, the availability of nutrients, including P and the plant uptake of P and growth. Such a practice can also be economically viable as it can reduce the use of P from inorganic sources, maintaining the world's reserve of P rocks and reducing the price of fertilizers and the environmental pollution often associated with the excessive application of inorganic N and P fertilizers. Little is known about how P, with N in proper form, added from inorganic and/or residue sources can affect wheat growth in the salt affected soils with no confounding pH or sodium adsorption ratio (SAR). Increasing microbial activity, N and P availability and wheat uptake of P by application of N and P from organic and inorganic sources may improve wheat growth and hence productivity under saline conditions. The overall aim of this study was to determine ways for enhancing the activity of microorganisms and increasing the availability of N and P, the uptake of nutrients, particularly P and the growth of wheat by management of fertilization from inorganic and organic sources in saline soils. This study therefore was conducted with the following aims: 1) to investigate the relationship between salinity and P availability; 2) to assess wheat response to combined application of N and P fertilizers under saline conditions; 3) to evaluate the effect of plant residue addition on N and P availability and microbial activity in salt affected soils; 4) to determine microbial response to addition of inorganic N rate and form, and how this will affect N and P availability in a saline soil, and 5) to determine the effect of P added from inorganic fertilizer and plant residue, compared to inorganic P fertilization, on microbial biomass and wheat nutrient composition and growth in a saline soil.

In saline soils, P availability can be affected by the salt type and concentration and soil texture. Three experiments were conducted to study the relationship between P availability, soil texture and salinity. The results of the first experiment in which soil was shaken with different concentrations of NaCl or CaCl₂ or Na₂SO₄, indicated that P solubility decreased with increasing concentration of Ca²⁺, but was not affected by Na⁺ salts. In the second experiment, P availability (after 24h shaking) decreased with

increasing salt concentration up to $EC_{1:5}$ 3.1 dS m⁻¹, increased with increasing P addition (0, 100, 200, 400, 600, 1200, 2500 and 5000 µg P g⁻¹ soil), and was generally higher in sandy soil than in sandy loam soil. In the third experiment (15 days incubation), it was found that P availability significantly decreased one day after P addition which was followed by a further decrease to day 5, but then remained unchanged until day 15. It can be concluded that P availability is reduced in presence of clay, and decreases with increasing concentration of salts, particularly Ca²⁺, and that the availability of P stabilizes in sandy and sandy loam soils within 2 weeks after addition of P from inorganic source.

Increasing N or P fertilization enhanced wheat growth in salt affected soils. Therefore combined application of N and P may enhance wheat growth in saline-non sodic soils with neutral pH. Three glasshouse experiments were carried out with the aim to determine the salinity range to be used in the subsequent experiments and to test the hypothesis that combined addition of N and P fertilizers can enhance wheat growth in a sandy loam soil with low SAR and neutral pH. The first two experiments were conducted in a sandy loam salinized to $EC_{1:5}$ of 0.18, 1.36, 2.00 and 2.67 dS m⁻¹ using NaCl and CaCl₂. The wheat varieties Janz and Krichauff died in all soils to which salt was added showing that these EC levels were too high. The third experiment was conducted with Krichauff in the sandy loam soil with EC_{1:5} 0.19, 0.32, 0.49, 0.67 and 0.86 dS m⁻¹, equivalent to EC_e 2.2, 4.4, 6.7, 9.2 and 11.8 dS m⁻¹, respectively, and with 0, 30 and 60 mg P kg⁻¹ soil and 50, 100 and 200 mg N kg⁻¹ soil. Salinity reduced plant dry matter at all N and P application rates. Increasing N application rates decreased growth at low and high salinity, whereas increasing P addition improved growth at all salinity levels. The highest shoot and root dry weights were obtained with 50 mg N and 60 mg P kg⁻¹ soil. Nitrogen and P fertilization did not increase wheat growth in soil with greater than $EC_{1:5}$ 0.67 dS m⁻¹, equivalent to EC_e 9.2 dS m⁻¹.

Plants are known to respond differently to N form. A glasshouse experiment was carried out to assess the effect of N form (NH_4^+ , NO_3^- or NH_4NO_3) added at 50, 100 and 200 mg kg⁻¹ soil, in addition to the control (no N), on nutrient composition and growth of Krichauff in a sandy loam soil with EC_{1:5} 0.21, 0.48 and 0.86 dS m⁻¹, equivalent to EC_e 2.8, 6.6 and 11.8 dS m⁻¹. Increasing soil salinity decreased shoot and root dry weights and shoot macro- and micronutrient concentrations with all forms of N. At every N addition rate and with increasing N addition from N50 to N200,

compared to NH_4^+ , the salinity of soil solution was far higher with NO_3^- and lowest with NH_4NO_3 . Shoot and root dry weights were highest with addition of 50 mg NO_3 -N or 100 mg NH_4 -N or as NH_4NO_3 at all salinity treatments. Concentrations of shoot P, Fe, Mn and Zn concentrations were greater with NH_4^+ and NH_4NO_3 compared to NO_3^- , but concentrations of shoot K and Ca were higher with NO_3^- than with NH_4^+ nutrition at all salinity treatments. At a given N rate, shoot and root dry weights were greatest with NH_4NO_3 in the saline sandy loam soil with up to $EC_{1:5}$ 0.67 dS m⁻¹.

Two experiments were conducted to evaluate the effect of plant residue addition on microbial activity and biomass, and N and P availability in salt affected soils. Although the same amounts of Na^+ and Ca^{2+} salts, $EC_{1:5}$ differed between tested soils due to differences between soils in clay content and water holding capacity. The first experiment aimed to assess the salinity range for microbial activity over 2 weeks in saline soils with different texture amended with glucose/nitrate (C/N ratio 16:1). The EC1:5 were 0.2, 1.26, 1.83, 2.28 and 2.99 dS m⁻¹ in the silty loam, 0.16, 1.10, 1.98, 2.33 and 3.18 dS m^{-1} in the sand and 0.19, 0.82, 1.75, 2.03 and 2.79 dS m^{-1} in the sandy loam. Soil respiration significantly decreased with increasing salinity in the glucose/nitrate amended soils, but was not completely inhibited even at highest salinity treatment. Cumulative CO₂-C increased over 2 weeks and was highest in the silty loam soil and decreased in the following order: silty loam soil < sandy loam soil < sandy soil. The second experiment was conducted to determine the effect of three different plant residues added at 2% (w/w) on microbial biomass and N and P availability over time (70 days) in saline sandy and sandy loam soils with low SAR and neutral pH. The $EC_{1:5}$ was 0.21, 1.08, 1.90, 2.63 and 2.89 dS m⁻¹ in the sand and 0.19, 0.87, 1.63, 2.32 and 2.49 dS m⁻¹ in the sandy loam. Microbial biomass C, N and P decreased with increasing soil salinity and were highest on day 10. With residue addition, microbial biomass C and P were significantly higher in the sandy than in the sandy loam soil, whereas no significant differences were found between soils for microbial biomass P at all salinity treatments. Under all salinity treatments, compared to other residues, highest biomass N was found in canola-amended sandy loam and in lupin-amended sandy soils. With increasing soil salinity, highest microbial P was found in the sandy soil amended with lupin residue. Nitrogen availability was generally higher in the sandy soil than in the sandy loam soil, whereas the opposite was found for P availability. Compared to canola and lucerne, N and P availability were highest in lupin amended sandy and sandy loam soil.

Two experiments were conducted to assess whether N addition (rate and form) can affect the microbial activity in presence of residues in a saline sandy loam soil. The first experiment aimed to evaluate the effect of N rate (0, 25, 50 and 100 mg N kg⁻¹ soil) added as NO_3^{-} on soil respiration over 2 weeks under non-saline conditions in presence of 2% lupin residues. The second was to determine the effect of N added at 50 mg N kg⁻¹ soil as NH₄⁺ or NO₃⁻ and lupin residue added at 2 and 4% (w/w) on microbial activity and biomass and N and P availability over 45 days in a sandy loam soil with $EC_{1:5}$ 0.21, 0.51 and 0.85 dS m⁻¹, equivalent to EC_e 2.8, 7.0 and 11.7 dS m⁻¹. Soil respiration and cumulative respiration were not significantly affected by N application rate over 2-week-incubation under non-saline conditions. Microbial biomass and N and P availability decreased with increasing salinity and were highest at 4% lupin residue. Soil respiration rate and cumulative CO₂-C and microbial biomass C, N and P were greater with addition of 50 mg N kg⁻¹ soil as NO₃-N compared to NH₄-N at every addition rate of lupin residues under saline conditions. Soil microbial biomass C, N and P were highest on day 15 and decreased over time, whereas N and P availability were lowest on day 15 and increased over time.

Since addition of inorganic N and P fertilizers improved the growth of wheat (cv Krichauff) in the saline sandy loam soil at SAR 1 and neutral pH, two glasshouse experiments were conducted to determine the effects of plant residue addition on the nutrition of wheat. The first experiment was conducted under non-saline condition to determine the effect of lupin residue rate (2% and 4% w/w) on wheat growth. The second experiment was conducted under saline conditions to determine the effect of P added as lupin residue (2%) and/or KH₂PO₄ (0, 20 and 40 mg P kg⁻¹ soil) with and without 50 mg N kg⁻¹ soil added as (NH₄)₂.SO₄ on microbial biomass, N and P availability, plant growth and nutrient composition in the saline sandy loam soil. The $EC_{1:5}$ were 0.23, 0.35 and 0.51 dS m⁻¹, equivalent to EC_e 3.1, 4.8 and 7.0 dS m⁻¹, respectively. In the first experiment under non-saline conditions, shoot dry weight was lower with addition of 4% than with 2% lupin residue with and without inorganic N. In the second experiment under saline conditions, microbial biomass C and N increased with increasing application of inorganic P, but was not as much as in presence of lupin residues. In presence of lupin residue, wheat growth increased with increasing addition of inorganic P under saline conditions. Compared to the soil with P from inorganic fertilizer and residues, inorganic P increased shoot and root dry weights and shoot P, K, Mn and Zn concentrations, but not N concentration. Addition of 50 mg inorganic N in presence of lupin residues significantly increased N and P availability and microbial biomass, but had no significant effect on wheat growth in a saline sandy loam soil.

The results showed that optimal application of N and P organic and inorganic fertilizers can improve N and P availability, microbial activity and wheat growth in salt affected soils. Highest wheat dry weight was achieved by application of 60 mg P kg⁻¹ soil in a sandy loam soil with $EC_{1:5}$ 0.67 dS m⁻¹, equivalent to EC_e 9.2 dS m⁻¹. Wheat growth was also improved with application of N-NH₄⁺ or as NH₄NO₃ at 100 mg N kg⁻¹ soil. These N and P fertilization rates can potentially enhance wheat growth in the sandy loam soil with up to $EC_{1:5}$ 0.67 dS m⁻¹, but with SAR 1 at neutral pH.

Plant residues increased microbial activity and N and P availability in the saline soils. In the soils used here, with residue addition wheat growth was P limited due to competition with microorganisms for available P. Therefore application of residues with inorganic P is necessary to satisfy wheat requirements of N and P in the saline sandy loam soil. In the saline sandy loam soil at SAR 1 and neutral pH, application of 2% lupin residues and 40 mg KH₂PO₄-P kg⁻¹ soil achieved highest microbial biomass, nutrient availability and wheat growth. However, wheat growth with these rates is not as high as with inorganic P at similar rate due to micronutrient deficiency in the saline soil with lupin residues.

Chapter 1 – Introduction

One of the critical challenges, worldwide, is the un-proportional growth of human population compared to that of food production. The maximization of crop production is therefore urgently required to remedy this imbalance. Globally, most of the suitable lands for agriculture have been cultivated and expansion into new areas to increase food production is rarely possible. Therefore increasing the unit productivity is the main aim in farming systems. Increasing crop production can only be achieved by applying suitable agricultural management practices, particularly in relation to maintenance or enhancement of soil fertility. Fertilization should gradually improve soil fertility and increase field crop yield. Soil fertility depends on many factors, the most important being soil nutrient concentration, as well as physical and chemical properties and the activity of microorganisms. The fertility of productive soils may be degraded over time due to one or a combination of the following factors: soil erosion, water shortage, irrigation using wastewater or over-fertilization. These factors may result in the salinization of soils leading to further degradation of soil fertility and cause environmental pollution threatening the health of plants, animals and humans.

Over 10% of the world's land is salt affected (Szabolcs, 1989). "Salt affected soils" is a broad term covering a range of soils defined as saline, saline-sodic and sodic. These soils contain soluble salts leading to an increase in electrical conductivity (EC). Soils having electrical conductivity over 4 dS m⁻¹ are saline (USDA, 1954). Sodic soils have exchangeable sodium percentage (ESP) greater than 15% or sodium adsorption ratio (SAR) higher than 13, and can be saline. Soil salinity can suppress plant growth via specific (ionic stress) and non-specific (osmotic stress) effects (Munns, 2002). Salinity also decreases the availability of nutrients, particularly P, and depresses microbial activity.

In addition to salinity, low P availability and uptake by plants are major limitations for plant growth in many soils around the world (Grattan and Grieve, 1999). Low P availability results from precipitation, transformation, fixation of P with soil minerals (Prafitt, 1978) and in presence of high amounts of soluble salts (Awad et al., 1990), and also occurs in the soil having low total P concentration. Salinity may also reduce the P flux through the xylem (Navarro et al., 2001), reducing plant P content and concentration. Increasing P availability and enhancing P nutrition of plants through fertilization may enhance plant salt tolerance and growth. In nutrient-deficient soils, P

fertilization can increase P availability, but may not efficiently enhance plant growth, unless plants are sufficiently supplied with nutrients, particularly N. In saline soils, fertilization of N in proper form may increase the availability and the uptake of nutrients including N (Gahoonia et al., 1992). Adequate supply of N, in combination with P, may influence plant-nutrient-salinity relationships (Martinez and Cerda, 1989) and thus enhance plant growth under saline conditions. This may however be effective only up to a certain salinity level above which the negative salinity effect is dominant.

Application of N and P from organic sources such as animal manure or plant residues can be economically viable and contribute to the stabilization or even the reduction of inorganic fertilizer market prices or cost for farmers. Also, it is of prime importance to incorporate organic materials to maintain soil fertility and productivity. Additionally, such a practice may also reduce the environmental pollution often associated with the excessive application of inorganic N and P fertilizers.

Plant residue is considered important for the maintenance of soil organic matter content (Varvel, 1994) and biological activity (Tian et al., 1992). It is also a major source of N and P (Bhatti et al., 2005; Masri and Rayan, 2006), and can therefore potentially contribute to the nutrition of plants (Richardson, 2004). Salt affected soils tend to have low organic matter content due to poor plant growth and thus low input of organic materials to soil. Plant residue addition improves the stability of soil structure by enhancing aggregation (Oades, 1993), which will improve soil porosity (Halkiah et al., 1981) and increase soil water infiltration, water holding capacity (Walker and Bernal, 2008) and hydraulic conductivity (Robindra et al., 1985). Enhancing soil structural stability may also accelerate leaching of Na⁺ (Lax et al., 1994) and decrease sodicity and salinity (Qadir et al., 2001).

The assessment of soil fertility including microbial activity to provide nutrients for plant growth is of utmost importance, especially with addition of plant residues, in salt-affected soils. Microorganisms have a central role in residue decomposition. Soil microbial characterization is therefore indispensable for knowing the soil nutrient level and its nutrient supply capability in saline soils. With increasing concentration of soluble salts water availability to microorganisms decreases and accumulation of Na⁺ and Cl⁻ ions increases in microbial cells (Killham, 1994; Zahran, 1997) reducing their activity to decompose plant residues. In saline-sodic or sodic soils, adsorption of Na⁺ on clay minerals may disperse the clay particles and reduce soil aggregation

allowing microorganisms to access organic matter previously occluded with aggregates (Nelson et al., 1996). Also, in saline soils with high pH (> 8), alkali can dissolve, disperse or cause chemical hydrolysis of the added as well as the native soil organic matter and enhance microbial activity (Li et al., 2007). In saline soils, in which salinity is the major limiting factor to microbial activity, plant residues can decompose and provide nutrients that may increase the tolerance of microorganisms to salinity.

Effects of plant residues on soil nutrient availability, microbial activity and crops differ and depend on their decomposition and nutrient release rates. Plant residues with high C/N and C/P ratios and high lignin and polyphenol contents decompose and release nutrients slowly (Melillo et al., 1982; Sivapalan et al., 1985; Tisdale et al., 1993; Green and Blackmer, 1995). These types of residues may have a slow effect on soil nutrients, but may indirectly, as mulching materials, affect crops. In contrast, residues with low C/N and C/P ratios and lignin and polyphenol contents decompose rapidly, and have a fast effect on soil nutrients and microorganisms under saline conditions.

Combined application of organic and inorganic fertilizers can also increase residue decomposition (Chen et al., 2007) under saline conditions. Inorganic N applied in different forms has a stimulating effect on microbial activity under non-saline conditions (Parkinson et al., 1971; Recous and Mary, 1990) and under saline conditions (Azam and Ifzal, 2006). In naturally saline-alkaline soils, addition of inorganic N enhanced microbial activity and residue decomposition (Luna-Guido and Dendooven, 2001; Conde et al., 2005; Dendooven et al., 2006). Inorganic P also has a positive effect on microorganisms in presence of farm yard or animal manures (Manna et al., 2006; Raiesi and Ghollarata, 2006). Application of inorganic fertilizers and plant residues can also enhance the plant use efficiency of nutrients from both plant residues and inorganic fertilizer (Quirk and Murray, 1991; Stark et al., 2007) under saline conditions.

In the literature there is no consistent response of plants to addition of P and N fertilizers under saline conditions; positive, negative or no effects have been reported. Some of these studies have been conducted in nutrient solutions, others in saline soils, where not only salinity, but also pH and level of SAR varied and plants were nutrient limited. Hence the results can not be attributed to salinity only. Also, there are no

reports on the effects of plant residue addition on the activity of microorganisms, the availability of nutrients, particularly N and P, and the nutrient composition and growth of wheat in salt affected soils. Additionally, information is scant on how addition of inorganic N or inorganic P can affect microbial activity and N and P availability in saline soils with low SAR and pH.

In this study, several experiments were conducted to investigate the hypothesis that addition of N and P from organic or inorganic sources will improve nutrient availability, microbial activity and wheat growth in saline soils. The general aim of this study was to evaluate the response of microorganisms and wheat to inorganic fertilizers and plant residues in saline soils.

Chapter 2 – Literature review

2.1 Salt affected soils

Salt affected soils are characterized by accumulation of soluble salts: cations such as calcium, magnesium and sodium, and anions such as carbonate, sulfate and chloride (Aydin et al., 2004). Salt affected soils cover a range of soils defined as saline, saline-sodic, and sodic (Abrol and Yadav, 1988; Szabolcs, 1989).

As reported (FAO/Unesco Soil Map of the World), over 10% of the world's land is salt affected and the total global area of saline soils is 397 million ha and that of sodic soils is 434 million ha (Table 2.1). Of the current 230 million ha of irrigated land, 45 million ha (19.5%) are salt affected soils and of the almost 1500 million ha of dryland agriculture, 32 million (2.1%) are salt affected soils to varying degrees by human-induced processes (Szabolcs, 1989; Oldeman et al., 1991).

Table 2.1 Regional distribution of salt affected soils, in million hectares

NOTE: This table is included on page 5 of the print copy of the thesis held in the University of Adelaide Library.

Source: FAO Land and Plant Nutrition Management Service

2.1.1 Origin and form

Salt affected soils are more extensive in arid and semi-arid regions compared to humid regions (Abrol and Yadav, 1988). However, the occurrence of saline soils is not limited to arid environments. Salt affected soils can be found in the tropical regions of Africa and Latin America, and in all continents and under almost all climatic conditions. Soil salinity problems can occur naturally (primary salinization), or as a result of human activities (secondary salinization) (Minhas and Sharma, 2003).

Primary salinization is the accumulation of salts in soils as a result of natural processes such as weathering of soil parent materials and the deposition of marine salts carried in wind and rain (Isbell et al., 1983). The phenomenon of primary salinization affects about 1.3 Gha out of the global land area of 13.4 Gha (Szabolcs, 1989). Secondary salinization, the gradual build up of salt in previously salt-free topsoil, occurs in irrigated areas as salt is introduced into the soil with every round of irrigation. Part of the salt is leached below the root zone, but parts remain in it. In Australia, the dry climate is the background of widespread salinity (Bertrand et al., 2003). The capillary rise of salt-rich groundwater driven by high evapotranspiration in summer brings salt to the surface (Lavado and Taboada, 1988). In irrigated areas, the increased use of saline water together with poor management practices aggravates the problem, particularly when there is not sufficient drainage of water exceeding plant requirements. Dryland salinity is another form of secondary salinization in soils. It occurs due to the removal of native deep-rooted perennial vegetation. Its replacement with shallow-rooted annual crops and pastures modifies the hydrological balance by increasing the quantity of recharge to groundwater as a result of reduced evapotranspiration. As the water table rises, salts stored within the soil profile are mobilized and carried toward the soil surface, resulting in salinization. At least 2.5 million hectares (5% of cultivated land) are currently affected by dryland salinity and approximately 5.7 million hectares of Australian farmland have a high salinity risk. This could rise to 12 million hectares (22%) in 50 years (Coram et al., 2001). Soils affected by secondary salinization account for about 76 Mha of 1.5 Gha cropped land, including 56 Mha of 280 Mha irrigated lands (Yeo, 1999). In addition, salts can come from vegetation growing on salt affected soils. Up to one quarter of plant dry weight is ash, and the large proportion of the ash may be soluble salts. Almost 200 kg ha⁻¹ of salts is brought annually to soil surface by vegetation (Russell, 1989). Although small compared to amounts of salts added by irrigation, plants can contribute to the accumulation of salts in the top soil over long periods of time.

The area affected by salts is expected to increase over time due to water shortage, and adoption of agricultural practices such as the use of wastewater in irrigation or over fertilization with absence or installation of inefficient drainage systems.

2.1.2 Characteristics of salt affected soils

Salinity and sodicity are major soil problems leading to the deterioration of soil fertility. The occurrence of each can change the properties of soils, but these two main groups of salt affected soils differ not only in the chemical characteristics, but also in the physical and biological properties.

2.1.2.1 Chemical and physical properties

In salt affected soils, soluble salts consisting of chlorides and sulphates of sodium, calcium and magnesium are present in high concentrations leading to an increase in electrical conductivity (EC). According to the classification of U.S. Salinity Laboratory (USDA, 1954) soils (saturated paste) with an electrical conductivity (EC) greater than 4 deciSiemens/meter (dS m⁻¹) are saline. In Australia, a classification was proposed to describe the level of salinity in soils based on the values of electrical conductivity in the soil saturation extract (abbreviated EC_e), or in the 1:5 extract (EC_{1:5}) (Table 2.2). Salinity is generally expressed in deciSiemens/meter (dS m⁻¹) or millimohs/centimetre (mhos cm⁻¹). Siemens per meter equals 0.1 mhos cm⁻¹. The ratio of the electrical current density to the electric field in the soil is expressed as the reciprocal value of its resistivity in mhos cm⁻¹ (or mmhos cm⁻¹) of soil extract or paste and generally refers to the salt/solute content of the soil.

Suggested term ^a	$\mathbf{EC}_{\mathbf{e}}$ range (dS m ⁻¹)	EC _{1:5} range $(dS m^{-1})$ in		
Suggested term		Sands	Loams	Clays
Non-saline	0 - 2	0-0.14	0-0.18	0-0.25
Low salinity	2 - 4	0.15 - 0.28	0.19 - 0.36	0.26 - 0.50
Moderate salinity	4 - 8	0.29 - 0.57	0.37 - 0.72	0.51 - 1.00
High salinity	8 - 16	0.58 - 1.14	0.73 – 1.45	1.01 - 2.00
Severe salinity	16 - 32	1.15 - 2.28	1.46 - 2.90	2.01 - 4.00
Extreme salinity	> 32	> 2.29	> 2.91	> 4.01

Table 2.2 Suggested Australian classification system for categorization of soil salinity

^a The non-saline and low-, moderate- and high-salinity categories are identical to those used by (Rogers et al., 2005). ^b EC_{1:5} range is calculated using a conversion factor selected for every soil texture (George and Wren, 1985).

Although increasing the salinity of the soil solution has a positive effect on soil aggregation and stabilization, it can damage soil structure with increasing Na⁺ concentration in the soil solution and accumulation on clay particles causing sodicity (Rengasamy, 2006). In sodic soils, carbonate and bicarbonate are the principal anions and Na⁺ is the dominant cation. Sodicity is based on Na⁺ concentration relative to other cations (i.e. Ca^{2+} , Mg^{2+} , K^+ , NH_4^+) adsorbed to soil particles on the exchange complex (Russell, 1961). In sodic soil, aggregate stability is reduced due to the increase in exchangeable sodium percentage (ESP) (Buckland et al., 2002). Soil clay particles have negative electrical charge attracting a large number of cations. The zone surrounding the cations at the negative charges on soil particles is termed "diffuse double layer." The concentration of cations is high within the diffuse double layer close to the surface of the clay particles, and decreases with distance. Clay particles are subject to forces that can move them together (aggregation) or apart (dispersion). In non-sodic soils, the accumulation of salts can affect soil physical properties by causing fine particles to bind together into aggregates. This process is known as flocculation (Pearson and Bauder, 2003), and is beneficial in terms of soil aeration (due to aggregate stabilization when wet), hydraulic conductivity (because soil pores remain open and water readily infiltrates into the soil) and root growth. Dispersion of clay particles occurs when diffuse double layer is distorted by large size of hydrated ions, or due to electrostatic repulsion (Qureshi and Barrett-Lennard, 1998). The reduction of thickness of diffuse double layer can maintain the clay particles close to each other. When dissolved in water, Na^+ ions are larger in size than K^+ , and Mg^{2+} is larger than Ca²⁺. Compared to small cations, large cations in the diffuse double layer push clay particles apart (Shainberg, 1992).

The structure of the soil is therefore degraded due to a high degree of clay dispersion (Gupta and Verma, 1983; Rengasamy et al., 2003). Sodicity may also be expressed as sodium adsorption ratio (SAR), which is the concentration of Na⁺ relative to other cations in soil solution. SAR is approximately equivalent to ESP for saturation extracts (Richards, 1954), and approximately half the value of ESP for 1:5 soil:water extracts (Rengasamy et al., 1984). Saline soils tend to have sodium adsorption ratios (SAR) less than 13 in their saturation extract and exchangeable sodium percentage (ESP) lower than 15. Sodic soils have SAR greater than 13 in their saturation extract and EC values less than 4 dS m⁻¹. The electrical conductivity (EC) of the saturation

extract of sodic soil is generally less than 4 dS m⁻¹, but may be greater with high CO_3^{2-} concentration in soil solution. The pH of the saturation paste of saline soil is less than 8.5.

In sodic soils, NaCO₃ is formed in the soil solution (Russell, 1961) causing the pH to rise above 9. High ESP and pH in the soil may reduce soil porosity and permeability (Pearson and Bauder, 2003; Rengasamy et al., 2003), thereby negatively affecting soil aeration and water conductivity (Acharya and Abrol, 1991, 1998) and establishing hypoxia (lack of oxygen). Sodic soils tend to have a dark, organic appearance due to the dispersion of clay and organic particles, whereas saline soils tend to have a white crusty surface due to the precipitation of salts (Lax et al., 1994). Saline-sodic soils have chemical and physical features that are intermediate between saline and sodic soils (USDA, 1984).

2.1.2.2 Biological properties

In addition to having adverse effect on most of the soil chemical and physical properties, salinity and sodicity also have negative effects on soil biota (Pankhurst et al., 2001) and biological processes. Increased accumulation of Na⁺ or Cl⁻ in microbial cells can influence cellular physiology and metabolic processes (Killham, 1994). Also, increased salt concentration in soil solution can restrict water availability to microorganisms (Sardinha et al., 2003). With increasing soil salinity with NaCl, the density of bacteria, fungi and actinomycetes decreased in unplanted soils (Przybulewska and Krompiewska, 2005; Nelson and Mele, 2007) as well in presence of plants (Uyanoz et al., 2006; Walker and Bernal, 2008). Sodium chloride reduced the activity of nitrogenase in some nitrogen fixers grown in pure culture (Hassouna et al., 1995) and reduced mycorhizal infection (Azcon and El-Atrash, 1996). Nitrification was inhibited by 50% and 70% in silty loam and sandy loam soils salinized to EC_e 5 and 10 dS m⁻¹, respectively (Kumar and Wagenet, 1985). Salinity had a negative effect on dehydrogenase activity (Batra and Manna, 1997), cellulase activity (Badran, 1994) and symbiotic nitrogen fixation (Cordovilla et al., 1999); enzymes and microorganisms that are essential for the maintenance of soil health.

Most of the microbial groups have species that show a tolerance for increased salinity in the environment (Zahran, 1997). Most of organisms (halophiles) require salt for growth, and they are classified slight, moderate, and extreme halophiles (Kushner, 1978). As reported in (Russell, 1961), other microbial groups described as halotolerant have no specific requirement for salt, other than the usual 100-200 m*M* NaCl, equivalent to approximately EC_e 5-10 dS m⁻¹, needed by all (nonhalotolerant) organisms. However, they are able to grow in saline conditions up to approximately EC_e 60 (slightly halotolerant), 150 (moderately halotolerant), or up to 260 (extremely halotolerant). Most of the bacterial species appeared to be susceptible to soil salinity in presence of high concentration of NaCl (Przybulewska and Krompiewska, 2005). Several fungal species were also found tolerant to salinity stress (Wichern et al., 2006). Two major adaptation mechanisms most of the microorganisms use to cope with increasing accumulation of solutes in the microbial cells were described in (Killham, 1994). The first is to exclude the incorporated ions (Na⁺ and Cl⁻) and replace them with other ions (e.g. NH_4^+ and $SO_4^{2^-}$) necessary for metabolism. The second is to produce organic compounds that can off-set the concentration gradient between soil solution and cell cytoplasm. Microorganisms may also change their morphology to tolerate increasing soil osmotic pressure (Zahran, 1997).

2.1.3 Salt effects on plant growth

Deterioration of the physical condition of the soil causes less air entering the soil, creating a poor environment for root penetration and waterlogging (due to poor drainage) increasing chances to seeds to rot and roots to die. Additionally, soil crusting and compaction of top- and subsoil reduces seedling emergence, increases runoff and erosion, and affects tillage and sowing operations. Obviously, accumulation of salts in the soil is a threat for plants. Crop species show a spectrum of responses to salt, although all show growth reduction (Brady and Weil, 1996). Generally the presence of high concentration of salt in the soil solution (salinity) reduces dry matter content, increases root:shoot ratio and diminishes leaf size and, eventually, crop yield is reduced. Salt effects are the combined result of the complex interaction among different morphological, physiological and biochemical processes in plants.

Salinity may directly or indirectly inhibit cell division and enlargement in the growing part of plant. Salts can suppress plant growth via specific and non-specific effects (Maas and Hoffman, 1977). Plant growth reduction due to non-specific salt effects is related to the osmotic potential of the soil solution (Maas and Hoffman, 1977). With the increase of salt concentration, the osmotic potential of the soil solution increases

(Bernstein et al., 1974; Maas and Hoffman, 1977) resulting in reduced availability of water to plant roots (Al-Karaki, 1997). On the other hand, high salt concentration can cause ion toxicity stress (Maas and Hoffman, 1977; Greenway and Munns, 1980). If excessive amounts of salt enter the plant in the transpiration stream, salt may be accumulated in cells of transpiring leaves leading to a premature death (Munns and Termat, 1986) and reductions in growth. This also negatively affects enzyme activity and membrane selectivity which eventually lead to the inability of cells to retain solutes, and hence the uptake and translocation of nutrients may be affected (Izzo et al., 1989). This is called the salt-specific or ion-excess effect of salinity (Greenway and Munns, 1980). Ion excess also results in reduction of leaf growth and, to a lesser extent, root growth (Munns, 1993; Garg and Gupta, 1997) thus the access of plants to water in the soil is decreased. Reduced shoot growth by salinity originates in growing tissues, not in mature photosynthetic tissues (Munns et al., 1982). Photosynthesis is reduced because it is affected by leaf expansion rate and area. Growth may be indirectly affected given that salts decrease the amount of photosynthates, water and other growth factors reaching the growing regions. This decrease may be due to stomatal closure or the direct effect of salt on the photosynthetic apparatus. Transport of photosynthates in the phloem may also be inhibited. As a result, leaves and stems of affected plants appear stunted. Plant response to salinity stress is dependent on the growth stage. Many investigators have reported retardation of germination and seedling growth at high salinity, while at later stages of growth, plants become more tolerant to salinity stress (Ramoliya and Pandey, 2003).

Salt accumulation in the soil can also negatively affect plant growth by reducing nutrient availability in the soil and decreasing nutrient uptake (Pessarakli, 1991; Grattan and Grieve, 1999) and causing nutritional imbalance in plants (Grattan and Grieve, 1999) (see below).

2.2 Nitrogen

Nitrogen is the most growth limiting element in agricultural systems (Graham and Vance, 2000). It is an essential component of proteins, nucleic acids, hormones and chlorophyll (Kozlowski, 1985; Hopkins, 1999). Nitrogen demand by plants is therefore high compared to other nutrients. Nitrogen enters the soil through biological fixation, atmospheric deposition or fertilization. Nitrogen is fixed in the soil clay lattices, oxidized and assimilated by plants or microorganisms, or lost from the system

(Evangelou, 1998). As much as 90% of soil N exists in the organic form (Pulford, 1991). During soil organic matter decomposition, organic N is then used and converted to mineral N (NH_4^+ , NO_3^- or NO_2^-) by soil microorganisms, mainly nitrosomonas and nitrobacter bacteria (Evangelou, 1998). Nitrogen in the soil is lost as a result of volatilization, denitrification, runoff and leaching. Of the total global N, 99.96% is in the atmosphere as N₂ gas and of the remaining 0.04%, only 6.5% is in forms that can be taken by plants and be used for physiological functions (Evangelou, 1998). The availability of N in soil is limited by the rates of organic matter decomposition unless availability is enhanced by biological N₂ fixation or additions of N fertilizers. Nitrogen availability is also dependent on the type of organic matter present, and on presence and activity of soil microorganisms.

