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Comparison of breeding strategies to improve salt tolerance of wheat (Triticum aestivum L.).

Ahsan, Muhammad.

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Comparison of breeding strategies to improve salt tolerance of wheat (*Triticum aestivum* L.)

Muhammad Ahsan

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Abstract

The research work reported in this thesis studied the effects of selfing and selection, and the effects of cross breeding on some salt-tolerance traits, yield and yield components of spring wheat under saline conditions. The study included some salt-sensitive and salt-tolerant spring wheat varieties. A series of pot experiments under both soil and hydroponic conditions was conducted under glasshouse conditions in the UK. Selections made from within varieties indicated the presence of intra-varietal variation under saline conditions. The results suggested that more salt-tolerant and high yielding lines can be selected from within existing varieties and by successive selfing it is possible to have more salt-tolerant pure lines. These can be cultivated as salt-tolerant varieties or can be manipulated further in breeding programmes. Few significant relationships were found between the traits studied in soil culture and hydroponic culture. These results suggest that tolerance of soil salinity and hydroponics salinity are independent and varieties evolved or selected under hydroponics might behave differently under soil salinity. Ion contents changed with age in the fourth leaf. The results showed that salt-tolerant varieties had low leaf Na^+, Cl^-, high K^+ content and high K^+/Na^+ ratio. They also had high yield under saline conditions. Low Na^+, Low Cl^-, high K^+, high K^+/Na^+ ratio were associated with high yield. Fewer infertile spikelets per spike, more fertile spikelets per spike, more grains per plant, more grains per spike, more grain weight per spike, more main tiller height and more straw weight per plant were also associated with high yield. A salt-tolerant variety was crossed with a high yielding variety to study the biometrical genetics of salt-tolerance. In a generation means analysis additive and dominance genetic effects were found to be involved in the inheritance of Na^+, K^+, Cl^- contents, K^+/Na^+ ratio, main tiller height, straw weight per plant, fertile spikelets per spike, number of grains per plant, grain weight per plant and grain weight per spike. This suggests that inheritance of these traits is relatively simple. In addition to additive and dominance effects, additive × additive genetic effects also involved in the inheritance of number of infertile spikelets per spike and number of grains per spike. However additive, dominance, and dominance × dominance genetic effects were also found to involved in the inheritance of spikes per plant and average grain weight per plant. In a generation variance analysis, it was shown that all these traits are mainly controlled by additive genetic effects. These results suggest that these traits may be easy to manipulate in a breeding programme. The interrelationships and similar gene action of these traits suggest that they might be controlled by some common genes.

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TO GOOD WISHES OF MY LATE FATHER, SUB. MUHAMMAD SIDDIQUE,

FOR MY SUCCESS IN EVERY SPHERE OF LIFE.

Contents

CHAPTER 1

INTRODUCTION

Levitt (1972, 1980) developed a definition for biological stress from physical science. Physical stress is any force applied to an object (for example, a steel bar); strain is the change in the object's dimensions (for example, bending) caused by the stress. He suggested that biological stress is any change in environmental conditions that might reduce or adversely change a plant's normal functions. Biological strain is the reduced or changed function. He also defined elastic biological strain as those changes in an organism's function that return to the optimal level when conditions are again optimum (that is, when the biological stress has been removed). If the function does not return to normal, the organism is said to exhibit plastic biological strain. Plant physiologists have emphasized such plastic strains as those caused by the stresses of frost, high temperature, limited water, or high salt concentrations. Elastic strain in plants includes stresses such as reduced photosynthesis in response to low light as it returns to normal with the return of high light levels.

Larcher (1987) noted that we can keep this distinction clearly in mind if we use certain modifiers for the term stress: stress factor = Levitt's stress and stress response = biological strain. Larcher pointed out that Levitt's concept works best when we are dealing with individual stress factors, although stress

responses are typically caused by more than one stress factor (Larcher *et al.*, 1990). For example there are several factors affecting stand establishment of which poor seedling emergence, soil crusts, poor seedling vigour, high temperature, salinity and drought are important (Wilson *et al.*, 1982; Peacock, 1982; Maiti *et al.*, 1984; Maiti, 1986; Soman and Peacock, 1985).

Wheat (*Triticum aestivum* L.) is grown on 213.52 million hectares in most parts of the world as a cereal crop (FAO, 1994) and provides food for one third of the 4.5 billion people (Johnson, 1984). Wheat is staple food in Pakistan, grown on 8.08 million hectares (FAO, 1994). Salt stress is a complex and major environmental factor which causes a considerable decrease in crop production (Shannon, 1985). Salinity is an ancient phenomenon and is a serious environmental constraint associated with arid and semi-arid agricultural systems (Rains, 1979; Downton, 1984). Most of these areas are confined to the tropics and Mediterranean regions. Salt stress is a common and important factor in deserts due to the presence of high salt concentrations in the soil (Flowers *et al.*, 1977). Soil salinity also restricts plant growth in many temperate regions besides deserts (Greenway and Munns, 1980). Millions of acres have become saline and gone out of production as salt from irrigation water has accumulated in the soil. A growing plant faces two problems in such areas, one of obtaining water from a soil of negative osmotic potential and

another the toxicity of sodium, carbonate, and chloride ions.

Saline soils include soils containing appreciable quantities of soluble salts to interfere with the growth of most crop plants but not containing enough exchangeable sodium to alter soil characteristics. The principal soluble anions are chloride, sulphate, bicarbonate and occasionally some nitrate. Technically, saline soil is a soil having a EC (electrical conductivity) greater than 4 m mhos and exchangeable sodium percentage less than 15. The pH is usually less than 8.5. Where as sodic soils are those which are not containing appreciable quantities of soluble salts. But sodic soils contains dominant sodium ions. The EC (electrical conductivity) less than 4 m mhos and exchangeable sodium percentage greater than 15. The pH is usually between 8.5 and 10.

There are many techniques which can be used for the reclamation of saline soils, such as leaching down the salts through excessive irrigations or by the addition of gypsum. Gypsum ($CaSO_4$) is sometimes used, providing both Ca^{2+} and some acidity, which helps in leaching out Na^+. Gypsum is only slightly soluble in water and therefore, large quantities of water must be added to soil amended with this material. However addition of $CaCl_2$ can be more economical for reclamation of sodic soils (Magdoff and Bresler, 1973). Sulphur is also sometimes applied. It becomes oxidized to produce sulphuric acid, which aids in Na^+ leaching. Sulphuric acid itself has been applied with some success. Another

technique for reclaiming sodic soils involves engineering approach. Installation of an efficient drainage system and the installation of tube wells have been effective in decreasing the deleterious effects of soil salinity on plants by providing good drainage in the root zone in arid and semi-arid areas. This approach has proved to be successful to reclaim the saline deserts, but it is not economical for developing countries to run such projects (Shannon, 1984). Therefore a genetic dimension is essential to overcome such soil problems. Plant scientists are seeking to modify plants to suit such adverse soil conditions while maintaining reliable yield. This approach is called a 'biological fix' and it has been emphasized as a possible means of utilising unexploited saline soil (Epstein *et al.*, 1980; Epstein and Rains, 1987).

The importance of the interaction between plant breeders and plant physiologists has been strongly emphasized by Blum (1988). He emphasized that there is very often a misunderstanding by the plant physiologist, who may define selection criteria which may be physiologically acceptable but totally unacceptable to plant breeders. The incomplete physiological knowledge of a plant breeder in selecting salt tolerant genotypes could lead to major waste of time and resources (Reitz, 1974).

A range of different techniques is needed for evolution and selection of genotypes resistant to different stress factors occurring simultaneously in the

field in the semi-arid tropics. Therefore the identification of a simple morpho-physiological trait related to resistance to several biotic and abiotic stresses is highly desirable in any crop improvement programme. In an early study Lyon (1941) observed genetic variability after studying the response to salinity of two tomato species and their F_1 progeny, but little attention was paid to his work. There was much research conducted on the effects of salinity on germination and growth of cereals during the first half of this century (Stewart, 1898; Loughrigde, 1901; Kearnery and Scofield, 1936; Magistad and Christiansen, 1944; U.S.A. Salinity Laboratory, 1947; Ayers *et al.,* 1952; Bernstein, 1961, 1963). However, there was no pressure as there is today on breeders and physiologists to exploit the potential of saline soil, nor for the selection and breeding of salt-tolerant crop varieties to produce better yield than the susceptible cultivars under saline conditions.

The main purpose of the experiments reported in this thesis was to compare two potential methods for increasing salt-tolerance.

1) By crossing a high yielding variety with a salt-tolerant variety.

2) By making selections from within existing varieties.

CHAPTER 2

REVIEW OF LITERATURE

2.1 Impacts of salinity on agricultural productivity

Soil salinity refers to the presence of excessive levels of dissolved, or readily dissolvable inorganic solutes in soil. Soil salinity may also be defined as the concentration of the mineral salts present in the soil water on a unit volume or weight basis (Tanji, 1990). Cations (Na^+, Ca^{2+}, Mg^{2+}, K^+) and anions (Cl^-, SO_4^{2-}, NO_3^-, and HCO_3^-) are the major components of soil salinity. Chlorides, sulphates and bicarbonates of sodium , calcium and magnesium are most commonly found in saline soils and irrigation water. The primary source of salinity is the continuous geochemical weathering of rock and soils.

Salinity problems are especially prevalent and serious in irrigated lands in many parts of the world. It has been estimated that nearly 40% of world's land surface can be categorized as having potential salinity problems (World Resources, 1987).