The presence of salts in the soil may indirectly affect N availability through inhibition of microbial N mineralization and immobilization processes and also by increasing the soil pH. In salt affected soils, N uptake decreased due to the suppressing effect of salinity on root growth and/or inhibition of NO_3^- uptake due to competition with Cl⁻ during uptake (Kafkafi et al., 1982). As shown by Helal and Mengel (1979), presence of NaCl at EC 8 dS m⁻¹ in solution culture results in low N incorporation into proteins and amino acids in barley. Furthermore, salinity may decrease nitrate reductase activity (Abdul-Kadir and Paulsen, 1982). Ammonium and nitrate are the most important inorganic N forms readily available to plants. In a loamy sand with ECe 12.1 dS m^{-1} , no significant differences were found between NH₄⁺ and NO₃⁻ nutrition for shoot biomass of wheat (Irshad et al., 2002). In studies conducted in solution culture, compared to NH₄-N, NO₃-N was taken up at higher rates by maize at EC 6-8 dS m⁻¹ (Lewis et al., 1989). Also, compared to NH₄⁺, NO₃⁻ nutrition increased the plant uptake of Ca²⁺, Mg²⁺, K⁺ and N and plant biomass under EC 10 dS m⁻¹ induced using NaCl (Mahmood and Kaiser, 2003). Moreover, in solution with EC 15 dS m⁻¹, NO_3 -N uptake by barley increased with increasing Ca^{2+} salts (Ward et al., 1986). With 12 mM N for wheat (Al-Mutawa and El-Katony, 2001) and 5 mM for pea (Frechilla et al., 2001) plant growth was greater with NO_3^- , compared to NH_4^+ nutrition, with increasing salinity by NaCl up to 150 mM, equivalent to 15 dS m⁻¹. In these studies, the lower root and shoot biomass and nutrient uptake with NH_4^+ , compared to $NO_3^$ nutrition, were attributed to plant toxicity with NH_4^+ when applied at 3-6 mM N. Ammonium, as a sole source of N applied at high concentrations, may be toxic for

plants (Britto and Kronzucker, 2002). However, some crops, such as wheat and maize, can produce high dry matter with addition of NH_4^+ , compared to NO_3^- , under saline conditions (Singh et al., 1992). Generally, plant uptake of NH_4^+ decreases the rhizosphere pH because of excretion of H⁺ protons, whereas NO_3^- uptake decreases soil pH through OH⁻ and HCO₃⁻ release (Gahoonia et al., 1992). Changes in soil pH, to a great extent, can affect the availability of P (Gahoonia et al., 1992) particularly in the saline soils (see section 2.3.2.2). Although most plant species can grow on either N form, supplying plants with mixtures of NO_3^- and NH_4^+ in better growth and enhanced nutrient accumulation than either form in hydroponics with EC 8 dS m⁻¹ (Ali et al., 2001) and in a loamy sand soil with EC_e 12.1 dS m⁻¹ (Irshad et al., 2002).

Nitrogen addition in either form can affect soil pH and P availability in soils. The maximum response to N fertilizers occurs when P is not limiting, and vice versa (Reichman and Grunes, 1966). Therefore adequate supply of N as well as P may enhance plant nutrient uptake and growth under saline conditions.

2.3 Phosphorus

Phosphorus (P) has many functions in organisms (plants, animals, microorganisms) and is one of 16 elements known to be essential for higher plants (Arnon, 1943). In plants, P has numerous physiological functions and is therefore required in large amounts for plant growth. Phosphorus plays a vital role in energy storage and transformation for plant metabolic processes (Brady and Weil, 1996). In the metabolic processes in plants, many P-containing compounds are synthesized. Phosphorus also has a structural role in nucleic acids, ATP, enzymes and phospholipids, which have basic roles in essential physiological processes including photosynthesis, respiration, cell division and cell enlargement (Marschner, 1995; Schachtman et al., 1998). Phosphorus is essential for seed production, plant root growth and early plant maturity and resistance to root rot diseases and winter kill. Inadequate supply of P to crops causes P deficiency leading to a reduction in leaf area and photosynthetic capacity (Rodriguez et al., 1998) and a decline of plant growth (Elliott et al., 1997).

2.3.1 Phosphorus sources and forms in soils

The total P content in the soil is commonly between 100 and 3000 mg of P kg⁻¹ (Tiessen and Moir, 1993), depending on the P content of the parent material and subsequent amelioration. Phosphorus exists in inorganic and organic forms in soils.

Researchers have used terms such as primary mineral, secondary mineral, labile, organic and occluded P to describe P forms in the soil.

Phosphorus in inorganic and organic compounds is continuously converted from one form to the other (Brady and Weil, 1996), and may be lost through erosion and leaching, be utilized by plants and microbes, enter the labile pool or be transformed into secondary P minerals. The P cycle and its dynamics, including the interchangeable sources in soil, are presented in Figure 2.1.



Adapted from Tisdale et al. (1993) and Brady and Weil (1996)

Figure 2.1 Phosphorus dynamics in soil

2.3.1.1 Soil inorganic P

In absence of P fertilization, P in soils is the result of weathering of P-containing minerals (Chang and Jackson, 1958). Apatite, a P-containing rock, is present in less weathered soils in various forms representing the primary P forms: (fluroapatite $[Ca_{10}F_2(PO_4)_6]$, hydroxyapatite $[Ca_{10}(OH)_2(PO_4)_6]$ and oxyapatite $[Ca_{10}O(PO_4)_6]$) of P. The forms of primary minerals which contain P are quite variable due to the ability

of phosphate to isomorphically substitute silicate in the structure (Lindsay et al., 1989). Other P compounds, called secondary P compounds, are formed during weathering of primary P forms or transformation of P fertilizers (Lindsay et al., 1989). The P anion is adsorbed on the surfaces of minerals, precipitated with Fe³⁺ and Al³⁺ hydroxides and carbonate or reacts with Ca²⁺ (Smeck, 1985; Brady and Weil, 1996; Hinsinger, 2001). The Ca-P mineral forms can contain varying amounts of carbonate, fluoride, sulphide, hydroxide and cations (Lindsay et al., 1989; Tiessen and Moir, 1993). In highly weathered soils, most P exists in non-labile, occluded or stable organic forms. The occluded P is called Rs-P because strong reducing agents are required to dissolve the coating materials and release the occluded P. Neutral and slightly acid soils usually contain all P forms in comparable amounts, whereas alkaline and calcareous soils contain mostly Mg-P and/or Ca-P (Sah and Mikkelsen, 1986; Samadi and Gilkes, 1998; Bertrand et al., 2003). In the soil solution P is in the form of $H_2PO_4^{-1}$ and HPO_4^{2-1} (Smeck, 1985). These forms are readily available to plants (Marschner, 1995). Labile inorganic P (Pi) consists of P adsorbed on surfaces of more crystalline P compounds, sesquioxides or carbonates. It can be readily available to microorganisms and plants in the short term. Non-labile P is considered to be tightly bound to soil particles. Phosphorus in both primary and secondary minerals is regarded as non-labile P and unavailable to plants (Cross and Schlesinger, 1995). In most agricultural systems, geochemical processes determine the long-term distribution of P in soils, but in the short term, biological processes influence P distribution (Walbridge, 1991; Cross and Schlesinger, 1995). In slightly weathered soils, the majority (80-90%) of released P from these compounds is converted into organic (P_0) form or adsorbed to clay minerals and the rest remains in the inorganic form (P_i) (Tisdale et al., 1993).

2.3.1.2 Soil organic P

Soil organic P (P_o) is derived from the decomposition of recently applied plant residues, native soil organic matter or microorganisms (Nziguheba et al., 1998; Mafongoya et al., 2000). Phosphorus availability from these sources ranges from that available in undecomposed plant residues and microbes, to stable compounds that have become part of soil organic matter. Hence environmental factors that lead to increased soil organic matter concentration increase soil organic P concentration and at the same time can potentially increase residue decomposition and microbial turnover, and thus P availability.

2.3.1.2.1 Organic forms of P

After mineralization, organic P (P_0) can be taken up by plants (Richardson, 2004). Organic P accounts for 20 to 80% of total P in the soil (Brady and Weil, 1996), about 50% of the organic P compounds have been chemically identified. The amount of organic P is positively correlated with the soil organic matter content. Organic P in soil is present in four main compounds (Cade-Menum et al., 2000). The first group represents 10-50% of soil organic P and includes orthophosphate monoesters $(ROPO_3^{2^2})$, where R is an unspecified organic moiety) such as inositol phosphates, mononucleotides and sugar phosphates (Newman and Tate, 1980; Tate and Newman, 1982). The second group comprises orthophosphate diesters ($R_1 OROPO_2^-$, where R and R_1 are unspecified organic moieties) such as phospholipids, RNA and DNA and accounts for 1-5% of soil organic P (Kowalenko and McKercher, 1971; Tate and Newman, 1982). The third group contains teichoic acids, which are acidic polysaccharides attached to the cell walls of gram-positive bacteria consisting of repeating units of either glycerol or ribitol that are connected by phosphate esters (Brock et al., 1984). The fourth group covers phosphates containing C-P bonds, which are very resistant to oxidation and hydrolysis (Corbridge, 1990). Small amounts of sugar phosphates, phosphoproteins, glycerophosphate and phosphonates can also be found (Tate, 1984), but have not yet been quantified. A large proportion of organic P in soil is present as uncharacterized high molecular weight substances (Dalal, 1977; Newman and Tate, 1980). The remaining Po probably occurs as insoluble complexes with organic matter (Tate, 1984). Organic P can be divided into labile and non-labile Po with labile Po being that extracted by bicarbonate (Olsen et al., 1960) while nonlabile Po is the rest of organic Po in the soil.

2.3.1.2.2 Microbial forms of P

Microbial biomass P is an important pool of organic P (Cross and Schlesinger, 1995). The microbial biomass is particularly important for P_0 turnover in the soil (Kwabiah et al., 2003; Oberson and Joner, 2004). The availability of soil P_0 to plants and microorganisms is controlled by the rate of P_i release by mineralization, rather than
the amounts of P_o present in soil (Ballard, 1980). The biological portion of the P cycle is controlled primarily by microbial decomposition, immobilization and mineralization, and secondarily by plant uptake (Bolan, 1991; Walbridge, 1991). Phosphorus in the microbial biomass accounts for 11-67 kg ha⁻¹ (up to 3.3%) of total P in soil (Brookes et al., 1984), and is believed to be a labile reservoir of P (Jenkinson and Ladd, 1981). Microbial P may represent 3 to 24% of total organic P in temperate arable soils and up to 20% in some grassland and forest soils (Brookes et al., 1984). Phosphorus in microorganisms is present as 30-50% RNA, 15-20% insoluble inorganic P and other organic compounds, and 5-10% DNA (Chauhan et al., 1981). The microbial organic P compounds include mainly ortho-meta sugars, adenosine phospholipids (Paul, 1986). Microbial P represents the actively cycling pool of P_o in the soil and is part of labile or readily available P_o.

2.3.2 Factors affecting P availability in the soil

The availability of P to plants depends on the concentration of orthophosphate ions in the soil solution (intensity factor), the reserve of P on the surfaces of soil particles and organic materials (quantity factors) and the potentiality, or capacity, which generally relates to the P sorption capacity (Tran et al., 1988).

The dynamics of P and in particularly the availability of P in the soil is mainly controlled by the following factors: clay (type, amount and aggregation), pH, Ca^{2+} , HCO_3^{-} , hydroxides of Al^{3+} and Fe^{3+} , organic matter and concentration of certain cations and anions.

2.3.2.1 Soil clay type, amount and aggregation

Phosphorus in the soil solution reacts with soil minerals and its sorption and desorption processes are influenced by soil aggregate size (Misra et al., 1988; Wang et al., 2001). Phosphorus reacts with other elements on surface of aggregates and over time slowly penetrates into aggregates (Ibrahim and Pratt, 1982). Lindquist et al. (1997) found that P penetrates only a thin outer-most layer of soil aggregates, suggesting that larger soil aggregates with relatively less surface area than small aggregates may reduce P fixation and result in increased availability of recently applied P. Phosphorus availability and rate of fixation is dependent on clay content (McPharlin et al., 1990). The type of clay minerals also affects P availability, retention and fixation. Montmorillonite and Kaolinite sorb P on their edge faces (White, 1980; Sposito, 1984).

2.3.2.2 Soil pH, calcium, carbonates and hydroxides of Al and Fe

The highest P availability occurs at the pH 6-7 while at higher or lower pH availability of P decreases (Prafitt, 1978). Phosphorus reacts with Al³⁺, Fe³⁺ and Ca²⁺ on the surface area of the clay minerals (White, 1980). In acidic soils, P is generally bound to hydrous oxides of Fe³⁺ and Al³⁺ and is gradually converted into crystalline Fe³⁺ and Al³⁺ phosphates (Bertrand et al., 2003); but its solubility increases with increasing soil pH. In contrast, in neutral, calcareous and alkaline soils, P precipitates with calcium in stable forms of Ca-P and is gradually converted into soluble hydroxyapatite or fluroapatite minerals (Samadi and Gilkes, 1998). The solubility of Ca-P increases with decreasing soil pH. Chang and Jackson (1958) showed that more P is available from Fe-P and Al-P than from Ca-P minerals due to higher specific surface activity of the former in many soils. The presence of oxide coatings on the surface of soil materials increases surface area and P retention (Jones and Uehara, 1973). Calcareous soils fix P in the form of Ca-P and hydroxyapatite on the surfaces of carbonate minerals (Prafitt, 1978). Ryan et al. (1985) found that due to the greater surface area to volume ratio, small particles of carbonates sorb more P than large particles.

2.3.2.3 Organic matter

The studies on P sorption in different soils showed a correlation not only between P and Al^{3+} , Fe^{3+} , soil pH, clay and carbonate content, but also between P and organic matter (Tisdale et al., 1993; Brady and Weil, 1996). It was reported that polygalacturonic acid from organic materials and root exudates can reduce P adsorption on soil clay minerals. Organic acids produced during organic matter decomposition can compete with P for binding sites in soil particles (Nziguheba et al., 1998). Other organic matter can also block CaCO₃ surfaces, thereby reduce P retention (Inskeep and Silvertooth, 1988).

2.3.2.4 Cations and anions

As mentioned above, Al^{3+} and Fe^{3+} precipitate P in acid soils whereas in alkaline soils P is precipitated with Ca^{2+} and Mg^{2+} and is generally sparingly soluble (Tisdale et al., 1993). High concentrations of Zn^{2+} also enhance P adsorption whereas high

concentrations of selenite and arsenate decrease P adsorption in soils with goethite (White, 1980). Phosphorus can be replaced by other anions such as SO_4^{2-} , SeO_3^{2-} , HCO_3^{-} , OH^- , MO_4^{2-} and polybasic organic anions (Prafitt, 1978).

2.3.2.5 Soil salinity and sodicity

Salt concentration and type, exchangeable sodium percentage (ESP) and $CaCO_3$ can influence P availability (Prafitt, 1978). Most of the soluble salts react with soil components or fertilizer P to form less soluble P compounds, and change the soil pH (Olsen et al., 1960). In soil with pH < 5.5, Al-P precipitate as the NaCl concentration increases in the soil up to ECe 20 dS m⁻¹ (Saini, 1970; Sharpley et al., 1992). Also with increasing NaCl from 10 to 100 mM in the solution culture, P activity decreases due to increased ionic strength, but increases with increasing P addition up to 130 mM P (Awad et al., 1990). In saline calcareous soils, high concentrations of Na⁺ and increased pH led to increases in the amount of NaHCO₃-extractable P, whereas in sodic soil the amount of extractable P was mainly affected by pH, with little effect of Na^+ (Rimmer et al., 1992). With increasing Ca^{2+} concentration, P can precipitate as poorly soluble Ca-P (Aslam et al., 1996), particularly in soils with CaCO₃ (Balba, 1980). In salinized soils, P availability decreased more with 75:25 compared to 25:75 Ca:Na and soil ionic strength generally increased with increasing salt addition (Curtin et al., 1993). Ryden et al. (1977) found that P retention increased with the increasing ionic strength of soil solution. In addition, when Na^+ replaces Ca^{2+} , Mg^{2+} and Al^{3+} at the exchange sites, the surface negative potential is increased leading to desorption of P (Curtin et al., 1993; Naidu and Rengasamy, 1993). In sodic soils, P may exist as Na-P, which is formed as a result of the presence of soluble sodium salts and which is highly soluble (Mashali et al., 1988).

2.3.3 Phosphorus uptake in the saline soils

Generally, P diffuses in soil and for its uptake roots must increase the soil volume explored. However, increasing soil salinity reduces root growth which in turn reduces the surface area of the roots in contact with P in the soil and may result in reduced P uptake. Through its influence on plant metabolic activities, salinity may also reduce the P flux through the xylem (Navarro et al., 2001), reducing plant P content and concentration. Phosphate and Cl⁻ can possibly be absorbed through the same mechanisms. Chloride may strongly reduce plant P uptake because of competitive

inhibition (Chabra et al., 1976; Manchanda et al., 1982). In saline solution with EC 15 dS m⁻¹, NaCl inhibited P uptake by cotton more at low P concentration (30 μ M) than at high P concentration (1 mM) (Martinez and Lachli, 1994). Phosphorus deficiency can decrease plant salt tolerance expressed as dry matter weight (Gibson, 1988). Compared to low (1 mM), addition of high P (10 mM) increased P uptake and salt tolerance of tomato in solutions with EC up to 10 dS m⁻¹ (Awad et al., 1990). In hydroponics, increasing addition of P from 3 to 60 µM P increased barley growth up to EC 30 dS m⁻¹ (Al-Karaki, 1997). In glasshouse and field trials, wheat and maize growth increased with increasing P fertilization up to 50 kg ha⁻¹ in a sandy soil irrigated with water having EC 15-19 dS m⁻¹ (Manchanda et al., 1982). Grain yield of rice (Jalil et al., 1979) and of wheat (Mehdi et al., 2003) responded positively to addition of 110 kg P₂O₅ (48.5 kg P) ha⁻¹, from different P sources, in a sandy loam soil with EC_e 6 dS m⁻¹ and SAR 28. Also, wheat straw and grain yield was highest at 150 kg P₂O₅, (64.5 kg P) ha⁻¹, in a clay loam with EC_e 5.6 dS m⁻¹ and SAR 14.02 and in a silty loam with EC_e 12 dS m^{-1} and SAR 24.02 (Abid et al., 2002). Additionally, wheat growth was highest with addition of 50 kg P ha⁻¹ in a sandy soil irrigated with water having 15-19 dS m⁻¹ (Manchanda et al., 1982). However, other researchers showed that high and non-limiting concentrations of P do not affect plant response to salinity (Langdale and Thomas, 1971; Rogers et al., 2003).

Therefore, increasing P as well as N fertilization may enhance nutrient availability and uptake and plant growth under salinity-induced nutritional deficiency and to a great extent, under osmotic stress, but may not be effective in soils where plant growth is negatively affected by sodicity or alkalinity.

Nitrogen and P in soil solution can be replenished by N and P fertilization from inorganic as well organic sources such as animal manure, green manure and plant residues (Bhatti et al., 2005).

2.4 Plant residues

Plant residues are essential for soil productivity through organic matter maintenance and as a source of nutrients (Kumar and Wagenet, 1985; Varvel, 1994). Plant residues added to the soil are transformed into CO₂, inorganic nutrients, microbial biomass and relatively stable humus (Shields et al., 1973; Berg et al., 1993). Soil organic matter maintains favourable soil physical, chemical and biological properties and releases Plant residues have positive effects on soil physical properties because they are the main source of organic matter (Hulugalle et al., 1986). The increase of organic matter content increases the water-stable aggregates (Quirk and Murray, 1991). Organic materials can also change bulk density (Barathi et al., 1974), porosity (Halkiah et al., 1981) and hydraulic conductivity of soil (Robindra et al., 1985). Plant residues have a significant influence on soil structural stability, and particularly sodic soils (Oades, 1993; Nelson et al., 1996). The structural stability of salt affected soils can also be improved indirectly because the addition of organic matter accelerates leaching of Na⁺, decreases the ESP and the EC, and increase soil water infiltration and water holding capacity (Lax et al., 1994; Qadir et al., 2001; Walker and Bernal, 2008).

Decomposition of plant residues not only increases soil organic matter content, but also increases nutrient availability in the salt affected soils.

2.4.1 Factors affecting residue decomposition

The main factors controlling residue decomposition are residue quality (chemical and physical properties), composition of the decomposer community and environmental factors such as temperature, moisture, pH, salinity and sodicity.

2.4.1.1 Residue quality

Differences between plant residues in decomposition rate are attributed to variation in residue traits such as leaf toughness, nitrogen, lignin, and polyphenol concentrations, and the carbon/nitrogen and lignin/nitrogen ratios which affect microbial activity and substrate utilization (Berg et al., 1993; Cadish and Giller, 1997; Perez-Harguindeguy et al., 2000).

Nitrogen concentration in plant residues is a limiting factor for residue decomposition, nutrient release and microbial activity and biomass (Zagal and Persson, 1994). Plant residues with high, compared to low N content show high decomposition rate and nutrient release (Swift et al., 1979). Also, residues with C/N higher than 20:1 decompose slowly leading to a slow release of nutrients and net N immobilization (Green and Blackmer, 1995). Additionally, increasing lignin concentration decreased decomposition of plant residues (Heal et al., 1997) and as a result N release from residues (Melillo et al., 1982). Also, increased concentrations of polyphenols and/or

silica reduce the decomposition rate of plant residues (Vallis and Jones, 1973; Sivapalan et al., 1985). Particle size also determines decomposition rate of residues. Ground or finely chopped residue material is more susceptible to microbial attack than large intact plant parts due to a greater surface area to volume ratio, better soil-residue contact (Jensen, 1994; Angers and Recous, 1997) and disruption of the lignified barrier in tissues (Sumerell and Burgess, 1989). However, fine particles are also more likely to be protected against decomposition through physical protection by clay and other particles (Stickler and Frederick, 1959). The ratio of carbon to P (C/P ratio) is also used as an index for determination of P mineralization. If the C/P ratio is less than 200, the decomposition rate of residues is high and hence P availability in soil solution increases, whereas in soils amended with residues with C/P ratio higher than 300, residue decomposition is slow and net P immobilization may occur (Tisdale et al., 1993). Thus plant residues with high C:N and C:P ratios, and high lignin and polyphenol contents have negative effects on nutrient availability, but indirectly affect nutrient availability via increasing soil organic matter content.

2.4.1.2 Microbial community

Plant residues are already colonized with microorganisms before senescence (Tester, 1988), but the diversity and population size of soil microorganisms, particularly bacterial and fungal biomass will increase as decomposition proceeds (Henriksen and Breland, 1999; Malosso et al., 2004). Microbial diversity and biomass greatly depend on the quality of added organic materials and the amount of nutrients released (Sajjad et al., 2002). Most of the readily available organic C added with residues is rapidly taken by soil microorganisms. Thereafter decomposition is dominated by turnover of microbial C and recalcitrant organic C (Ladd et al., 1985). This may therefore influence microbial diversity. The composition of the microbial community may also vary with changes in soil properties such as pH, texture salinity, sodicity and nutrient status.

2.4.1.3 Environmental conditions

It is generally known that microorganisms have a central role in residue decomposition. Environmental factors affecting residue decomposition are mostly similar to factors limiting microbial activity. These factors are water content, temperature, pH, salinity and sodicity.

2.4.1.3.1 Soil texture and water content

Generally, soil texture and structure affect pore space, bacterial and faunal populations and their activity in soil. Fine-textured soils have a larger proportion of small pores than coarse-textured soils. Habitable pore space accounts for a small portion of total pore space and affects bacterial and faunal distribution in soils (Bamforth, 1988). Bacteria probably have the most favourable environment in aggregates with small diameter pores (Foster, 1988). With increasing soil aggregation, C in organic matter can be bound to clay minerals and be inaccessible to degrading organisms and their enzymes (Berg and McClaugherty, 2003). Turnover of microbial biomass C and N is faster in coarse-textured soils than in fine-textured soils (van Veen et al., 1985). Therefore, soil texture has a significant role in residue decomposition and microbial community structure, activity and biomass. However, Plante et al. (2006) found no effect of the soil texture on decomposition of particulate organic matter.

2.4.1.3.2 Soil pH, salinity, alkalinity and sodicity

Soil pH, salinity, alkalinity and sodicity may affect microbial activity and residue decomposition. Negative effects have been shown on microbial activity (Sarig et al., 1996; Zahran, 1997; Rietz and Haynes, 2003; Sirulink et al., 2007), carbon mineralization (Frankenberger and Bingham, 1982; Nelson et al., 1996; Nelson and Mele, 2007) and nitrogen mineralization (Pathak and Rao, 1996) in salt affected soils amended with plant residues. However, with increasing addition of NaCl or Na₂SO₄ at similar levels of Na⁺ up to 142 mM kg⁻¹ soil in a soil with 17% clay, pH 8.1, EC_e 2.17 dS m⁻¹, 2% maize straw increased microbial activity and biomass (Li et al., 2006a; Li et al., 2006b). Microbial activity is often higher in presence of SO_4^{2-} compared to Cl⁻ (Garcia and Hernandez, 1996; Li et al., 2006a) due to stimulation of microorganisms by S due to the essentiality of SO_4^{2-} for microbial cells for the synthesis of amino acids, vitamins and other organic compounds; chloride on the other hand is not required by most microorganisms, and tends to build up to toxic levels. Also, increased soil pH up to 10 (Li et al., 2007) increased residue decomposition and microbial activity and biomass in naturally alkaline saline soils (Li et al., 2007). Additionally, microbial activity increased with addition of different plant residues added at 1% in a soil with 61.2% clay, ESP 21.4%, $EC_{1:10}$ 32.9 dS m⁻¹ and

pH 6 (Nelson et al., 1996), or at 0.2% in a soil with 17.1% clay and ESP 25, EC_{1:5} 1.5 dS m⁻¹ and pH 9.1 (Clark et al., 2007). With increasing addition of NaCl salts, Na⁺ ions, relative to Ca²⁺ and/or Mg²⁺, may occupy a large number of exchange sites on clay particles. Increased Na⁺ adsorption on clay minerals leads to dispersion of clay particles and reduced soil aggregation allowing microorganisms to decompose organic matter particles more easily (Nelson et al., 1996). Also, with increasing soil pH, microbial activity may be stimulated as alkali can dissolve, disperse or cause chemical hydrolysis of the added as well as the original soil organic matter and because dissolved organic matter can be easily decomposed by microorganisms (Laura, 1973, 1974; Nelson and Mele, 2007). On average, microbial activity is highest near neutral pH (Grundmann et al., 1995). Bacterial activity is highest in soil with high pH, whereas at low pH fungal activity is highest (Allison, 1973). With increasing soil pH, residue decomposition and nutrients release increased in salt affected soils (Li et al., 2007). However, at high pH 8.9 microbial activity decreased with increasing salinity to above ECe 5.7 dS m⁻¹ in soils amended with 1% maize or pea straws (Muhammad et al., 2006).

2.4.2 Phosphorus availability in soils with plant residues

Although organic inputs generally can not provide sufficient amounts of P for plant nutrition due to the low P content, these inputs have the potential to increase P availability and concentration in the soil solution in soils with low P availability (Irshad et al., 2005). The dynamics of P_0 in soil depends on the interactions between microbes, organic matter and plants (Stewart and Tiessen, 1987). Microbial P can be mobilized through microbial turnover. Microorganisms, particularly P solubilising bacteria and fungi, can also contribute to the dissolution of precipitated P compounds causing an increase in P availability and thus uptake by plants (Richardson, 2004). Additionally, microorganisms can break down organic P compounds increasing P in soil solution.

As mentioned above, residue C composition is an important regulator for decomposition processes (Swift et al., 1979), and can control the production of P solubilising compounds. The use of materials with different C/P ratio can affect the microbial immobilization and remobilization of inorganic P during their decomposition (Umrit and Friesen, 1994). Decomposition products (organic acids and anions) can alter the availability of native soil P (Oladeji et al., 2006). In amended

soils, increases in P availability are usually attributed firstly to an increase in net negative charges on soil colloids that reduces adsorption of applied P (Guppy et al., 2005), and secondly to organic anions (low molecular weight organic acids and humic and fulvic acids) formed during decomposition of residues, or manures that can compete with P for the same adsorption sites in the soil (Iyamuremye et al., 1996). The decrease in P sorption may be related to the amount of Al^{3+} released from soil colloids by the dissolved organic carbon, suggesting that complexation of Fe³⁺ and Al³⁺ from oxides may also contribute to the decreased P sorption (Ohno and Erich, 1997). Organic compounds released during plant residue decomposition can also result in soil acidification increasing P availability in the soil (Hammond et al., 1986; Troeh and Thompson, 1993). Additionally, decomposing organic matter can affect the availability of P through changing microbial activity and the rate of uptake of P_i by the microbial biomass (Kwabiah et al., 2003). It has been shown that addition of plant residues increased N and P availability and thus growth of sorghum (Kurdali, 2004) barely (Liang et al., 2005), sugar beet (Uyanoz et al., 2006) and wheat (Walker and Bernal, 2008) in salt affected soils.

2.5 Combined application of inorganic N and P and plant residues

Combined application of organic and inorganic fertilizers may increase residue decomposition (Chen et al., 2007) and plant use efficiency of nutrients from both soil organic matter and inorganic fertilizer (Oue'draogo et al., 2006; Stark et al., 2007). The activity and thus biomass of soil microorganisms may be limited by nutrient availability (Gray, 1976), which may, in turn, reduce residue decomposition and nutrient release into the soil solution. Plant residue with high C/N and C/P ratios do not contain sufficient amounts of nutrients for rapid decomposition, therefore addition of inorganic N and/or inorganic P may enhance residue decomposition in the soil.

Inorganic N may also have a stimulating effect on microbial activity. Increased microbial biomass (Parkinson et al., 1971) and/or changes in fungal community composition (Hudson, 1971) have been reported after N addition. Addition of inorganic N also increased residue decomposition rate and nutrient release (Carreiro et al., 2000; Moran et al., 2005; Sirulink et al., 2007). It was reported that immobilization of NH_4^+ is faster than NO_3^- (Azam et al., 1993). Greater immobilization of NH_4^+ than NO_3^- during straw decomposition has been also shown (Jansson, 1958; Recous and Mary, 1990), and is attributed to differences in microbial

community structure, and because with NO3⁻ microorganisms may require more energy for the conversion into NH_4^+ and for synthesis (Azam et al., 1993). This is also attributed to the preferential assimilation of NH_4^+ by the heterotrophic microflora. However in field studies (Bowden et al., 2004; Burton et al., 2004) and in glasshouse experiments (Micks et al., 2004), N application depressed or did not influence soil respiration, especially when it was added as NH_4NO_3 due to decreased pH when availability of C is low (Amador and Jones, 1993; Thirukkumaran and Parkinson, 2000), or osmotic stress (Lucken et al., 1962; Martikainen et al., 1989). Also, in a Nrich forest soil, microorganisms did not respond to addition of inorganic N (Bowden et al., 2004). Bjarnason (1987) found no difference between N forms on rates of immobilization and remineralisation. In naturally saline-alkaline soils, positive effects of N in either forms on microbial biomass, soil respiration and N availability have been observed with either glucose or organic materials (Luna-Guido and Dendooven, 2001; Conde et al., 2005; Dendooven et al., 2006). In contrast, Azam and Ifzal (2006) found that NaCl was inhibitory to microorganisms in presence of NO3⁻ compared to NH_4^+ in a glucose-amended soil.

Microbial responses to addition of inorganic P fertilizer have been reported to be either neutral (Amador and Jones, 1993), negative (Flanagan and Van Cleve, 1983), or positive (Manna et al., 2006; Raiesi and Ghollarata, 2006). It was also shown that inorganic P to a maximum of 44 kg P ha⁻¹, with or without farmyard manure, increased the phosphatase activity and microbial biomass P (Manna et al., 2006). In contrast, triple super phosphate had no effect on soil respiration or on litter decomposition (Thirukkumaran and Parkinson, 2000). Intensive use of chemical fertilizers has been reported to depress microbial activity (Manna et al., 2006). Compared to the soil with high P content (1473 mg kg⁻¹ soil), addition of 32.6 m*M* P in soils with low P content (231 mg kg⁻¹ soil) resulted in increased soil respiration (Amador and Jones, 1993). Nziguheba et al. (1998) found that, compared to the sole application of 15 kg P ha⁻¹ from either source, 7.5 kg from organic sources, Tithonia leaves containing 0.27% P, and 7.5 kg from Triple super phosphate (20% P), P availability and reduced P adsorption to clay minerals.

2.6 Proposal and aims of study

Although many studies have been published on the decomposition of plant residues and N release and availability to growing plants in salt affected soils, phosphorus has received less attention.

Phosphorus fertilization is often necessary to improve plant growth, produce economic yields and establish nutrient balance in the soil for sustainable agriculture. Therefore it can be assumed that P fertilization is as important as N fertilization to wheat in saline soils. Previous studies showed different plant responses to N, added in different forms, under saline conditions. But, no consideration was given to P availability and plant P uptake which may also be important to ensure optimal use of N in salt affected soils.

Organic matter decomposition has been studied in naturally saline soils with either high pH or SAR. As identified in the literature review, the term "salt affected soils" covers a range of saline-sodic soils (saline, saline-sodic and sodic soils). There are few studies on organic matter decomposition in saline soils with low SAR, CaCO₃ and neutral pH. Information related to P and N release from plant residues in saline soils is also scarce. Several questions are also arising regarding the relationship between organic matter and P as well as N availability; and how this can affect the growth of wheat in saline soils.

The investigations conducted in this work focus on N and P nutrition of wheat in saline soils and, on the other hand, on organic matter decomposition and effect on the fertility (microbial biomass and N and P availability) in saline soils.

For studies related to nutrition of wheat in saline soils, the following questions were addressed, firstly, what is the relationship between P and salts and how is the availability of P influenced in different textured-saline soils (Chapter 4)? Secondly, what are the separate and the combined effects of N and P on the wheat growth in a saline soil (Chapter 5)? Thirdly, is wheat growth affected by the form of applied N in saline soils (Chapter 6)?

For studies on organic matter, four questions were addressed. Firstly, how is microbial activity affected by increasing salinity, and how this is modulated by soil texture and organic carbon content (Chapter 7)? Secondly, can plant residues affect microbial biomass and N and P availability in saline soils (Chapter 7)? Thirdly, how does the addition of inorganic N affect microbial biomass and change the availability

of N and P in a saline soil amended with residues; and do N forms $(NH_4^+ \text{ or } NO_3^-)$ differ in their effect (Chapter 8)? Lastly, how effective are residues compared to the inorganic fertilization in supplying wheat with N and P and how do mixtures of residues and inorganic fertilizers affect microbial biomass and growth of wheat in saline soils (Chapter 9)?

Using saline soils with low SAR and pH, the aims of this study were:

- 1. To optimize P and N fertilization rates for wheat growth,
- 2. To study the effect of residues with different properties on N and P availability and microbial activity and biomass,
- 3. To evaluate the effect of inorganic N rate and form on plant residue decomposition, microbial activity and biomass and N and P availability, and
- To determine the effects of P from inorganic and/or organic sources with and without inorganic N on microbial biomass, N and P availability and wheat growth.

Chapter 3 – General materials and methods

3.1 Soil analyses

Soils were collected from three sites, two of which are near Monarto, South Australia (latitude 35°05'S, longitude 139°06'E and elevation 166 m), 60 km south-east from Adelaide and located on the eastern margins of the south Mount Lofty Ranges (Chittleborough et al., 1976). Monarto soils (virgin sites that have never been fertilized) were colluvium and alluvium red duplex (Chittleborough et al., 1976) and classified as red chromosol (Isbell, 1996) or Rhodoxeralf in the USDA soil classification system (USDA, 1999). The soil from the third site of the non-sodic Urrbrae series, South Australia (Grace et al., 1995) was classified as red-brown chromosol and Rhodoxeralf in the USDA classification.

At each site, several sub-samples (0-15 cm depth) were bulked to give a composite sample. The soil was air-dried and passed through a 2 mm sieve prior to the establishment of the experiments. The particle size fractions of the soil were characterized according to Klute (1986). Soil pH and electrical conductivity (EC) were measured in 1:5 soil-water suspension after 1 hour end-over-end shaking at 25 °C. Soil bulk density was determined after weighing soil samples taken by 5 x 5 cm (diameter x height) metal rings. Total soil C and N were determined using a LECO dry combustion apparatus. Total P was measured colorimetrically according to Westernman (1990) after digestion with acid mixture (6:1 HNO₃:HClO₄). Total organic C was determined after Walkley and Black (Jackson, 1958). Available P was extracted by the anion exchange resin membrane after Kouno et al. (1995) (see section 3.1.5.) and P was colorimetrically determined after Murphy and Riley (1962). Soil available N (NH₄⁺ and NO₃⁻) was extracted in a 1:10 with 2 *M* KCl mixture with 1 hour shaking (Anderson and Ingram 1993) and colorimetrically measured (Kempers, 1986) by a Skalar autoanalyzer. The characteristics of collected soils from Monarto and Urrbrae are presented in Table 3.1.

Parameter	Monarto	Urrbrae	Monarto
Particle fractions (%)			
Sand Silt Clay	97.5 0.0 2.5	62.5 27.5 10.0	75.0 5.0 20.0
Soil texture	Sandy	Silty loam	Sandy loam
pH _{1:5}	6.83	6.69	7.08
$EC_{1:5} (dS m^{-1})$	0.04	0.17	0.03
CEC (meq 100 g ⁻¹ soil)	9	16	11
ESP (exchangeable sodium percentage)	0.5	2.2	2
Bulk density (g cm ⁻³)	1.14	1.64	1.23
Water holding capacity (%)	7	23	13
Exchangeable cations (meq 100 g ⁻¹ soil)			
$\begin{array}{c} Ca^{2+} \\ Mg^{2+} \\ Na^{+} \\ K^{+} \end{array}$	4.8 0.8 0.2 0.1	7.5 2.0 0.2 0.2	7.0 1.7 0.2 0.1
Soluble cations (meq 100 L ⁻¹)			
Ca^{2+} Mg ²⁺ Na ⁺ K ⁺	3.2 0.6 0.7 0.2	6.3 1.9 1.0 1.0	5.0 1.6 1.6 0.1
Total C (%)	0.34	1.20	0.62
Organic C (%)	0.26	0.91	0.55
Microbial C (mg kg ⁻¹ soil) *	141	na	263
Total N (%)	0.02	0.13	0.03
Available N- NH_4 (mg kg ⁻¹ soil)	12.61	na	14.46
Available N- NO_3 (mg kg ⁻¹ soil)	3.60	na	8.01
Microbial N (mg kg ⁻¹ soil) *	10.05	na	26.57
Total P (mg kg ⁻¹ soil)	41	461	148
Total organic P (mg kg ⁻¹ soil)	5.04	103	59.35
Available P (mg kg ⁻¹ soil)	0.87	22.80	2.30
Microbial P (mg kg ⁻¹ soil) *	3.16	3.80	3.44

Table 3.1 Characteristi	ics of soils collect	ed from 3 differer	nt sites in South Australia
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Each value represents the mean of 3-4 replicates; na, not available

* Measurements conducted after 2 weeks incubation in moist soil

3.2 Soil salinization

Salinity in the soil was induced using a combination of NaCl and CaCl₂.2H₂O solutions in order to maintain a sodium adsorption ratio (SAR) 1 in all salinity treatments. SAR was low in the soil in order to avoid detrimental effects of sodicity on soil properties (poor soil structure causing low soil-hydraulic conductivity and oxygen availability).

The concentration of Na⁺ was also maintained low in the soil to avoid the toxic effect of Na⁺ on plants. Generally, two equations [SAR = Na $/\sqrt{Ca}$ (mmol L⁻¹) and salt concentration (mg kg⁻¹ soil) = EC (dS m⁻¹) x 640] were used to calculate the amount of Na⁺ and Ca²⁺ salts to be added to the soil to establish SAR 1 (assuming that salts have 100% solubility) at different levels of salinity. For homogenous distribution of all salts in the soil, nutrient solution (described in next section) containing Na^+ and Ca²⁺ salts for a given salinity level was added and thoroughly mixed with the soils using a cement mixer. Soils of each salinity treatment were packed into plastic bags and pre-incubated at 85% of soil water holding capacity (WHC) at 25 °C in the dark for 2 weeks. The incubation will allow the microbial activity to reach the equilibrium after rewetting of air-dry soil. Additionally, if salt is added before the incubation, this may also allow the adaptation of soil microorganisms to a given salinity treatment. Samples were taken prior to the establishment of experiments for electrical conductivity (EC) measurement. Soil salinity is estimated by measuring electrical conductivity (EC) in 1:5 soil-water suspension. This represents a high dilution of saline solution because soil water is lower than 50% in the field, so EC in the field soil water will be more than 10 times the $EC_{1:5}$. The measured $EC_{1:5}$ and the calculated EC (mM), EC_e (dS m⁻¹), as a function of EC_{1:5}, EC (dS m⁻¹), as a function of EC_e, and osmolarity (mosmoles L⁻¹) of the soil solution are given in the respective chapters. Osmolarity is a measure of osmoles of solute per kilogram of solvent (water in the present study). The osmolarity of soil soltuion is calculated as follows: osmolarity (mosmoles L^{-1}) = solute concentration (mM) x number of solute ions. For example, the osmolarity of soil solution with 1 mM NaCl is 2 mosmoles L⁻¹, whereas with 1 mM CaCl2 it is 3 mosmoles L^{-1} .

3.3 Nutrient solution

A nutrient solution was prepared for studies on wheat growth and microbial biomass in saline soils. A nutrient solution was prepared containing the following (mg kg⁻¹ soil): 71.5 Ca (CaCl₂.2H₂O) , 37.0 Mg (MgSO₄.7H₂O), 78.1 K (K₂SO₄), 0.04 mg Fe (Fe-EDTA), 0.147 Mn (MnSO₄.4H₂O), 0.499 Zn (ZnSO₄.7H₂O), 0.050 Cu (CuSO₄.5H₂O), 0.087 B (H₃BO₃), 0.277 Mo (Na₂MoO₄.2H₂O), 0.084 Co (CoCl₂.6H₂O). In some experiments, N (in different forms) and P as KH₂PO₄ were separately prepared and in others were added to the above nutrient solution. Rates of N and P addition will be given in the respective chapters.

3.4 Soil respiration

A closed chamber method was used to measure soil respiration of C-CO₂. A PVC container with mesh at the bottom was filled with 20 g soil (dry weight equivalent). The container was then transferred into a Mason jar containing a small container with 8 ml de-ionized water and closed with an air-tight lid with a septum. The jars were incubated in the dark in a constant temperature room $(28\pm3 \text{ °C})$ throughout the experiments. Released C-CO₂ was measured using a gas analyzer (Servomex 1450 series foodpack) by withdrawing air with needle going through the lid's septum. After each measurement, jars were opened to replace the CO₂-enriched air inside the jars with fresh air. The baseline CO₂ concentration was measured immediately after closing the jars. The frequency of measurement will be described in the respective chapters. To express released C-CO₂ as μ g C-CO₂ g⁻¹ soil h⁻¹ (hour), a spreadsheet developed by Jeff Baldock, CSIRO Land & Water, South Australia, was used. The total amounts of mineralized C derived from plant and soil is described in the results as cumulative respiration.

3.5 Microbial biomass C and N

Microbial biomass C and N were measured by the chloroform fumigation extraction method (Brookes et al., 1985). Two sub-samples of 10 g soil were prepared. One sub-sample was fumigated with chloroform in a desiccator for 24 hours. After addition of 40 ml 0.5 M K₂SO₄, fumigated and non-fumigated samples were placed on an end-over-end shaker for 1 hour and filtered through Whatman no. 42. Extracts were diluted with MilliQ water when required and analyzed for dissolved organic C and N using Skalar autoanalyzer. The concentration of microbial C and N was calculated

from the difference between the concentrations in extracts of fumigated (flushed and microbial C and N) and non-fumigated (flushed C and N) soil samples.

3.6 Microbial biomass P and available P

Microbial biomass P and available P were extracted using an anion exchange resin membrane (Kouno et al., 1995), with hexanol as fumigant. Anion exchange resin strips were pre-conditioned by shaking twice in de-ionized water for 1 hour and then in 4% NaHCO₃. Two sub-samples, each of 2 g soil (dry weight basis), were weighed in separate containers and 30 ml de-ionized water and one 12 x 12 cm resin strip (BDH Laboratory supplies, Poole, PH15 1TD, England) were added to each. To extract microbial P, 1 ml hexanol was added to one sub-sample. The samples were placed on an end-over-end shaker for 16 hours. Resin strips were then washed with de-ionized water to remove adhering soil particles. Adsorbed P was eluded from the resin membranes by shaking the strips for 1 hour in a 0.1 *M* NaCl/HCl solution (29.22 g NaCl and 49.1 ml HCl in 5 L de-ionized water). Extracted P was measured at 712 nm according to Murphy and Riley (1962). Microbial biomass P was calculated as the difference between P concentration of fumigated (microbial and soluble P) and non-fumigated (soluble P) sub-samples.

3.7 Plant residues

Three plant residues namely: lucerne (*Medicago sativa*), canola (*Brassica napus*) and lupin (*Lupinus albus*, L.), collected from Roseworthy, the University of Adelaide, were used in the experiments. The plant residues were oven dried (70 °C) and ground to 1.5 mm. Carbon and N contents in the residues were determined using a LECO dry combustion apparatus. Total P content was measured colorimetrically according to Westernman (1990) after acid digestion (6:1 HNO₃:HClO₄⁻). The residues differed in their properties (Table 3.2). Concentrations of lipids, lignin and cellulose in the residues were added and thoroughly mixed with the soil manually in experiments with small amounts of soil or using a cement mixer for experiments involving large amounts of soil. The application rate was based on total P content in the plant residues.

	Total C	Total N	Total P	C:N	C:P	Lipids	Lignin	Cellulose
Residue	(%)	(%)	(g kg ⁻¹)	ratio	ratio	(%)	(%)	(%)
Lucerne	41.51	1.94	1.2	21.4	345	11.6	9.9	64.1
Canola	40.31	1.22	1.1	33.0	366	11.6	6.5	74.8
Lupin	40.60	2.64	1.1	15.4	369	10.8	4.0	75.3

Table 3.2 Properties of three different plant residues used in experiments

3.8 Plants

Two wheat (Triticum aestivum L.) varieties (Janz and Krichauff) were used in the experiments. These two varieties are commonly grown in South Australia. The seeds were obtained from the Wheat Breeding Group of the University of Adelaide. The seeds were soaked in 12% hypo-chloride solution diluted 3 times with de-ionized water for 20 minutes. After washing with running de-ionized water until the bleach smell disappeared, the seeds were placed in Petri dishes to germinate. Four day-old seedlings were transplanted at 2-cm depth into the soil. The soil weight for a every experiment is mentioned in the respective chapter. After 5 days, seedlings were thinned to three plants per pot. Soil moisture was maintained at 85% WHC through out the experiments by adjusting the weight of the pot with de-ionized water. Plants were grown in a glasshouse with a daily light intensity ranging between 230-679 umol S⁻¹ m⁻². The temperature varied between 25 and 40 °C. The temperature range for a given experiment is mentioned in the respective chapters. At harvest (24 to 30 days after planting), shoots were cut 0.5 cm above the soil surface. Shoots were carefully washed with de-ionized water. Roots were washed out of the soil by running de-ionized water. Fresh plant material was dried in an oven at 70 °C for 48 hours to determine dry weights. Shoot samples were ground and digested using described methods (Section 3.7) for determination of N and P concentrations in the plant (dry weight). In some experiments, inductively coupled plasma-atomic emission spectrometry (ICP) was used to measure total Ca, Mg, K, Fe, Mn, Zn and Cu in plant digests.

3.9 Statistical analysis

Analysis of variance was carried out using Genstat 8 (Genstat 8, 2005, Lawes Agricultural Trust, Rothamsted Experimental Station). The data was also subjected to

Tukey test, when significant differences between treatments were found. In tables, values are means. In most experiments, values in the same column, or row, followed by the same letter are not significantly different at $P \le 0.05$ according to Tukey test. The abbreviation "ns" means not significant. Bars in histograms show standard deviation (STD).

Chapter 4 - Phosphorus availability in saline soils with different texture

4.1 Introduction

Phosphorus supply to plants depends on the concentration of P in the soil solution. The most important soil properties involved in regulating P availability are total P (Brady and Weil, 1996), soil pH, concentration of Fe³⁺, Al³⁺ and Ca²⁺ (Parfitt, 1978), amount and type of clay minerals (White, 1980), Fe³⁺ and Al³⁺ hydroxides (Carreira and Lajtha, 1997) and organic matter (Xie et al., 1991). For example, P is adsorbed to OH⁻ groups of Al³⁺ ions on the surface of clay minerals (Parfitt, 1978). Therefore P sorption is related to the size of surface area of clay minerals occupied by Al-OH groups. Other environmental factors governing P availability in the soil are soil moisture and temperature. In addition, management techniques (P fertilizer type, method of placement, and rate of application) can affect P availability in agricultural soils. Beside the aforementioned factors, salt concentration and type, exchangeable sodium percentage (ESP) and CaCO₃ can influence P availability (Parfitt, 1978). With increasing Ca²⁺ concentration in the medium, P can precipitate as poorly soluble Ca-P. Additionally, increased concentration of Na⁺ and Ca²⁺ in soil solution can lead to increased soil ionic strength (Curtin et al., 1993) which may affect P availability and P uptake by plants.

Because P availability in saline soils with different clay content can be limited by various factors, P supply and availability to plants in saline soils is not well understood which makes the estimation of P fertilization requirement difficult.

It is therefore necessary to study the relationship between soil texture and P availability and to elucidate the relationship between salt (concentration and type) and P availability in the soil.

In order to understand these relationships, three experiments were conducted under laboratory conditions. The major aim of the experiments was to investigate P availability over time after addition of several rates of P at different salinity levels in soils with different texture.

4.2 Materials and methods

4.2.1 Experiment 1

To study the effect of salt type and concentration on P solubility from KH_2PO_4 , six individual solutions denoted S1, S2, S3, S4, S5 and S6 of each NaCl, CaCl₂ and Na₂SO₄ at different concentrations were prepared as shown in Table 4.1.

	Та	Target EC (dS m ⁻¹) in solution								
	2	4	8	12	15	20				
Salt type	alt type Salt concentration (g L ⁻¹)									
	S1	S2	S3	S4	S 5	S6				
NaCl	1.176	2.34	4.68	7.02	8.77	11.70				
CaCl ₂	1.110	2.22	4.44	6.66	8.32	11.10				
Na ₂ SO ₄	1.420	2.84	5.68	8.52	10.65	14.20				

Table 4.1 Concentration (g L⁻¹) of three salt types in 6 separate solutions

The salt concentrations were selected to represent a series of saline conditions of the same EC. The targeted EC values were 2, 4, 8, 12, 15 and 20 dS m⁻¹. In the following chapters, the effect of P fertilization on wheat growth will be examined at these levels. After preparing a stock solution of KH_2PO_4 (1000 mg P L⁻¹), 20 µg P was added to 30 ml (0.66 µg P ml⁻¹) of each solution with 3 replicates per treatment. These samples were shaken end-over-end for 1 hour and then filtered through Whatman no. 42. The amount of soluble P was then measured as previously described.

4.2.2 Experiment 2

To investigate P availability with increasing addition of P in saline soils with different texture (sandy soil and sandy- loam soil), salinity was established in the soils to 3 EC levels using a mixture of NaCl and CaCl₂.H₂O. The average EC_{1:5} values of both soils at given salinity treatments were 0.05, 1.7 and 3.1 dS m⁻¹. Three g soil was spiked with KH₂PO₄ at 0, 100, 200, 400, 600, 1200, 2500 and 5000 μ g P g⁻¹ soil. There were 4 replicates for each combination of soil x salinity x P concentration. After P addition, soils were shaken on an end-over-end shaker for 24 hours in 30 ml de-ionized water. Available P was then extracted using resin membrane and measured after Murphy and Riley (1962).

4.2.3 Experiment 3

Another experiment was set up to evaluate P availability over time in saline sandy and sandy loam soils. Salinity was imposed using the same salt combinations as in the experiment described above. The average $EC_{1:5}$ values were approximately 0.05, 0.98, 1.70 and 3.10 dS m⁻¹. For each soil, KH₂PO₄ was added to 50 g soil at 0, 10, 20, 40, and 60 µg P g⁻¹ soil with 4 replicates per treatment. Throughout the experiment soil moisture was maintained at 85% WHC using de-ionized water. Phosphorus availability was measured in the soils on days 1, 5, 10 and 15.

4.2.4 Statistical analysis

Analysis of variance was carried out using Genstat 8. The data was also subjected to Tukey test, when significant differences between treatments were found. Bars in histograms show standard deviation (STD).

4.3 Results

4.3.1 Effect of salt concentration on P solubility

In the salt-free de-ionized water (control) approximately, 97% of added P was recovered. There was no significant effect to increasing concentration of Na⁺ salts (NaCl and Na₂SO₄) on P solubility (data are not shown), but P solubility significantly ($P \le 0.05$) decreased with increasing Ca²⁺ concentration from S1 to S6 solution (Figure 4.1).



Figure 4.1 Phosphorus concentration ($\mu g \text{ ml}^{-1}$) in 6 solutions with 20 μg P and different concentrations (S1, S2, S3, S4, S5 and S6) of CaCl₂. Columns annotated with the same letter are not significantly different at $P \le 0.05$ according to Tukey test.

At S1, 1.8% more P was extracted from solutions with Na⁺ compared to solutions with Ca²⁺ salts, while at S4, extracted P was 6.4% higher in Na⁺ than in Ca²⁺ solutions. At the highest salinity treatment (S6) 87.7, 77.6 and 89.9% of added P were recovered from NaCl, CaCl₂ and Na₂SO₄ solutions, respectively.

4.3.2 Phosphorus availability in saline soils

Available P 1 day after P addition in saline sandy and sandy loam soils is shown in Table 4.2. In both soils, P availability significantly ($P \le 0.05$) increased with increasing P addition. However, within a given P treatment, P availability decreased with increasing salt concentration ($P \le 0.05$). Recovery of P was higher from sandy soil than from sandy loam soil.

Table 4.2 Available P ($\mu g g^{-1}$ soil) 1 day after addition of 8 concentrations of P-KH₂PO₄ in sandy and sandy loam soils with a mixture of NaCl and CaCl₂ at three salinity levels

	Salinity level	Added P ($\mu g g^{-1}$ soil)									
Soil	$(EC_{1.5} \text{ dS m}^{-1})$	0	100	200	400	600	1200	2500	5000		
	(201.) us in)		Available P (µg g ⁻¹ soil)								
Sandy	0.05	1.2	100	194	390	591	1075	1549	2451		
Sandy loam		2.4	95	185	372	556	1033	1500	1975		
Sandy	1.7	0.16	66	163	259	342	559	852	1159		
Sandy loam		0.13	61	120	237	320	539	826	1109		
Sandy	3.1	0.02	29	61	102	138	296	572	720		
Sandy loam		0.07	22	44	99	127	212	367	487		

4.3.3 Phosphorus availability over time in saline soils with different texture

With increasing P addition, soil P availability significantly increased (Figure 4.2). Phosphorus availability significantly decreased from day 1 to day 5 after which it remained unchanged until the end of the experiment (Figure 4.3). At a given P

concentration, P availability decreased with increasing salt concentration in the soil. With no salt addition (S1), P availability was higher in the sandy soil compared to the sandy loam soil at all times (Figure 4.4).



Figure 4.2 Available P (μ g g⁻¹ soil) on day 15 after addition of 5 concentrations of P-KH₂PO₄ in a sandy loam soil with a mixture of NaCl and CaCl₂ at 4 salinity levels (S1, S2, S3 and S4).



Figure 4.3 Available P (μ g g⁻¹ soil) on days 1, 5, 10 and 15 after addition of 60 μ g P g⁻¹ soil in a sandy loam soil with a mixture of NaCl and CaCl₂ at 4 salinity levels (EC_{1:5} 0.05, 0.98, 1.70 and 3.10 dS m⁻¹ denoted S1, S2, S3 and S4, respectively).



Figure 4.4 Available P (μ g g⁻¹ soil) on days 1, 5, 10 and 15 after addition of KH₂PO₄ at 0, 10, 20, 40 and 60 μ g P g⁻¹ soil in sandy and in sandy loam soils with no salt added (S1)

4.4 Discussion

Phosphorus extractability from saline soils and solutions was not affected by Na^+ salts, (NaCl or Na_2SO_4) at the concentrations used in the present experiments (data not shown). This can be explained by the high solubility of Na-phosphate. Curtin et al. (1993) showed that P availability increases with increasing sodium adsorption ratio (SAR) in the soil. In soils containing high Na^+ concentrations, Na^+ ions replace exchangeable Ca^{2+} ions on the particle surface allowing the dissolution of P bound to soil particles via Ca^{2+} .

In this study, in solutions with similar EC, less P was extracted from Ca^{2+} compared to Na⁺ solutions (Figure 4.1). The decreased P availability after addition of Ca-salts can be explained by the low solubility of Ca-phosphates. Therefore with increasing Ca^{2+} concentration in the medium, less P remains in solution. Moreover, addition of salt increases the soil ionic strength (Curtin et al., 1993). With increasing soil ionic strength, P sorption tends to increase and hence P availability decreases.

At each P addition rate, P availability was lower in the sandy loam than in the sandy soil (Table 4.2). One day after addition of 100 μ g P g⁻¹ soil, the percentage of P recovered was approximately 80, 48 and 18% in the sandy loam compared to 85, 53 and 24% in the sandy soils at S1, S2 and S3, respectively. Since the pH of both soils

was similar, the lower P availability in the sandy loam is probably due to its higher content of clay (20%) compared to the sandy soil (3%). Clay can adsorb P via OH⁻ groups attached to Al^{3+} on the edges of clay lattices and to layers of cations adsorbed to clay surfaces (Parfitt, 1978; Holford, 1997). The sandy loam soil also contains higher organic matter content (0.55% C) than the sandy soil (0.26% organic C). Organic matter can bind P (White, 1980) and result in stabilization of organic P complexes over time.

The availability of P decreased in the first 5 days after addition of 10 to 60 μ g P g⁻¹ soil of KH₂PO₄ in saline soils (Figure 4.4). The initial decline can be attributed to rapid P sorption by soil Al³⁺ and Fe³⁺ on clay and organic matter surfaces. Increased microbial biomass in the soil could also have contributed to the reduced availability of P in both soils over 15 days (see Chapters 7 and 8).

4.5 Conclusions

The availability of P is reduced with increasing concentration of salts, particularly of Ca^{2+} , but the concentration of available P stabilizes within 2 weeks after addition of P -KH₂PO₄. The greater P availability in the sandy soil than in the sandy loam soil is probably due to the higher content of clay minerals and organic matter in the sandy loam soil.

The relationship between soil salinity and P availability described here is important for the interpretation of the data in the following chapters.

Chapter 5 - Wheat growth in a saline sandy loam soil as affected by N and P

application

5.1 Introduction

Salt accumulation has a negative effect on soil chemical, physical and biological properties (Szabolcs, 1989). Salinity can also suppress plant growth via specific and non-specific effects (Maas, 1996). Plant growth reduction due to non-specific salt effects is related to the total soluble salts or osmotic potential of the soil solution (Maas, 1996). With the increase of salt concentration, the osmotic potential of the soil solution decreases (Bernstein et al., 1974; Maas and Hoffman, 1977) resulting in reduced availability of water to plant roots (Al-Karaki, 1997) and thus decreased nutrient uptake (Pessarakli, 1991; Grattan and Grieve, 1999). Limited water uptake, due to increased soil osmotic stress, adversely affects dry matter partition, cell extension, leaf photosynthesis and transpiration (Greenway and Munns, 1980; Frechilla et al., 2001). The specific salt effect (ionic stress) is related to increased plant uptake of Na⁺ and/or Cl⁻ (Greenway and Munns, 1980; Maas, 1996). Ionic stress occurs when excessive amounts of Na^+ and Cl^- enter the plant in the transpiration stream and accumulate to high levels in cells of transpiring leaves reducing enzyme activity (Jeschke, 1984) membrane selectivity (Bohra and Dorffling, 1993) and plant metabolism (Marschner, 1995) and leading to a premature death (Munns and Termat, 1986). The assimilation is also reduced because of the effects of salt on leaf expansion rate. The osmotic and the ionic stresses may result in a nutritional imbalance in the plants (Grattan and Grieve, 1999).

As described in the literature review (Chapter 2), most of the soluble salts can change the soil pH and, in addition to soil minerals, react with fertilizer P making it less available to plants (Sharpley et al., 1992; Curtin et al., 1993; Naidu and Rengasamy, 1993). Salinity can also indirectly affect N availability through inhibition of microbial N mineralization and immobilization processes. Salinity also can decrease N uptake (Kafkafi et al., 1982) and P uptake (Chabra et al., 1976; Manchanda et al., 1982) by affecting root growth and/or through inhibition of NO₃⁻ and P uptake by Cl⁻. In salinized solutions, NaCl up to 80 m*M* (EC 8 dS m⁻¹) decreased N incorporation into proteins (Helal and Mengel, 1979) and up to 150 m*M* (EC 15 dS m⁻¹) decreased P uptake, content and concentration in plants (Martinez and Lachli, 1994). Salinity may

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also reduce P flux through xylem by influencing plant metabolic activities (Navarro et al., 2001). Nitrogen and P deficiency can decrease the salt tolerance of plants (Gibson, 1988).

In the saline soils of arid and semi-arid regions, wheat is grown widely, because it is moderately tolerant to salinity (Maas, 1996). In wheat, the cause of reduced growth by salinity was attributed mainly to the osmotic effect. Termat and Munns (1986) showed that under saline conditions, reduced growth of wheat is also due to reduced rate of transport of essential nutrients to the shoot. Fertilization can significantly influence plant response to salinity (Champagnol, 1979; Grattan and Grieve, 1999).

Studies conducted in solutions with salinity up to EC 15 dS m⁻¹ (Cox and Reisenauer, 1973; Ali et al., 2001; Al-Mutawa and El-Katony, 2001) or in the loamy sand with up to EC_e 12.1 dS m⁻¹ (Irshad et al., 2002) indicated that N applied at 100 kg ha⁻¹ can alleviate salinity stress. Wheat growth responded positively for P addition at 48 kg ha⁻¹ from different P fertilizers in a sandy loam soil with EC_e 6 dS m⁻¹ and SAR 28 (Mehdi et al., 2001). Also, wheat straw and grain yield were highest at 150 kg P₂O₅ (64.5 kg P) ha⁻¹, in the silt clay loam with EC_e 5.6 dS m⁻¹ and SAR 14.02 and in the silty loam with EC_e 12 dS m⁻¹ and SAR 24.02 (Abid et al., 2002). Additionally, wheat growth was highest with addition of 50 kg P ha⁻¹ in a sandy soil irrigated with water having 15-19 dS m⁻¹ (Manchanda et al., 1982). However, other researchers showed that high and non-limiting concentrations of P do not affect plant response to salinity (Langdale and Thomas, 1971; Rogers et al., 2003). It is known that at a given level of salinity the growth and yield of crops are lower if nutrients are limiting (Hassan et al., 1970).

Nitrogen and P are generally required in large amounts by plants (Marschner, 1995). Maximum response to fertilization of plants grown in saline soils can be expected when plants are given sufficient amounts of nutrients, particularly N and P. Increasing P fertilization with high N rates may enhance P uptake, and thus increase plant growth under saline non-sodic conditions. Information is needed to investigate the response of wheat to N and P fertilization in the sandy loam along a gradient of salinity levels under non-sodic (SAR 1) or alkaline conditions.

A glasshouse experiment was carried out with the aim to test the hypothesis that addition of N and P fertilizers can enhance wheat N and P uptake and growth under saline, non sodic, conditions.

5.2 Materials and methods

A preliminary experiment was conducted to evaluate the growth of wheat (cv Janz) in a sandy loam soil at different levels of salinity.

The soil collected from Monarto, South Australia with characteristics shown in Table 3.1 was used in this study. After air drying the soil and sieving it to 2 mm, four salinity treatments were established in the soil using mixtures of NaCl and CaCl₂.2H₂O. Selection of salinity treatments was based on information collected from the literature and through personal communication with Dr Rengasamy. Sodium and Ca²⁺ salts were calculated to maintain SAR 1 in all salinity treatments (Table 5.1).

 Table 5.1 Amounts of NaCl and CaCl₂.2H₂O added to a sandy loam soil to impose 4 salinity treatments with SAR 1

		NaC	1	CaCl ₂ .2H		[₂ O	Osmolarity	EC _{1:5}	ECe	EC
Salt	mg	тM	mM	mg	mM	mM	osmoles L ⁻¹			
level	$k \sigma^{-1}$	1 = -1 = $1 = -1$ in soil		$k a^{-1}$	soil	in soil	in soil	dS m ⁻¹		
	ĸg	son	soln	kg soll		soln	soln			
S1	0	0	0	0	0	0	0	0.18	2.4	4.8
S2	306	5.23	47.6	4025	27.38	249.0	824	1.36	18.7	37.4
S 3	399	6.82	62.1	6849	46.59	423.5	1394	2.00	27.6	55.2
S 4	475	8.11	73.9	9694	65.94	599.5	1946	2.67	36.8	73.6

Soil soln = soil solution. The concentration (m*M*) was estimated in the soil solution. $EC_{1:5}$ was measured in the soil after incubation prior to establishment of the glasshouse experiment. EC_e was calculated based on the relationship between the texture and the moisture of the sandy loam (Slavich and Petterson, 1993). EC was calculated knowing that the EC of the soil solution at field capacity is approximately double the EC_e (Richards, 1954).

5.2.1 Nutrient solution

Nutrients were added in the form of a nutrient solution to give the following concentrations (mg kg⁻¹ soil): 33 N (NH₄NO₃), 71.5 Ca (CaCl₂.2H₂O), 37.0 Mg (MgSO₄.7H₂O), 78.1 K (K₂SO₄), 0.04 Fe (Fe-EDTA), 0.147 Mn (MnSO₄.4H₂O), 0.499 Zn (ZnSO₄.7H₂O), 0.050 Cu (CuSO₄.5H₂O), 0.087 B (H₃BO₃), 0.277 Mo (Na₂MoO₄.2H₂O) and 0.084 Co (CoCl₂.6H₂O). Salts were dissolved in the nutrient solution, which was then thoroughly mixed with the soil using a cement mixer. The

amended soils were moisten at 50% WHC and incubated in plastic bags for 2 weeks at 25 ± 4 °C. Phosphorus was applied as lucerne residue at the equivalent of 20 mg kg⁻¹. Treated soil (4 replicates) was then filled into plastic pots (1 kg) and transferred to a glasshouse (25 °C).

5.2.2 Plants

Seeds of wheat (cv Janz) were surface sterilized as described in Chapter 3 and germinated in Petri dishes. On day four, 8 germinated seeds were transplanted into the soil at 2-cm depth. Four days after transplanting, the plants were thinned to three plants per pot. The day temperature in the glasshouse ranged between 28-30 °C. De-ionized water was added daily in order to maintain soil moisture at approximately 85% WHC. Despite daily watering, lower soil moisture could not be prevented. On day 8, plants showed wilting symptoms and most of them died in the following days except in the control (Plate 5.1). The experiment was therefore terminated on day 12. The salinity levels chosen were considered to be below the critical concentration for wheat plants (Dr Rengasamy, Personal Communication). Therefore it was concluded that Janz may be particularly salt-sensitive.



Plate 5.1 Wheat growth in a sandy loam soil with $EC_{1:5}$ 0.18, 1.36, 2.00 and 2.67 dS m⁻¹

A new glasshouse experiment was set up, this time with wheat cv Krichauff, which is said to be salt tolerant. In addition, the aim of this experiment was to study the effects of N and P fertilizers on wheat growth under saline conditions. Nutrients and salts were added similarly as in the previous experiment except that N was excluded from the nutrient solution and P was added as KH_2PO_4 , not in the form of residues.

After the 2-week pre-incubation of amended saline soil, N was applied at (equal amounts of N from each source; 1:1) as $Ca(NO_3)_2.4H_2O$ and KNO_3 at 50 (N50), 100 (N100) and 200 (N200) mg N kg⁻¹ soil, along with P, which was applied at 0 (P0), 30 (P30) and 60 (P60) mg kg⁻¹ soil, in the form of KH_2PO_4 . Soil was filled in plastic pots (600 g capacity) with 5 holes (1-mm diameter) covered with mesh at the bottom. The holes were created to allow air exchange and prevent anoxia. Pots were transferred to the glasshouse. Seeds of wheat (cv Krichauff) were germinated as described above. After transplanting, pots were watered daily with de-ionized water up to 85% WHC by weighing the pot. However, as Janz in the previous experiment, Krichauff died in at all treatments with added salt.

The experimental set up of the 3rd experiment was the same as in experiment 2, but the salinity levels were lower (Table 5.2). From day 8 to the end of the experiment, the plants were irrigated 2-3 times a day to maintain 85% WHC. The frequent watering was necessary because the temperature in the glasshouse often reached 33 °C.