Pakistan has extensive areas of saline soils. According to the Soil Survey of Pakistan, about 5.7×10^6 hectares are salt affected and 2.13×10^6 hectares are waterlogged (Rafique, 1975). But, Muhammad (1978, 1983) estimated that $4.85\text{-}7.91 \times 10^6$ hectares land is saline and $1.16\text{-}6.17 \times 10^6$ hectares are water logged. Qayyum and Malik (1988) reported a 20 billion rupees economic

loss per annum because of salinity due to the decrease in agricultural production. The soil salinity problem is not confined to developing countries , since areas in developed countries such as the USA and Australia are also affected by salinity. In the United States, about one third of the irrigated land is affected by salt (Postel, 1989). In the case of the Colorado river system, in 1982 the annual damage to agriculture amounted to US $ 113 X 10^6 and it was expected to increase to over $ 250 x 10^6 (in constant US dollars) by the year 2000 (Holburt, 1984). Watkins *et al.* (1991) reported $ 37 million per year losses to agricultural farm land from salinity.

The world has about 13.2×10^9 hectares land. Approximately 7×10^9 hectares are potentially productive but only 1.5×10^9 hectares are cultivated (Massoud, 1981). Szabolcs (1989) estimated about 10% of the total arable land are saline and sodic soils and these exist in 100 countries. According to estimate of Tanji (1990), 0.34×10^9 hectares (23%) of the cultivated land are saline and 0.56×10^9 hectares (37%) are sodic.

2.2 Effects of salinity on ion uptake

Many workers have shown that salinity decreases K^+ uptake and increases uptake of Na^+ and Cl^- of crop plants. Such responses have been found in wheat (Salam, 1993, Sastry and Prakash, 1993) and in barley (Gorham, *et al.*, 1994). Salt-tolerance in wheat is associated with accumulation of inorganic ions (Na^+,

K^+ and Cl^-). Salam *et al.* (1992) found a highly significant negative correlation between Na^+ and Cl^- contents and yield. Youngest leaf K^+/Na^+ ratio showed a very high positive correlation with yield and its components. They concluded that salt-tolerance is under genetic control. Gorham and Wyn Jones (1990) reported that high leaf K^+/Na^+ ratio is associated with salt-tolerance and this character is genetically controlled in durum wheat. They also reported development of salt-tolerant lines from a Chinese Spring × *Agropyron junceum* [*Elymus farctus* spp. *bessarabicus*] hybrid. Begum *et al.* (1992) reported that salt stress increased accumulation of Na^+ and Cl^-, while it decreased K^+ accumulation in germinated wheat seeds.

There are a number of reports indicating that additional calcium in the hydroponic culture medium can influence responses to salt. Addition of Ca^{2+} reduces the effects of salinity on plants (LaHaye and Epstein, 1969; Cramer *et al.* 1990). Uptake of Ca^{2+} was not hindered by high Na^+ in soils (Waisel, 1972). Rengel (1992) reported the beneficial effects of Ca^{2+} on Na^+ uptake through roots. Colmer *et al.* (1994) reported that supplemental Ca^{2+} reduced Na^+ accumulation, and maintained the levels of K^+ in *Sorghum bicolor* root tips. Hyder and Greenway (1965) observed that elevated levels of external Ca^{2+} can increase both growth and Na^+ exclusion of plant roots exposed to NaCl stress (LaHaye and Epstein, 1971). Lauchli (1990) also found under saline conditions

that roots supplied with elevated levels of external Ca^{2+} are often able to maintain their K^+ concentrations, whereas roots supplied with lower Ca^{2+} frequently cannot maintain their K^+ concentrations.

It has also been suggested that Ca^{2+} displaces Na^+ from the plasmalemma of salt-stressed root cells, thus increasing the influx of ions into the cytoplasm (Cramer *et al.*, 1985; Lynch *et al.*, 1987). Helal and Mengel (1981) found that plants grown at high light intensity treatment were more able to exclude Na^+ and Cl^- and accumulate nutrient cations (Ca^{2+}, K^+, Mg^{2+}) than plants grown under low light intensity.

Ehret *et al.* (1990) found that amendment of the saline solution with Ca^{2+} increased the $Ca^{2+}/(Na^+ + Mg^{2+})$ ratio and ameliorated the effects of salt, but more so in wheat than in barley. At least part of the difference in salt tolerance between the two species must therefore relate to species differences in the interaction of salinity and Ca^{2+} nutrition. The greater response of wheat to Ca^{2+} was not due to a lower Ca^{2+} status in leaf tissue. On the contrary, although Ca^{2+} amendments improved tissue $Ca^{2+}/(Na^+ + Mg^{2+})$ ratios in both species, salinized wheat had equivalent or higher Ca^{2+} content, and high $Ca^{2+}/(Na^+ + Mg^{2+})$ ratios than did barley. The higher Ca^{2+} requirement of wheat is apparently specific to a saline condition. At low salinity, wheat growth was not reduced as extensively as that of barley as $Ca^{2+}/(Na^+ + Mg^{2+})$ ratio decreased.

High night time humidity dramatically improved wheat growth under saline condition, but increasing the Ca^{2+} concentration of the saline solution had no effect on growth in the high humidity treatment. These results confirm the importance of Ca^{2+} interaction with salinity stress, and indicate differences in species response.

Interactions of Ca^{2+} with other ions at high salinity are also known to occur and low Ca^{2+}/Na^{+} concentration ratios result in reduced growth and in some cases tissue Ca^{2+} deficiencies (Kent and Lauchli, 1985; Maas and Grieve, 1987; Muhammad *et al.*, 1987; Grieve and Maas, 1988). Recently it has been shown that high ionic strength of saline solutions displaces Ca^{2+} from the membranes of root cells (Cramer *et al.*, 1985; Lynch *et al.*, 1987; Lynch and Lauchli, 1988), contributing to salinity-induced Ca^{2+} deficiencies. Salt tolerance is not associated with Na^{+} accumulation in maize (Alberico and Cramer, 1993; Cramer *et al.*, 1994a). There are significant effects of salinity on ion accumulation in and transport from the roots of maize. Na^{+} and Cl^{-} are increased and K^{+} and Ca^{2+} are decreased by NaCl salinity. Supplemental Ca^{2+} increases Ca^{2+} and K^{+} and decreases Na^{+} accumulation and transport. There are no apparent effects of Ca^{2+} on Cl^{-} accumulation and transport (Cramer *et al.*, 1994a).

There was no inter-relationship between Mg^{2+} and Ca^{2+} concentrations in halophytes (Joshi, 1986). Albert and Popp (1977) found that plants growing under saline conditions accumulated more Mg^{2+} than K^+. Similarly Joshi and Bhoite (1988) found the proportionate accumulation of all ions in decreasing order: $Cl^- > Na^+ > Mg^{2+} > Ca^{2+} > K^+$ in soils as well as in vegetative parts of the halophytic grass (*Aeluropus lagopoides* L.), but Albert and Popp (1977) suggested that monocotyledonous halophytes accumulated more K^+ than Na^+.

2.3 Mechanisms of salt tolerance

Mechanisms of salt-tolerance in halophytes and glycophytes have been reviewed by many workers (Bernstein and Hayward, 1958; Strogonov, 1964; Gauch, 1972; Levitt, 1972; Greenway, 1973; Mass and Nieman, 1978; Cramer *et al.*, 1985). There are many hypotheses developed concerning the mechanisms by which ions may inhibit growth. It is believed that Na^+ and Cl^- can have direct toxic effects on various metabolic processes (Flowers *et al.*, 1977; Greenway and Munns, 1980). Exclusion of these ions is correlated to salt tolerance (Greenway, 1973; Jeschke, 1984; Yeo and Flowers, 1984; Lauchli, 1984, 1986; Lauter and Munns, 1986; Subbarao *et al.*, 1990a; Omielan *et al.*, 1991; Schachtman *et al.*, 1991). But in contrast to these findings, many halophytes take up much larger amounts of Na^+ and Cl^- than non-halophytes (Flowers *et al.*, 1977; Greenway and Munns, 1980; Jeschke,

1984; Lauchli, 1984). Halophytes can tolerate high concentrations of Na^+ and Cl^- by removing the toxic ions away from important metabolic processes (Flowers *et al.*, 1977; Greenway and Munns, 1980; Jeschke, 1984; Lauchli, 1984). The interaction of Na^+ and Ca^{2+} on plant growth and ion relations is well established (Rengel, 1992). The replacement of K^+ by Na^+ has been closely related to salinity tolerance, although the decline of K^+ level below a specific level could be an indication of deficiency (Marschner, 1971). Subbarao *et al.* (1990a) reported that salinity tolerance in pigeonpea based on Na^+ and Cl^- exclusion, and a high K^+/Na^+ ratio.

According to several workers (Christiansen and Lewis, 1982; San, 1982; Staples and Toenniessen, 1984; Shannon, 1985) different species groups have developed polymorphisms for adaptation to saline and other problem soils. A polymorphism is a major category of discontinuous variation within a species, which is controlled by suppergenes, inversions or loci and where allelic substitutions tend to bring about marked differnces in phenotype. However mechanisms imparting resistances to salinity and other soil stresses are yet to be properly understood and reliable markers (mutations that mark the existance of given genes and which can be identitified reliably) need to be made available (Rana, 1986). Greenway and Munns, (1980) reported many examples in which the mechanism of salt tolerance varied from cultivar to cultivar within species, although in general

mesophytes exclude ions when subjected to saline environments.

2.4 Effects of salinity on nutrient uptake

Salinity decreases root growth and nutrient uptake is hindered in crop plants (Levitt, 1972). Improved soil fertility led to more uptake of nitrogen, phosphorus and potassium (Garg *et al.*, 1990). Nitrogen is the key element of many cell components, such as amino acids, proteins, nucleic acid, porphirins, cytochromes etc. (Ullrich, 1992). Nitrogen uptake is affected by salt stress (Lips *et al.*, 1990). There are varying reports of the effects of salinity on nitrate (NO^-_3) uptake. Salinity strongly inhibited NO_3^- uptake , but the effect of Cl^- did not seem to be competitive (Ward *et al.*, 1986; Botella *et al.*, 1994). Leidi *et al.* (1991) found that the inhibition of NO_3^- uptake by Cl^- in peanuts (salt sensitive crop) was far more clear than in cotton (salt resistant crop). It has been variously reported that Cl^- increased the net uptake rate of NO_3^- (Smart and Bloom, 1988), had little effect (Rao and Rains,1976), had no effect on NO_3^- efflux (Smith, 1973; Glass *et al.*, 1985) and affected NO_3^- efflux (Deanne-Drummond and Glass, 1982). Salinity and low temperature alter nutrient uptake by plants (Gunvor *et al.*, 1990). Joshi *et al.* (1980) suggested that wheat genotypes tolerant to salinity and sodicity were characterized by lower Na^+:K^+ values in contrast to the sensitive ones. However Garg *et al.* (1990) observed that Na^+:K^+ ratio under saline conditions remained markedly less in high fertility as

compared to low fertility pots in both tolerant and sensitive wheat varieties. This indicated a positive salinity-fertility interaction in tolerant as well as sensitive wheat varieties.

2.5 Effects of salinity at different growth stages

The response of crop plants to salt stress at different growth stages is different (Maas and Grieve, 1994). Maas *et al.* (1986) reported that sorghum was more sensitive during vegetative and early reproductive stages than at flowering and grain filling stages. Similar results were found in wheat (Maas and Poss, 1989a), and cowpea (Maas and Poss, 1989b). Similarly, Francois *et al.* (1994) reported the effects of salinity at different growth stages in wheat. They found that continuous salinity throughout the growing season significantly reduced all growth and yield components. Salinity imposed prior to terminal spikelet differentiation reduced the number of spikelets per spike and the number of tillers per plant, whereas salinity imposed after terminal spikelet differentiation significantly reduced only kernel number and weight. Salinity causes a great reduction in vegetative as well as in reproductive growth. The reduction was through a decrease in tillers per plant and leaf area (Gorham *et al.*, 1985; Sharma and Kumar, 1985).

Supplemental Ca^{2+} can decrease Na^{+} and increase K^{+} concentrations and Ca^{2+} uptake. It is ultimately associated with an increase in plant growth. So

it is questioned by some reviewers whether Na^+, Cl^- or other ions are the predominant factors limiting plant growth (Bernstein and Hayward, 1958; Maas and Nieman, 1978; Munns and Termaat, 1986; Cheeseman, 1988). There are a few studies indicating that Na^+ accumulation is not correlated with the growth inhibition of some species (Lauchli, 1984; Alberico and Cramer, 1993; Cramer *et al.*, 1994a). Cramer (1993) concluded that the growth of maize under saline conditions is primarily limited by osmotic not ionic effects.

Munns and Termaat (1986) reported that short-term salinity may limit plant growth by inhibiting leaf expansion, whereas long-term stress may limit growth by inhibiting the carbon supply. In addition, relative salt tolerance between genotypes can change with time (Lynch *et al.*, 1982; Rawson *et al.*, 1988), and may result from different mechanisms needed for short-term and long-term salt tolerance. The early seedling stage is the most sensitive to salinity (Kaddah and Ghowail, 1964; Maas *et al.*, 1983). Cheeseman (1988) reported that reduction of growth by plants exposed to salinity is often much greater than the reduction in photosynthesis. He suggested that carbon allocation may be an important factor in salt tolerance.

Cramer *et al.* (1994b) found that relative growth rate (RGR) and leaf area ratio (LAR) were inhibited by salinity in the early stages of stress and were associated with differences in salt tolerance. Net assimilation rate (NAR) was

not significantly affected by salinity nor was it correlated with the differences in salt tolerance between hybrids. In the later stages of salinity stress, both RGR and LAR of Na^+ and Ca^{2+} treated plants were very similar to the controls. Thus it appears that the early differences in leaf expansion established early differences in plant size, which resulted in differences in final total dry matter production, despite similar RGR at later stages of growth. This study indicates that it is important to consider the early effects of salinity on plant growth when considering the long-term salt tolerance of the plant.

2.6 Effects of salinity on germination

Generally salinity inhibits seed germination (Jibury *et al.*, 1986; Yasseen *et al.*, 1989; Kumer *et al.*, 1988; Mondal *et al.*, 1988; Navetiyal *et al.*, 1989; Alwan *et al.*, 1989; Begum *et al.*, 1992). Salinity at 4.5 m mhos cm^{-1} did not affect germination, but 8.9 m mhos cm^{-1} salinity level inhibits germination (Narele *et al.*, 1969). Germination of wheat seed was decreased in the presence of salt (Babu and Kumar, 1975). Kabar (1986) reported delayed germination under salinity.

Uhvits (1946) reported that high concentration of NaCl decreased the germination of alfalfa seeds. Dell' Aquila and Spada (1993) reported in two different salt sensitive wheat genotypes under salinity stress a general decrease or disappearance of polypeptides specific to the radicle emergence phase in the

salt-sensitive genotype and a new synthesis of polypeptides that were not found during water imbibition and are common to both genotypes. They also found a differential synthesis of polypeptides that are unique to each cultivar. Upon return to water, salt-induced proteins ceased to be synthesized while proteins associated with an advanced germination phase were actively produced. So they suggested that the expression of salt stress proteins is related to the adaption process of seeds to salinity as well as to the genetic constitution of selected salt-tolerant genotypes.

Dass and Jain (1988) found that *Ziziphus rotundifolia* was tolerant to irrigation water salinity up to 4.5 and 6.5 m mhos EC at germination and seedling growth stages respectively. *Ziziphus spinachisti* and *Ziziphus mauritiana* cv Tikadi were moderately tolerant up to 2.5 m mhos EC, *Ziziphus hummularia* was sensitive to salinity. Poljakoff-Mayber *et al.* (1994) reported that the effect of salinity on imbibition is largely osmotic, but germination is inhibited, apparently, by the combined osmotic and " ionic " effects, especially at high NaCl concentrations. Inhibition of germination by high NaCl concentrations is relatively more severe in scarified then in intact seed, indicating that the seed coat acts as a partial barrier to Na^+ influx in *Kosteletzya virginica* (Malvaceae). Somers (1982) also reported that *Kosteletzya virginica* is more tolerant to salinity during germination than at the seedling stage. Singh *et al.* (1985)

reported that germination of wheat decreased with increase in the salinity level (Shah *et al.*, 1973) as well as decreased coleoptile length in wheat.

2.7 Effects of multiple stress on growth

Chapin *et al.* (1987) reported that in their life cycle plants encounter multiple stress factors whose interacting effects may be far from additive. In some cases however preconditioning to one stress factor may even increase the tolerance of a plant to a different stress factor imposed simultaneously or at a later time (King and Nelson, 1987; McBirde, 1987).

Oertli (1960) reported that when Azalea (*Rhododendron-spp.*) are grown at high temperature they are relatively more sensitive to salinity then when grown at lower temperature. These observations point to possibly different mechanisms of action for salinity and low temperature stress for growth of barley. Tyler *et al.* (1981) reported that subjecting winter wheat to salt stress reduced the rate of cold acclimation by the plants. Mozafar and Oertli (1990) concluded that barley was relatively tolerant to both low temperature and high salinity and when preconditioned to low levels salinity becomes more sensitive to subsequent low temperature stress. Plants preconditioned to higher levels of salinity, however, tolerated the low temperature shock much better.Their growth was not reduced further by low temperature stress. These observations point to possibly different mechanisms of action for salinity and low temperature stress

for growth of barley and wheat.

Borochov' Neori and Shani (1995) studied and reported the effects of temperature on salt treated melon plants. They found that growth of salt-grown seedlings was considerably inhibited at 20°C. At higher temperatures growth was enhanced more in the salt-treatment than in the control. Plant growth under saline conditions was shown to be very sensitive to air temperature (Gale, 1975). Salinity damage increased under hot, as compared with cool, conditions of growth. Various other reports on salinity and frost tolerance (Boussiba *et al.*, 1975; Schmidt *et al.*, 1986; Syversten and Yelenosky, 1988; O'Connor *et al.*, 1991) showed that similar mechanisms may operate in the two processes, but the molecular basis of the cross-tolerance was not established.

2.8 Effects of salinity on yield and various yield components

Evans *et al.* (1975) and Kirby (1988) have reported that the yield components of wheat depend on the growth of the contributing organs which develop at different phenological stages. Environmental stresses affect total grain yield differently depending on when they occur (Friend 1965; Langer and Ampong, 1970; Halse and Weir, 1974; Frank *et al.*, 1987). Salinity had different effects on yield components depending on when plants are stressed (Maas and Grieve, 1990). Environmental stresses shorten the duration of spikelet differentiation, resulting in fewer spikelets per spike (Friend, 1965;

Oosterhuis and Cartwright, 1983; Frank *et al.*, 1987). Salt stress causes a similar response (Grieve *et al.*, 1993). Grain filling and maturation is accelerated in some cereal crops by salt stress (Francois *et al.*, 1986, 1988). Straw yield was more sensitive to salt than grain yield (Pearson, 1959; Francois *et al.*, 1986, 1989).

Francois *et al.* (1994) reported significant reductions in straw yield, total above ground biomass, number of spikelets per spike, number of kernels per spike, individual kernel weight, number of tillers per plant and number of tiller spikes in wheat with continuous salinity throughout the growing period. In another study Francois *et al.* (1986) reported that there was no decrease in tiller number in a wheat variety grown under saline conditions, but vegetative growth and yield were reduced. Qureshi *et al.* (1990) found that salinity stress after emergence i,e. before tillering and at the booting stage, was more injurious than at later stages in wheat and caused a drastic decrease in grain yield. Abrol and Bhumbla (1971) reported substantial reductions in crop yield with increasing exchangeable sodium (Swarup, 1981). Maas (1993) reported that foliar injury, and reductions in growth and fruit yields of citrus appear to be related to the accumulation of Cl^- rather than Na^+.