NaC		1	Ca	aCl ₂ .21	H ₂ O	Osmolarity	EC _{1:5}	ECe	EC		
Salt	mg mM		m <i>M</i>	mg	тM	m <i>M</i>	mM osmoles L^{-1}				
Level	$k \alpha^{-1}$	coil	in soil	ka ⁻¹	oil	in soil	in soil	$dS m^{-1}$			
	ĸg	8011	soln	kg son		soln	soln				
S1	0	0	0	0	0	0	0	0.19	2.6	5.2	
S2	79	1.35	12.3	268	1.82	16.6	74	0.32	4.4	8.8	
S3	117	2.00	18.2	588	4.00	36.4	145	0.49	6.7	13.4	
S4	146	2.49	22.8	918	6.24	56.8	216	0.67	9.2	18.4	
S 5	170	2.90	26.6	1255	8.53	77.6	286	0.86	11.8	23.6	

Table 5.2 Amounts of NaCl and CaCl₂.2H₂O added (in addition to the basal nutrient solution) to each kg soil to impose 5 salinity treatments

Soil soln = soil solution. $EC_{1:5}$ was measured in the soil after incubation prior to establishment of the glasshouse experiment.

5.2.3 Analyses

On day 14, soil available P was measured in the soil (samples were taken from 0-2 cm a cork borer). At harvest (day 24), shoots were cut off approximately 0.5 cm from the soil surface, and roots were washed from soil by running tap water. Plant shoots and roots were dried and then weighed. Nitrogen and P concentrations in the shoots were measured as described in Chapter 3.

5.2.4 Design and statistical analysis

The experiment was set up in a completely randomised design (5 salinity treatments, 3 P and 3 N treatments) with three replicates. Three-way analysis of variance was carried out using Genstat 8. The abbreviation "ns" means not significant. Bars in histograms show standard deviation (STD).

5.3 Results

5.3.1 Nitrogen effect on soil salinity

In experiment three, salinity was not measured in the soil after addition of N fertilizers from KNO_3 and $Ca(NO_3)_2.4H_2O$ sources, but only after a 2-week incubation time of the sandy loam with Na⁺ and Ca²⁺ salts and nutrients, but without N (described in Section 5.2.1). As calculated, with increasing N addition, the salinity of soil solution increased (Table 5.3).

Table 5.3 Calculated EC (dS m^{-1}) of soil solution as affected by addition of KNO₃ and Ca(NO₃)₂.4H₂O at different application rates

	KNO ₃			$Ca(NO_3)_2.4H_2O$			Osmolarity	EC ^b
N level ^a	mg	тM	mM	mg	тM	mM	mosmoles L ⁻¹	dS m ⁻¹
	kg	soil	soil soln	kg	soil	soil soln	soil soluti	on
N50	180	1.79	16.2	210	0.89	8.1	57	5.5
N100	360	3.57	32.5	421	1.78	16.2	114	11.1
N200	721	7.14	64.9	842	3.57	32.5	227	22.2

Soil soln = soil solution.

^a Nitrogen was applied at 1:1 (equal amounts of N from each source).

^b EC is in the soil solution, and was estimated using the following equation: salt concentration (mg L^{-1}) = EC (dS m⁻¹) x 640].

5.3.2 Effects of N, P and salinity on wheat growth

Individual effects of salts, N and P were significant for shoot and root dry weights, root/shoot ratio and shoot N and P concentration. The interaction between N, P and salinity was significant for all variables, except available P (Table 5.4).

Interactions	Shoot dry Weight	Root dry weight	Root/Shoot Ratio	Plant N	Plant P	Soil available P
Salinity	0.01	0.01	3.33	0.20	0.07	0.31
Ν	0.01	0.01	2.58	0.31	0.05	0.24
Р	0.01	0.01	2.58	0.31	0.05	0.24
Salinity x N	0.01	0.01	5.77	0.70	0.12	0.54
Salinity x P	0.01	0.01	5.77	0.70	0.12	0.54
N x P	0.01	0.01	4.47	0.54	0.09	0.41
Salinity x N x P	0.02	0.02	9.99	1.22	0.22	ns

Table 5.4 Results of statistical analysis; least significant differences (LSD) and significance at 5% level, of main and interactive effects of P, N and salinity on wheat growth, N and P concentration and available P in the soil

Values of LSD are significant at $P \le 0.05$; ns, not significant

5.3.3 Nitrogen and P effects on shoot dry weight under saline conditions

Shoot and root dry weights had a similar trend, but with different magnitudes with N, P and salinity treatments. At all P and N treatments, shoot dry weight decreased with increasing soil salinity. Results of only shoot dry weight as affected by N fertilization at 50, 100 and 200 mg N kg⁻¹ soil with 60 mg P kg⁻¹ soil under saline conditions (5 levels; Table 5.2) are shown in Figure 5.1.



Figure 5.1 Shoot dry weight (g pot⁻¹) as affected by the osmolarity (mosmoles L⁻¹) of soil solution as calculated after KNO₃ and Ca(NO₃)₂.4H₂O addition at 50, 100 and 200 mg N kg⁻¹ soil in the sandy loam with 60 mg P kg⁻¹ soil and Na⁺ and Ca²⁺ salts at 5 salinity levels defined in Table 5.2.

At a given salinity treatment shoot dry weight significantly decreased with increasing N addition. In contrast, at a given salinity treatment shoot dry weight significantly increased with increasing P addition (Figure 5.2). Under saline conditions, highest shoot dry matter was obtained with the combination N50 and P60.

With N50, effect of P30 and P60 on shoot and root dry weights (DW) is shown in Figure 5.2. In S1, compared to P0, shoot DW (Figure 5.2) was approximately 20 and 25% higher and root DW (Figure 5.2) was approximately 8.5 and 11% higher with addition of P30 and P60, respectively.



Figure 5.2 Dry weight of shoot and root (g pot⁻¹) with addition of 0, 30 and 60 mg P and 50 mg N kg⁻¹ in a sandy loam soil at five salinity levels.

Relative to S1, with P30 shoot dry weight was approximately 50, 35, 23 and 15%, whereas with P60 shoot dry weight was 90, 80, 50 and 20% at S2, S3, S4 and S5, respectively. Root dry weight was higher with P60 compared to P30 at all salinity treatments, but differences between P30 and P60 were not as high as in shoot dry weight at every salinity treatment.

5.3.4 Shoot nitrogen concentration

Salinity had no effect on shoot N concentration. Shoot N concentration significantly increased with increasing N fertilization from N50 to N200 in all P treatments. At a

given addition rate of N, shoot N concentration was higher when P was added compared to the control (P0) with no clear difference between P30 and P60 (Figure 5.3).



Figure 5.3 Shoot N concentration (mg g^{-1} DW) as affected by addition of P at 0, 30 and 60 mg P kg⁻¹ and N at 50, 100 and 200 mg kg⁻¹ soil in a sandy loam soil with five salinity treatments.

5.3.5 Shoot phosphorus concentration

Phosphorus concentration in plant shoots significantly ($P \le 0.05$) increased with the addition of P fertilizers. Increasing the addition rate from P30 to P60 increased shoot P concentration (Figure 5.4). Compared to N50, shoot P concentration was significantly lower at N100 and N200. At N100 and N200, shoot P concentration decreased with increasing soil salinity from S2 to S5.



Figure 5.4 Shoot P concentration (mg g^{-1} DW) as affected by addition of P at 0, 30 and 60 mg and N at 50, 100 and 200 mg kg⁻¹ soil in a saline sandy loam soil.

5.3.6 Soil available P

Phosphorus availability decreased with increasing soil salinity (Table 5.5). Available P increased significantly ($P \le 0.05$) with addition of P fertilizers and was higher at P60 than at P30. Nitrogen addition did not significantly affect P availability. At a given N level, available P decreased with increasing salinity both at P30 and P60.

P rate	N rate	S1	S2	S3	S4	S 5
(mg kg	¹ soil)	A	vailabl	e P (mg	kg ⁻¹ soi	1)
0		0.26	0.12	0.08	0.06	0.06
30	50	13.58	11.67	10.36	9.99	4.86
60		34.11	32.48	30.08	28.20	26.36
0		0.33	0.14	0.10	0.09	0.09
30	100	13.92	12.88	12.68	11.42	8.20
60		37.06	35.69	34.61	30.77	28.77
0		0.37	0.34	0.26	0.22	0.15
30	200	14.53	13.89	13.54	12.65	8.01
60	_00	38.45	37.78	36.00	34.92	30.91

Table 5.5 Available P (mg kg⁻¹ soil) 14 days after addition of 0, 30 and 60 mg P and 50, 100 and 200 mg N kg⁻¹ soil in a sandy loam soil with five salinity treatments
5.4 Discussion

The results emphasize the complex relationship between salinity and N and P fertilization and show that plant response is highly dependent upon the level of salinity and P and N fertilization rates in the growth medium.

In the first two experiments, the values of $EC_{1:5}$ in all treatments without added salts (shown in Table 5.1) indicated that salinity estimated approximately EC_e 18.7, 27.6 and 36.8 dS m⁻¹ in S2, S3 and S4, respectively, was too high for plants. Therefore plants did not respond to addition of fertilizers, except in the control (S1).

In experiment three, as expected, salinity reduced plant dry matter at all N and P application rates (Figures 5.1 and 5.2). Several studies showed that generally N addition (Nour et al., 1989; Ragheb et al., 1993; Soliman et al., 1994; Singh and Sharma, 2001) and particularly at 100 mg kg⁻¹ soil can alleviate salt stress and enhance the plant growth in saline solutions with up to EC 15 dS m⁻¹ (Cox and Reisenauer, 1973; Ali et al., 2001; Al-Mutawa and El-Katony, 2001) and in a loamy sand soil up to EC_e 12.1 dS m⁻¹ (Irshad et al., 2002). In contrast, the results of present study showed that increasing amounts of N, from N50 to N200, had a detrimental effect on shoot dry weight in the soil with no Na^+ or Ca^{2+} salts added (S1; EC_e 2.6 dS m⁻¹), and that the effect became more pronounced with increasing soil salinity (Figure 5.1). A positive interactive effect of N and P on plant growth can therefore only be seen at lowest N rate (N50). In agreement with Villa-Castorena et al. (2003) in this study N fertilizers strongly contributed to soil salinity. Calculations (Table 5.3) indicated that solutions of N had EC 5.5, 11.1 and 22.2 dS m⁻¹ in N50, N100 and N200, respectively. Also, at the end of the experiment in the soil with no P added (P0), measurement of EC in S1 indicated that in N100 and N200 EC_{1:5} were 0.24 and 0.32 dS m⁻¹, equivalent to EC_e 3.3 and 4.4 dS m⁻¹, at N100 and N200, respectively. Hence, plants were possibly P limited, and the increase in the soil EC may have over-ride the positive effect of N on plant growth. However, with no Na⁺ and Ca²⁺ salts added (S1), EC_{1:5} values in soil with N100 and N200 fall in a salinity range indicating that salinity is low (George and Wren, 1985; Rogers et al., 2005), and therefore might not limit the plant growth. However, the plant growth reduction with increasing N addition can not be explained by an increase in EC alone, but also because of the low P concentration (0.11 mg P g⁻¹ DW) in the plant which is clearly shown in Figure 5.4. With or without sufficient P concentration in the plant, other explanations for the negative effect of high N addition rates include the high temperature in the glasshouse during the experiment as shown in Lawlor et al. (1988) or reduced uptake of micronutrients, particularly Zn, as found by Warren and Adams, 2002).

It appears that the addition of 50 mg N kg⁻¹ soil in addition to the natively available N (14 mg N-NH₄ and 8 mg N-NO₃) covered N requirements at the initial growth stage of Krichauff. In this study, salinity did not affect shoot N concentration, with increasing N addition (Figure 5.3). Effect of rate and form of N on wheat growth will be investigated in more detail in the following chapters.

Sufficient uptake of P by plants is of utmost importance; however, its significance for growth under saline conditions is unclear (Champagnol, 1979). Several studies showed that salinity decreased P availability (Sharpley et al., 1992; Curtin et al., 1993; Naidu and Rengasamy, 1993), therefore it affects P uptake and plant growth. In the present study, P availability decreased with increasing soil salinity (discussed in Chapter 4). However, the availability of P increased with increasing P fertilization and was at sufficient levels (Singh et al., 2000) in the soil (Table 5.5).

Several studies showed that P fertilization had a positive effect on crop growth in saline soils (Manchanda et al., 1982; Al-Karaki, 1997; Abid et al., 2002). Other researchers showed that high and non-limiting concentrations of P do not affect plant response to salinity (Langdale and Thomas, 1971; Rogers et al., 2003). The present study demonstrated that with increasing P fertilization, shoot and root dry weights increased in a sandy loam with up to ECe 9.2 (Figure 5.2), irrespective of salinity from N fertilizers (Figure 5.3). The results shown by Al-Karaki (1997), Navarro et al. (2001) and Rogers et al. (2003) indicated that shoot P concentration decreased with increasing soil salinity. In these studies NaCl was the only source of salinity. It is possible that in combination with increasing osmotic stress, increased uptake of Na⁺ and/or Cl⁻, to levels that can be toxic, reduced plant growth and therefore inhibited P uptake. Another possible reason for the low P uptake is inhibition of P uptake by Cl⁻ (Chabra et al., 1976). In the current study, Na⁺ was used to impose soil salinity, but its concentration in the soil (it can be calculated from information given in Table 5.2) remained at a level that would theoretically have no negative effect on the soil structure or cause toxicity to plants. Also, shoot P concentration was higher at P60 compared to P30. Additionally, with N50 shoot P concentration increased, but only to up to S3, and with N100 and N200, shoot P concentration decreased with increasing soil salinity from S2 to S5 with all P treatments (Figure 5.4). Moreover, at a given N fertilization, shoot P concentration was higher at P60 compared to P30. In the soil with added Na⁺ and Ca²⁺ salts (S2-S5), N fertilizers applied at rates higher than N50 contributed to the salinity of soil solution with approximately 11.1 and 22.2 dS m⁻¹ at N100 and N200, respectively. Apparently under these conditions osmotic stress would be dominant and causes growth depression in wheat. With N50, the results indicate that plants required greater P than that found with P30 soil under saline conditions, although shoot P concentration was optimal with P30 and P60 (Reuter and Robinson, 1997). The fact that P fertilization improved plant growth (Figure 5.2) suggests that in the sandy loam soil, P was limiting for wheat growth.

Mehdi et al. (2001) reported that addition of only 48 kg ha⁻¹ from different P fertilizers increased enhanced wheat growth in the sandy loam with EC_e 6 dS m⁻¹ and SAR 28. In this study, increasing P addition up to 60 mg kg⁻¹ soils greatly improved wheat growth in the sandy loam under saline non-sodic (SAR 1) conditions. This effect was found only to a certain level of soil salinity (S4; EC_{1:5} 0.67 dS m⁻¹ equivalent to EC_e 9.2) which was induced by Na⁺ and Ca²⁺ salts, but the actual osmolarity in the soil solution, as affected by Na⁺ and Ca²⁺ salts and N50, was approximately 273 mosmoles L⁻¹. Beyond this salinity level, the negative effect of salinity became too strong and therefore increasing P addition above 30 mg kg⁻¹ soil is insignificant.

5.5 Conclusions

In the sandy loam soil with SAR 1, the growth of wheat was decreased by the presence of salts in the soil and completely inhibited with salinity $EC_{1:5}$ 1.36 dS m⁻¹. This depression of wheat growth started in the soil with 131 mosmoles L⁻¹ in the soil solution, and was aggravated with N fertilization at rates higher than 50 mg kg⁻¹ soil. Increasing P addition increased wheat growth, but P fertilization beyond a 273 mosmoles L⁻¹ (soil solution), or at least $EC_{1:5}$ 0.67 dS m⁻¹, equivalent to EC_e 9.2, did not improve plant growth. In this study, in the sandy loam soil, highest wheat growth was found at 60 mg P kg⁻¹ soil with 50 mg N kg⁻¹ soil.

Unexpectedly, N addition at 100 and 200 mg N kg⁻¹ soil had a negative effect on plant growth. In this study, negative effects such as increased soil salinity due to N addition from several sources at a particular rate may have resulted in the growth reduction of wheat. This is further investigated in the following experiments.

Chapter 6 - Wheat growth in a saline sandy loam soil as affected by N form and application rate

6.1 Introduction

Addition of nutrients in forms efficiently used by plants can reduce the amount of required fertilizers and minimize environmental risks. Also, adequate supply of N can promote plant growth and increase crop production. This could increase economic returns of crops in salt affected soils. In Chapter 5, it was shown that with increasing salinity in the soil, addition of 60 compared to 30 mg P kg⁻¹ increased plant N and P concentrations and wheat growth. It was also shown that addition of several N salts at rates higher than 50 mg N kg⁻¹ soil negatively affected wheat growth.

In saline soils, the proper use of N fertilizers could reduce the adverse effects of salinity on plant growth and improve N content of plants. The form in which N is applied to salt-stressed plants may influence plant-nutrient-salinity relationships (Martinez and Cerda, 1989). Unlike many other nutrients, plants can take up N from the soil in two ionic forms, NH_4^+ or NO_3^- (Traore and Maranville, 1999). The N form taken up by plants depends on the available N form, soil characteristics and plant species, variety and age.

Nitrogen form can affect the availability of N to plants as a result of differences in mobility of each form in soil solution. It also can affect the availability of other nutrients such as P. Addition of NH_4^+ increased the capacity of plant roots to take up P from soils (Hofmann et al., 1994). NH_4^+ uptake by plants decreased rhizosphere pH because of excretion of H^+ from growing plants (Gahoonia et al., 1992). High concentration of NH_4^+ ; however, especially when it is the only N source in the soil, can be toxic to plants (Van der Eerden, 1982; Gerendas et al., 1997; Britto and Kronzucker, 2002). In these studies, the lower root and shoot biomass and nutrient uptake with NH_4^+ , compared to NO_3^- nutrition, were attributed to plant toxicity with NH_4^+ when applied at 3-6 m*M*. Under non-saline conditions, most plant species exhibit preference for NO_3^- compared to NH_4^+ nutrition (Al-Mutawa and El-Katony, 2001). Compared to NH_4^+ , NO_3^- nutrition increased the uptake of cations such as Ca^{2+} , Mg^{2+} and K^+ (Cox and Reisenauer, 1973; Mahmood and Kaiser, 2003). Nitrate nutrition can also cause an increase in the rhizosphere pH through OH⁻ and HCO₃⁻ release. These mechanisms can enhance mobilization of soil P in acidic soils (Gahoonia et al., 1992).

Nitrogen uptake can therefore play an important role in enhancing the uptake of nutrients (Warren and Adams, 2002), and hence salt tolerance of plants. In studies conducted in solutions, compared to NH₄-N, NO₃-N was taken up at higher rates by maize in saline solution with EC 6-8 dS m⁻¹ (Lewis et al., 1989). Also, compared to NH₄⁺, NO₃⁻ nutrition increased the plant uptake of Ca²⁺, Mg²⁺, K⁺ and N and plant biomass at EC 10 dS m⁻¹ induced using NaCl (Mahmood and Kaiser, 2003). Moreover, in solution with EC 15 dS m⁻¹, NO₃-N uptake by barley increased with increasing Ca²⁺ salts (Ward et al., 1986). Additionally with 2 m*M* N for wheat (Al-Mutawa and El-Katony, 2001) and 5 m*M* for pea (Frechilla et al., 2001). compared to NH₄⁺, plant growth was greater with NO₃⁻ nutrition with increasing salinity by NaCl up to 150 m*M*, equivalent to 15 dS m⁻¹.

Although most crops can grow on either N form, supplying plants with mixtures of NO_3^- and NH_4^+ often results in better vegetative growth and enhanced nutrient accumulation than either form alone (Haynes and Goh, 1978) in hydroponics with EC 8 dS m⁻¹ (Ali et al., 2001) and in a loamy sand soil with EC_e 12.1 dS m⁻¹ (Irshad et al., 2002).

Most studies on the effect of N form on plant growth under saline conditions have been conducted in hydroponics (Lewis et al., 1989; Ali et al., 2001; Al-Mutawa and El-Katony, 2001; Inal and Tarakcioglu, 2001), where nutritional elements as well as Na⁺ salts were soluble and completely available to plants. With no comparison with other N rates, Irshad et al. (2002) concluded that with 100 mg N kg⁻¹, there is no significant differences between NH_4^+ and NO_3^- fertilizers for wheat growth. Crops and plant species differ not only in their response to osmotic or ionic stress, but also in their ability to take up sufficient amounts of essential nutrients. Salinity negatively affected the growth of plant roots and reduces nutrient (N, P, K, etc.) uptake, translocation and assimilation (Grattan and Grieve, 1999). Nitrogen addition in different forms can enhance plant growth under saline conditions, but plants may not respond positively to N addition, if they are also limited by other nutrients, particularly P (Reichman and Grunes, 1966).

In the study of Irshad et al. (2002) as compared to other N fertilizers, wheat growth was highest with NH_4NO_3 , but shoot P concentration fell in a range (1.3-1.8 mg g⁻¹ DM) under all N treatments indicating that wheat was P deficient. The conditions

differ between soil in terms of pH, nutrient mobility and availability, soil structure and plant nutrient and water uptake, and are also completely different from those in solution culture. The negative effect of the high N rates (100 and 200 mg N kg⁻¹ soil) on plant growth (Chapter 5) could be due to the osmotic stress caused by excess N fertilizers in general or may be specific to NO_3^- . There is no information available about wheat response to N addition in different forms and at different application rates in the sandy loam under saline non-sodic or alkaline conditions. Therefore the aim of the experiment described in this chapter was to assess the effect of N added as NH_4^+ , NO_3^- or in combination as NH_4NO_3 on wheat growth in a saline sandy loam soil with SAR 1 and neutral pH.

6.2 Materials and methods

A sandy loam soil collected from Monarto, South Australia (Chapter 3) was used in this study. After air drying the soil and sieving it to 2 mm, three salinity treatments were established in the soil using mixtures of NaCl and CaCl₂.2H₂O (Table 6.1). Similar salinity treatments had been used in Chapter 5. A nutrient solution, containing, in addition to 60 mg P (KH₂PO₄) kg⁻¹ soil, all essential nutrients without N; Section 5.2.1, was added as described in Chapter 5. Salts were dissolved in the nutrient solutions which were then thoroughly mixed with the soil using a cement mixer. The amended soils were moistened to 50% WHC and incubated in plastic bags for 2 weeks at 25 ± 4 °C.

Table 6.1 Amounts of NaCl and CaCl ₂ .2H ₂ O added to a sandy loam soil and values of
$pH_{1:5}$, EC _{1:5} and concentrations (mg or m <i>M</i>) of salts kg ⁻¹ soil and in the soil solution

		NaC	1	Ca	Cl ₂ .2H	2 0	Osmolarity	EC _{1:5}	EC _{1:5} EC _e	
Salt	mg	тM	mM ^a	mg	тM	тM	mosmoles L ⁻¹	dS m ⁻¹		
level	1× a ⁻¹	soil	soil	1×a ⁻¹	soil	soil	in soil soln			
	ĸg	son	soln	кg	son	soln	III SOII SOIII			
S1	0	0	0	0	0	0	0	0.21	2.8	6.32
S2	117	2.00	18.2	588	4.00	36.4	145	0.48	6.6	6.14
S3	170	2.90	26.6	1255	8.53	77.6	286	0.86	11.8	6.07

^a The concentration (m*M*) was estimated in the soil solution; soil soln = soil solution. $EC_{1:5}$ and $pH_{1:5}$ were measured in the soil after incubation prior to establishment of the glasshouse experiment.

6.2.1 Nitrogen treatments

Solutions of KNO₃, $(NH_4)_2$.SO₄ or NH_4NO_3 were added to incubated soil at 0, 50, 100 and 200 mg N kg⁻¹ soil denoted N0, N50, N100 and N200. Plastic pots were filled with 500 g of treated soil and then transferred to the glasshouse. Seeds of wheat (cv Krichauff) were germinated as described (Section 3.3). After transplanting, plants were watered daily with de-ionized water to 85% WHC.

6.2.2 Analyses

On day 23, soil pH and EC were measured. On day 24, shoots were cut off approximately 0.5 cm from the soil surface, and roots were washed from soil by running tap water. Plant-shoots and roots were dried and then weighed. Shoots were ground and digested as described in Section 3.7 for determination of total N, P, K, Ca, Fe, Mn, Cu and Zn.

6.2.3 Design and statistical analysis

There were four addition rates of three N forms in combination with three salinity treatments replicated three times and arranged in a randomized complete block design. For collected data, two- and three-way as well as analysis of variance as well as Tukey test were carried out using Genstat 8. In tables, values are means. Values in the same column followed by the same letter are not significantly different at $P \le 0.05$ according to Tukey test. The abbreviation "ns" means not significant. Bars in histograms show standard deviation (STD).

6.3 Results

6.3.1 Effects of N form and N addition rate on wheat growth parameters under non-saline conditions

In the control soil, without added Na^+ and Ca^{2+} salt, the interaction between N form and N application rate was significant for dry weights of shoot and root (Table 6.2). While the N addition rate had a significant effect on all growth parameters, N form and the interaction of N form with N rate were not significant for root/shoot ratio.

Shoot and root dry weights were lowest in the control (N0). With addition of N50, shoot and root dry weights increased significantly ($P \le 0.05$), but no significant

differences were found between N forms. At N100, dry weights of shoots and total plant were significantly lower with NO_3^- than with NH_4^+ or NH_4NO_3 with no significant differences between NH_4^+ and NH_4 -NO₃. Increasing N addition as NO_3^- and NH_4^+ from N50 to N100 significantly increased root dry weight compared to NH_4NO_3 . Root dry weight was higher with NH_4^+ than with NO_3^- nutrition. Total plant, shoot and root dry weights significantly decreased when N addition rate increased from N100 to N200. At N200, no significant differences were found between $(NH_4)_2$.SO₄ and NH_4NO_3 except in root dry weight, but all growth parameters were lower with KNO₃.

6.3.2 Effects of salinity, N form and N addition rate on wheat growth

Three-way analysis of variance showed that the interaction between salinity and N rate as well as the main effects of salinity, N form and N addition rate had a significant effect ($P \le 0.05$) on shoot dry weights (Table 6.2). In contrast, the interactions between salinity and N form and between salinity, N form and N addition rate were not significant for root dry weight and root/shoot ratio (Table 6.2).

Response of root dry weight to N treatments was similar to shoot dry weight at all salinity treatments. At all salinity treatments, shoot dry weight with NH_4^+ was greatest at N100, whereas with NO_3^- highest dry weight was achieved with N50. At all N application rates, shoot dry weight was higher with $(NH_4)_2$.SO₄ than with KNO₃ at S2 and S3 (Figure 6.2). Additionally, at S2 and S3, plants supplied with NH_4NO_3 had higher shoot and root dry weights compared to $(NH_4)_2$.SO₄ and KNO₃.

Table 6.2 Wheat growth parameters as affected by addition of (NH₄)₂.SO₄, KNO₃ and NH₄NO₃ at 0, 50, 100 and 200 mg N kg⁻¹ soil in a

Fontilizon	Nunata	Shoot	dry we	eight	Root d	ry weig	ht	Root/shoot ratio		
(N form)	$(mg kg^{-1} coil)$	(g pot	⁻¹)		$(g pot^{-1})$)				
	(ing kg son)	S1	S2	S3	S1	S2	S3	S1	S2	S3
$(NH_4)_2.SO_4$	0	0.15 ^a	0.11 ^a	0.07^{a}	0.17 ^a	0.14 ^a	0.10^{a}	1.13 ^b	1.27 ^a	1.42
	50	0.49^{b}	0.39 ^b	0.17 ^b	0.27 ^{de}	0.25^{b}	0.12^{ab}	0.56^{b}	0.64^{b}	0.95
	100	0.64 ^c	0.44^{c}	0.22^{c}	0.30 ^{ef}	0.18 ^a	0.15^{b}	0.47^{ab}	0.59 ^c	0.68^{t}
	200	0.53^{b}	0.21 ^d	0.15^{b}	0.18^{a}	0.14^{a}	0.09 ^a	0.33 ^a	0.46^{bc}	0.60^{t}
KNO ₃	0	0.15 ^a	0.11 ^a	0.06^{a}	0.19 ^{ab}	0.15 ^a	0.09^{ab}	1.27 ^b	1.36 ^a	1.50°
	50	0.50^{b}	0.36 ^b	0.13 ^b	0.28 ^{de}	0.23^{ab}	0.12^{c}	0.56^{b}	0.63 ^b	0.92^{t}
	100	0.46^{b}	0.28°	0.10^{ab}	0.25 ^{cd}	0.16^{a}	0.07^{ab}	0.54^{b}	0.57^{b}	0.70°
	200	0.34 ^c	0.15 ^a	0.09 ^a	$0.12^{\rm f}$	0.09 ^c	0.06^{a}	0.35 ^a	0.61 ^c	0.66
NH ₄ NO ₃	0	0.17^{a}	0.12^{a}	0.08^{a}	0.17^{a}	0.16 ^a	0.12^{a}	1.00^{b}	1.33 ^a	1.50°
	50	0.49^{b}	0.42^{b}	0.25^{b}	0.33 ^{fg}	0.30°	0.20^{b}	0.67^{b}	0.71 ^b	0.80^{t}
	100	0.69 ^c	0.45^{c}	0.25^{b}	0.35 ^g	0.23 ^b	0.18^{b}	0.50^{b}	0.51 ^c	0.72
	200	0.52^{b}	0.30 ^d	0.16 ^c	0.22^{bc}	0.17^{a}	0.11 ^a	0.43 ^{ab}	0.57^{b}	0.68
					LS	$\mathbf{D} (P \leq 0$.05)			
N rate		0.034			0.013			0.229		
N form		0.029			0.011			ns		
N rate x N form		0.059			0.023			ns		
Salinity	0.019			0.012			0.122			
N rate X sali	0.024			0.016			0.228			
N form X sal	inity	ns			ns			ns		
N rate X N f	orm X salinity	0.047			ns			ns		

sandy loam soil without added salt

Values in the same column followed by the same letter are not significantly different at $P \le 0.05$ according to Tukey test. ns, not

significant.

Relative shoot dry weight (in % of S1) at S2 and S3 in response to N additions in different forms is shown in Figure 6.1. In all N treatments shoot dry weight decreased with increasing salinity. Compared to S1, addition of N50, N100 and N200 as NH_4^+ at S2 resulted in approximately 20, 30 and 40% reduction in shoot dry weight, respectively.

The addition of NH_4NO_3 resulted in approximately 5% higher dry weight than $(NH_4)_2$.SO₄ at N50 and N100. There were no differences between NH_4^+ and NH_4NO_3 at N200. Compared to S1, shoot dry weight in S2 with NO_3^- was reduced by 30, 40 and 55% at N50, N100 and N200, respectively. The same trend of shoot dry weight occurred in S3, but shoot dry weight was lower than in S2 (Figure 6.1).



Figure 6.1 Relative shoot dry weight (%) at S2 and S3 compared to S1 in a sandy loam soil

6.3.3 Nitrogen effect on soil salinity

Salinity in the soil solution, calculated and expressed in osmolarity or as EC, as affected by addition of different N forms is shown in Table 6.3. Regardless of N form, increasing N addition from N50 to N100 resulted in 100% increase of the salinity of the soil solution, and likewise with increasing N addition from N100 to N200. At every level of N addition, the osmolarity as well as the EC of soil solution was highest with N addition as KNO₃ and decreased in the following order: KNO₃, (NH₄)₂.SO₄ and NH₄NO₃.

Table 6.3 Salt concentration, expressed in osmolarity (mosmoles L^{-1}) and in EC (dS m ⁻¹)
$^{1}\mbox{)},$ in a sandy loam soil amended with KNO3, (NH4)2.SO4 or NH4NO3 added at 50, 100
and 200 mg N kg ⁻¹ soil

Fortilizor	Ν	Weight	Concentr	ation	Osmolarity ^a	EC ^b
r ei tilizei	mg	kg⁻¹ soil	mM kg ⁻¹ soil	$mM L^{-1}$	mosmoles L ⁻¹	dS m ⁻¹
KNO ₃	50 360		3.57	32.5	64.9	0.56
	100	721	7.14	64.9	129.8	1.12
	200	1443	14.28	129.8	259.5	2.25
(NH ₄) ₂ SO ₄	50	235	1.78	16.2	48.7	0.36
	100	471	3.57	32.5	97.4	0.73
	200	943	7.14	64.9	194.7	1.47
NH ₄ NO ₃	50	142	1.78	16.2	32.4	0.22
	100	285	3.57	32.5	64.9	0.44
	200	571	7.14	64.9	129.8	0.89

^a Osmolarity is in the soil solution.

^b EC is in the soil solution, and was estimated using the following equation: salt concentration (mg L^{-1}) = EC (dS m⁻¹) x 640].

6.3.4 Soil $EC_{1:5}$ and $pH_{1:5}$ as affected by induced salinity and N form and application rate

Soil electrical conductivity (EC) increased with increasing addition of N fertilizers, whereas soil pH decreased (Table 6.4). In S1, the $EC_{1:5}$ was higher with $(NH_4)_2.SO_4$ and KNO_3 than with NH_4NO_3 . In S2 and S3 at all N addition rates, $EC_{1:5}$ was highest with KNO_3 and lowest with NH_4NO_3 .

Generally, soil pH decreased with increasing soil salinity at a given N treatment (Table 6.4). At a given salinity treatment, soil pH decreased with increasing addition of $(NH_4)_2$.SO₄ and NH₄NO₃. Also, compared to N50 soil pH increased with increasing N addition as KNO₃.