Salt stress led to a great reduction in grain yield (Joshi *et al.*, 1979; Ashraf and McNeilly, 1988; Qureshi *et al.*, 1990). Yeo (1983) reported that

this is due to the fact that energy required for the maintenance of ion gradients and osmotic adjustment is mainly obtained at the expense of growth. Salt stress induced early maturity (Ayer *et al.*, 1952) and enhanced leaf senescence (Iqbal, 1992) could also result in reduced grain yield, because of the decrease in grain filling and leaf area duration. Grieve *et al.* (1992) found that there is a 12-15% increase in yield under saline conditions, as compared to control, when only grain on the main spike was considered. So salt stress stimulated increase in yield is attributed to the increase in kernel weight of the central spikelet. In wheat decrease in yield is mainly due to decrease in tillering and fewer kernels per spike (Gorham *et al.*, 1985; Maas and Grieve, 1990).

Cordovilla *et al.* (1994) reported that in *Vicia faba*, dry matter yield of both shoot and root decreased significantly at 75 and 100 mol m^{-3} salt concentrations ,however salinity affected shoot growth more than root growth. Garg *et al.* (1990) found that improved soil fertility significantly increased the yield of both salt tolerant (Kharchia-65) and sensitive (HD-2009 and HD-4502) wheat varieties under saline water (10 dSm^{-1}) irrigation. Oertli (1976) reported that overall effect of salt stress is to decrease productivity. Srivatsava *et al.* (1988) found reduction in dry matter accumulation and yield at elevated levels of soil salinity and alkalinity. Grain size was less affected unless both salinity and alkalinity increased.

2.9 Genetics of salt tolerance in crop plants

Lauchli (1976) strongly emphasized the importance of genetic differences in ion uptake or transport for a successful breeding programme for salt-tolerance. Inter and intra-specific variation in salt-tolerance has been reported by several workers (Epstein *et al.*, 1980; Qureshi *et al.*, 1980; Rashid, 1986). Gorham (1990a) reported that an enhanced K^+/Na^+ discrimination character is present in most D and U genome *Aegilops* species, but it is not present in the S genome species of the *Aegilops* section *sitopsis*. These species are thought to have contributed to the evolution of the B and G genomes of wheat. The enhanced K^+/Na^+ discrimination character is also present in the A genome of diploid wheats (*Triticum boeoticum* Boiss; *T. monococum* L. and *T. urartu* tum.), and is expressed in amphidiploid wheats and diploid wheats (Gorham, 1990b; Gorham *et al.*, 1991). Gorham (1994a) reported that some wild species in the family Malvaceae are resistant to drought, salinity and hot climates. These species may be a useful source of genes for tolerance to abiotic stress and these genes might be incorporated into commercially important members of the family Malvaceae (Cotton and Okra). One aspect of stress tolerance found in the Malvaceae is appearance of glycinebetaine. It was found at quite high concentrations in several *Gossypium* species, and increased in response to salt and drought stress.

Abel (1969) reported that a single Cl^- exclusion gene (Ncl) made the soybean (*Glycine max*) cultivar Lee more tolerant. Subbarao *et al.* (1990b) confirmed the potential for genetic introgression of salinity tolerance in pigeonpea based on Na^+ and Cl^- exclusion, and high K^+/Na^+ ratio. Ayers *et al.* (1952) found significant genotype x salinity interactions between two cultivars of wheat and three cultivars of barley in saline irrigation treatments ranging from 5000 to 20000 mg L^{-1} added salt (~ 7.8 to 31.2 dSm^{-1}). In contrast to this, Lehman *et al.* (1984) found that only 3 out of 14 characters in six rice (*Oryza sativa* L.) cultivars showed significant cultivars x salinity interaction under the lower salinities of 1.4, 3.0 and 6.0 dSm^{-1}. The estimation and reliability of how a character is related to resistance to an environmental stress depends on how far this character is heritable. Therefore estimation of heritability has a great value in the prediction of the effects of selection (Johnson *et al.*, 1955). Teran *et al.* (1990) reported high heritability and genetic advance in germination percentage of sorghum genotypes treated with NaCl.

Ashraf *et al.* (1986c) reported under saline conditions high heritability estimate, both in narrow and broad sense, and the broad sense heritabilities were above 0.80 for all the four grass species studied. In another study Ashraf *et al.* (1987) also reported high heritability estimates for tolerance to NaCl ranging from 31-62% for realised and 50-98% for narrow sense estimated by the

parent-progeny regression method. Maiti *et al.* (1994) observed highly significant differences among glossy sorghum lines for different variables for temperature, drought and salinity stresses. They also reported high heritabilities for shoot dry mass (74%), root dry mass (64%) and root length (40%) under salinity stress. Therefore they concluded that genetic variability and high heritability of some of the resistance traits in glossy sorghum lines offers good scope for the selection of lines in the genetic improvement of sorghum in semiarid tropics.

Jones (1984) studied the segregating generations of a cross between salinity tolerant and susceptible cucumber plants using the extent of leaf necrosis as the index of salt tolerance. He suggested that resistance is controlled by a single, dominant, major gene. Narrow sense heritability for resistance ranged from 0.41-0.86. Al-Khatib *et al.* (1994) reported that salinity tolerance was heritable in lucerne, with broad- and narrow-sense heritabilities at 0.76 and 0.61 respectively. They also found that there were no reciprocal differences.

Fooland and Jones (1991) studied reciprocal F_1, F_2 and BC_1 populations of tomato and partitioned variation in salt tolerance during germination into embryo, endosperm and maternal (testa and cytoplasmic) components and reported that in generation mean analysis there were no significant embryo (additive, dominance or epistatic) effects on germination performance under salt

stress. Highly significant endosperm additive and testa dominance effects were detected. Narrow-sense heritability estimates were moderately high. In another study Fooland and Jones (1992) reported heritability of germination performance under salinity in tomato (*Lycopersicon esculentum*). They found moderately high heritabilities estimates (r = 0.58-0.78) and expected rapid response to selection in early segregating generations for this important seed trait in tomato.

2.9.1 Varietal differences in response to salinity

Genotypic differences in tolerance may be attributed to genetic variation in ion exclusion by roots, translocation of salts into and through the xylem, retention of ion in tissues, mobility of ions in the phloem and the efficiency of the metabolic utilisation of ions under saline conditions (Epstein, 1972). Prakash and Sastry (1992) found significant differences between wheat genotypes and the parameters studied. They also found significant differential responses of genotypes to NaCl concentrations.

Salt tolerance of nine spring wheat cultivars was assessed at germination, and at maturity using solution and sand culture techniques. There was no consistent correlation between tolerance assessed at these two growth stages in any of the cultivars except Wembley. But Wembley was very sensitive as compared with the other cultivars. A general selection criterion based upon

whole plant performance for assessment of salt tolerance in wheat has been proposed (Ashraf and McNeilly, 1988). Salam (1993) also ranked four wheat varieties as Lyp-73 > Pato > Tobari 66 > Blue Silver for Na^+ exclusion at the lower level of salinity but he reported that at high level of salinity the order changed markedly and Tobari 66 accumulated the least amount of Na^+ and Cl^- in its leaves.

In barley cultivars, varietal differences in Na^+ and Cl^- uptake have been found by many workers (Ayers *et al.*, 1952; Greenway, 1962; ; Wyn Jones and Storey, 1978; Epstein *et al.*, 1980; Rawson *et al.*, 1988; Richards *et al.*, 1987). Varietal differences in foliar uptake of Na^+ and K^+ have been reported by Gorham *et al.* (1984) and Papa *et al.* (1993). Dua and Bhattacharyya (1988) reported the response of pearl millet hybrids and populations to salinity stress. They found populations were relatively more tolerant to salinity than hybrids for grain yield. Tall and long ear populations were better suited in saline soils, but hybrids gave higher absolute yield than populations. Bold seeded and bristled hybrids were highly salt tolerant. They suggested that salt tolerant hybrids can be developed from inbred lines of salinity tolerant populations.

Won *et al.* (1992) studied 4 rice cultivars differing in their sensitivity to salt and reported that salt-tolerant cultivars had lower Na^+ and higher K^+ contents and lower Na^+:K^+ ratio than susceptible cultivars, but there was no

difference in Ca^{2+} and Mg^{2+} contents. Igartua *et al.* (1994) reported large genotypic differences in grain sorghum for salt tolerance at germination and emergence stages, which were not related to variability of seeds, and poorly related to seed weight.

It has been suggested that the salt resistance of Rangpur lime compared with Etrog citron is associated with the differential accumulation of Cl^- in leaf and stem tissue (Grieve and Walker, 1983; Walker and Douglas, 1983; Storey and Walker, 1987). In contrast the adverse effects of high NaCl in citrus have been associated with the foliar accumulation of Na^+ (Behboudian *et al.*, 1986; Lloyd *et al.*, 1987). It therefore follows that salt resistance in citrus is associated with the exclusion of both Na^+ and Cl^- (Grieve and Walker, 1983).

2.9.2 Variability for salt tolerance within variety

Wheat is a self-pollinated crop in which natural cross pollination involving 1 to 4% of flowers may occur. Wheat is partly self-sterile from chromosomal irregularities, or from adverse environment and this sometimes leads to extensive cross-pollination (Leonard and Martin, 1963). These authors also reported a maximum of 34% cross-pollination in a strain of Fulcaster wheat in Virginia and approximately six times as much natural cross-pollination in the secondary heads as in the primary heads of five wheat varieties. Systematic work to examine genetic variability within crops is still in its infancy

(Srivastava and Jana, 1984). Joshi (1992) reported significant variation for eight attributes including grain yield and four indexes,both under normal and saline conditions in Kharchia wheat (*Triticum aestivum* L.) collections. Allard and Bradshaw (1964) reported that populations which have more variability of heterogenous gametes can better withstand salinity. These populations were more tolerant at flowering through population buffering mechanisms. In wheat variety Blue Silver intra-varietal variation for Na^+ accumulation and salt tolerance occurs (Rashid, 1986).

Shah (1987) examined the second selfed generation of wheat and reported variation in Na^+ and Cl^- uptake. Similarly, Salam (1993) generally concluded that there is genetic variation in ion uptake within Blue Silver wheat variety.

2.10 Response of crop plants under different salinity forms

Ashraf *et al.* (1986d) reported the tolerance of inland and sea cliff populations of *Holcus lanatus* and *Agrostis stolonifera* to soil salinity and salt spray. There were no differences between ecotypes in sensitivity to soil salinity, but there were differences in response to salt spray, leading to the conclusion that resistances to the two forms of salt application are independent.

Gorham *et al.* (1994b) reported that barley varieties differed in foliar uptake of sodium and chloride than uptake through roots. Storey (1995)

described the ion relations of two citrus genotype (sensitive and resistant) under conditions of high NaCl concentrations and found that calculated rates of net Na^+, K^+ and Cl^- uptake and transport were higher in plants grown in solution culture than those of plants grown in sand culture for both genotypes.

2.11 Selection criterion for salt tolerance

Various workers evaluated different characters for their potential as selection criteria for salt tolerance. For early screening of wheat genotypes, germinability at high salt concentration (Roy, 1991) and seedling dry and fresh weight at different levels of salinity (Prakash and Sastry, 1992) along with Na^+ and K^+ contents are useful criteria for salt tolerance. Ashraf and McNeilly (1988) proposed a general selection criterion for salt tolerance as they suggested the use of whole plant performance for assessment of salt tolerance in wheat. Growth response to salinity is very important and can be regarded as a basis for evolution of tolerance (Kuiper *et al.*, 1988; Weimberg and Shannon, 1988). Greenway and Munns (1980) suggested values of ion content as a selection criteria in non-halophytes.

Seed to seed screening is reported to be satisfactory and suggested for breeding salt tolerant lines (Epstein, 1976; Epstein and Norlyn, 1977). Kelman and Qualset (1991) suggested that selection in low salinity environments would produce cultivars with high yield potential for environments

with moderate salinity stress (soil conductivity of ~7dSm-1), as may be prescribed with a controlled saline irrigation cropping system for wheat.

Falconer (1960) suggested that the relative efficiency of selection in the moderately saline and non saline environments can also be approached using the concept of genetic correlation. Cramer *et al.* (1994a) found that salt tolerance of maize was not correlated with the [Na^+] concentration in the shoot and suggested that this is not a useful selection criterion for salt tolerance of maize. Matveev and Vakulenko (1990) reported that selection for grain weight per plant, grain weight per main ear and grain number per ear in wheat under saline conditions was found to be more advisable.

2.12 Basis for current work

Biotic approaches to overcoming salinity problems have recently received considerable attention from many workers throughout the world. There are three major approaches to the problem available for improving the salinity of existing crop species. Firstly, salinity tolerance of crop species can be improved by examining variation within existing crop cultivars and selecting promising lines/genotypes (Srivastava and Jana, 1984; Kingsbury and Epstein, 1984). Secondly, variable material can be produced by artificial crossing of self-pollinated species or by which occurs naturally in out-crossing species and again the most promising lines multiplied for further selection (Ashraf *et al.*, 1986a).

Thirdly, the tolerance of crops may be improved if genes from a wild relative can be transferred to the cultivated species either by conventional crossing techniques, or if possible through genetic engineering. Wild relatives of crop plants provide a rich source of novel variation which can be introduced into crops. One of the major limitations in transferring genes for stress tolerance is the lack of good tests for tolerance which is largely due to the fact that the physiological mechanisms involved are not fully understood. There is also a great lack of knowledge of the control of these genes at the molecular level (Forster, 1992).

Wheat is grown in the crop rotation in the San Joaquin Valley. The degree of grain and biomass yield reduction per unit increase in soil electrical conductivity in San Joaquin and Impend Valley of California have been well documented (Ayers *et al.*, 1952; Francois *et al.*, 1986; Rhoades *et al.*, 1988). Richards *et al.* (1987) found significant differences between the slopes of regression lines relating grain yield to soil salinity for a diverse set of wheat cultivars in soil salinity levels ranging from 5-20 dSm^{-1}.

Kelman and Qualset (1991) reported that under saline conditions genetic variances were significant and genotype x environment interaction variances were not significant for grain and biomass yield and harvest index in wheat. Broad-sense heritabilities estimated each year were low for grain yield

(0.30 and 0.10) and biomass (0.07 and 0.02). At the high saline irrigation treatment levels differences between Anza and Cajeme-71 became more apparent. These differences may relate to the differing pattern of dry matter accumulation in the two cultivars in that Anza accumulates while Cajeme-71 loses vegetative dry matter after anthesis.

Munns and Termaat (1986) hypothesized that plant responses to salinity in the long term (week, months) are largely dependant on the balance between new leaf production and death of older leaves, because of the accumulation of salts. Salam (1993) concluded that Na^+, K^+, and Cl^- accumulation, K^+/Na^+ ratio and osmotic pressure in wheat are all heritable traits in wheat under saline conditions.

Wheat is regarded as moderately salt tolerant among glycophyte species. Salt tolerant cultivars show selective uptake of K^+ both at plant (Erdei and Trivedi, 1989) and callus levels (Trivedi *et al.*, 1991). Plants may be ion excluders or ion includers depending on their responses to salinity. These properties tend to change in the same species at different levels of salinity. The salt tolerant species can grow at higher levels of salinity compared to sensitive species (Flowers *et al.*, 1977). Greenway and Munns (1980) reported that monocotyledonous species can be considered as moderately resistant to salinity

stress. Nagy *et al.* (1995) found that maize proved to be more susceptible than sorghum to drought and salt stresses.

Wheat productivity plays a vital role in stabilizing the economy of an agricultural country such as Pakistan. Pakistan spends a large amount of foreign exchange every year on importing wheat. In Pakistan wheat is grown on 7.8 M ha (Economic Survey of Pakistan, 1991-1992), out of which approximately 2.9 M ha are salt affected (Qureshi *et al.*, 1990), and the area of saline arable land is growing at the rate of 250 acres per day (Rozema *et al.*, 1990). According to an estimate losses in wheat yield due to salinity damage range from 36% to 67% (on slightly affected soils to moderately affected soils respectively) (Qayyum and Malik, 1988).

It is well documented that improving salt tolerance to increase economic yield can be accompanied by genetic manipulations which are normally accomplished through hybridization and selection. Genetic diversity is the foundation of all plant breeding programmes. Systematic work to examine genetic variability within crops is still in its infancy (Srivastava and Jana, 1984), but it is evident from previous work that there are inter-specific (Maas and Hoffman, 1977; Kingsbury and Epstein, 1984; Shah *et al.*, 1987) and intra-specific variations for salt tolerance (Ashraf and McNeilly, 1988; Rashid, 1986; Singh *et al.*, 1988).

The estimation of heritability has a great value in prediction of the effect in selection (Johnson *et al.*, 1955). Teran *et al.* (1990) reported high heritability and genetic advance in germination percentage of sorghum genotypes treated with NaCl. Heritability estimates in forage and wheat grasses (Ashraf *et al.*, 1986a, 1986b and 1987) and *Sorghum bicolor* (L.) Meench (Azhar, 1988) also indicated that salt tolerance is a heritable trait and there is potential for progress through selection.

In view of above evidence the current studies were planned to extend this approach in wheat. The aim of this research was to investigate and compare two methods of improving salt tolerance of wheat. One by making selections from within existing varieties on the basis of yield per plant and K^+/Na^+ ratio. Secondly by breeding (crossing nearly homozygous high yielding lines with low yielding tolerant lines) to produce new combinations which will be used in further studies to determine reliable information about the genetic basis of salt tolerance. This accurate and precise information could be helpful in developing wheat varieties which can give reliable yield under saline conditions. The cultivars tested were selected on the basis of their contrasting origins and salt-tolerance. They were:

1) Alexandria, a pure breeding variety with high potential for yield under non-saline conditions. This was supplied by Twyford Seeds, Oxon, UK.

2) Kharchia-65, an Indian landrace that has been shown to be salt-tolerant (Prakash & Sastry, 1992).

3) KRL1-4, a pure breeding line which is a selection from Kharchia-65 with improved salt-tolerance. This was supplied by Dr. S. Quarrie, Cambridge Laboratory, Norwich, UK.

CHAPTER 3

STUDY OF INTER- AND INTRA-VARIETAL VARIATIONS IN WHEAT (*Triticum aestivum* L.) UNDER SALINE AND NON-SALINE CONDITIONS

3.1 INTRODUCTION

Existence of genetic variability (inter- or intra-varietal) is the pre-requisite for any breeding programme to improve crop plants. Varietal differences in salt tolerance have been reported for many crops including wheat (Ashraf and McNeilly 1988), barley (Ayers *et al.*, 1952; Epstein *et al.*, 1980; Greenway, 1962; Rawson *et al.*, 1988; Richard *et al.*, 1987; Wyn Jones and Storey, 1978). Varietal differences in foliar uptake of Na^+ have been reported by Papa *et al.* (1993).

Intra-varietal variation for salt tolerance has also been reported by many workers: in rice by Flowers and Yeo (1981), Yeo *et al.* (1988); in wheat varieties by Salam (1993) and Joshi (1992).

In view of the previous studies of all these workers, the present study was planned to extend this approach in wheat. The aims of this study were to:

1) Identify variability in physiological and morphological traits within and between varieties and land races.

2) Identify lines suitable for inclusion in later experiments to investigate

and compare different breeding strategies.

3) Determine the effects of the growing system on individual plant performance, specifically by comparing yield and ion uptake of plants growing in the inside and outside rows of pots.

It is imperative to use near homozygous lines from varieties to generate such information which will give more precise and accurate information about the genetic basis of salt-tolerance (Jones and Qualset, 1984). These information could be great value for developing wheat varieties which can yield reliably on saline soils.