Fertilizer	Application rate		EC _{1.5}			pH1.5		
(N form)	$(mg kg^{-1} soil)$	S1	S2	S3	S1	S2	S3	
	0	0.23 ^a	0.40^{a}	0.60^{a}	6.26 ^a	6.20^{a}	6.05 ^a	
(NHJ) SO	50	0.26^{ab}	0.44^{b}	0.67^{b}	6.02^{b}	5.82 ^b	5.70^{b}	
(1114)2.504	100	0.29^{c}	0.52^{c}	0.77 ^c	5.65 ^c	5.56 ^c	5.50 ^c	
	200	0.51 ^d	0.68^{d}	0.93 ^d	5.38 ^d	5.52 ^d	5.44 ^d	
	0	0.23 ^a	0.40^{a}	0.60^{a}	6.26 ^a	6.20^{a}	6.05 ^a	
KNO2	50	0.23 ^a	0.47^{b}	0.71 ^b	6.58 ^b	6.32 ^b	6.11 ^{ab}	
III (03	100	0.27 ^b	0.56^{c}	0.80^{c}	6.76 ^c	6.45 ^c	6.24 ^c	
	200	0.49 ^c	0.72 ^d	0.92^{d}	6.99 ^d	6.56 ^d	6.32 ^d	
	0	0.23 ^a	0.40^{a}	0.60^{a}	6.26 ^b	6.20^{a}	6.05 ^a	
NH NO	50	0.23 ^a	0.41^{ab}	0.62^{ab}	6.32 ^a	6.11 ^b	6.04 ^a	
11141103	100	0.25^{ab}	0.45^{c}	0.65^{b}	6.14 ^c	6.00°	5.95 ^b	
	200	0.33 ^c	0.53 ^d	0.72^{c}	5.97 ^d	5.89 ^d	5.79 ^c	
				LSD (P	≤0.05)			
N rate		-	0.05			0.07		
N form			0.11			0.11		
Salinity			0.14			0.05		
N rate X N f	orm		0.08		0.14			
N rate X sali	nity		0.10		0.12			
N form X sal	linity		0.18		0.12			
N rate X N fe	orm X salinity		0.21			0.19		

Table 6.4 Soil $EC_{1:5}$ and $pH_{1:5}$ as affected by induced salinity at three levels denoted S1, S2 and S3 and $(NH_4)_2$.SO₄, KNO₃ and NH₄NO₃ added at 0, 50, 100 and 200 mg N kg⁻¹ soil

For every N form, values in the same column followed by the same letter are not significantly different at $P \le 0.05$ according to Tukey test.

6.3.5 Effects of salinity, N form and N addition rate on shoot N, P, K and Ca

concentrations

Concentrations of N, P, K and Ca in the plant as affected by N forms and addition rate under saline conditions are presented in Table 6.5. In general, there were no significant differences between N forms for shoot N concentration at a given N rate. Increasing N addition increased shoot N concentration at a given salinity treatment. Salinity also significantly affected shoot N concentration. With increasing soil salinity from S2 to S3, shoot N concentration decreased. Nitrogen addition also had no significant effect on shoot P concentration. However, P concentration was greater with (NH₄)₂.SO₄ and NH₄NO₃ compared to KNO₃ nutrition. In N200, shoot P concentration decreased with increasing salinity from S2 to S3. Potassium concentration increased significantly ($P \le 0.05$) with increasing salinity and N addition; it was greater with NO₃⁻ compared to NH₄⁺.

Increasing soil salinity and N addition rate significantly ($P \le 0.05$) increased shoot Ca²⁺ concentration. The Ca²⁺ concentration was higher in plants fertilized with KNO₃ and NH₄NO₃ than in plants fertilized with (NH₄)₂.SO₄.

6.3.6 Effects of salinity, N form and N addition rate on shoot Fe, Mn, Cu and Zn concentrations

The concentration of micronutrients was significantly ($P \le 0.05$) affected by salinity, N form and N addition rate (Table 6.6). Salinity decreased Fe concentration in the shoots. At a given salinity, Fe concentration significantly increased with addition of N to up to N100 as NH₄⁺ and to up to N200 with NH₄-NO₃, but significantly decreased with increasing N addition with KNO₃ from N0 to N200. Shoot Mn concentration was affected similarly as Fe. Shoot Cu and Zn concentrations significantly ($P \le 0.05$) increased in the shoots with increasing N addition and soil salinity. With N100 and N200, shoot Cu and Zn concentrations were lower with KNO₃ than with (NH₄)₂.SO₄ and NH₄NO₃.

	N P K							Ca					
N application rate	Salinity			NH ₄									
$(mg kg^{-1} oil)$	level	NH ₄	NO ₃	-									
(ing kg soir)				NO ₃									
							$mg g^{-1}$	DW ^a					
	S1	13.7	13.6	13.7	5.1	5.1	5.0	26	27	27	2.6	2.5	2.7
0	S2	17.6	17.6	17.5	5.7	5.6	5.5	29	30	30	3.6	3.5	3.7
	S3	19.4	19.4	19.4	6.0	5.9	6.0	32	32	33	8.6	8.5	8.3
	S1	28.7	28.7	28.3	6.4	5.1	5.4	32	34	34	2.3	2.8	2.7
50	S2	30.9	29.5	31.8	6.8	5.6	6.3	35	36	36	3.1	4.1	4.3
	S3	23.3	27.6	23.2	6.8	5.4	5.8	35	40	37	6.4	8.4	8.5
	S1	32.3	31.1	32.5	6.3	5.4	5.8	36	37	37	1.7	3.3	2.8
100	S2	33.2	32.1	30.8	6.9	5.8	6.5	38	40	37	3.2	5.4	4.6
	S3	30.3	28.2	31.3	6.5	4.4	5.0	38	42	39	4.7	9.9	9.5
	S1	35.9	33.9	35.2	6.4	5.4	5.9	38	40	38	1.4	4.7	3.4
200	S2	37.6	36.9	36.3	7.2	4.4	6.1	39	41	39	2.5	9.6	4.7
	S3	33.0	34.1	33.0	5.9	4.2	5.5	42	41	41	4.3	14.8	12.0
						I	LSD (P	≤ 0.05	5)		-		
N rate			2.2			0.41			1.92			0.21	
N form			ns			0.52			ns			ns	
Salinity			1.5			0.39			1.50			0.19	
N rate x N form			ns			0.38			1.13			0.27	
Salinity x N rate			2.75			0.38			1.13			0.27	
Salinity x N form			ns			ns			ns			0.23	
Salinity x N rate x N	form		ns			ns			ns			0.47	

Table 6.5 Shoot N, P, K and Ca concentrations (mg g^{-1} DW) as affected by (NH₄)₂.SO₄, KNO₃ and NH₄NO₃ added at 0, 50, 100 and 200 mg N kg⁻¹ soil in a saline sandy loam soil

^a DW, dry weight. ns, not significant.

			Fe			Mn			Cu			Zn	
N application rate	Salinity			NH ₄			NH ₄			NH ₄			NH ₄
$(mg kg^{-1} soil)$	level	NH_4	NO ₃	-	NH_4	NO ₃	-	NH_4	NO ₃	-	NH_4	NO ₃	-
				NO ₃			NO ₃			NO ₃			NO ₃
							mg kg ⁻	1 DW ^a					
	S1	350	360	337	105	106	104	6.1	6.0	6.1	21	20	21
0	S2	227	225	214	98	99	98	8.9	9.0	8.8	38	37	37
	S3	190	183	170	56	56	55	9.0	9.0	9.0	43	40	43
	S1	340	353	360	114	98	174	8.6	8.5	8.6	44	35	37
50	S2	292	171	295	104	88	98	9.2	9.3	9.2	44	39	44
	S3	242	138	258	99	59	95	9.6	8.5	8.5	39	35	39
	S1	404	227	390	122	91	186	8.9	8.2	8.7	45	38	40
100	S2	341	144	350	99	75	93	9.4	9.1	9.5	39	36	39
	S3	291	113	286	95	56	89	10.8	6.2	8.1	35	34	33
	S1	340	194	403	94	86	88	9.3	7.9	9.5	40	32	42
200	S2	192	149	169	91	74	84	9.6	7.2	9.5	36	29	33
	S3	151	121	151	85	52	80	8.1	5.7	8.1	32	25	29
						Ι	LSD (P	≤ 0.05)				
N rate			30.0			4.11			0.51			2.11	
N form			ns			4.11			ns			1.91	
Salinity			25.0			3.51			0.31			1.54	
N rate x N form			49.1			4.26			0.66			ns	
Salinity x N rate			ns			4.26		0.66				2.87	
Salinity x N form			ns		3.69			ns		ns			
Salinity x N rate x N	form		ns			7.38			ns			ns	

Table 6.6 Shoot Fe, Mn, Cu and Zn concentrations (mg kg⁻¹ DW) as affected by $(NH_4)_2$.SO₄, KNO₃ and NH₄NO₃ added at 0, 50, 100 and 200 mg N kg⁻¹ soil in a saline sandy loam soil

^a DW, dry weight. ns, not significant.

6.4 Discussion

The results of the present study confirmed the results of Chapter 5, and are came in accordance with the results of others (Ali et al., 2001; Inal and Tarakcioglu, 2001; Irshad et al., 2002) that wheat growth decreased with increasing soil salinity at all N treatments.

Without added Na⁺ and Ca²⁺ salts (S1), N addition significantly increased dry weights of shoot and root (Table 6.2). However, no significant differences were found between N forms at N50 (Table 6.2) indicating that plants were neither nutrient limited nor salinity stressed, at least at this rate of N application.

In contrast to the findings of Cox and Reisenauer (1973), Lewis et al. (1989), Al-Mutawa and El-Katony (2001), Frechilla et al. (2001), Inal and Tarakcioglu (2001) and Mahmood and Kaiser (2003) the results of this study indicated that, compared to N-NO₃, N-NH₄⁺ resulted in higher shoot and root dry weights at rates higher than N50. Increasing N addition may have resulted in increasing plant protein content and photosynthetic capacity (Cox and Reisenauer, 1973) stimulating plant growth. It has been shown that the transfer of C compounds to the roots of plants grown on NH₄⁺ is generally higher than in plants grown on NO_3^- (Lewis et al., 1990). Possibly such a mechanism has occurred in this study and therefore root biomass was greater with NH_4^+ compared to NO₃⁻ addition (Table 6.2). At N100, compared to NO₃⁻, shoot N and K concentrations were the same and shoot P and Ca concentrations were higher with NH_4^+ addition. However, these nutrients were at sufficient concentrations and therefore were not directly associated with poor plant growth. In other studies, lower plant dry weight of NO₃⁻ compared to NH₄⁺-fed plants was found to be related to low rates of NO₃⁻ absorption (Lavoie et al., 1992), inefficient assimilation owing to nitrate reductase activity (Poonnachit and Darnell, 2004), or deficiency of micronutrients, particularly Zn (Warren and Adams, 2002).

In agreement with the results presented in Chapter 5, addition of N as NO_3^- at levels higher than N50 decreased growth. In S1, addition of more than N50 as $NO_3^$ significantly decreased shoot Fe, Mn and Cu concentrations, whereas with NH_4^+ , the opposite is true (Table 6.7). Also at every addition rate of N, the concentrations of Fe, Mn, Cu and Zn were higher with NH_4^+ than with NO_3^- . However, micronutrient concentrations fell in a range indicating that plants were not deficient (Reuter and Robinson, 1997). Soil pH and salinity can be limiting factors to plant growth as they affect most of chemical reactions in the vicinity of plant roots as well as water and nutrient uptake. On the last day of the experiment, the results shown in Table 6.4 indicated that in S1 at all salinity treatments, the EC_{1:5} with $(NH_4)_2SO_4$ was nearly the same as with KNO₃. These results in Table 6.4, although indicative, may not reflect the actual salinity that the plants had been subjected to during the experiment. Calculations of the salinity of soil solution with addition of N100 (Table 6.3) indicated that in S1 with NO₃⁻ plants were subjected to salinity far higher than with NH₄⁺; approximately 130 mosmoles L⁻¹ with NO₃⁻ compared to 97 mosmoles L⁻¹ with NH₄⁺. With no added Na⁺ and Ca²⁺ salts (S1), it appears that, compared to NH₄⁺, with NO₃⁻ addition at all application rates, particularly rates higher than N50, wheat growth was limited because of osmotic stress.

Villa-Castorena et al. (2003) showed that increasing application of $(NH_4)_2$.SO₄ contributed to soil salinity and negatively affected wheat growth. Also, Barker et al. (1966) and Cox and Reisenauer (1973) found that NH_4^+ nutrition reduced the uptake of Ca²⁺, Mg²⁺ and K⁺. In this study, compared to NH₄-N100, plant growth was lower at N200 (Figure 6.1). At N200, shoot micronutrient concentrations were lower compared to N100 (Table 6.7). Also, in S1 at N200 as NH_4^+ salinity was higher compared to N100, and at harvest, EC_{1:5} of soil solution reached 0.51 dS m⁻¹, whereas with N100 it was only 0.29 dS m⁻¹ (Table 6.4). It seems that with addition of N at rates greater than N100 increased osmotic pressure and decreased uptake of nutrients, particularly micronutrients and possibly water, or combination may have decreased wheat growth.

The form in which N is supplied to plants growing under osmotic stress can be important (Martinez and Cerda, 1989). In hydroponics, growth of maize (Lewis et al., 1989), wheat (Al-Mutawa and El-Katony, 2001) and pea (Frechilla et al., 2001) increased with increasing N addition up to 12 m*M* N, and compared to NH_4^+ was greater with NO_3^- nutrition with increasing salinity by NaCl up to 15 dS m⁻¹.

In this study, under saline conditions, dry weights of root and shoot increased with addition of N in all tested forms. Also, shoot dry weight was lower with NO_3^- than with NH_4^+ . In addition, observations on plant micronutrient composition under saline conditions were similar as under S1; lower with NO_3^- than with NH_4^+ nutrition at all N treatments (Table 6.6). The positive effect of NH_4^+ compared to NO_3^- on plant growth under saline conditions could be due to the reduced energy requirement for

utilization of NH4⁺ as compared to NO3⁻ in protein synthesis (Cox and Reisenauer, 1973), and/or enhanced uptake of nutrients, particularly micronutrients, and thus increased root growth (Table 6.2). Addition of Na⁺ and Ca²⁺ salts established high salinity; 145 and 286 mosmoles L^{-1} in the soil solution; equivalent to EC_e 6.6 and 11.8 dS m⁻¹ in S2 and S3, respectively. According to the calculations (Table 6.3), at N100 in S2, with NH4⁺ the plants were subjected to soil solution salinity of approximately 242 mosmoles L^{-1} , approximately EC_e 11.0 dS m⁻¹, whereas with NO₃⁻ soil salinity was 275 mosmoles L⁻¹, approximately EC_e 12.5. Relative to S1 at N200, NH₄⁺ shoot dry weight was 60% in S2; EC_{1.5} 0.67 dS m⁻¹, equivalent to EC_e 9.38 dS m⁻¹, whereas with NO₃⁻ it was only 45% in S2; EC_{1:5} 0.72 dS m⁻¹, equivalent to EC_e 9.93. At all salinity treatments, with NH₄⁺ and NH₄NO₃, dry weights were maximal at N100, whereas with NO₃⁻ highest dry weight was at N50. This emphasizes the positive effect of fertilization with N100 as NH₄⁺, compared to NO₃⁻, on the enhancement of plant nutrient composition and growth under saline conditions. Lewis et al. (1989) found that with increasing soil salinity concentration of Cl⁻ increases in the plant and causes plant growth decline at a certain level. In the present study, in addition to increased osmotic stress and decreased concentration of micronutrients, possibly increased uptake of Cl⁻ may have resulted in reduced plant growth with addition of NO₃⁻ at rates higher than N50.

Under saline conditions, dry weights of shoot and root were greatest with addition of NH_4NO_3 at every addition rate of N with highest weight at N100 (Figure 6.3). This is probably because the concentration of all nutrients was highest with NH_4NO_3 at all N treatments. Also, at N100, compared to NH_4^+ and NO_3^- , soil salinity (osmolarity, $EC_{1:5}$ and EC_e) was lowest with NH_4NO_3 (Tables 6.3 and 6.4).

6.5 Conclusions

It could be concluded that application of 100 mg N kg⁻¹ as NH_4^+ compared to NO_3^- fertilizers is beneficial for wheat nutrient uptake and growth in the sandy loam with salinity $EC_{1:5}$ 0.67 dS m⁻¹, equivalent to EC_e 9.3 dS m⁻¹. Addition of NH_4NO_3 , compared to NH_4^+ or NO_3^- , is beneficial and therefore strongly recommended for wheat growth in the sandy loam soil with up to EC_e 9.3 dS m⁻¹.

Chapter 7 - Microbial activity and biomass and N and P

availability as affected by different plant residues in saline soils

with different texture

1.1 Introduction

The decomposition of plant residues and the release and consequently the availability of N and P to the plants are mediated by soil microorganisms (Tian et al., 1992; Fang et al., 2007). Salinity has been widely reported to have a negative effect on microbial and enzyme activities (Rietz and Haynes, 2003; Sardinha et al., 2003) and on soil microbial biomass (Yuan et al., 2007). This effect is often greater in soils naturally saline than in artificially salininized soils.

Increased organic matter input from trees, compared to crops, (Kaur et al., 2000), addition of C-glucose (1000-2000 mg kg⁻¹ soil) (Luna-Guido and Dendooven, 2001; Dendooven et al., 2006), or increased soil pH up to 10 (Li et al., 2007) increased microbial activity and/or biomass in naturally alkaline saline soils. Also in sodic soils, microbial activity increased with addition of different plant residues added at 1% in a soil with 61.2% clay, ESP 21.4%, $EC_{1:10}$ 32.9 dS m⁻¹ and pH 6 (Nelson et al., 1996), or at 0.2% in a soil with 17.1% clay and ESP 25, EC_{1:5} 1.5 dS m⁻¹ and pH 9.1 (Clark et al., 2007). Additionally, with increasing addition of NaCl or Na₂SO₄ at similar levels of Na⁺ up to 142 mM kg⁻¹ soil in a soil with 17% clay, pH 8.1, EC_e 2.17 dS m⁻¹, 2% maize straw increased microbial activity and biomass (Li et al., 2006a; Li et al., 2006b). With increasing soil pH, microbial activity may be stimulated as alkali can dissolve, disperse or cause chemical hydrolysis of the added as well as the original soil organic matter and because dissolved organic matter can be easily decomposed by microorganisms (Laura, 1973, 1974; Nelson and Mele, 2007). However, at high pH (8.9) microbial activity decreased with increasing salinity (ECe above 5.7 dS m⁻¹) in soils amended with 1% maize or pea straws (Muhammad et al., 2006). With increasing addition of NaCl salts, Na⁺ ions, relative to Ca²⁺ and/or Mg²⁺, may occupy a large number of exchange sites on clay particles. Increased Na⁺ adsorption on clay minerals led to dispersion of clay particles and reduced soil aggregation allowing microorganisms to attack small as large organic matter particles increasing organic matter decomposition (Nelson et al., 1996). Also, compared to Cl⁻ salts, sulphate salts increased microbial activity and organic matter decomposition (Garcia and Hernandez, 1996; Li et al., 2006a) due to better use of SO_4^{2-} by microbial cells in the synthesis of amino acids, vitamins and other organic

Available information about salinity effect on soil microbial properties is controversial. The interactions between salinity, sodicity and pH can confound the final conclusion about residue decomposition and its effects on soil microbial properties and nutrient availability under osmotic stress conditions.

Substrate quality is one of the most important factors influencing the decomposition of added residues, microbial biomass and nutrient release (Mueller et al., 1998). Tian et al. (1992) found a negative correlation between nutrient release and C:N ratio, lignin content and polyphenol concentration. However, Kwabiah et al. (2003) found no clear relationship between lignin and soluble polyphenolic concentrations and available P due to the overwhelming effect of total P in the residues on P availability. Clark et al. (2007) showed that N release is highly dependent on the soluble C/total N ratio in the amendment in sodic soils. Muhammad et al. (2006) found no correlation between respiration and salinity in soils amended with maize and pea straw. Also, microbial activity differs between soils with different texture (Ladd et al., 1981; Ladd et al., 1985), and is dependent on the amount (Zunino et al., 1982) and the type (Kunc and Stotzky, 1974) of clay minerals in non-saline and in naturally alkaline salt-affected soils (Li et al., 2007). The studies on residue decomposition in saline soils have been conducted in soils differing not only in soil physical and chemical properties, but also in the type of applied residues, which may differ in chemical properties. More studies are therefore needed to characterize the soil and the substrate parameters that can influence plant residue decomposition under only osmotic stress conditions. The aim of this study was to determine the effects of plant residues, differing in quality, on microbial biomass and N and P availability along a gradient of salinity in sandy and sandy loam soils with SAR 1 and neutral pH.

7.2 Materials and methods

7.2.1 Preliminary experiment

A preliminary experiment was conducted to assess the salinity threshold level for microorganisms in the tested soils using glucose and KNO₃.

A sandy and a sandy loam soil were collected from two sites in Monarto, South Australia, that had not been fertilized. The collected soils had neutral pH, low ESP and a low concentration of organic matter, salt, CaCO₃ and available N and P. Soil was also collected from a third site, Urrbrae, South Australia and used in this study. The properties of the soils used in this study are shown in Table 3.1.

7.2.1.1 Soil salinization and incubation

After air drying and sieving the soil to 2 mm, five salinity treatments were imposed in the soil using mixtures of NaCl and CaCl₂.2H₂O. Solutions of Na⁺ and Ca²⁺ salts were thoroughly mixed with the soils using a cement mixer. Sodium and Ca²⁺ salts were calculated to maintain SAR 1 at all salinity treatments (Table 7.1). Soils were incubated in plastic bags at 50% WHC at 25 °C in the dark for 2 weeks. Samples were taken prior to the establishment of the experiment for determination of EC_{1:5}, available N (NH₄⁺ and NO₃⁻), available P and microbial biomass C, N and P.

Table 7.1 Amounts of NaCl and CaCl₂.2H₂O added in three soils with different textures to impose 5 salinity levels, each has SAR 1

		Na	Cl		CaCl	Osmolarity	
Salt loval	mg	тM	mM	mg mM		m <i>M</i>	mosmoles L ⁻¹
Salt level	kg ⁻¹	soil	in soil solution	kg⁻¹	soil	in soil solution	in soil solution
S1	0	0	0	0	0	0	0
S2	306	5.23	47.6	4025	27.38	249.0	842
S3	399	6.83	62.1	6849	46.59	423.5	1394
S 4	475	8.12	73.9	9694	65.94	599.5	1946
S 5	570	9.74	88.7	13985	95.14	864.8	2771

Table 7.2 Measured $EC_{1:5}$ and calculated EC_e and EC (dS m⁻¹) after addition of a mixture of NaCl and CaCl₂.2H₂O at 5 salinity levels (preliminary experiment)

	Silty	loam	soil	Sa	ndy so	oil	Sandy loam soil					
Salt level	EC _{1:5}	ECe	EC	EC _{1:5}	ECe	EC	EC _{1:5}	ECe	EC			
	$dS m^{-1}$											
S1	0.20	1.9	3.8	0.16	3.6	7.2	0.19	2.6	5.2			
S2	1.26	12.2	24.4	1.10	24.9	49.8	0.82	11.3	22.6			
S3	1.83	17.3	34.6	1.98	44.9	89.8	1.75	24.1	48.2			
S4	2.28	21.6	43.2	2.33	52.8	105.6	2.03	28.0	56.0			
S 5	2.99	28.4	56.8	3.18	72.1	144.2	2.79	38.5	77.0			

 $EC_{1:5}$ was measured after soil incubation prior to establishment of the experiment under controlled conditions. EC_e is calculated from $EC_{1:5}$ using a conversion factor as shown by (Slavich and Petterson, 1993). EC values are electrical conductivity in soil solution, and calculated knowing that the EC of the soil solution at field capacity is approximately double the EC_e (Richards, 1954).

7.2.1.2 Addition of glucose and KNO₃

A solution of C as glucose ($C_6H_{12}O_6$) and N as KNO₃ (2500 µg C and 160 µg N g⁻¹ soil; C/N ratio 16:1) was added to soils (85% WHC) at all salinity levels. PVC containers with mesh at the bottom were filled with 20 g soil (dry weight equivalent). Each container was then transferred into a Mason jar (described in Chapter 3). Soil respiration was measured over 14 days and calculated as described in Chapter 3.

Table 7.3 EC (dS m^{-1}) in the soil solution as affected by glucose and KNO₃

Salt type	Weight	EC
	(g kg ⁻¹ soil)	$(dS m^{-1})$
$C_6H_{12}O_6$	5.000	71.0
KNO ₃	1.155	16.4

EC is based on the salt weight, not the electrical charge, and was estimated using the following equation: salt concentration (mg L^{-1}) = EC (dS m⁻¹) x 640]. Therefore data are approximate values of EC in the soil solution.

7.2.2 Main experiment

For the evaluation of the effect of addition of different plant residues on N and P availability and microbial biomass in saline soils, an experiment was carried out over 50 days under glasshouse conditions. Sandy and Sandy loam soils from Monarto were air dried, sieved, salinized (5 levels) and stored in plastic bags in the dark for 2 weeks as described above. The $EC_{1:5}$ was measured in the soils before establishment of the glasshouse experiment. The average values of $EC_{1:5}$ are shown in Table 7.4.

		Sandy		Sandy loam						
Salt level	EC _{1:5}	ECe	EC	EC _{1:5}	ECe	EC				
	dS m ⁻¹									
S1	0.21	4.7	9.4	0.19	2.6	5.2				
S2	1.08	24.5	49.0	0.87	12.0	24.0				
S3	1.90	43.1	86.2	1.63	22.4	44.8				
S 4	2.63	59.7	119.4	2.32	32.0	64.0				
S 5	2.89	65.6	131.2	2.49	34.3	68.7				

Table 7.4 Measured $EC_{1:5}$ and calculated EC_e and EC (dS m⁻¹) after addition of a mixture of NaCl and CaCl₂.2H₂O at 5 salinity levels (main experiment)

7.2.2.1 Plant residues

Residues of lucerne (*Medicago sativa*), canola (*Brassica napus*) and lupin (*Lupinus albus*, L.), collected from Roseworthy experimental station, the University of Adelaide, were oven dried (70 °C) and ground to 1.5 mm. Total concentrations of C, N and P were measured in the plant residues as described in Chapter 3. The residues differed in their nutrient, lipids, lignin and cellulose contents and C/N and C/P ratios (Chapter 3; Table 3.2). Residues were added at 2% (w/w), corresponding to approximately 20 mg P kg⁻¹ soil, and thoroughly mixed with the soil using a cement mixer. Amended and unamended (control) soils were placed in 1 kg pots, transferred to the glasshouse and watered to 85% WHC in each soil.

7.2.2.2 Measurements

Samples of 2 g soil (dry weight equivalent) were taken on days 10, 30, 50 and 70 after the establishment of the experiment for the determination of extracted available P and microbial P. For the measurement of extracted available N (NH_4^+ and NO_3^-), 5 g soil (dry weight equivalent) was used. Microbial C and N were determined on days 0, 10 and 50 in the extract of 2 sub-samples of 10 g soil (dry weight equivalent), one of which was chloroform-fumigated. Sampling of soil, methods of extraction and calculations are described in Chapter 3.

7.2.2.3 Design and statistical analysis

The experiment was set up in a completely randomised design [2 soils, 5 salinities, 3 plant residues, 5 sampling times (3 times for microbial biomass C and N) treatments] with four replicates. Analysis of variance was carried out using Genstat 8. In tables, values are means. Values in the same column followed by the same letter are not

significantly different at $P \le 0.05$ according to Tukey test. The abbreviation "ns" means not significant. Bars in histograms show standard deviation (STD).

7.3 Results

In the experiment with addition of glucose and KON_3 , the individual effects and the interactions between soil, salinity and time were significant for soil respiration rate and cumulative respiration (Table 7.5).

Table 7.5 Results of statistical analysis; least significant differences (LSD) and significance at 5% level, of soil respiration and cumulative respiration over time as affected by glucose and KNO₃ in saline soils with different texture

Interactions	Respira	tion rate	Cumulative respiration			
inter actions	LSD	P	LSD	Р		
Soil	0.01	**	0.39	**		
Salinity	0.01	**	0.51	**		
Time	0.20	**	0.94	**		
Soil X salinity	0.12	**	0.88	**		
Soil X time	0.03	**	1.62	**		
Salinity X time	0.14	**	2.09	**		
Soil X salinity X time	0.25	**	3.62	**		

7.3.1 Respiration rate in saline silty loam, sandy loam and sandy soils amended with glucose and KNO₃

The time course of the respiration rate (first 70 hours and from day 6 to day 14 of incubation) in a glucose and KNO_3 -amended silty loam, sandy loam and sandy soils with similar five salinity treatments is shown in Figure 7.1.

Generally, the respiration rate significantly changed over time in all soils with the greatest rates in the silty loam soil at all salinity treatments. With increasing soil salinity, respiration rate significantly decreased in the amended soils. Significant differences in respiration rate between soils remained until the end of the experiment. In silty loam soil, respiration rate strongly increased in the first hours after glucose addition reaching a peak of approximately 7 μ g g⁻¹ soil h⁻¹ at 32h. This increase was followed with a dramatic decrease until the end of the experiment. In the sandy loam soil, the highest respiration rate in this soil remained unchanged until 68h and then slowly decreased until the end of the experiment. In the sandy soil, respiration rate was very low until 68h then increased with highest respiration rates (approximately 2 μ g g⁻¹

soil h⁻¹) after 1 week, followed by a slow decrease until the end of the experiment. From the beginning until 68h incubation, the respiration rate decreased with increasing salinity.



Figure 7.1 Respiration rate (μ g CO₂-C g⁻¹ soil h⁻¹) over time [hour (h) and day (d)] as affected by glucose and KNO₃ in saline silty loam, sandy loam and sandy soils

7.3.2 Cumulative respiration in saline silty loam, sandy loam and sandy soils

amended with glucose and KNO₃

Cumulative respiration significantly increased over time (Figure 7.2) with the highest rates in soils without salt. Soils differed in cumulative CO₂-C release. From the beginning of the experiment until 68h, cumulative CO₂-C release increased in the silty loam soil with significant differences between salinity treatments. Cumulative respiration decreased with increasing salinity. After approximately 32h, the cumulative respiration at S1 in the silty loam soil were 78, 55, 44 and 38%, whereas after 68h they were 84, 74, 72 and 69% at S2, S3, S4 and S5, respectively. Up to 68h, cumulative respiration slowly increased in the sandy loam soil and remained low in the sandy soil with no significant differences between salinity treatments. From 68h to 231h, cumulative respiration increased most strongly in S1 in all soils and there were significant differences between salinity treatments in silty loam and sandy soils, but not in the sandy loam soil. After approximately 231h, compared to S1, cumulative respiration in the silty loam soil was 89, 80, 77 and 71% at S2, S3, S4 and S5, respectively. From 231h until the end of the experiment, cumulative respiration increased in all soils and was high in the following order: silty loam, sandy loam and sandy soils.



Figure 7.2 Cumulative CO₂-C ($\mu g g^{-1}$ soil) over time (h) after addition of glucose and KNO₃ in silty loam, sandy loam and sandy soils with five salinity treatments

7.3.3 Available N and P in saline sandy and sandy loam soils amended with

different crop residues on day 50

The availability of N and P increased, and the microbial biomass decreased (Section 7.3.4.2) in the amended saline soils over time. In this section, day 50 was selected to show N and P availability in relation to the residue type and salinity level.

Soil available N (NH_4^+ and NO_3^-) and P significantly increased with addition of plant residues in both soils. The availability of N and P was also affected by soil type, residue type, salinity level and their interactions (Table 7.6).

7.3.3.1 Available NH_4^+ and NO_3^-

Compared to unamended soil, residue addition significantly increased NH_4^+ and NO_3^- concentrations. In amended soil, NH_4^+ and NO_3^- concentrations decreased with increasing soil salinity. In the sandy loam soil at S1, compared to unamended control, addition of lucerne residues increased NH_4^+ concentration 17 fold, while canola and lupin caused 9 and 11 fold increase, respectively. The trend in the sandy soil was similar to that in the sandy loam soil except that the NH_4^+ concentration increased 5, 3 and 5.5 times compared to the control with the addition of lucerne, canola and lupin residues, respectively. Nitrogen availability was higher in the sandy soil than in the sandy loam in the soil with no added salts (S1). With increasing salinity, no clear differences were found between residues, but higher NO_3^- concentration was found in the sandy than in the sand loam. In all salinity treatments highest NH_4^+ concentration in the sand than in the sand loam at all salinity treatments.

The ratio of NH_4/NO_3 significantly increased with increasing soil salinity. The NH_4/NO_3 ratio was higher in the sandy than in the sandy loam soil with no residue addition, whereas with residue addition, the opposite was true. At all salinity treatments, the highest NH_4/NO_3 ratio was found with addition of lupin residue in both soils.

7.3.3.2 Available P

Compared to the unamended soils, P availability increased with addition of plant residues with significant differences between plant residues in both soils at all salinity treatments. The availability of P decreased with increasing soil salinity. At all salinity treatments, P availability was higher in the sandy loam than in the sandy soil. In the sandy loam soil, canola residues resulted in highest P availability at all levels of salinity. In the sandy soil, lupin addition resulted in highest P availability to S3.

		$N-NH_4^+$		N-NO ₃		Total N		MII + NO		Р		
Residue	mg N kg ⁻¹ soil					nH ₄ / nO ₃ ratio		mg P kg ⁻¹				
	Level	GT	G	GT	G	GT	G	CT	G			
N		SL 14	8	SL 10	ð	SL	5	SL 1.15	8	SL 1.44	8	
No	SI	14	42	12	12	26	54	1.15	3.44	1.44	1.11	
residue	S 2	17	40	9	8	26	49	2.04	4.60	1.26	1.04	
	S3	24	44	7	6	31	51	3.79	6.87	0.81	0.96	
	S4	27	42	3	3	31	46	7.50	11.78	0.54	0.83	
	S5	26	37	1	1	26	38	37.67	54.37	0.13	0.12	
Lucerne	S1	239	214	154	172	394	386	1.55	1.25	4.39	3.81	
	S2	91	136	56	76	147	213	1.84	1.72	4.09	3.60	
	S3	76	124	24	44	100	168	3.22	2.83	2.00	2.76	
	S4	66	101	18	29	84	130	3.74	3.56	1.62	2.07	
	S5	56	86	11	23	74	109	3.20	4.01	0.94	1.18	
Canola	S1	124	130	101	124	226	254	1.23	1.05	7.27	2.60	
	S2	116	108	53	71	170	180	2.18	1.54	5.13	2.26	
	S 3	101	91	36	53	138	145	2.82	1.79	3.08	1.74	
	S4	94	82	29	39	123	121	3.39	2.17	1.83	1.39	
	S5	90	79	20	30	111	109	4.37	2.58	0.81	1.21	
Lupin	S1	147	231	165	177	312	409	0.90	1.30	4.23	4.58	
•	S2	135	221	55	75	191	296	2.45	2.93	3.62	4.13	
	S3	122	165	26	53	148	219	4.64	3.12	3.12	3.34	
	S4	116	144	14	34	131	179	8.05	4.20	1.63	2.00	
	S5	112	123	10	22	122	146	11.71	5.56	0.60	0.50	
	LSD (P < Compared by Compare				$(P \leq $	0.05)						
Soil		1.6 2.13		2.61		ns		0.01				
Salinity	Salinity		2.6 3.1		37 4.13		13	1.56		0.02		
Residue		2.3		3.01		3.69		1.39		0.02		
Soil X Salinity		3.7		ns		5.84		ns		0.03		
Soil X Resi	Soil X Residue		3.3		4.26		5.22		1.97		0.03	
Salinity X	Salinity X Residue		5.2		6.73		8.26		ns		0.05	
Soil X Residue X		7.3 ns		IS	11.68		ns		0.06			
Salinity										0.00		

Table 7.6 Available N-NH₄⁺ and N-NO₃⁻, total inorganic N and available P (mg kg⁻¹) and NH₄⁺/NO₃⁻ ratio on day 50 after addition of lucerne, canola and lupin residues in sandy loam (SL) and sandy (S) soils with five salinity treatments

7.3.4 Effect of plant residue addition on microbial biomass C, N and P in saline soils

With the addition of residues, the pattern of microbial biomass C, N and P was similar in both soils at all sampling times, but with a difference in magnitude between residue types (described in section 7.3.4.2). Lucerne was taken as an example to show the difference between amended and unamended saline soils for microbial biomass C, N and P on day 50.