3.2 MATERIALS AND METHODS

Three wheat varieties KRL1-4 (a selection from within Kharchia, reported to be more salt-tolerant and agronomically superior to Kharchia, supplied by Dr. S. Quarrie, Cambridge Laboratory, Norwich, UK), Kharchia-65 (salt tolerant reported by Prakash & Sastry (1992)) and Alexandria (unknown) were tested in this experiment to determine the extent of any inter- and intra-varietal variation in salt tolerance. The experiment was conducted in a glass-house at the University College of North Wales, College Farm, Aber, Bangor from March to July 1993. Temperature in the green-house was not controlled. No supplementary lighting was used in the experiment.

3.2.1 Raising the seedlings and transplanting

Seeds of the three varieties were germinated on capillary matting in a growth-room set at 20°C starting on 18-3-1993. The light intensity in the growth-room was 200-300 μ mol m^{-2} s^{-1} PAR at leaf surface. Seedlings were transplanted into hydroponic culture in three pots on 26-3-1993. 25 l pots (52 × 35 × 16 cm) were used in this experiment. For air supply, 7 mm holes were made in the pots (two in the front, one in the right side and one in the left side). One 9 mm hole were also made in every pot in the front to allow for solution changes. The holes were plugged with rubber bungs to facilitate easy changes of nutrient solutions and to fix air supply needles (No. 16: Terumo Europe, Belgium). The pots were arranged along the sides of work benches to facilitate easy access for maintenance and sampling. Silicon tubing (Scientific Services, Chester, UK) was used as it automatically seals holes created by needles in it. The silicon tubing (5 mm internal diameter and 8 mm outer diameter) was fixed along the work benches and connected to the air regulator. Air from the silicon tubing to the pots was supplied through narrow (0.58 mm internal diameter and 0.96 outer diameter) polythene capillary tubing (Portex Ltd. Hythe, Kent, England). The capillary tubing was cut into appropriate lengths and then fixed with needles at both ends, one end inserted into the silicon tubing and the other into the bung fitted in the pots (Figure 3.1). There were 45 plants per variety

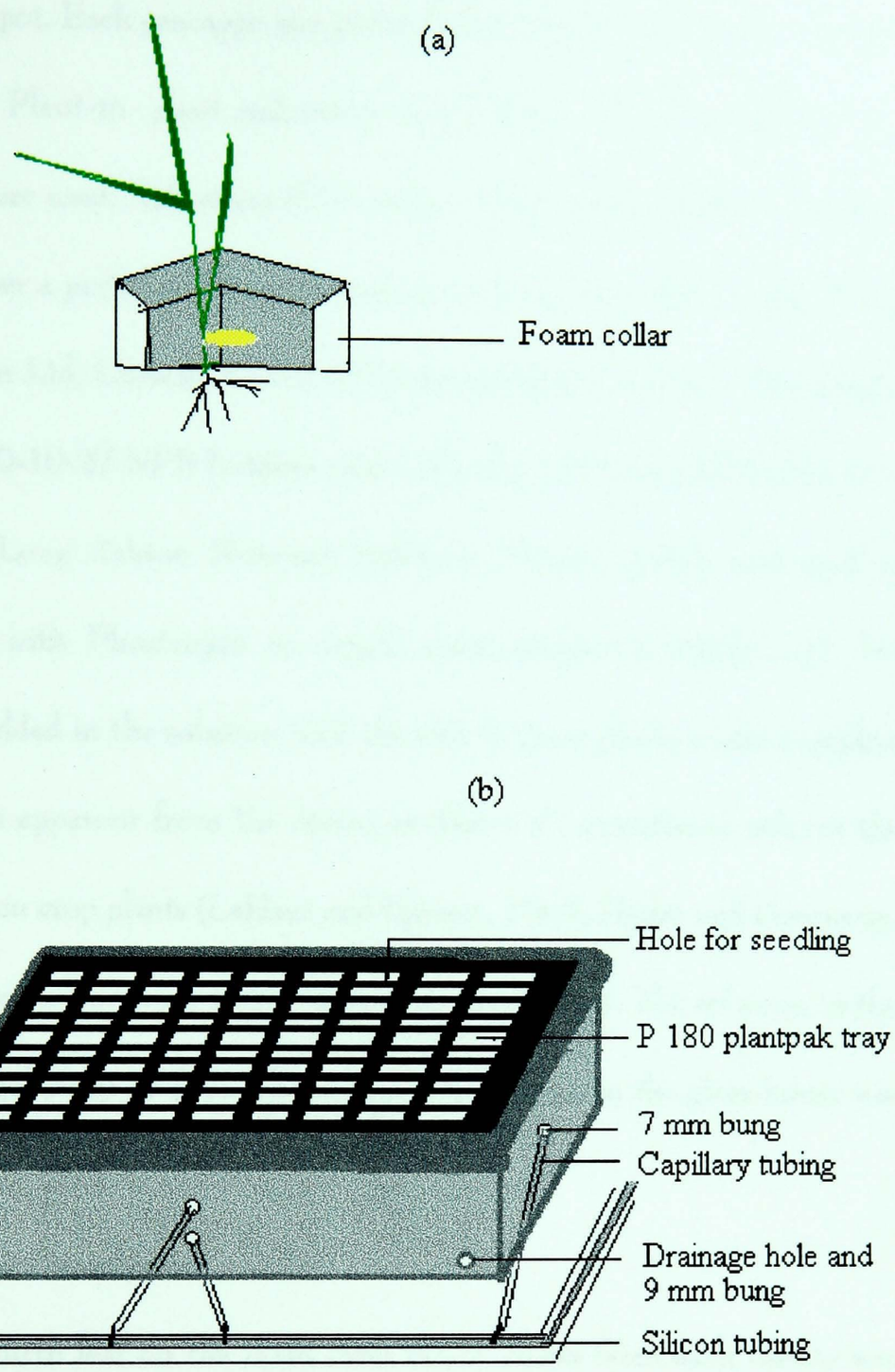


Figure 3.1. (a) Expanded diagram of supportive foam collar around seedling. (b) Growth container showing aeration lines, drainage hole and lid.

grown in one pot. Each genotype was grown in single pot to avoid inter-varietal competition. Plant-to- plant and row-to-row distance of 7.0 cm and 6.0 cm respectively were used. Salt stress (125 mol m^{-3} NaCl) was introduced in three increments over a period of five days starting on 5-4-1993. Phostrogen (0.5 g l^{-1}, Phostrogen Ltd, Corwen, Clwyd, UK) was applied to each pot. Phostrogen is a blended 10-10-27 NPK fertiliser with 1.3% Mg. 0.4% Fe and 0.02 % Mn. A modified Long Ashton Nutrient Solution (Hewitt, 1966) was used in combination with Phostrogen to supply micro-nutrients (Table 3.1). No calcium was added in the solution with the idea to grow plants under complete stress, as it is apparent from the literature that Ca^{2+} sometimes reduces the effects of salt on crop plants (LaHaye and Epstein, 1969; Hyder and Greenway, 1965; Alberico and Cramer, 1993; Cramer *et al.*, 1994a). The solution in the pots was changed every 15 days. The average temperature in the glass-house was 21.5±0.31°C.

3.2.2 Chemical analysis.

The fourth leaf on the main stem of 27 plants from each variety was sampled on 21-4-1993 when it was fully expanded. The leaves from the plants were sampled randomly. The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The samples were placed in Eppendorf tubes and stored in a freezer set at -10°C. Stress was removed to allow the plants to

recover and produce sufficient quantities of seed to be harvested for further studies. Cell sap was extracted by crushing frozen leaf tissue in Eppendorf tubes using a metal rod with a tapered end. Small holes were made in the base of the tube and it was placed in the open top of another empty Eppendorf tube. Sap was extracted by centrifugation at 8500 rpm and collected in the second tube (Gorham *et al.,* 1984). The cell sap was diluted with distilled water for the estimation of Na^+ and K^+ content using as atomic absorption spectrophotometer151 (Model 151, Instrumentation Laboratory) and K^+/Na^+ ratio was determined.

Table 3.1. Composition of modified Long Ashton Nutrient Solution (Hewitt, 1966) used to supply micro-nutrients.

Micro-nutrient	**Stock solution (g liter^{-1})**	**Volume of stock for one litre nutrient solution (cm^{-3})**
$MnSO_4.4H_2O$	22.3	0.1
$CuSO_4.5H_2O$	2.5	
$ZnSO_4.7H_2O$	2.9	
Fe EDTA (Monosodium complex)	37.3	0.5
H_3BO_3	31.0	0.1
$Na_2MoO_4.H_2O$	1.2	0.1

3.2.3 Final harvest

The experiment was harvested at maturity on 12-7-1993. All 27 plants

from which the fourth leaf had been detached were harvested. The effects of removing this leaf on yield are discussed in Chapter 6. Main tiller height, number of spikes per plant, straw weight per plant, number of infertile spikelets per spike and number of fertile spikelets per spike were recorded. Threshing was done by hand and grain weight per plant, number of grains per spike, and average grain weight were determined.

3.2.4 Statistical analysis

Statistical analyses were performed by using the Minitab and SYSTAT statistical packages. Analysis of variance (ANOVA) was used to assess the significance of differences between the means of the varieties. Where differences between means were found to be significant ($P<0.05$) an LSD test was applied at the 5% level of significance.

LSD was calculated as $\sqrt{2EMS/N} \times t_{df\ 5\%}$

Where: EMS= error mean square from the analysis of variance and N=number of values for each variety.

The values of all traits recorded on plants growing in the inside and outside rows of pots for were compared using Students t test.

The coefficient of variation for all parameters was calculated as $\sigma^2/\bar{x}$. To test the question of whether intrinsic variation in ion content, yield and its components varied between genotypes. The coefficients of variation were

compared using the procedure of Lewontin (1966 . For each trait and variety the individual values were converted into logarithms and the variance of these values was computed. To compare pairs of varieties (X and Y) an F ratio was calculated as: $s^2_{\log X} / s^2_{\log Y}$ (Lewontin, 1966).

Where:

$s^2_{\log X}$ = Variance of the logarithms of genotypes X;

$s^2_{\log Y}$ = Variance of the logarithms of genotypes Y; and $s^2_{\log X}$ is the larger of the two variances.

This F ratio calculated for any two genotypes was then compared with the tabulated F ratio in tables of the F distribution, with N_{X-1} and N_{Y-1} degrees of freedom.

Where: N_{X-1} = Degrees of freedom of X genotype.

N_{Y-1} = Degrees of freedom of Y genotype.

If the calculated F ratio was greater than the tabulated F ratio it was concluded that intrinsic variability in the traits differed between genotypes.

3.3 RESULTS

3.3.1 Varietal differences

Significant differences were found between the varieties in ion uptake (Table 3.2). KRL1-4 was found to be salt tolerant and had significantly ($P<0.05$) low Na^+, high K^+ and higher K^+/Na^+ ratio than Alexandria and

Kharchia-65. There were no significant differences in Na^+, K^+ content and K^+/Na^+ ratio between Alexandria and Kharchia-65.

There were significant differences ($P<0.05$) in yield and its components between varieties (Table 3.3). Alexandria had significanly ($P\leq0.001$) higher grain weight per plant than KRL1-4 and Kharchia-65. This was due it having significantly ($P\leq0.001$) more grains per spike and more fertile spikelets per spike. Kharchia-65 had significantly ($P\leq0.001$) more spikes per plant than Alexandria and KRL1-4. Alexandria also had significantly ($P\leq0.001$) more spikes per plant and greater main tiller height than KRL1-4. KRL1-4 was significantly ($P\leq0.001$) lower in straw weight per plant than Alexandria and Kharchia-65, but Alexandria had significantly ($P\leq0.001$) greater straw weight per plant than Kharchia-65. Kharchia-65 had ($P\leq0.001$) fewer infertile spikelets than Alexandria and KRL1-4.

3.3.2 Variability within varieties

The range between minimum and maximum values of individual plants shows that there was a large amount of variability within Alexandria, KRL1-4 and Kharchia-65 for Na^+, K^+ contents and K^+/Na^+ ratio (Table 3.4). However there were no significant differences found in coefficients of variation for ion uptake between these varieties (Table 3.5).

The range between minimum and maximum values shows that there was

Table 3.2. Means, S.E and least significant differences for leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio in three wheat varieties under saline conditions.

Trait	Genotypes						
	KRL1-4		Alexandria		Kharchia-65		
	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Na^+	137	6.6	172	9.8	162	7.3	22.4**
K^+	152	9.7	115	5.3	97	4.5	19.2***
K^+/Na^+	1.1	0.05	0.7	0.04	0.6	0.03	0.1***

** = P<0.01, *** = P<0.001

Note: Analyses of variance for these traits are presented in appendices 3.1-3.3.

Table 3.3. Means, S.E and least significant differences for grain weight per plant and various yield components in three wheat varieties under saline conditions.

Trait	Genotypes						
	KRL1-4		Alexandria		Kharchia-65		
	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Grain weight per plant (g)	2.2	0.2	5.7	0.3	3.6	0.4	0.8***
Main tiller height (cm)	66.3	0.8	91.6	1.4	77.2	1.3	2.7***
Number of spikes per plant	2.0	0.1	3.1	0.1	3.9	0.3	0.5***
Straw weight per plant (g)	2.1	0.1	5.2	0.3	3.0	0.2	0.6***
Infertile spikelets per spike	2.0	0.1	1.0	0.1	0.6	0.1	0.4***
Fertile spikelets per spike	14.7	0.3	20.0	0.2	12.4	0.2	0.6***
Number of grains per spike	36.3	1.2	51.7	1.9	25.3	0.8	3.9***
Average grain weight (mg)	29.8	1.3	35.5	0.8	34.9	1.3	3.0**

** = P<0.01, *** = P<0.001

Note: Analyses of variance for these traits are presented in appendices 3.4-3.11.

Table 3.4. Minimum and maximum values of leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio in three wheat varieties under saline conditions.

Trait	Genotypes					
	KRL1-4		Alexandria		Kharchia-65	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Na^+	87	217	70	300	82	274
K^+	108	385	67	203	56	156
K^+/Na^+	0.7	1.8	0.4	1.4	0.4	1.1

Table 3.5. Coefficients of variation (CV %) and variances of logarithms (s^2_{log}) in parentheses for leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio in three wheat varieties under saline conditions.

Trait	Genotypes		
	KRL1-4	Alexandria	Kharchia-65
	CV%	CV%	CV%
Na^+	25.0(0.010404)	29.6(0.017636)	23.4(0.010545)
K^+	33.1(0.010384)	24.0(0.009545)	24.0(0.010531)
K^+/Na^+	25.0(0.012277)	27.7(0.013202)	26.1(0.010778)

There were no significant differences between any coefficient of variation.

also large variability within Alexandria, KRL1-4 and Kharchia-65 for grain weight per plant and most of its components (Table 3.6).The landrace Kharchia-65 had significantly greater variability in grain weight per plant, main tiller height, number of spikes per plant, and average grain weight than the pure breeding line Alexandria. Alexandria had significantly higher greater variation in number of infertile spikelets per spike and less for main tiller height and average grain weight than KRL1-4. Kharchia-65 had significantly greater variation in main tiller height and number of infertile spikelets per spike than KRL1-4(Table 3.7).

The frequency distributions of each variety for Na^+ content, K^+/Na^+ ratio and grain weight per plant are given in Figures 3.2, 3.3 and 3.4. These illustrate the wide distribution of values for each trait observed within each variety.

3.3.3 Comparison between plants growing in inside and outside rows

To assess whether individual plant performance was affected by position within the pot, the data were analyzed to compare the means of plants growing in inside and outside rows. Plants growing in the outside rows of pots might have received more light and have had higher transpiration and hence ion uptake and growth than plants growing in the inside rows of pots. In the case of Kharchia-65, there were no significant differences ($t_{(cal)} < t_{(tab)}$) between inside and

Table 3.6. Minimum and maximum values of grain weight per plant and various yield components in three wheat varieties under saline conditions.

Trait	Genotypes					
	KRL1-4		Alexandria		Kharchia-65	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Grain weight per plant (g)	0.6	3.9	2.0	9.7	0.6	8.0
Main tiller height (cm)	58.0	74.5	84.7	96.5	57.2	87.8
Number of spikes per plant	1.0	4.0	2.0	5.0	2.0	7.0
Straw weight per plant (g)	0.4	3.5	2.4	8.1	0.7	5.8
Infertile spikelets per spike	0.5	3.0	0.0	2.7	0.0	2.0
Fertile spikelets per spike	12.5	17.0	17.5	22.0	10.0	13.7
Number of grains per spike	24.0	48.0	26.0	70.0	11.0	31.3
Average grain weight (mg)	19.7	46.5	27.9	43.9	18.3	45.9

Table 3.7. Coefficients of variation (CV %) and variances of logarithms (s^2_{log}) in parentheses for grain weight per plant and various yield components in three wheat varieties under saline conditions.

Trait	Genotype		
	KRL1-4	Alexandria	Kharchia-65
	CV%	CV%	CV%
Grain weight per plant (g)	39.6(0.035834)	31.6(0.023531)[a]	51.7(0.065096)[a]
Main tiller height (cm)	5.9(0.000676)[a]	3.7(0.000266)[a]	9.0(0.001685)[a]
No of spikes per plant	25.9(0.013433)	22.5(0.009643)[a]	34.6(0.022572)[a]
Straw weight per plant (g)	28.0(0.026212)	34.0(0.040602)	40.9(0.039351)
Infertile spikelets per spike	34.5(0.033966)[ab]	76.2(0.082197)[a]	86.9(0.078411)[b]
Fertile spikelets per spike	9.1(0.001624)	6.9(0.000979)	6.8(0.000938)
Grains per spike	17.2(0.005929)	19.5(0.008780)	15.6(0.007265)
Average grain weight (mg)	22.6(0.009761)[a]	12.3(0.002851)[ab]	18.7(0.007813)[b]

Note: Values with the same letter are significantly different at 5% level of significance. Values without letters are not significantly different.