Generally, compared to the unamended soils, microbial biomass C, N and P was higher in the soil with lucerne residues, but decreased with increasing soil salinity (Table 7.7). With no residue addition, microbial C and N were higher in the sandy loam than in the sandy soil, whereas no significant difference between the two soils was found for microbial P. Under non-saline conditions, microbial biomass C, N and P were higher in sandy loam than in sandy soils when amended with lucerne residues.

Compared to soils without residue, with residue addition microbial biomass C was approximately 3.5 and 10 times higher in both soils at S1 and S5, respectively. Compared to S1, with residue addition microbial biomass C was approximately 50 and 69% at S2, and 25 and 36% at S5 in the sandy loam and sandy soils, respectively.

Table 7.7 Microbial C, N and P on day 50 after addition of lucerne residues in sandy and sandy loam soils with 5 salinity treatments

		Micro	bial C	Micro	bial N	Microbial P			
Dosiduo	Salinity Level	mg kg ⁻¹							
Kesiuue		Sandy loam	Sandy	Sandy loam	Sandy	Sandy loam	Sandy		
No residue	S1	175	140	25	10	1.85	1.76		
	S2	115	110	11	8	0.95	0.76		
	S 3	91	77	8	5	0.79	0.74		
	S4	61	44	5	3	0.52	0.56		
	S5	17	16	1	2	0.17	0.21		
Lucerne residue	S1	620	503	107	100	9.05	9.02		
	S2	307	348	48	53	6.30	6.34		
	S 3	252	288	37	44	3.59	3.66		
	S4	223	237	35	37	2.28	1.37		
	S5	154	182	22	29	1.21	1.26		
	LSD ($P \leq 0.05$)								
Soil		ns		ns		ns			
Salinity		6.8		1.89		0.26			
Residue		4.3		1.20		0.37			
Soil X Salinity		9.6		2.68		ns			
Soil X Residue		6.1		1.69		ns			
Salinity X Residue		9.6		2.68		0.52			
Soil X Residue X Salinity		13.6		ns		ns			

ns, not significant

Compared to unamended soils, with addition of lucerne residue microbial biomass N was approximately 4 and 10 fold higher at S1, and 14 and 22 fold higher at S5 in the sandy loam and sandy soils, respectively.

With residue addition, no significant difference between soils was found for microbial biomass P at all salinity treatments. Compared to unamended soil, with lucerne residue addition, microbial biomass P was approximately 4, 5 and 10 times higher at S1, S3 and S5, respectively.

7.3.4.1 Microbial C, N and P in sandy and sandy loam soils after addition of different plant residues (day 10)

Differences between plant residues in microbial biomass C, N and P were similar in both soils over time, but greatest on day 10. This sampling date is taken in this section to describe the differences between residues and between soils under saline conditions. Generally, there were significant differences between soils and residues for microbial N and P, and slight differences for microbial biomass C. Microbial biomass C, N and P were also significantly affected by residue addition and salt concentration and their interaction.

7.3.4.1.1 Microbial biomass C

Microbial biomass C decreased with increasing soil salinity and was higher in amended than unamended soils (Figure 7.3). Without addition of amendments, microbial biomass C was higher in sandy loam than in sandy soil at all salinity treatments. In S1, with residue addition, microbial biomass C was significantly higher in sandy soil than in sandy loam soil. With increasing soil salinity, microbial biomass C decreased and was slightly lower in the amended sandy loam than in the amended sandy soil. In the sandy soil, microbial biomass C was highest with lupin residues, but no significant differences were found between lucerne and canola residues at all salinity treatments. In the sandy loam soil, no significant differences were found between plant residues at all salinity treatments.



Figure 7.3 Microbial C (mg kg⁻¹ soil) on day 10 after addition of lucerne, canola and lupin residues in saline sandy and sandy loam soils with five salinity treatments

7.3.4.1.2 Microbial biomass N

Microbial biomass N was significantly higher in amended than unamended soils, and decreased with increasing salt concentration (Figure 7.4). In all salinity treatments, microbial biomass N was higher in the sandy than in the sandy loam soils. In the sandy soil, microbial biomass was approximately 20-40 mg N kg⁻¹ soil higher with canola than with lucerne and a similar difference was found between lupin and canola residues at all salinity treatments. In the sandy loam soil, microbial biomass N was highest with canola residues at all salinity treatments.



Figure 7.4 Microbial N (mg kg⁻¹ soil) on day 10 after addition of lucerne, canola and lupin residues in saline sandy and sandy loam soils with five salinity treatments

7.3.4.1.3 Microbial biomass P

Compared to the sandy and sandy loam soils with no residue, microbial biomass P significantly increased with residue addition in both soils, but decreased with increasing soil salinity (Figure 7.5). Microbial biomass P was higher in the sandy loam soil than in the sandy soil, but only at S1 and S2; at higher salinity treatments the opposite was true. At salinity treatments higher than S2, no significant differences were found between residues for microbial biomass P in the sandy loam soil. In the sandy soil, lowest microbial P was found with canola residues and highest microbial P was found with lupin residues at all salinity treatments, except the control S1.



Figure 7.5 Microbial P (mg kg⁻¹ soil) on day 10 after addition of lucerne, canola and lupin residues in saline sandy and sandy loam soils with five salinity treatments

7.3.4.2 Changes of microbial biomass C, N and P over time in saline soils as affected by addition of plant residues

Microbial biomass C, N and P changed significantly over time (Figure 7.6). Microbial biomass was generally higher in amended soils than in unamended soils, but was strongly affected by soil texture and, to some extent, by residue type. The trend of microbial biomass in soils amended with lucerne, canola and lupin residues is the same, but with different magnitudes (described in previous sections). Changes over time of microbial biomass C, N and P will therefore be shown in both soils with only one crop residue: lupin at all salinity treatments. Lupin residue was chosen as an example:

because its low C/N ratio and lignin content make it easily decomposable, thus has the greatest positive effect on nutrient availability for plants.

7.3.4.2.1 Microbial biomass C over time

Without addition of salt (S1), microbial biomass C increased in both soils in the first 10 days and then remained unchanged until day 50 in the sandy loam soil, but decreased from day 10 to day 50 in the sandy soil. Microbial biomass C was higher in sandy soil than in sandy loam soil at all times. In S1, compared to unamended soil, microbial biomass C was 2 and 4 times higher on day 10 in sandy loam and sandy soil, respectively. With increasing salinity, microbial biomass C significantly decreased in both soils with and without residue. On day 10, microbial biomass C decreased by about 100 mg C with each increase in salinity level in the sandy loam and by about 60 mg C kg⁻¹ soil in the sand.

7.3.4.2.2 Microbial biomass N over time

Microbial biomass N followed the same trend of biomass C; increasing in the first 10 days and then decreasing slightly to day 50. Microbial N was higher in sandy than in sandy loam soil at all salinity treatments. In both soils, biomass N decreased with increasing salinity with about 15 mg N kg⁻¹ soil with every increase in soil salinity.

7.3.4.2.3 Microbial biomass P over time

Microbial biomass P strongly increased in the first 10 days in both soils with higher biomass P in the sandy loam than in the sandy soil at S1 and S2 and higher biomass P in the sandy soil than in sandy loam soil at salinity treatments above S2. In both soils, microbial biomass P decreased with increasing soil salinity. Microbial P decreased from day 10 to day 50 in both soils. On day 50, microbial biomass P was higher in sandy soil than in sandy loam soil at all salinity treatments.



Figure 7.6 Microbial biomass C, N and P before residue addition and on days 10 and 50 after addition of lupin residues in saline sandy loam and sandy soils with five salinity treatments

7.4 Discussion

In response to increased soil salinity, changes of cellular physiology and metabolic processes of microorganisms as well as the death of several microbial groups have been reported (Killham, 1994; Zahran, 1997; Rietz and Haynes, 2003; Nelson and Mele,

2007). In the preliminary experiment, respiration (Figure 7.1) was observed in the sandy loam with salinity in the soil solution up to approximately EC 77 dS m⁻¹ (S5; Table 7.2), whereas in the initial plant experiment (Chapter 5), plants did not grow in the same soil at approximately EC 37.4 dS m⁻¹ (S2; Table 5.1 and Plate 5.1). Therefore the results in this chapter indicate that, compared to wheat and probably many other plants, microorganisms can adapt to salinity (see mechanisms in Section 2.1.2.2).

The negative effect of salinity on respiration in this study is in accordance with several studies (Laura, 1974; McCormick and Worlf, 1979; Azam and Muller, 2003), which showed that salinity significantly decreased respiration in a range of soils. Reduced soil respiration with increasing salinity can be attributed to increasing soil osmotic pressure restricting microbial access to soil water. Accumulation of Na⁺ and Cl⁻ ions in microbial cells to levels that can be toxic can negatively affects the physiology and metabolic processes of microorganisms.

The reduction of soil respiration over time has been explained by a reduction of water availability (Sardinha et al., 2003), aeration (Agehara and Warncke, 2005) and/or availability of nutrients, particularly C, N and P (Gray, 1976; Azam et al., 1993; Cheshire and Chapman, 1996). In the present study, the pattern of CO₂ evolution was similar in all soils at each salinity treatment, but with different magnitudes over time (Figure 7.1). The higher initial respiration rates can be explained by the addition of soluble C-glucose and N-KNO₃. Rates of respiration decreased in the second week. In this experiment, amended soils were well aerated and watered (85% WHC; after every measurement). Therefore, aeration is unlikely to have caused the decrease in respiration over time. Also, the results shown in Figure 7.2 indicated that approximately 7% (175 μ g CO₂-C g⁻¹ soil) of added C was respired in the first 8 days (note that this does not take into account CO₂ from soil native C compounds). This makes it unlikely that the decrease in respiration in the second week was due to C limitation, but rather due to the changes in microbial community structure, microbial death or N and P limitation, but none of these hypotheses can be confirmed in this study.

The higher concentration of soil C and availability of N and P (Chapter 3; Table 3.1) to microorganisms might be one reason for highest respiration in the silty loam.

Data shown in Tables 7.2 and 7.3 indicated that at all salinity levels, salinity of the soil solution was highest in the sand, and decreased in the order sand > sandy loam > silt loam. Hence, the low respiration in the sand can be due to the higher salinity stress in
this soil compared to the other two soils. Additionally, differences in microbial biomass size as well as the microbial community structure might also be reasons for the quicker response and the greater respiration in the silty loam than in the sandy and sandy loam soils.

The results of the main experiment showed that, compared to non-amended soils, N and P availability (Table 7.6) and microbial biomass (Table 7.7) were higher with plant residue addition which can be explained by release of nutrients during decomposition of plant residues. In the amended soils, concentrations of NH_4^+ and NO_3^- were lower with increasing soil salinity (Table 7.6). The lower NH_4^+ concentration can be due to the death of ammonifiers with increasing soil salinity (Pathak and Rao, 1996; Irshad et al., 2005). Ammonium adsorption and fixation to soil particles, or conversion into other N forms such as NH_3 could also decrease NH_4^+ concentration (Petit et al., 1998). In a salinized loamy sand soil amended with manure, NO3⁻ concentration increased over time; was higher at week 8, compared to week 1, but decreased with increasing salinity, and was detected at EC 113 dS m⁻¹ (Irshad et al., 2005). Similarly, in the present study, NO3⁻ concentration increased over time and decreased with increasing salinity, and was detected in both soils at S5; approximately 67.8 and 131.1 dS m⁻¹ in the sandy loam and in the sand, respectively. The reduction of NO₃⁻ concentration with increasing salinity can be attributed to inhibition of nitrifiers (Azam and Ifzal, 2006), but also indicates that nitrification was not completely inhibited. In non-amended soil, nitrification was inhibited by 50% and 70% in silty loam and sandy loam soils salinized to ECe 5 and 10 dS m⁻¹ (Kumar and Wagenet, 1985). In the present study, with no added amendement nitrification was inhibited by only 25% in the sandy loam with EC_e 12 dS m⁻¹ and 43% in the sand with EC_e 24.5 dS m⁻¹. Compared to S1, with residue addition available NO_3^{-1} was approximately 80% lower in both soils at S5, and available NH4⁺ was approximately 73% lower in the sandy loam and 60% lower in the sand. This indicates that ammonifiers, compared to nitrifiers, to some extent, can adapt, and are more tolerant to salinity. This is supported by the fact that NH₄/NO₃ ratio increased with increasing soil salinity (Table 7.6).

Phosphorus availability was higher in the amended than in the non-amended soils (Table 7.6), to a great extent, due to the release of P from the added residues. Availability of P also decreased with increasing salinity, which is expected due to possible chemical reactions; Ca-P precipitation, or P fixation to clays directly or via clay-organic matter complexes (White, 1980). In the sand, although the actual EC was

higher than in the sandy loam (Table 7.4), concentrations of Ca^{2+} from added salts in all salinity treatments was the same as in the sandy loam, given the salts have been 100% dissolved. The interactions between plant residue and salinity and soil texture may have resulted in higher release of soil native P in the sandy loam than in the sand, but this can not be confirmed in this study.

Compared to non-amended soils, microbial biomass was greater with amendements (Table 7.7). The increase of microbial biomass size could have occurred due to availability of greater amounts of labile C and soluble N and P associated with amendments (Li et al., 2006a; Li et al., 2006b) which were immobilized increasing the microbial activity and thus salt tolerance.

Earlier studies (Chaussod et al., 1986; Gregorich et al., 1991; van Gestel et al., 1991; Amato and Ladd, 1992) showed that in non-saline soils, microbial biomass was greater in clayey soil than in sandy soil with residue addition. In the present study, with increasing salinity, greater microbial biomass C (Figure 7.3) and N (Figure 7.4) were found in the amended sandy soil than in the amended sandy loam soil. Most likely different types of microorganisms were involved in the decomposition processes as the microbial biomass was generally lower in the sandy loam than in the sandy soil.

In the sandy loam soil, a physical protection resulting from the adsorption of substrates to surfaces, or from their location within soil aggregates at sites inaccessible to microorganisms (Jenkinson, 1977) could have occurred, and although high salinity and because of better accessibility to organic matters, nutrient release and microbial biomass were higher in the sand (Tables 7.5 and 7.6). While the size of microbial biomass C and N was higher in the sandy soil, microbial biomass P was greater in the sandy loam (Figure 7.6). It might be that there were more P accumulating microbial species in the sandy loam soil than in the sandy soil.

As shown in Figure 7.6, microbial biomass C, N and P increased in the first 10 days after residue addition due to the flush of high amounts of readily-soluble (available) compounds from the residues into the soil, and decreased from day 10 to day 50 probably due to the death of several microbial groups as a result of limited availability of nutrients (C, N and P) (Chauhan et al., 1981).

In amended soil, the type of amendment and its interaction with the soil salinity and texture affected nutrient availability. It should be stressed that the plant residues used here were not P-rich and contained almost the same amount of P (0.11%), but differed in N content, C:N and C:P ratios and lignin and cellulose contents. The trend in the

sandy soil was similar to the sandy loam that under saline conditions; N availability was significantly higher in the lupin amended soil than in the soil with the other residues. The high decomposition rates of lupin residues under non-saline and saline conditions, compared to the other residues, can be due to its high N and low lignin contents (Table 3.2). This suggests that the stressed microbes in the saline soils were not able to take advantage of the higher nutrient availability from the lupin residues. Therefore, the type of the residue did not affect the microbial biomass size under saline conditions. However, at all salinity levels, compared to the non-amended soils, with residue addition the increase in soil microbial biomass and the lower reduction rate of biomass size indicates that residue addition enhanced the salt tolerance of microorganisms.

7.5 Conclusions

Plant residue addition increased the availability of N and P, and microbial biomass more in the sand than in the sandy loam under saline conditions. Microbial biomass size was negatively affected by soil salinity. Ammonification and nitrification processes were however not completely inhibited up to approximately EC_e 65.6 dS m⁻¹, equivalent to 131 dS m⁻¹ (in soil solution), in the sand and EC_e 34.3 dS m⁻¹, equivalent to 68 dS m⁻¹ (in soil solution), in the sand loam suggesting that at least some ammonifiers and nitrifiers were tolerant to the saline conditions. The salt tolerance of microorganisms was enhanced by residue addition, but differences between plant residues were override by soil salinity at SAR 1 and neutral pH.

Chapter 8 – Effects of inorganic N and lupin residue on soil

respiration, microbial biomass and N and P availability in a saline

sandy loam soil

2.1 Introduction

Improving soil microbial, physical and chemical properties is of utmost importance to enhance plant growth and hence productivity in the salt affected soils. Addition of fertilizers (organic and inorganic) is a key factor in the nutrient management for the promotion of agricultural sustainability in saline soils.

The vital role of microorganisms in the soil biochemical processes, including organic matter decomposition and nutrient transformation and cycle, and its contribution to the agricultural sustainability has been shown (Smith and Paul, 1990; Cookson et al., 2007). With increasing salinity, microbial activity decreased thus nutrient release from residue decreased. With consideration to the results of Chapters 5 and 6 and 7, addition of 2% lupin residue can provide approximately 30 and 100% of wheat requirements of P (60 mg kg⁻¹ soil) and N (100 mg kg⁻¹ soil), respectively, in the sandy loam soil. Therefore, it can be assumed that the rate of residue decomposition, as well as P availability, will be high at the salinity levels that wheat can tolerate, addition of 2% or more of lupin residues may increase microbial activity and turnover and thus N and P availability. Thus it could be hypothesized that addition of plant residues at 2% or more can relatively enhance wheat growth under saline conditions.

Salinity has been widely reported to have a negative effect on microbial and enzyme activities (Rietz and Haynes, 2003; Sardinha et al., 2003) and microbial biomass (Yuan et al., 2007). The activity and the microbial biomass can also be severely limited by nutrient availability and as a result many soil organisms may have very low metabolic rates or survive in dormant or resting phases (Gray, 1976).

Inorganic N has a stimulating effect on microbial activity by increasing microbial biomass (Parkinson et al., 1971). Under N limiting conditions, addition of inorganic N increased the decomposition rate of plant residues in soils (Carreiro et al., 2000; Moran et al., 2005; Sirulink et al., 2007). Jones and Shannon (1999) showed that addition of inorganic N had little effect on the microbial use of N from organic sources in amended soils. It was reported that immobilization of NH_4^+ is faster than NO_3^- (Azam et al., 1993). Also, greater immobilization of NH_4^+ than NO_3^- during straw decomposition has

been shown (Jansson, 1958; Recous and Mary, 1990). This is attributed to differences in microbial community structure and because with NO_3^- microorganisms may require more energy for the conversion into NH_4^+ and for synthesis (Azam et al., 1993), and also due to the preferential assimilation of NH_4^+ by the heterotrophic microflora. However in field studies (Bowden et al., 2004; Burton et al., 2004) and in glasshouse experiments (Micks et al., 2004), N application depressed or did not influence soil respiration, especially when it was added as NH_4NO_3 due to decreased pH and limited availability of C (Amador and Jones, 1993; Thirukkumaran and Parkinson, 2000), or increased osmotic potential by addition of fertilizer salts. Bjarnason (1987) found no difference between N forms for rates of immobilization and remineralisation.

Under saline conditions, N as NH_4^+ or NO_3^- applied up to 200 mg kg⁻¹ soil increased microbial biomass, soil respiration and N availability in naturally saline alkaline soils amended with either glucose or organic materials (Luna-Guido and Dendooven, 2001; Conde et al., 2005; Dendooven et al., 2006). In presence of glucose, compared to NO_3^- , microbial response was higher with NH_4^+ under saline conditions due to the higher sensitivity of microorganisms assimilating NO_3^- to NaCl, but N remineralisation is faster with NO_3^- than NH_4^+ (Azam and Ifzal, 2006). Also, in naturally-alkaline soils, cumulative soil respiration increased with addition of N-NO₃ in presence of glucose (Dendooven et al., 2006).

Addition of plant residues has significantly increased microbial activity and/or biomass in saline soils (Li et al., 2006a; Li et al., 2006b) and in sodic soils (Nelson et al., 1996; Clark et al., 2007). Microbial immobilization of N may be greatly dependent on the availability of C. Nitrogen as NO_3^- was not immobilized when added at 100 mg kg⁻¹ soil without addition of a C source (Wickramasinghe et al., 1985). Azam et al. (1988) observed N-NO₃ disappearance during incubation with glucose, cellulose and sucrose.

Addition of N in organically-amended soil with high pH can lead to loss of N-NH₄ through NH₃ volatilization (Luna-Guido and Dendooven, 2001) and negatively affect certain fungal and actinomycete groups, but can increase the hydrolyzation and hence decomposition of organic matter (Rai and Srivastava, 1982). Microbial activity and thus decomposition of organic matter in naturally-saline soils is often greater than in soils with induced salinity since microorganisms in naturally saline soils had time to adapt to the osmotic stress (McClung and Frankenberger, 1987; Beltran-Hernandez et al., 1999). The studies described above were conducted in soils naturally saline which

are, in most cases also alkaline and/or sodic, or in salinized soils with NaCl that can indirectly increase soil SAR. The high Na⁺ adsorption on clay minerals in sodic soils leads to dispersion of clay particles and reduced soil aggregation allowing microorganisms to attack small as large organic matter particles increasing organic matter decomposition (Nelson et al., 1996). Hence, salinity, alkalinity and sodicity can have contrasting effects on microbial activity. There is no information about the effect of N and lupin residues on microbial biomass and activity, and N and P availability in saline soils that are not alkaline or sodic.

The objectives of this study were therefore to determine the effects of addition rate of lupin residues and inorganic N on soil microbial respiration and biomass, and to investigate how N and P availability and microbial activity and biomass are affected by the form of N and its interaction with lupin residues in the sandy loam under saline non-sodic or alkaline conditions.

8.2 Materials and methods

8.2.1 Preliminary experiment

A preliminary experiment was set up to determine soil respiration in a sandy loam soil amended with inorganic N and lupin residues under non-saline conditions. The sandy loam soil from Monarto (Table 3.1) was used in this study.

8.2.1.1 Soil preparation and nutrient solution

The preparation of the soil was as described in Chapter 6. A nutrient solution was added to give the following concentrations (mg kg⁻¹ soil): 71.5 Ca (CaCl₂.2H₂O), 37.0 Mg (MgSO₄.7H₂O), 78.1 K (K₂SO₄), 0.04 Fe (Fe-EDTA), 0.147 Mn (MnSO₄.4H₂O), 0.499 Zn (ZnSO₄.7H₂O), 0.050 Cu (CuSO₄.5H₂O), 0.087 B (H₃BO₃), 0.277 Mo (Na₂MoO₄.2H₂O) and 0.084 Co (CoCl₂.6H₂O). The nutrient solution was added to the soil to have the same nutrient conditions as in the experiments with wheat (Chapters 5 and 6). Phosphorus was not added to the nutrient solution because lupin residues were to be the sole P source.

8.2.1.2 Nitrogen and organic matter treatments

After a 2-week incubation time at 50% WHC, solutions of KNO_3 were added to the soil at 25 (N25), 50 (N50) and 100 (N100) mg N kg⁻¹ soil. A treatment without N addition (N0) was also included. Lupin residues (Chapter 7.1), ground to 1.5 mm, were added at

2% (w/w) to the soil. The amended soil was moistened to 85% WHC. The study was conducted with 4 replicates per treatment.

8.2.1.3 Incubation and Measurements

A PVC container with mesh at the bottom was filled with 20 g amended soil (dry weight equivalent). Each container was then transferred into a jar as described in section 3.1.3 and incubated 14 days in the dark in a constant temperature room (28 ± 3 °C). Released CO₂-C was measured (at intervals over 14 days) and calculated as described in Chapter 3.

8.2.2 Main experiment

For the evaluation of the effect of inorganic N addition on microbial biomass and N and P availability in amended-saline soil, an experiment was conducted in a constant temperature room $(28\pm3 \text{ °C})$ for 45 days. The sandy loam soil of Monarto was air dried, sieved, treated with solutions of salts and nutrients, and stored in the dark for 2 weeks as described above.

8.2.2.1 Soil salinization

Mixtures of NaCl and CaCl₂.2H₂O were added to induce 3 levels of salinity in the sandy loam soil (Table 8.1). In three salinity treatments, the average values of $EC_{1:5}$ were 0.21, 0.51 and 0.85 dS m⁻¹ in S1, S2 and S3, respectively. The levels of salinity were based on the results of Chapters 5 and 6: at these levels wheat (cv Krichauff) positively responded to the addition of inorganic N and P.

Table 8.1 Amounts of NaCl and CaCl₂.2H₂O added to a sandy loam soil and soil osmolarity (mosmoles L^{-1}) and EC_{1:5} EC_e and EC in soil solution (dS m-1)

	NaCl		Ca	Cl ₂ .2H	$_{2}\mathbf{O}$	Osmolarity	EC _{1:5}	ECe	EC		
Salt	mg	тM	тM	mg	тM	тM	mosmoles L ⁻¹				
Level	kg^{-1} soil		soil	kg ⁻¹ soil		soil	in soil soln	dS m^{-1}			
	0		soln	0		soln					
S1	0	0	0	0	0	0	0	0.21	2.8	5.6	
S2	117	2.00	18.2	588	4.00	36.4	145	0.51	7.0	14.0	
S 3	170	2.90	26.6	1255	8.53	77.6	286	0.85	11.7	23.4	

Soil soln = soil solution. The concentration (m*M*) was estimated in the soil solution. $EC_{1:5}$ was measured in the soil after incubation prior to establishment of the glasshouse experiment. EC_e was calculated based on the relationship between the texture and the moisture of the sandy loam (Slavich and Petterson, 1993). EC was calculated knowing

8.2.2.2 Addition of inorganic N and lupin residue

After the 2-week moist incubation, solutions of KNO3 and (NH4)2.SO4 were added to the soil at 0 (N0) and 50 (N50) mg N kg⁻¹ soil to determine whether the form or the application rate of N affects microbial biomass and activity in a saline sandy loam soil. To study to what extent increasing addition of organic matter can affect microbial biomass and activity and N and P availability, lupin residues were ground to 1.5 mm, added at 0, 2% and 4% (w/w) and thoroughly mixed with the soil. After addition of N and lupin residues, plastic and PVC containers were filled with 100 and 20 g (dry weight equivalent) of amended saline soil, respectively, and then transferred to an incubation room with temperature of 28 ± 3 °C. Pots with 100 g soil were used to measure available N and P and microbial biomass C, N and P at different sampling intervals, whereas 20 g soil samples were used for measurements on soil respiration. The pots with 100 g soil were left uncovered while the PVC container of 20 g soil was placed in a Mason jar with lid. Four replicates for every treatment were prepared. Throughout the experiment (45 days), the moisture of the soil in the uncovered pots and in the PVC containers was maintained at 85% WHC by adjusting container weight with de-ionized water routinely.

8.2.2.3 Measurements

Soil respiration was measured (as described in Chapter 3) at intervals over 45 days, while microbial biomass C, N and P and the availability of P were measured in soil samples before the addition of amendments and on day 15, 30 and 45 after the establishment of the experiment. Available N was measured before addition of amendments and on days 20 and 40 after residue and inorganic N addition, and calculated as the sum of available NH_4^+ and NO_3^- . Soil sampling and methods of extraction, measurement and calculation are described in Chapter 3.

8.2.2.4 Design and statistical analysis

In the main experiment, there were two application rates of two N fertilizers in combination with three salinity and three application rates of lupin residue treatment replicated four times and arranged in a completely randomised design. Analysis of variance was carried out using Genstat 8. In tables, values are means. Values in the

same column followed by the same letter are not significantly different at $P \le 0.05$ according to Tukey test. The abbreviation "ns" means not significant. Bars in histograms show standard deviation (STD).

8.3 Results

8.3.1 Effect of inorganic N on respiration rate in amended non-saline soil

For the preliminary experiment under non-saline conditions, the two-way analysis of variance indicated that in the lupin amended sandy loam N application rate had no significant effect on the soil respiration 14 days (Figure 8.1).





8.3.2 Effect of inorganic N and lupin residue on the respiration rate and the cumulative respiration in the saline soil

The results of the statistical analysis for main and interactive effects of N form and addition rate, salinity and lupin residues on soil microbial biomass, and N and P availability are shown in Table 8.2. Statistical analysis was applied at particular times to correlate the soil microbial biomass with nutrient availability.

Generally, addition of lupin residue had a significant effect on respiration rate and cumulative respiration in saline soil. The analysis of variance showed that salinity and its interaction with N addition rate or N form had no significant effect on soil respiration in the non-amended and amended soil throughout the experiment (45 days).

Nitrogen form and addition rate had a significant effect on soil respiration and cumulative respiration in the lupin-amended saline soil from day 16 to 45, but not in the first 2 weeks of incubation.

Table 8.2 Results of statistical analysis; least significant differences (LSD) and significance at 5% level, of main and interactive effects of N form and addition rate, salinity and lupin residue on soil respiration rate and cumulative respiration in a saline sandy loam soil

	Soi	l respira	ation	Cumulative respiration			
Interactions	Day	Day	Day	Day	Day	Day	
	1-15	16-30	30-45	1-15	16-30	30-45	
N rate	ns	0.04	0.02	ns	83	95	
N form	ns	ns	ns	ns	103	113	
Residue	1.3	0.06	0.03	ns	95	94	
Salinity	ns	ns	ns	119	ns	ns	
N rate X residue	ns	ns	ns	ns	ns	ns	
N form X residue	1.1	0.05	0.02	ns	80	75	
N rate X salinity	ns	ns	ns	99	ns	164	
N form X salinity	ns	ns	ns	ns	ns	ns	
residue X salinity	ns	ns	0.04	ns	165	164	
N rate X residue X salinity	ns	ns	0.05	ns	202	284	
N form X residue X salinity	ns	ns	ns	354	309	339	
ng not significant							

ns, not significant.

8.3.3 Effect of lupin residue on respiration rate in the non-saline sandy loam soil

The time course of respiration of sandy loam soil amended with 0, 2 and 4% lupin residue is shown in Figure 8.2. Generally, the respiration rate was low in the unamended soil with an average of 3 μ g CO₂-C g⁻¹ soil hour⁻¹ over the 45 days of incubation. Soil respiration significantly increased with addition of lupin residue. On day 3 of incubation, compared to the unamended soil, CO₂ evolution rates were approximately 15 and 30 times higher with addition of lupin residues at 2 and 4%, respectively. Soil respiration rates decreased over time in the amended soil and were 17 and 36 μ g CO₂-C soil hour⁻¹ by the end of the first week with lupin residues at 2 and 4%, respectively. The respiration rate continued to decrease over time with highest rates in the soil amended with 4% lupin residue.



Figure 8.2 Respiration rate (μ g CO₂-C g⁻¹ soil hour⁻¹) over 45 days in a non-saline sandy loam soil with 0, 2 and 4 g lupin residue 100 g⁻¹ soil

8.3.4 Effects of N on cumulative respiration in a saline sandy loam soil with lupin residue

The trend of cumulative respiration with NH_4^+ was similar to NO_3^- in the unamended soil at all salinity levels. Therefore the soil amended with 2% lupin residue was selected as an example to show the effect of N rate and form on cumulative respiration in a saline sandy loam soil at salinity level S2. Cumulative respiration in a saline sandy loam soil amended with 50 mg N (NH_4^+ or NO_3^-) mg⁻¹ soil and 2% lupin residue is shown in Figure 8.3.

Cumulative respiration increased rapidly in the first 20 hours to approximately 770 μ g CO₂-C in the soil without N addition and with 50 mg NH₄-N and 1900 μ g CO₂-C g⁻¹ soil with 50 mg NO₃-N. Cumulative respiration increased linearly until the end of the experiment. Expressed in percentage of the total soil C content (native soil C and added O.M), cumulative respiration was 18, 26 and 34% in the soil with NO₃⁻ and only 10, 17 and 24% in the soil without N or with NH₄-N on days 7, 21 and 42, respectively.



Figure 8.3 Effect of N form (NH₄⁺ and NO₃⁻) and addition rate (50 mg N kg⁻¹ soil) on cumulative respiration (μ g CO₂-C g⁻¹ soil) over time (first 90 hours and from day 7 to 45 of incubation) in a sandy loam soil amended with 2% lupin residue at S2

8.3.5 Cumulative respiration in a saline sandy loam soil as affected by addition rate of lupin residue and the form of inorganic N

The interactive effects between lupin residues rates and inorganic N rate and form on cumulative respiration under saline conditions (S3) are shown in Figure 8.4. Cumulative respiration in the soil without N and was similar to the soil with NH_4^+ or NO_3^- .

Cumulative respiration increased with increasing addition rate of lupin residue. The cumulative respiration in the soil with 4% lupin residue and 50 mg N kg⁻¹ soil was twice as high as the rates in the soil amended with 2% lupin residue and 50 mg N kg⁻¹ soil and approximately 10 times higher than in the soil with only 50 mg N kg⁻¹. With 4% residue, cumulative respiration was greater with NO_3^- than with NH_4^+ throughout the experiment and the difference became greater over time. Lowest cumulative respiration was in the soil without residues, with or without NO_3^-N .



Figure 8.4 Cumulative respiration (μ g CO₂-C g⁻¹ soil) at S3 over 45 days in a sandy loam soil amended with lupin residue added at 2 and 4%, and N as NH₄⁺ or NO₃⁻ at 50 mg N kg⁻¹ soil

8.3.6 Microbial biomass C, N and P and N and P availability in a saline soil amended with inorganic N and lupin residue

Results of statistical analysis of main and interactive effects of N form and addition rate, salinity and lupin residue on N and P availability and microbial biomass C, N and P in a saline sandy loam soil are shown in Table 8.3.

Statistical results indicated that N addition significantly affected N availability and microbial biomass C and N, but had no significant effect on microbial P and available P. The results also indicated that the form of N had a significant effect on all measured parameters, except P availability. Residue addition and salinity individually also had significant effects on N and P availability, and on microbial biomass (C, N and P). In contrast to the individual effects, all interactive effects, except for plant residue with N rate and form or with salinity, were not significant for the measured soil properties.

N rate X residue

N form X residue

N rate X salinity

N form X salinity

Residue X salinity

N rate X residue X salinity

N form X residue X salinity

saline sandy loam soil											
]	Day 15	5	Day 30			Day 45				
Interactions	Mic P	Mic C	Mic N	Mic P	Mic C	Mic N	Mic P	Mic C	Mic N		
N rate	ns	18	2.3	ns	14	2.3	ns	11	2.3		
N form	0.5	14	2.2	0.3	10	2.1	0.2	13	2.1		
Residue	0.7	20	2.6	0.6	16	2.7	0.3	13	2.7		
Salinity	0.7	20	2.6	0.6	16	2.7	0.3	13	2.7		

3.2

3.0

ns

ns

4.5

ns

ns

0.9

0.6

ns

ns

1.0

ns

ns

19

18

28

ns

ns

ns

ns

3.3

ns

ns

ns

4.6

ns

ns

ns

0.4

ns

ns

0.6

ns

ns

16

19

ns

19

22

ns

ns

3.3

4.2

ns

ns

4.6

ns

ns

Table 8.3 Results of statistical analysis; least significant differences (LSD) and significance at 5% level of main and interactive effects of N form and addition rate, salinity and lupin residue on N and P availability and microbial biomass C, N and P in a saline sandy loam soil

8.3.6.1 Microbial biomass C, N and P over time as affected by addition of inorganic N and lupin residue in a saline sandy loam soil

ns

1.2

ns

ns

The changes of microbial biomass C, N and P over time in the amended soil are shown in Figure 8.5. After 2-week incubation (before addition of N and residue amendments), microbial biomass C was 180, 143 and 98 mg C kg⁻¹ soil, biomass N was 5, 8 and 11 mg N kg⁻¹ soil and biomass P was 2.9, 2.5 and 2.3 mg P kg⁻¹ soil at S1, S2 and S3, respectively. Microbial biomass C, N and P increased to up to day 15 and were higher in the amended soil than in the unamended soil. After day 15, microbial biomass C, N and P decreased gradually over time with significant differences between treatments. Increasing soil salinity decreased microbial biomass C, N and P over time. The effect of amendment addition on microbial biomass C, N and P was more pronounced at S2 and S3 than at S1.

8.3.6.1.1 Microbial biomass C

Microbial biomass C as affected by residue rate and N rate and form under saline conditions is shown in Figure 8.5a. With every increase in soil salinity, microbial biomass C decreased by about 150-200 mg C kg⁻¹ soil, with or without amendments at all times. On day 15, compared to the un-amended soil with or without salt, microbial biomass C was approximately 1, 2, and 3 fold higher in the soil with 50 mg N kg⁻¹ soil,

2% and 4% lupin residue, respectively. Addition of inorganic N increased microbial biomass C by approximately 150-200 mg C kg⁻¹ soil in the soil with 0, 2, and 4% residues. Microbial biomass C was approximately 50 to 100 mg C kg⁻¹ higher in NO_3^- than in NH_4^+ amended soil with lupin residues at all salinity treatments. Microbial biomass decreased from day 15 to day 30. On day 45, microbial biomass C was approximately 10% lower than the biomass at day 30 in all salinity and amendment treatments.

8.3.6.1.2 Microbial biomass N

Microbial biomass N in the saline sandy loam soil as affected by addition of N and lupin residue over time is shown in Figure 8.5b. Similar to the trend of microbial biomass C, the highest microbial biomass N was found on day 15. On day 15, in presence of inorganic N, addition of lupin residue at 2% resulted in approximately 32, 36 and 39% increase of the microbial biomass N, while with 4% residues microbial biomass N was 77, 86 and 83% greater than in the soil without residues at S1, S2 and S3, respectively. With lupin residue, microbial biomass N was 70-100 mg N kg⁻¹ higher with NO₃⁻ than with NH₄⁺ at all salinity treatments. In all N and residue treatments, with every increase in soil salinity, microbial biomass N decreased by about 20-40 mg N kg⁻¹. On day 30, microbial biomass N was approximately 60, 55 and 50% of the microbial biomass found on day 15 at S1, S2 and S3, respectively. On day 45, microbial N was approximately 10% lower than the biomass N of day 30 at all salinity treatments.

8.3.6.1.3 Microbial biomass P

Microbial biomass P increased with residue addition and was highest on day 15 (Figure 8.5c). Microbial biomass P decreased with increasing salinity, was not affected by N addition and significantly increased by addition of lupin residue. In all N and residue treatments, with every increase in soil salinity, microbial biomass P decreased by about 10-15 mg P kg⁻¹ soil. While 2% lupin residue increased microbial biomass P to approximately 7 fold of that in the unamended soil, microbial biomass P was about 1.6 times higher with 4% than with 2% residues at all salinity treatments. On day 30, microbial biomass P decreased with increasing salinity in the amended soil, but was lower than on day 15. On day 45, microbial biomass P was approximately 15% lower



than at day 30 and was approximately 60, 50 and 45% of the microbial P on day 15 at S1, S2 and S3, respectively.

(c)

Figure 8.5 Microbial biomass carbon (a), nitrogen (b) and phosphorus (c) on days 15, 30 and 45 after addition of inorganic N as NH_4^+ or NO_3^- at 50 mg N kg⁻¹ soil and lupin residues at 2 and 4% in a sandy loam soil with three salinity treatments

8.3.6.2 Microbial C/N ratio in an amended saline sandy loam soil over time

Microbial C/N ratio was affected by time in all treatments, but not by soil salinity (Table 8.4). Microbial C/N ratio increased over time in the unamended soil.

Table 8.4 Microbial C/N and C/P ratios on day 15, 30 and 45 in a saline sandy loam soil amended with with lupin residue added at 2 and 4%, and N as NH_4^+ or NO_3^- at 50 mg N kg⁻¹ soil

Treatment Solinity loyal C/N ratio C/P ratio	C/P ratio			
D15 D30 D45 D15 D30	D45			
Control S1 12 14 16 131 76	91			
(no lupin residue) S2 12 14 14 102 70	71			
(no nitrogen) S3 12 14 16 84 69	89			
S1 12 10 12 231 106	95			
N-NH ₄ S2 13 11 14 187 102	93			
S3 12 10 19 132 91	83			
S1 12 10 12 235 117	112			
N-NO₃ S2 12 10 14 212 109	102			
S3 12 10 16 189 90	87			
S1 11 10 14 35 28	51			
2% lupin residue S2 11 10 13 40 23	39			
S3 11 9 13 49 23	32			
S1 7 8 8 30 32	40			
4% lupin residue S2 8 7 9 31 27	36			
S3 8 9 10 32 27	32			
2% Junin residue S1 8 9 7 41 40	51			
$+ N-NH_4$ S2 8 9 7 59 32	47			
S3 8 9 9 103 32	49			
2% lupin residue S1 6 9 7 47 38	48			
+ $S2$ 6 9 7 54 33	43			
N-NO₃ S3 6 9 6 65 26	33			
4% lupin residue S1 7 9 8 36 36	46			
+ S2 7 9 8 39 34	49			
N-NH ₄ S3 7 9 10 44 27	39			
4% lupin residue S1 6 9 8 36 33	43			
+ S2 6 9 8 49 32	39			
N-NO₃ S3 6 9 7 45 27	35			
$LSD (P \le 0.05)$	2.00			
N rate 0.34 0.38 0.75 5.92 1.90	3.88			
N IOFM ns ns 0.79 15.93 2.23	ns			
residue 0.39 0.44 0.87 6.82 2.26 Solicity 0.87 0.87 0.82 2.26	4.48			
Samuely ns ns 0.87 ns 2.20 N note Y model 0.68 0.76 1.50 11.84 2.01	ns			
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	ns			
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	ns			
N form X salinity 115 115 115 115 115 115 115 115	115 5 86			
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	J.00			
N rate X residue X salinity $n_{\rm S}$ $n_{\rm S}$ $n_{\rm S}$ $n_{\rm S}$ $n_{\rm S}$ $n_{\rm S}$	ne			
N form X residue X salinity ne ne ne ne ne	ns			

In the first 15 days, microbial C/N ratio was not affected by addition of N only, but it was lower in soil with residue with inorganic N. On day 30, the microbial C/N ratio increased in the unamended soil, decreased in the soil with either 2% lupin residue with or without N or inorganic N only or with both and increased in the soil with inorganic N and 4% lupin residue. On day 45, microbial biomass C/N ratio increased in the unamended soil and in the soil amended with inorganic N or with 2% lupin residue and remained unchanged in the soil amended with the combination of inorganic N and lupin residue.

8.3.6.3 Microbial C/P ratio in an amended saline sandy loam soil over time

At all sampling dates, the microbial C/P ratio decreased with increasing soil salinity. With every increase in salinity, the microbial C/P ratio decreased by about 20-25% in the control soil (no residue) and by about 50-60% in presence of only inorganic N. Microbial C/P ratio was 2 times greater with inorganic N and much lower with residue compared to the unamended soil in all salinity treatments. At all salinity treatments, compared to soil with residue, C/P ratio was higher with addition of inorganic N. At all salinity treatments, microbial C/P ratio decreased from day 15 to day 30 in the unamended and amended soils. From day 30 to day 45, microbial C/P ratio decreased in the soil amended with inorganic N only and increased in the unamended soil and in the soil with lupin residue, with and without N.

8.3.6.4 Cumulative respiration (C_{cum}) to microbial biomass C (C_{mic}), N (N_{mic}) and

P (P_{mic}) 45 days after addition of lupin residue and inorganic N

The ratio of C_{cum} to C_{mic} , N_{mic} or P_{mic} was significantly affected by the individual and the interactive effects of soil salinity, lupin residue addition and inorganic N in the soil (Figure 8.5). All ratios of cumulative respiration to microbial C, N and P significantly increased with increasing soil salinity with or without amendments. Compared to nonamended soil, with N addition C_{cum} to N_{mic} ratio was at least 6 times higher at all salinity treatments with greater ratio in presence of NH_4^+ compared to NO_3^- . Also, N addition C_{cum} to P_{mic} ratio was greater, and it was higher in presence of NO_3^- compared to NH_4^+ . Compared to the non-amended soil, with 2% residue addition C_{cum} to C_{mic} or N_{mic} were greater and C_{cum} to P_{mic} was lower. With lupin residues, C_{cum} to C_{mic} was not affected and C_{cum} to N_{mic} or P_{mic} decreased with addition of inorganic N.

Table 8.5 Ratios of cumu	lative respiration	(C _{cum}) to mic	robial biomas	s C (C _{mic}), N						
(N _{mic}) and P (P _{mic}) 45 day	s after addition of	f lupin residu	e added at 2 a	and 4%, and N as						
NH_4^+ or NO_3^- at 50 mg N kg ⁻¹ soil in a sandy loam soil at three salinity levels										

Treatment	Salinity level	C _{cum} / C _{mic}	C _{cum} / N _{mic}	C _{cum} / P _{mic}
Control	S1	6	7	519
(no lupin residue)	S2	8	9	554
(no nitrogen)	S3	9	13	807
	S1	5	55	9543
N-NH ₄	S2	5	75	55333
	S 3	6	120	185480
	S1	3	43	6837
N-NO ₃	S2	4	60	58051
	S3	5	79	20857
	S1	9	125	474
2% lupin residue	S2	14	176	541
	S3	20	256	656
	S1	11	89	422
4% lupin residue	S2	13	109	480
	S 3	16	167	523
2% lunin residue	S1	10	76	824
\pm N-NH.	S2	11	103	1684
1 11-11114	S3	15	140	2743
2% lupin residue	S1	9	60	722
+	S2	15	98	1462
N-NO ₃	S3	19	116	1894
4% lupin residue	S1	10	85	847
+	S2	11	93	1073
N-NH ₄	S3	15	142	1460
4% lupin residue	S1	10	82	776
+	S2	11	91	946
N-NO ₃	S3	17	125	1532
		I	$LSD (P \le 0.05)$)
N		0.79	7.2	ns
Residue		0.91	8.3	15546
Salinity		0.91	8.3	ns
N x residue		ns	12.5	ns
N x salinity		ns	12.5	ns
Residue x salinity		1.58	ns	ns
N x residue x salinity		ns	ns	ns

8.3.7 Nitrogen availability over time in a saline sandy loam soil amended with inorganic N and lupin residue

In general, extractable N increased with addition of inorganic N and/or lupin residue in the first 20 days at all salinity levels (Table 8.6). On day 20, N availability in the soil with 2% lupin residue only was as high as with 50 μ g NO₃-Ng⁻¹ soil. Nitrogen availability was 47, 44 and 41% higher in the soil amended with 4% than that with 2% lupin residue at S1, S2 and S3, respectively. Also, N availability in the NH₄⁺ amended soil was 50, 34 and 24% higher compared to the unamended soil and was 41, 32, and 19% greater with NO₃⁻ than with NH₄⁺ at S1, S2 and S3, respectively. At all salinity levels, addition of inorganic N did not significantly increase N availability in the lupin amended soil and even resulted in reduced N availability as compared to the addition of lupin residue only. On day 40, N availability increased by 39, 27 and 20% compared to the N availability on day 20 in the soil amended with 50 µg NH₄-N g⁻¹ soil at S1, S2 and S3, respectively.

Compared to NH_4^+ , N availability was approximately 30% higher with NO_3^- at all levels of salinity. In the soil with lupin residue only, the availability of N did not change significantly from day 20 to day 40. With the combined addition of inorganic N and lupin residue, N availability was approximately 2 fold higher on day 40 compared to day 20 with higher availability in the NO_3^- than in the NH_4^+ amended soil at all salinity treatments. The ratio NH_4 /NO₃ increased with increasing soil salinity.

8.3.8 Phosphorus availability over time in a saline sandy loam soil amended with inorganic N and lupin residue

Phosphorus availability was significantly affected by salinity and the addition of lupin residue and N (Table 8.5). Without lupin residue, the N form had no effect on P availability under saline conditions through out the experiment.

In all N and residues treatments, P availability decreased by about 18% with every increase of soil salinity. On day 15 at all salinity treatments, compared to the control, P availability was approximately 4.5 and 8 times higher with addition of 2 and 4% residue, respectively. On day 30, with addition of 2% residue P availability was approximately 37, 24 and 16% higher compared to day 15 at S1, S2 and S3, respectively. On day 45, P availability was twice as high than on day 30 in the soil amended with either 2 or 4% lupin residue with higher P concentration in the NO₃⁻ than in the NH₄⁺ amended soil at all salinity levels.

		Av	ailabl	e P	Availa	able N	NH ₄ /NO ₃	
Treatment	Salinity level		m	g kg ⁻¹	soil			
	-	d15	d30	d45	d20	d40	d20	d40
Control	S1	0.37	0.23	0.23	22	25	0.1	0.1
(no lupin residue)	S2	0.29	0.24	0.15	29	26	0.4	0.2
(no nitrogen)	S 3	0.20	0.22	0.16	29	27	0.9	0.1
	S1	0.40	0.28	0.11	33	46	0.1	0.2
N-NH ₄	S2	0.35	0.30	0.02	36	46	0.5	0.1
	S3	0.30	0.28	0.01	39	47	1.5	0.1
	S1	0.42	0.27	0.14	45	59	0.1	0.1
N-NO ₃	S2	0.31	0.28	0.02	54	60	0.3	0.1
	S3	0.23	0.30	0.06	55	61	0.7	0.3
	S1	1.62	2.23	4.34	48	56	4.3	0.1
2% lupin residue	S2	1.31	1.63	3.31	56	57	5.7	0.2
	S3	1.24	1.44	2.67	57	58	8.2	0.7
	S1	2.75	4.47	7.51	71	66	5.4	0.2
4% lupin residue	S2	2.42	3.44	6.89	79	74	5.8	0.5
_	S3	2.34	3.11	5.63	80	79	7.6	1.1
20/ Junin regidue	S1	1.77	2.05	4.78	37	70	1.2	0.3
1 N NU	S2	1.37	0.97	2.32	44	75	2.2	1.0
+ 11-11114	S3	1.22	0.62	1.44	45	77	4.7	1.6
2% lupin residue	S1	1.76	2.70	5.37	47	77	0.3	0.1
+	S2	1.39	1.46	3.38	57	79	0.8	0.2
N-NO ₃	S3	1.25	1.17	2.18	58	82	1.4	0.7
4% lupin residue	S1	2.50	3.96	8.50	44	94	2.3	0.6
+	S2	2.44	3.05	6.51	50	97	3.2	1.6
N-NH ₄	S3	2.31	2.64	4.63	52	103	7.5	1.7
4% lupin residue	S1	2.49	4.04	9.36	50	100	0.8	0.1
+	S2	2.41	3.45	7.28	58	105	1.3	0.4
N-NO ₃	S3	2.30	2.89	5.01	59	117	2.3	0.8
				LSI	$D(P \leq 0)$).05)	-	
N rate		ns	ns	ns	2.56	2.24	0.09	0.05
N form		ns	0.11	0.14	1.66	1.34	0.4	0.2
residue		0.03	0.14	0.21	2.95	2.58	0.5	ns
Salinity	0.03	0.14	0.21	2.95	2.58	0.5	ns	
N rate X residue	ns	ns	ns	3.62	ns	0.7	0.3	
N form X residue	ns	ns	0.24	2.88	2.32	0.8	0.3	
N rate X salinity	0.05	ns	0.36	ns	ns	0.8	0.4	
N form X salinity			ns	ns	ns	ns	0.8	0.5
residue X salinity	ns	0.24	0.36	ns	ns	1.0	ns	
N rate X residue X	ns	ns	ns	ns	ns	1.1	0.6	
N form X residue X	salinity	ns	ns	ns	ns	ns	1.3	0.8

Table 8.6 Phosphorus and N availability over time in a saline sandy loam soil amended with inorganic N at 50 mg kg⁻¹ soil as NO_3^- or NH_4^+ and/or lupin residues at 2 and 4%

8.4 Discussion

The results of the main experiment came in agreement with Li et al. (2006a) and Li et al. (2006b) showing that, compared to the non-amended soil, microbial activity and biomass were higher with lupin residue addition under saline conditions. Residues contain nutrients and the higher nutrient (C, N and P) amount added with 4%, compared to 2% residues, could be the major reason for the enhancement of the activity and the size of the microbial biomass. This is supported by the fact that compared to the un-amended soil, respiration rates were 15 and 30 times higher with addition of lupin residues at 2 and 4%, respectively (Figure 8.1). Also, the microbial biomass was almost 2 times higher in the soil with 4% than with 2% lupin residues (Figure 8.5).

It has been shown (Rietz and Haynes, 2003; Sardinha et al., 2003) and confirmed in Chapter 7 that soil respiration decreased with increasing soil salinity, because of increased soil osmotic pressure and/or ion accumulation in microbial cells. It has been also found that bacteria (Przybulewska and Krompiewska, 2005) and fungi (Wichern et al., 2006) are susceptible to saline conditions (10-20 dS m⁻¹). Statistical results in this study indicated that soil respiration and cumulative respiration were not negatively affected by salinity (Table 8.2). The highest salinity treatment in the present study was $EC_{1:5} 0.85 dS m^{-1}$, approximately EC 23.4 dS m⁻¹ in the soil solution. Salinity up to this level might have suppressed certain microbial groups, but not to an extent that affected soil respiration.

With addition of residues or inorganic N, respiration rate and cumulative respiration increased (Figures 8.2 and 8.3) indicating that nutrients were limiting to microbial activity. However, because of the lack of C, microorganisms did not greatly respond to N addition as high as to residue addition, which was reflected by the cumulative respiration (Figure 8.3) in the main experiment. In the preliminary experiment, under non-saline conditions, it is possible that decomposition rate of lupin residue resulted in sufficient N being available for microorganisms, and that therefore N application did not influence the microbial activity (Figure 8.1) which is in consistence with the findings of Bowden et al. (2004).

Under non-saline conditions, Davidson et al. (1991) and Drury et al. (1991) found that large portions (30%) of added NH_4^+ were unavailable due to abiotic processes such as NH_4^+ fixation. Vega-Jarquin et al. (2003) reported that approximately 25% of added NH_4^+ was lost through volatilization as NH_3 and the same amount fixed in the soil

matrix under saline-alkaline conditions. In the soil with or without residue, adsorption of a large portion of the added NH_4^+ to soil matrix and/or loss of NH_3 could have occurred explaining the similar soil respiration rates with and without inorganic N-NH₄. However, this can not be confirmed in this study, because N availability although lower in the soil with NH_4^+ than with NO_3^- , it was still high enough to affect microbial activity or biomass (Table 8.6).

Under saline conditions, N applied in NH_4^+ or NO_3^- increased microbial biomass, soil respiration and N availability in naturally saline alkaline soils amended with either glucose or organic materials (Luna-Guido and Dendooven, 2001; Conde et al., 2005; Dendooven et al., 2006). Also, in presence of glucose, compared to NO₃, microbial response was higher with NH₄⁺ under saline conditions (Azam and Ifzal, 2006). In contrast, the present study showed that microbial activity (Figure 8.2) and biomass (Figure 8.5) were greater in the soil with NO_3^- than with NH_4^+ . Also, the results shown in Table 8.6 indicated that NH₄/NO₃ ratio increased with increasing salinity which suggests that the activity of ammonifiers was higher than the nitrifiers. Therefore, in presence of lupin residue, soil respiration and cumulative respiration were higher in the soil with NO_3^- than with NH_4^+ in all salinity treatments. The most likely reason why microbial activity and biomass increased under saline conditions with N supply is that microorganisms were N limited in the soil with lupin residue, which might was rich in C, but contains concentrations of N that are less than optimal for building microbial biomass. Also, under saline conditions there could be a shift in the microbial community towards organisms that are more efficient in utilizing NO₃⁻ compared to NH_4^+ .

At every salinity treatment, microbial biomass increased with addition of lupin residue and inorganic N. However, with every increase in soil salinity there was a decrease of microbial biomass C, N and P with higher reduction rate in the unamended soil compared to the soil with lupin residue and inorganic N (Figure 8.5). This indicates that lupin residue and inorganic N have enhanced the salt tolerance of microorganisms, regardless the type of the microbial groups, and that tolerance was highest in presence of residue and N, particularly NO₃⁻. Compared to the soil with lupin residue only under saline conditions, C_{cum} to N_{mic} or P_{mic} decreased with addition of inorganic N, indicating that N addition increased the microbial biomass N more than the activity (respiration) (Table 8.5). The availability of N and P has been doubled by increasing residue addition to 2 and 4% (Table 8.6). The data shown in Figure 8.3 and Table 8.3 indicated that the availability of N and P was correlated to the changes of microbial biomass size over the time. The microbial biomass was highest in the first 2 weeks and may have been higher in the first days after residue addition. Later, the size of microbial biomass decreased and N and P availability increased until day 45. It can be said that microorganisms have immobilized high amounts of N and P released from residue, and that due to microbial turnover or community change N and P availability increased.

On day 15 with no salt added in the soil with 2% residue, corresponding to approximately 22 mg P kg⁻¹ soil, 19 mg P was accounted for as microbial biomass and 1.62 mg P was available in the soils solution. On day 45, approximately 9 mg P was in the microbial biomass and 4.34 mg P was available. The unaccounted amount of P between day 15 and day 45 after residue addition, presumably, was adsorbed to the clay in the soil via OH⁻ groups, which can be attached to Al⁺³ on the edges of clay lattices, and/or to layers of cations adsorbed to clay surfaces (Prafitt, 1978). Additionally, soil organic matter can bind P (White, 1980), resulting in stabilization of organic P complexes over time. These assumptions can not be confirmed in this experiment, but to a great extent, can be supported by the results of Chapter 4 (Figure 4.4). A similar decrease in P availability as S1 was observed, but to a higher magnitude, under the saline conditions (S2 and S3), but the decrease was greater, which is likely due to Ca-P precipitation in addition to P adsorption to the clay. In relation to microbial response to N form and residue addition rate, N and P availability were highest in the soil with 4% lupin residues and 50 mg NO₃-N kg⁻¹ soil.

8.5 Conclusions

The practical significance of this study is that with addition of lupin residue and inorganic N the salt tolerance of microorganisms will increase. As a consequence microorganisms are able to decompose added residues, and increase N and P availability under saline conditions. Higher addition rates of lupin residue increased this effect. Also, addition of at 50 mg N kg⁻¹ as NO_3^- , compared to NH_4^+ , can be recommended for greater effect on microbial salt tolerance, and N and P availability in the saline sandy loam soil with SAR 1 and neutral pH.

Chapter 9 – Separate and interactive effects of organic and inorganic

fertilizers on microbial biomass C and N and wheat growth in a saline

sandy loam soil

9.1 Introduction

Most of soils, particularly salt affected soils, are low in available P. Losses through erosion, leaching and fixation may reduce P availability, and therefore P fertilization to sustain crop productivity is inevitable. Salinity, coupled with low N and P availability, are soil constraints for crop production. The improvement of soil properties including N and P availability and the enhancement of plant N and P uptake through fertilization may enhance plant growth under saline conditions. The addition of N and P from organic sources such as plant residues can be used to replace large amounts of N and P from inorganic fertilizers and costs for farmers, and also reduce the environmental pollution often associated with the excessive application of inorganic N and P fertilizers.

While organic inputs often are poorer sources of N and P than inorganic fertilizers, they affect many soil properties controlling nutrient cycling, availability and crop production (Palm et al., 1997; Oladeji et al., 2006). Under non-saline conditions, the organic compounds released from decomposing materials may complex inorganic P, thereby moving it into organic pools, which can be taken up by plants after mineralization of the organic compounds (Khasawneh and Doll, 1978; Kpomblekou and Tabatabai, 1994). This may also be the case in saline soils, but has not been investigated.

It has been shown that addition of organic materials such as plant residues and animal manures to saline soils can increase not only the microbial activity (Liang et al., 2005), but also the availability of N, P and other elements (Lax et al., 1994; Walker and Bernal, 2008) and the growth of barely, sugar beet and wheat (Liang et al., 2005; Uyanoz et al., 2006; Walker and Bernal, 2008).

It has been reported that the amount of plant residues has a significant effect on crop productivity (Holland and Coleman, 1987; Adeoye, 1990; Tossah, 2000). With addition of residues with low P content (Chapter 7) and with increasing residue application rate (Chapter 8), P availability increased compared to the control.

However, P availability in the saline sandy loam soil with 4% lupin residues (Chapter 8) was still low, and based on the results of Chapters 5 and 6, probably insufficient for the enhancement of wheat growth in a saline sandy loam soil.

Nziguheba et al. (1998) found that, compared to the sole application of 15 kg P ha⁻¹ from either source, 7.5 kg from organic sources, Tithonia leaves containing 0.27% P, and 7.5 kg from Triple super phosphate (20% P), P availability and reduced P adsorption to clay minerals. Combined application of organic and inorganic fertilizers may increase residue decomposition (Chen et al., 2007), plant use efficiency of nutrients from both soil organic matter and inorganic fertilizer (Oue´draogo et al., 2006; Stark et al., 2007) and enhance wheat growth in the short term under saline conditions.

Application of P from organic and inorganic sources can be an option to enhance soil microbial activity and biomass P. Microbial biomass P can potentially contribute to plant P nutrition over time. Effects of P fertilizer on soil microorganisms have been reported to be either neutral (Amador and Jones, 1993), negative (Flanagan and Van Cleve, 1983), or positive (Manna et al., 2006; Raiesi and Ghollarata, 2006). Phosphorus fertilization may affect soil microbial respiration and biomass depending on soil P status. Intensive use of chemical fertilizers has been reported to depress microbial activity (Manna et al., 2006). Compared to the soil with a high P content (1473 mg kg⁻¹ soil), addition of 32.6 mM P to soils with low P content (231 mg kg⁻¹ soil) resulted in increased soil respiration (Amador and Jones, 1993). Triple super phosphate had no effect on soil respiration or on litter decomposition (Thirukkumaran and Parkinson, 2000). Also, application of P decreased the activities of phosphatase, sulfatase and urease (Haynes and Swift, 1988). Since litter decomposition is mediated by microorganisms, it is possible that P fertilization can increase litter decomposition, especially in the sandy loam soil with low P content and availability.

Addition of inorganic N was shown to play an important role in the decomposition of plant residues in non-saline soils (Carreiro et al., 2000; Moran et al., 2005; Sirulink et al., 2007) as well in naturally saline-alkaline, soils (Luna-Guido and Dendooven, 2001; Conde et al., 2005). For soils with adequate endogenous concentrations of N and P, adding further N and P did not stimulate respiration, and adding N and P with glucose did not enhance respiration more than adding glucose alone (Smith, 2005). In

Chapter 8, it was shown that microorganisms therefore can utilize inorganic N to supplement the nutrients in litter being decomposed. There is no information about cycling of nutrients, particularly P, and the combined application of N and P from organic and inorganic sources and how this can affect microbial biomass, nutrient availability and wheat growth in the saline sandy loam soil.

The present study was undertaken to evaluate the effect of the following treatments on N and P availability, microbial biomass and wheat growth in a saline sandy loam soil (1) P added in inorganic form (2) P added from organic and inorganic sources and (3) N added with inorganic and organic P.

9.2 Materials and methods

9.2.1 Preliminary experiment

A preliminary experiment was set up under to determine the effect of plant residues and their addition rate on wheat growth, and to examine the effect of N addition with plant residues on N and P availability, and wheat growth in a sandy loam soil under non-saline conditions. To compare the effect of lupin residue on wheat growth with the optimal P addition rate, a treatment of 60 mg P kg⁻¹ soil added as inorganic P was also included. This rate had achieved the highest shoot dry weight of wheat (cv Krichauff) under non- and saline conditions (Chapter 5). The sandy loam soil from Monarto (Table 3.1) was used.

9.2.1.1 Soil fertilization and wheat transplanting

The soil was dried, crushed, sieved, amended with macro- and micronutrients without N and P and then incubated as described in previous chapters. After incubation, the soil was treated with 0 and 50 mg N kg⁻¹ soil as $(NH_4)_2$.SO₄, 2% lupin residues, 4 % lupin residues and 60 mg P-KH₂PO₄ kg⁻¹ soil. The controls received no amendments or only inorganic N. Nitrogen form and rate of application were selected based on the results of Chapters 6 and 8 which indicated no significant difference between N forms at 50 mg N kg⁻¹ soil for wheat growth (Chapter 6) or for P availability in a sandy loam soils with lupin residues. The results also indicated higher shoot and root dry weights with addition of NH₄-N than with NO₃-N at the same rate under saline conditions (Chapter 6). Nitrogen addition rate was also selected based on the results of Chapters 7 and 8 which indicated, compared to the soil with no residue, 2% lupin residue increased N availability to approximately 50 and 300 mg N kg⁻¹ soil after 20

and 50-day-incubation, respectively. Therefore, for an experiment of 3-4 weeks duration, N availability with 50 mg N kg⁻¹ soil and with addition of 2% lupin residue would be similar. Addition of 2 and 4% lupin residues is equivalent to addition of approximately 22 and 44 mg inorganic P kg⁻¹ soil, respectively. Plastic pots were filled with 500 g of treated soil and immediately planted with germinated wheat (cv Krichauff) seeds (six seedlings thinned out to four after four days). The pots were placed in a glasshouse with day time temperature of 35 ± 3 °C and light intensity of 520-860 µmolS⁻¹m⁻². Plants were daily watered to 85% WHC.

9.2.1.2 Measurements

Available P and N concentrations in the soil were determined on day 20. On day 30, plant shoots were cut off at 0.5 cm from the soil surface, washed with running deionized water, placed in the drying oven for 48 hours at 70 °C and then weighed.

9.2.1.3 Design and statistical analysis

There were 2 application rates of N (no N and 50 mg N kg⁻¹ soil) in combination with 4 P treatments (control, 2% lupin residue, 4% lupin residue and 60 mg P kg⁻¹ soil) replicated four times and arranged in a completely randomised design. Analysis of variance was carried out using Genstat 8.

9.2.2 Main experiment

9.2.2.1 Soil fertilization and salinization and wheat transplanting

The sandy loam soil from Monarto was dried, amended with macro- and micronutrients less N and P, salinized with mixtures of NaCl and CaCl₂.2H₂O (Table 9.1) and incubated in the dark for 2 weeks at 25 ± 4 °C.

Table 9.1 Amounts of NaCl and CaCl₂.2H₂O added to a sandy loam soil and values of $EC_{1:5}$ and concentrations (m*M*) of salts kg⁻¹ and in the soil solution

	NaCl		Ca	Cl ₂ .2H	2 0	Osmolarity	EC _{1:5}	ECe	EC		
Salt	mg	тM	тM	mg	тM	тM	mosmoles L ⁻¹				
Level	kg ⁻¹ soil		soil	kg ⁻¹ soil		soil	in soil soln	dS m ⁻¹			
			soln			soln	III SOII SOIII				
S1	0	0	0	0	0	0	0	0.23	3.1	6.2	
S2	117	2.00	18.2	588	4.00	36.4	145	0.35	4.8	9.6	
S3	170	2.90	26.6	1255	8.53	77.6	286	0.51	7.0	14.0	

 $\mathrm{EC}_{1:5}$ was measured after incubation prior to establishment of the experiment under controlled conditions

Salinity treatments were selected based on the results of Chapters 6 and 8, which indicated that at these EC values, wheat and soil microorganisms can positively respond to addition of inorganic and organic amendments.

Using a cement mixer, the soil was treated with inorganic N added as $(NH_4)_2.SO_4$, inorganic P added as KH_2PO_4 and organic P added as lupin residue as shown (Table 9.2). The form of N and the addition rates of N and P were selected based on the results of Chapter 6, while the results of the preliminary experiment were considered for the addition rate of lupin residue. After addition of fertilizers, plastic pots with several holes (1 mm size) in the bottom were filled with 500 g of treated soil and then transferred to the glasshouse (37±3 °C). The experiment was conducted during summer and the daytime temperature was 37±3 °C.

Table 9.2 Combinations of organic and inorganic N and P fertilizers applied in a saline sandy loam soil for the evaluation of wheat and microbial biomass growth

	Treatments									
Ν	Р	Lupin residues								
(mg	kg ⁻¹ soil)	(%)								
	0									
50	20	0								
50	40	0								
	60									
	0									
0	20	2								
	40									
	0									
50	20	2								
	40									

From the preliminary experiment (Table 9.2), it appeared that wheat growth is very weak when sown on the same day of plant residue addition which could have been due to nutrient immobilization by the microbial biomass or infection of the seedlings by minor pathogens. Therefore this delayed planting was done to reduce the competition of microbes and optimize N and P availability to plants. The results of Chapters 7 and 8 indicated that microbial biomass is maximal in the first 10 to 14 days after addition or plant residues and that N and P availability increases

concomitantly with a decline in microbial biomass. Therefore, the soil was left unplanted for 14 days while maintaining the soil moisture at 85% WHC. Six germinated seeds of wheat (cv Krichauff) were transplanted 14 days after amendment addition and thinned out to four plants four days later. The time of transplanting was chosen because it was assumed they will rely on seed nutrient content in the first 7 to 10 days and then start to take up nutrients from the soil. After planting, the pots were watered daily to 85% WHC.

9.2.2.2 Measurements

Nitrogen and P availability and microbial biomass C and N were determined before the addition of amendments (Table 9.4) and on days 15 (wheat sowing day) and 30 after amendment addition. At harvest (30 days), plant shoots were cut off at 0.5 cm from the soil surface and roots were washed out of the soil and washed with running de-ionized water. Shoots and roots were dried and then weighed. Shoots were ground and digested for determination of total N, P, K, Ca, Fe, Mn, Cu and Zn.

9.2.2.3 Design and statistical analysis

The experiment was set up in a completely randomised design (3 salinity, 3 P and 2 N treatments) with three replicates. Three-way analysis of variance was carried out using Genstat 8.

9.3 Results

9.3.1 Preliminary experiment

The statistical analysis (Table 9.3) indicated that N and P availability and wheat growth were significantly affected by the addition rate of lupin residues. The results also indicated that addition of inorganic N with plant residues or inorganic P did not significantly affect P availability, but significantly increased N availability and shoot dry weight.

9.3.1.1 Availability of soil N and P and shoot dry weight as affected by organic and inorganic fertilizers in a non-saline sandy loam soil

In the non-saline soil, compared to the control soil, P availability was approximately 3 and 9 fold higher with lupin residues added at 2 and 4%, respectively. With addition of 60 mg inorganic $P kg^{-1}$ soil, P availability was up to 5 times higher than in

the soil with 4% lupin residues. Compared to the control, N availability was 5 and 11 times higher with addition of 2 and 4% lupin residues without inorganic N, whereas with inorganic N addition, N availability was about 2 and 3 fold higher with residues added at 2 and 4%, respectively. In presence of residues or inorganic P, shoot dry weight was higher with addition of inorganic N than without N. Compared to 2%, shoot dry weight was low with 4% lupin residue irrespective of inorganic N addition. In presence of inorganic N, shoot dry weight significantly decreased with increasing residue addition. With addition of inorganic N, compared to the control, shoot dry weight was approximately 34 and 182% greater with addition of 2% lupin residues and 60 mg P kg⁻¹ soil, respectively.

	Avail	Available P		able N	Shoot dry weight		
Treatment		mg kg	⁻¹ soil		g plant pot ⁻¹		
	- N	+ N	- N	+ N	- N	+ N	
Control	0.38	0.46	5	24	0.11	0.23	
2% lupin residue	1.08	1.14	24	44	0.26	0.31	
4% lupin residue	3.49	3.62	56	68	0.13	0.21	
60 mg P	17.06	11.98	5	20	0.20	0.65	
			LSD	$(P \leq 0.0)$	05)		
¹ Residue rate	0.	64	5.69		0.02		
N addition	n	IS	4.	65	0.02		
Residue rate X N addition	n	IS	n	IS	n	IS	
² Fertilizer type	2.	89	3.	49	0.02		
N addition	n	IS	4.93		0.01		
Fertilizer type X N addition	n	IS	ns		0.03		

Table 9.3 Phosphorus and N availability (mg kg⁻¹ soil) and shoot dry weight (g pot⁻¹) in a sandy loam soil amended with inorganic P or lupin residues at different rates with and without inorganic N

¹ Statistical analysis for the interaction between residue addition rate and N addition ² Statistical analysis for the effects of N addition with lupin residue or with inorganic P

9.3.2 Main experiment

9.3.2.1 Effects of N and P fertilization on plant dry weights and shoot nutrient concentration, N and P availability and microbial biomass C and N under saline conditions

The statistical analysis shown in Tables 9.4 and 9.5 indicated that P addition and salinity treatments significantly affected N and P availability, microbial biomass C and N and plant growth parameters. While the interaction between P addition rate and salinity was not significant for N availability, microbial biomass and shoot N and

P, it significantly affected P availability and shoot dry weight with addition of inorganic N with or without plant residues. Addition of inorganic N with P from inorganic fertilizer and lupin residue sources significantly increased N and P availability and microbial biomass C and N at all salinity treatments, but only after the first 2 weeks, and did not significantly affect shoot N and P concentrations or shoot and root dry weights. In presence of inorganic N, the interaction between salinity and P from inorganic fertilizer and plant residues had no significant effect on N and P availability and any plant growth parameters properties, but was significant for microbial biomass C and N under saline conditions.

9.3.2.1.1 Shoot and root dry weight and shoot N and P concentrations

The results shown in Table 9.4 indicated that, regardless the type of amendment, shoot and root dry weights and shoot P concentration significantly decreased with increasing soil salinity and that shoot N concentration was not affected. Shoot and root dry weights and shoot P concentration significantly increased with increasing addition of inorganic P with or without residues at all salinity treatments. In relation to shoot P concentration, total plant dry weight increased with increasing P addition up to 60 mg kg⁻¹ soil (Figure 9.1). Compared to the soil with lupin residue and inorganic P without inorganic N, N addition had no effect on shoot and root dry weights. At all P addition rates, lupin residue addition to inorganic P decreased shoot P concentration at all salinity treatments.



Figure 9.1 Relationship between total plant dry weight (g pot⁻¹) and P concentration (mg g⁻¹ DM) in the sandy loam amended with A.) inorganic P and N at 0, 20, 40 and 60 mg and 50 mg N kg⁻¹ soil, B.) lupin residue (2%) and inorganic P (0, 20 and 40 mg kg⁻¹ soil) and C.) plant residue (2%), inorganic N (50 mg N kg⁻¹ soil) and inorganic P (0,20 and 40 mg kg⁻¹ soil) at 3 salinity levels (S1, S2 and S3)

			D	N	Shoot	Root		
Treatment		Salinity	1	19	dry weight	dry weight		
Treatment		level	mg g	⁻¹ DM	g p	ot^{-1}		
		S1	1.45	28	0.23	0.17		
P0 + N50	T1	S2	1.35	26	0.18	0.14		
		S3	1.26	28	0.15	0.12		
		S1	4.08	33	0.51	0.24		
P20 + N50	T2	S2	3.55	34	0.30	0.19		
		S3	2.69	34	0.24	0.14		
		S1	5.47	32	0.59	0.34		
P40 + N50	T3	S2	5.15	31	0.43	0.22		
		S3	4.39	33	0.34	0.17		
		S1	6.98	31	0.66	0.38		
P60 + N50	T4	S2	6.32	33	0.48	0.25		
		S3	5.95	31	0.40	0.21		
]	$LSD \ (P \le 0.05)$	5)		
P rate			0.39	2.25	0.02	0.02		
Salinity			0.34	ns	0.01	0.02		
P rate X salinity			0.41	ns	0.03	ns		
		S 1	2.62	34	0.23	0.17		
2% O.M +P0	T5	S2	1.98	31	0.18	0.14		
		S3	1.52	37	0.14	0.11		
		S1	3.53	36	0.36	0.21		
2% O.M + P20	T6	S2	3.28	37	0.31	0.17		
		S3	2.79	39	0.22	0.13		
		S1	4.29	39	0.42	0.26		
2% O.M + P40	T7	S2	3.89	37	0.35	0.20		
		S3	3.51	39	0.28	0.17		
			$LSD (P \le 0.05)$					
P rate			0.41	ns	0.02	0.01		
Salinity			0.09	ns	0.02	0.01		
P rate X salinity	T	1	ns	ns	0.04	ns		
		S1	1.90	38	0.31	0.14		
2% O.M + N50 + P0	T8	S2	1.68	39	0.18	0.11		
		S3	1.46	40	0.15	0.05		
		S 1	3.80	38	0.40	0.17		
2% O.M + N50 + P20	T9	S2	2.67	39	0.27	0.14		
		S3	2.19	41	0.23	0.09		
		S1	4.96	39	0.44	0.21		
2% O.M + N50 + P40	T10	S2	3.43	41	0.36	0.18		
		S 3	3.35	41	0.26	0.14		
		0.77]	$LSD (P \le 0.05)$	5)			
P rate			0.32	ns	0.02	0.01		
Salinity			0.32	ns	0.02	0.01		
P rate X salinity			ns	ns	ns	ns		

9.3.2.1.2 Shoot K, Ca, Fe, Mn, Zn and Cu concentrations

Concentrations of K, Ca, Fe, Mn, Zn and Cu in the shoots as affected by P from inorganic and/or lupin residues added at different rates with and without inorganic N in a saline sandy loam are shown in Table 9.5.

Generally, the concentrations of all nutrients, except Fe, significantly decreased with increasing salinity in soil with or without lupin residues. Also, compared to the controls, addition of inorganic P significantly increased the concentration of all nutrients in the shoot.

Table 9.5 Concentrations K, Ca (mg g^{-1} DW) Fe, Mn, Zn and Cu (mg kg^{-1} DW) in shoots as affected by P from inorganic and/or lupin residues added at different rates with and without inorganic N in a sandy loam with three salinity levels

Treatment		Salinity	K	Ca	Fe	Mn	Zn	Cu
		level	mg g	¹ DM		mg kg	5^{-1} DN	1
		S1	23	2.1	256	76	22	5.7
P0 + N50	T1	S2	25	2.5	265	66	17	5.1
		S 3	21	2.8	262	59	14	4.8
		S1	42	3.2	313	96	36	9.3
P20 + N50	T2	S2	35	3.5	317	78	27	7.3
		S 3	29	4.0	318	69	21	6.5
		S1	51	3.7	388	107	43	11.8
P40 + N50	T3	S2	41	4.3	361	88	32	8.3
		S 3	35	4.9	332	78	27	7.6
		S1	55	3.5	383	118	44	11.6
P60 + N50	T4	S2	46	4.2	354	97	36	9.4
		S3	40	5.0	329	85	32	5.2
				LS	SD (P	≤ 0.05	5)	
P rate			0.17	1.7	21	3.1	2.0	0.38
Salinity			0.15	1.5	ns	2.7	1.7	0.33
P rate X salinity			ns	3.0	ns	ns	ns	0.67
		S1	26	3.1	375	77	25	4.3
2% O.M +P0	T5	S2	20	3.5	365	47	21	4.1
		S 3	19	4.0	359	39	18	4.1
		S1	39	3.4	384	88	33	6.4
2% O.M + P20	T6	S2	30	3.9	368	61	26	5.5
		S 3	26	4.6	370	52	23	5.1
		S1	43	4.1	389	100	38	8.9
2% O.M + P40	T7	S2	35	4.5	379	75	32	7.6
		S3	30	5.0	376	64	27	6.8
		L	SD (P	≤ 0.05	5)			
P rate			0.12	1.3	ns	3.5	1.6	0.38
Salinity			0.12	1.3	ns	3.5	1.6	0.38
P rate X salinity			ns	ns	ns	ns	ns	ns

At each addition rate of P, shoot K, Mn, Zn and Cu concentrations were greater in the soil with P from inorganic source only (T1-T4) than in the soil with P from inorganic fertilizer and lupin residue sources (T5-T7).

9.3.2.1.3 Nitrogen and P availability over time

Availability of N and P (mg kg⁻¹ soil) in a saline sandy loam soil on days 15 and 30 after addition of P from inorganic and/or lupin residues with and without inorganic N are shown in Table 9.6. Before addition of N and P from inorganic fertilizer or residue sources, concentrations of soil available P were 2.11, 1.92 and 1.35 mg and of available N 13, 11 and 12 mg kg⁻¹ soil at S1, S2 and S3, respectively.

With increasing salinity, P availability decreased at all treatments. With addition of inorganic N, the availability of N was not affected by soil salinity, whereas with no inorganic N addition (T5-T7), salinity decreased N availability.

On day 15, compared to before amendment addition, N availability was higher and P availability was lower with addition of only 50 mg N kg⁻¹ soil. Compared to the control, addition of 20, 40 and 60 mg P kg⁻¹ soil significantly increased P availability 10, 20 and 30 fold, respectively, whereas it had no effect on N availability. At every rate of P addition, P availability was higher and N availability was approximately 30-50% lower with addition of P from fertilizer (T2-T4), compared to fertilizer and residues (T5-T7).

On day 15, compared to the control (T5), P availability was 2 and 3 fold higher with addition of 20 and 40 mg P kg⁻¹ soil, respectively. Also, compared to the control (T7), N availability decreased with addition of inorganic P (T6-T7). Phosphorus availability did not significantly change from day 15 to day 30 in T5-T7, but decreased in T1-T4. Addition of inorganic N to residues increased N and P availability at all rates of inorganic P.

Treatment		Salinity level	Available P		Available N Microbial C				Microbial N	
			mg kg ⁻¹ soil							
			D15	D30	D15	D30	D15	D30	D15	D30
P0 + N50	T1	S1	0.55	0.58	37	7	396	257	27	16
		S2	0.40	0.81	34	8	326	251	22	19
		S 3	0.32	0.78	33	10	230	152	15	23
P20 + N50	T2	S1	5.05	3.99	34	5	456	217	39	20
		S2	4.84	3.37	36	6	365	249	29	22
		S3	4.43	2.47	40	8	299	318	21	26
P40 + N50	Т3	S1	11.91	7.24	27	5	592	284	51	24
		S2	10.90	6.82	30	6	419	325	39	26
		S 3	10.47	5.08	35	7	331	351	27	32
P60 + N50	T4	S1	16.69	12.55	35	6	752	321	62	32
		S2	14.11	10.80	35	7	598	359	52	34
		S 3	12.92	9.39	29	7	517	376	39	37
			LSD ($P \le 0.05$)							
P rate Salinity P rate X salinity			0.9	1.1	ns	1.09	39	20	4.02	1.55
			0.3	0.45	ns	0.94	33	23	3.48	1.34
			1.2	1.8	ns	ns	41	36	ns	1.60
2% O.M + P0	Т5	S1	4.30	4.91	93	33	822	369	79	54
		S2	3.64	4.47	81	23	703	448	68	62
		S3	3.39	3.99	73	18	588	544	54	76
2% O.M + P20	T6	S1	7.78	8.93	72	24	933	322	90	39
		S2	7.23	7.93	71	18	833	352	78	51
		S3	7.04	7.61	62	14	727	426	66	60
2% O.M + P40	Τ7	S1	13.27	12.89	57	19	1025	255	99	31
		S2	11.15	11.55	54	13	933	297	83	39
		S3	10.04	10.13	47	11	798	346	73	49
P rate Salinity P rate X salinity		$LSD (P \le 0.05)$								
			1.0	1.1	3.77	3.2	36	22	4.56	4.27
			0.2	1.1	3.77	3.2	30	22	4.56	4.27
		C 1	118	5.42	06	25	42	224	112	115
2% O.M + N50 + P0	Т8	S1 S2	4.10	3.45	90	20	999	272	04	44 50
		52 52	2.19	4.90	00	32 20	900	372	94 75	50
		S5 61	5.10 9.42	4.30	01	29	014	430	120	21
2% O.M + N50 + P20	Т9	51	8.43 7.07	10.20	84 82	32 20	114/	280	120	20
		52 52	7.97	10.39	82 77	29	10/1	327	102	50 51
2% O.M + N50 + P40	T10	55	12.62	0.23	77	27	928	399	91	24
		51	12.03	13.28	/6	25	1223	226	130	24
		52 52	11.01	12.01	15	23	1130	245	117	30
		33	11.12	11.1/	05		1020	288	102	39
P rate			1.1	1.1	5.47	2.3	<u>- 0.05)</u> 41	16	6.21	3.18
Salinity			0.3	1.1	ns	ns	41	16	6.21	3.18
P rate X salinity			ns	ns	ns	ns	55	33	7.11	5.23

Table 9.6 Availability of N and P (mg kg⁻¹ soil) and microbial biomass C and N (mg kg⁻¹ soil) in a saline sandy loam soil amended with P from inorganic and/or lupin residues added at different rates with and without inorganic N
9.3.2.1.4 Microbial biomass C and N over time

Before addition of N and P from inorganic fertilizer or residue sources, concentrations of microbial biomass C were 172, 159 and 131 mg and of microbial biomass N were 7, 6 and 9 mg kg⁻¹ soil at S1, S2 and S3, respectively. The results shown in Table 9.6 indicated that microbial biomass C and N were higher on day 15 than on day 30 and higher on both sampling dates compared to before N and P fertilizers and/or residue addition. In the unamended and amended soils, with every increase in soil salinity level, microbial biomass decreased by approximately 90-130 mg C and 10-12 mg N kg⁻¹ soil.

On day 15, compared to the control (T1), microbial biomass was 50-60 mg C and 6-10 mg N higher with addition of 20 mg P and 50 mg N kg⁻¹ soil (T2). Microbial biomass C and N increased with increasing addition of inorganic P and were highest with addition of 60 mg P kg⁻¹ soil. At every addition rate of inorganic P, microbial biomass C was 1.5 time greater and microbial biomass N was 50-60% higher in presence of lupin residue at all salinity treatments. Addition of inorganic N to lupin residues increased microbial biomass C and N irrespective of addition rates of inorganic P.

9.4 Discussion

In agreement with other studies (Lax et al., 1994; Walker and Bernal, 2008), the results shown in Table 9.3 (preliminary experiment) indicated that, compared to the control soil, residue addition increased N and P availability. This effect increased with increasing addition of plant residues in the soil. This, as discussed in Chapter 8, could be due to the release of greater amounts of soluble and labile C, N and P added with 4% compared to 2% residue. It was shown in Chapter 8 that microbial biomass C, N and P increased with increasing addition of lupin residues at all salinity treatments. Compared to 2%, 4% residue can result in higher microbial activity, and possibly greater immobilization of N and P, reducing wheat uptake of N and P by wheat. The findings of McLaughlin and Alston (1986) indicated that in presence of plant residues, micro-organisms depleted the soil available P causing a reduction of wheat uptake of P and hence dry weight. In the preliminary experiment, although available concentrations of N and P were rather low, N and P availability were higher in the soil with 4% compared to 2% residue. Therefore it is unlikely that the reduced

plant growth at 4% residues was due to N or P limitation. Other factors such as pathogen infestation or micronutrient deficiency can affect root growth and nutrient uptake in presence of residues.

In this study, addition of 50 mg N kg⁻¹ soil and 2 or 4% residue or 60 mg inorganic P kg⁻¹ soil significantly increased shoot dry weight under non-saline conditions (Table 9.3). This suggests that exogenous inorganic N can supplement the released nutrients from residues and enhance plant N uptake and growth.

Shoot dry weight was higher in the soil with 60 mg P than with 2% and 4% lupin residues irrespective of inorganic N addition. This might be due to the lower availability of P in presence of lupin residues compared to P from inorganic source (Table 9.3), which possibly resulted in lower P uptake and hence plant growth. This is in agreement with the findings of Ghoshal (1975) which indicated that addition of a C source to the soil decreased uptake of P by rye plants. Most probably this was due to enhanced microbial activity and thus microbial P immobilization or slower release of P from residues. It should also be noted that in this experiment inorganic P was added at a higher rate (60 mg) compared to residue P rates.

Addition of fertilizer P to a maximum of 44 kg P ha⁻¹, with or without farmyard manure, increased the phosphatase activity and microbial biomass P (Manna et al., 2006). In the present study (main experiment), with or without lupin residues, microbial biomass C and N were higher at high than at low addition rate of inorganic P, and higher in presence of residues at all salinity treatments (Table 9.4). Amador and Jones (1993) reported that, compared to a soil with high P content (1473 mg kg⁻¹ soil), addition of approximately 32 m*M* enhanced the soil respiration in the soil with low P content (231 mg kg⁻¹ soil). The sandy loam soil used in this study had total content of 148 mg P kg⁻¹ soil with low availability of P and N. Compared to the soil with P from lupin residue only (T5; control), N availability decreased with increasing addition of inorganic P (Table 9.4). Also, at every addition rate of inorganic P, N availability was 30-50% higher in soil with residue (T5-T7), compared to the soil without lupin residues (T2-T4) at all salinity treatments. This suggests that P was limiting for microbial growth, residue decomposition and nutrient release in this soil irrespective of salinity level.

At all salinity treatments, in presence of lupin residues, with addition of inorganic P, N availability decreased and microbial biomass increased (Table 9.5). The increased biomass N is probably the result of improved microbial growth in presence of lupin residues. The fact that N availability decreased with every increase in inorganic P addition in the soil with lupin residue and 50 mg inorganic N (Table 9.4) indicates that P is limiting for microbial growth and N immobilization.

It was reported that P uptake, content and concentration decreased in plants with increasing salinity (Champagnol, 1979; Grattan and Grieve, 1992). However, in other studies (Manchanda et al., 1982; Al-Karaki, 1997; Abid et al., 2002), as well as in this study under saline conditions up to EC_e 7 dS m⁻¹, P fertilization up to 60 mg P kg⁻¹ soils increased plant P uptake (Table 9.4) and positively affected plant growth (Figure 9.1). It should be stressed that plants may respond positively to P fertilization when no other nutrients or salinity stress are limiting plant growth under saline conditions. In this study under saline conditions, shoot P concentration was at deficient (1.3 mg g⁻¹ DM) level at T1 (control), marginal (3 mg g⁻¹ DM) level at T2 and sufficient (4-5 mg g⁻¹ DM) level at T3 and T4, whereas shoot N concentration was sufficient at T1-T4 as indicated in the work of Elliott et al. (1997). Under saline conditions, increased shoot P concentration may have contributed to the growth stimulation by increasing water-use efficiency, stomatal conductance (Brück et al., 2000) and photosynthesis (Hu and Schmidhalter, 2005).

Although P availability with inorganic P was not affected by residue addition, plant P concentration was lower with residues (Table 9.4). This suggests that in presence of residues, plants could not access the available P to a similar extent as with inorganic P only. This can be due to the lower root growth in presence of residues (Table 9.4).

Although organic matter application can increase the total concentration of micronutrients in the soil (Sloan et al., 1998) availability of Cu, Zn and Mn decreased (McGrath et al., 1988). This was attributed to sorption (Brar and Sekhon, 1976) and chelation (DeRemer and Smith, 1964) of micronutrients by organic matter. Reduced availability of Zn and Mn was also attributed to microbial immobilization (Zamani et al., 1984; Zamani et al., 1985). The results shown in Tables 9.4 and 9.5 indicated that addition of lupin residues (T5-T7) decreased shoot P, K, Mn, Zn and Cu compared to inorganic P fertilization without residue (T2-T4) at all salinity treatments. Also, in presence of lupin residue, concentrations of Mn and Cu were at deficient levels in the shoots. In addition to micronutrient immobilization after residue addition, the lower concentration of micronutrients could also be due to the lower root growth in presence of residues.

9.5 Conclusions

Addition of inorganic P increased P availability and microbial biomass in the saline soil, even in presence of lupin residues. The combined application of P from inorganic fertilizer (40 mg kg⁻¹) and lupin residue (2%) sources is beneficial for the enhancement of soil microbial biomass, and can improve the soil N and P availability and wheat growth in the sandy loam with up to EC_e 7 dS m⁻¹. However, it is not as effective as inorganic P fertilization alone because residue addition inhibited root growth and reduced micronutrient and P uptake. Addition of inorganic N to P from inorganic and lupin residue improved the chemical and microbial properties of salt affected sandy loam soils, but had no significant effect on wheat growth.

Chapter 10 – General discussion, conclusions and suggestions for future work

10.1 General discussion

The experiments conducted in this study were related to wheat growth in the saline non-alkaline or sodic soils. They aimed at assessing salinity threshold of wheat with optimization of N and P fertilization, and establishing an appropriate method for enhancing the fertility, nutrient availability and microbial activity of saline soils with minimization of confounding effects of increasing pH and changes in soil structure stability that accompanied increases in salinity in many other studies.

The application of P fertilizers may increase wheat growth under saline conditions, when the soil osmotic stress is not severe. In studies on P availability in salt affected soils, P availability decreased with increasing concentration of salts of Ca^{2+} , not Na^+ , and was lower in the sandy loam than in the sandy soil (Chapter 4). Regardless the type of salt, P might was sorbed with Fe³⁺ and Al³⁺ hydroxides (White, 1980). Also, organic matter content, although low, could have chelated P (Nziguheba et al., 1998) reducing its availability more in the sandy loam than in the sandy soil. With increasing soil salinity, soil ionic strength may have increased P sorption thus reduced P availability (Ryden et al., 1977). In the soil with Ca^{2+} , compared to Na⁺ salts, P availability may have decreased due to precipitation as Ca-P (Prafitt, 1978). Clay particles may also have adsorbed P via OH groups attached to Al³⁺ on the edges of clay lattices or by replacing hydroxyl groups on the edges of layers (McPharlin et al., 1990). Saline soils containing clay minerals, Ca^{2+} salts and other minerals might fix and precipitate P fertilizers into less available forms.

In the studies on P fertilization in saline soils, P availability increased with increasing addition rate of P (Chapter 4). The efficient use of P fertilizers in saline soils may increase P uptake and wheat growth (Abid et al., 2002; Mehdi et al., 2003), but the effect depends on the level of soil salinity, and whether other nutrients are limiting plant growth or not. In the present study, salinity threshold and effects of P fertilization and N application rate and form were determined for only one wheat variety (Krichauff) in a P-deficient sandy loam soil. Wheat growth decreased with increasing salinity, but with increasing N and P availability through fertilization shoot N and P concentrations increased in the saline sandy loam soil (Chapter 5). The study also showed that shoot and root dry weights increased as a result of increasing

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application of P fertilizers at all salinity treatments. Phosphorus fertilization increased the salt tolerance of wheat, and plant dry weight was highest with application of 60 mg P kg⁻¹ soil in the sandy loam with up to EC_e 9.2 dS m⁻¹, but SAR was only 1.

The form in which N is applied to salt-stressed plant may influence nutrient availability (Warren and Adams, 2002) and plant-nutrient-salinity relationships (Martinez and Cerda, 1989). In studies on N fertilization in saline soils (Chapters 5 and 6), N availability was not affected by salinity, but increasing N fertilization increased salinity. At all N addition rates, soil salinity was highest with NO₃⁻. The growth of wheat was higher with addition of N as NH₄⁺, compared to NO₃⁻, at all application rates of N. Nitrate added at rates higher than 50 mg N kg⁻¹ soil decreased plant growth and shoot concentrations of P, Fe, Mn, and Cu even under non-saline conditions. On the other hand, shoot and root dry weights were highest at 100 mg N as NH₄⁺ or NH₄NO₃ under non- and saline conditions. This suggests that N should not be added to saline soil as NO₃⁻ at rates greater than 50 mg kg⁻¹ soil, and that NH₄⁺ and NH₄NO₃ are better N sources, and can be applied at 100 mg N kg⁻¹ soil for wheat in the sandy loam with up to EC_e 9.3 dS m⁻¹.

Application of plant residues may increase microbial activity and N and P availability and wheat growth in salt affected soils. In the studies with different residue types (Chapter 7), microbial activity decreased with increasing soil salinity, but was not inhibited to up to $EC_{1:5}$ 2.80 dS m⁻¹. Residue addition increased microbial activity and thus biomass. This suggests that addition of plant residues can enhance the salt tolerance of microorganisms; thus residue decomposition, microbial turnover and N and P transformation can proceed up to $EC_{1:5}$ 2.8 dS m⁻¹ increasing the availability of N and P in the saline soils. In addition, this highlights the high salt tolerance of microorganisms compared to plants. In this study, microbial biomass was greater in the sandy than in the sandy loam soil with the same salinity treatments. It may be that nutrients attached to added residues were accessed by microorganisms to a greater extent in the sand than in the sandy loam. Therefore it can be suggested that microbial biomass is higher in the sandy than in the sandy loam soil, which can increase nutrient release from added residues and increase N and P availability.

Microbial activity and biomass and N and P availability were higher at high than at low residue application rates, and were much higher with N addition to residues under saline conditions (Chapter 8). Also, microbial activity and biomass were higher with addition of 50 mg N kg⁻¹ as NO_3^- , compared to NH_4^+ , in presence of residue under saline conditions. In presence of residues, microorganisms might have been N limited under saline conditions. Addition of inorganic N to residues might have enhanced the salt tolerance of microorganisms and increased their activity to decompose added plant residues, thus microbial biomass and N and P availability increased. This suggests that in contrast to plants NO_3^- application is efficient for microbial use to increase residue decomposition and nutrient availability in the sandy loam under saline conditions.

Under non-saline conditions, shoot and root dry weights of wheat (cv Krichauff) were greater at 2% compared to 4% lupin residues (Chapter 9). The reduced plant growth at higher residue addition rates could be due to greater N and P uptake into the microbial biomass. The experiment with P from inorganic fertilizer or residues showed that while residue addition increases microbial activity and biomass, they are not as effective as P from fertilizer source for plants (Chapter 9). This could be due to reduction of root growth or micronutrient deficiencies in presence of residues.

10.2 Conclusions

This study confirmed previous studies, but also provided new information about nutrient cycling and plant growth in saline soils. In agreement with the earlier studies, the experiments showed that P and N addition can increase wheat growth in saline soils, but only up to a certain salinity level above which the negative effect of salinity on plant growth is dominant. For the wheat cultivar and the soils used here this threshold level was $EC_{1:5}$ 0.67 dS m⁻¹, equivalent to EC_e 9.38. Compared to wheat, microbial growth and activity were more tolerant to salinity. For the first time, this study showed N and P release and uptake into the microbial biomass in saline soils with plant residues or inorganic P. The results showed that decomposition of plant residues showed that plant residues are not as efficient in increasing plant growth and nutrient uptake as inorganic P. Moreover, plant residue addition decreased micronutrients uptake by plants. These results suggest that plant residues can not replace inorganic P fertilizers in saline sandy loam soil. However, plant residues and

the resulting increased microbial biomass could act as long-term nutrient supply pool and may have other beneficial effects such as improving soil structural stability.

10.3 Suggestions for future work

The present study addressed several aspects of N and P fertilizer management, microbial activity and wheat growth in the saline-non sodic sandy loam soil. Only one wheat variety (Krichauff) was used. Possibly, with a large number of wheat varieties, greater differences in response to N and P fertilization could have been identified. It should however be mentioned that one wheat variety was used to determine the hypothesis that with increasing P fertilization, wheat growth could be improved when plants are not deficient for nutrients, particularly N, in saline soils with no sodicity or pH problems. Increasing P fertilization was significant for the examined variety in the tested soil. Although fertilizers have contributed to the soil EC, increasing P fertilization to rates higher than 60 mg kg⁻¹ soil may enhance the growth of the tested wheat variety under saline conditions. Salinity decreased P availability. Phosphorus utilization by other wheat varieties may be greater than Krichauff at the rates examined in this study. Application of N at rates higher than 100 mg N kg⁻¹ soil increased soil salinity and negatively affected wheat growth. Wheat varieties may respond differently to the form and the application rate of N under saline conditions. Also, plant may respond differently according to the growth stage. In addition, N at higher rates may have a positive effect on wheat growth if applied differently, for example in split application.

Inorganic N was applied with lupin residue, which had low C/N ratio, and both increased microbial activity and biomass. Inorganic N application might be beneficial for decomposition of plant residues with high C/N ratio under saline conditions.

Short-term experiments were conducted to study plant residue effect on nutrient availability and wheat growth. Residue addition at investigated application rates supplied wheat with its N requirements. Although high compared to low application rate of residues increased N and P availability, it had a negative effect on wheat growth. Wheat may differently respond to residue addition if planted at later stages of residue decomposition. Plant residue used in this study had a low P content. Therefore application of inorganic P with residues was necessary to increase P availability and wheat growth in the saline soils. Plant residues with higher P content could be added

at similar application rates to achieve the same amount of P added. With less carbon added, the competition by the microbial biomass for nutrients would decrease, which could increase plant nutrient availability. However, how much P was taken from residues by plants or microorganisms under saline conditions could not identified. To asses how much plant residue or inorganic P fertilizer is taken up by plants, dual labelling experiments with ³²P-labelled inorganic P and ³³P-labelled residues could be conducted.

More investigations are needed for efficient management of P fertilization and wheat growth in saline non-sodic soils. Future studies could focus on the following:

- Phosphorus utilization efficiency by different wheat varieties at low and high application rates of P in saline soils with different texture,
- Effect of split application of ammonium fertilizers at different rates on wheat growth under saline conditions,
- Wheat response to foliar application of N under saline conditions,
- Comparison between plant residues with high and low P content on microbial activity, P availability and wheat growth in saline soils with different texture,
- Decomposition of plant residues with high C/N ratio as affected by inorganic N addition in saline soils,
- Effect of frequent applications of P-rich plant residues on P and micronutrients availability and wheat growth in salt affected P-deficient soils,
- Effect of planting time on wheat growth in saline soils with residues low or high P content,
- Identification of the source of P in plants by using isotopically labelled fertilizers.

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Plate 12.1 Wheat growth as affected by KNO_3 addition at 50, 100 and 200 mg N kg⁻¹ soil under non-saline conditions (Chapter 5)



Plate 12.2 Wheat growth as affected by KNO_3 added at 50 mg N and KH_2PO_4 added at 0, 30 and 60 mg P kg⁻¹ soil under saline conditions (Chapter 5)



Plate 12.3 Wheat growth as affected by KNO_3 added at 50 mg N and KH_2PO_4 added at 60 mg P kg⁻¹ soil in sandy loam soil with five salinity levels denoted S1, S2, S3, S4 and S5 (Chapter 5)



Plate 12.4 Wheat growth as affected by $(NH_4)_2$.SO₄ added at 0, 50, 100 and 200 mg N kg⁻¹ soil under saline conditions (Chapter 6)



Plate 12.5 Wheat growth as affected by $(NH_4)_2$.SO₄, KNO₃ and NH₄NO₃ added at 100 mg N kg⁻¹ soil in the saline sandy loam (Chapter 6)



Plate 12.6 Measurement of soil respiration (Chapters 7 and 8)


Plate 12.7 Wheat growth as affected by P from inorganic fertilizer (P_i) and/or plant residues (P_r) added at 50 mf N and 60 mg kg⁻¹ soil with and without 50 mg N as (NH_4)₂.SO₄ in a saline sandy loam soil (Chapter 9)