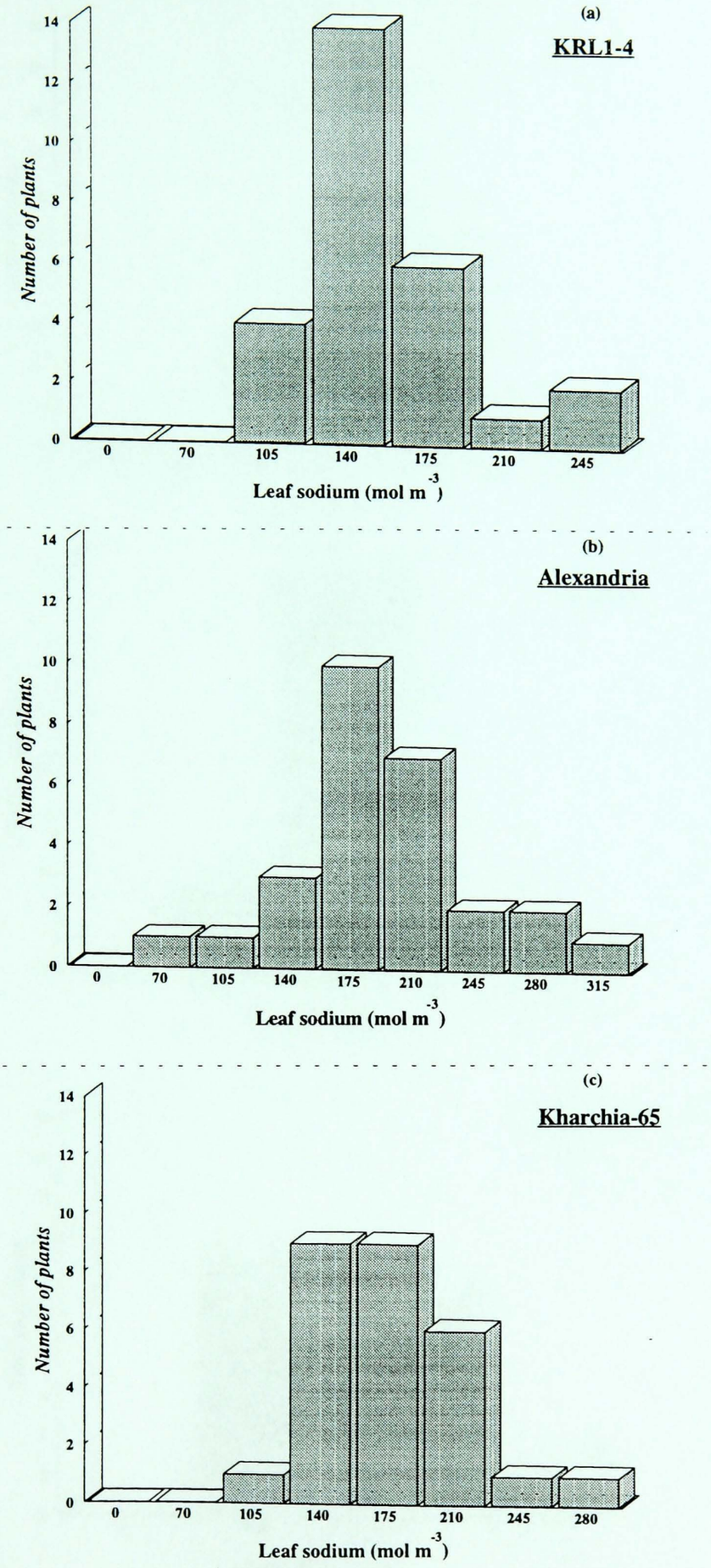


Figure 3.2 Frequency distribution of the three wheat varieti for sodium ion content under saline conditions.

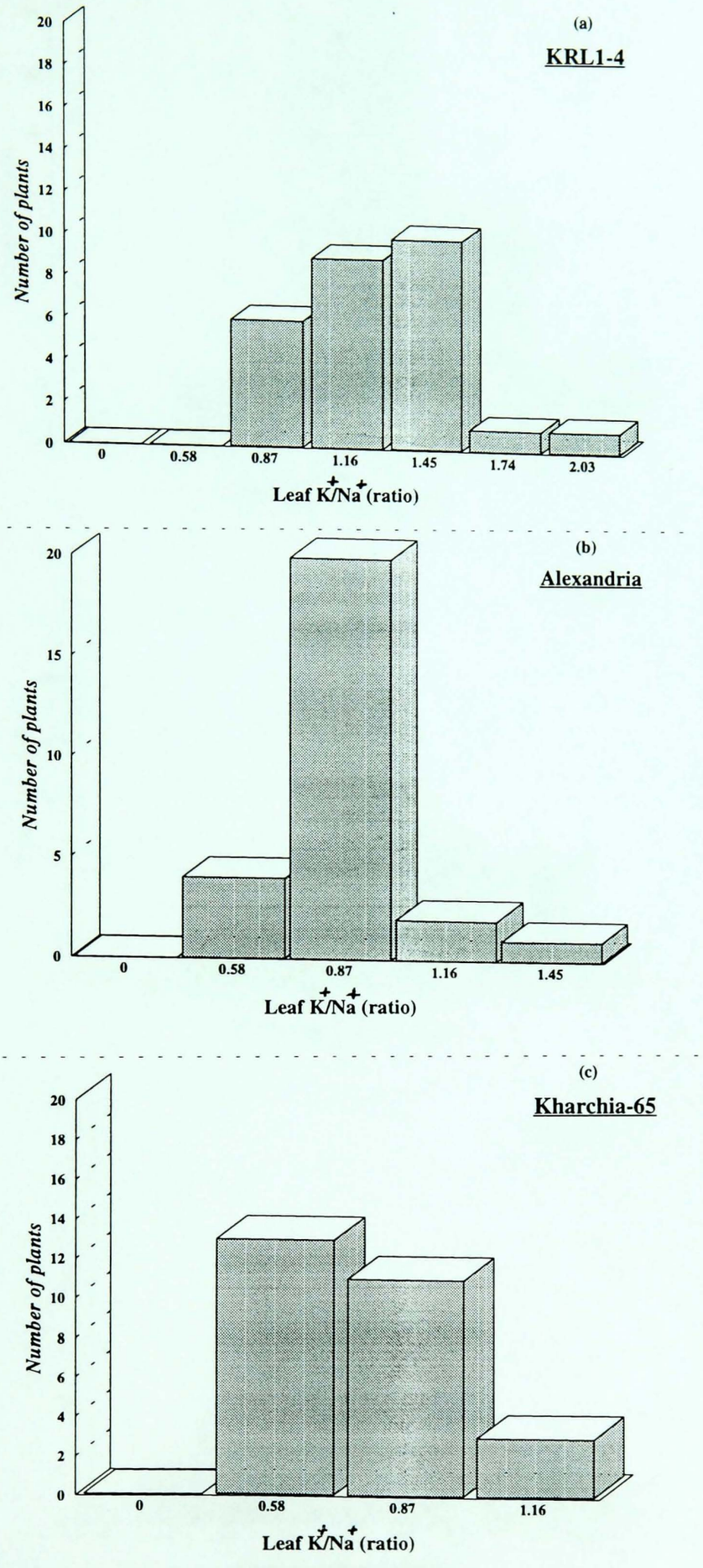


Figure 3.3 Frequency distribution of the three wheat varieties for K^+/Na^+ ratio under saline conditions.

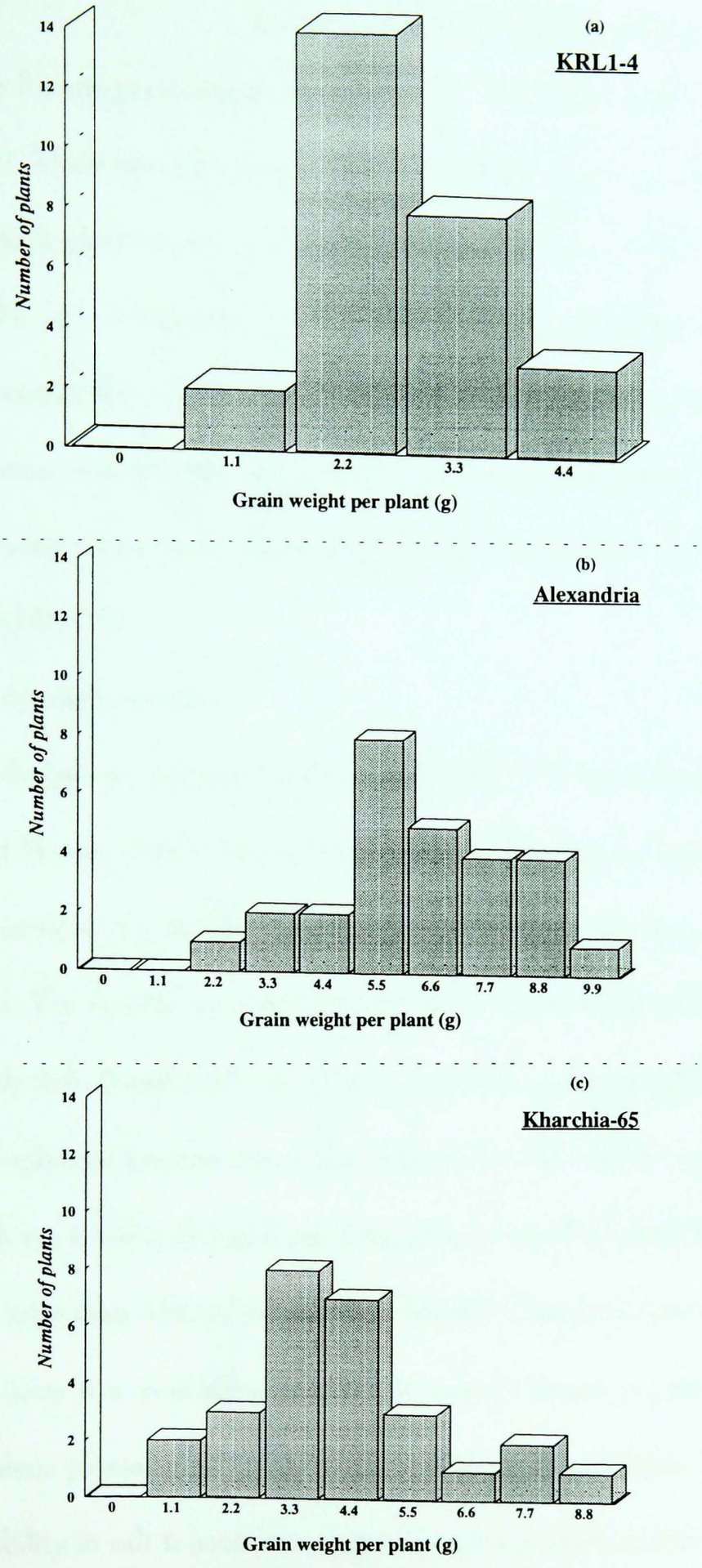


Figure 3.4 Frequency distribution of the three wheat varieties for grain weight per plant under non-saline conditions.

outside plants for any parameter studied except for main tiller height (Tables 3.8 and 3.11). There were also no significant differences ($t_{(cal)} < t_{(tab)}$) between inside and outside plants in any parameter in KRL1-4 (Tables 3.9 and 3.12). However in the case of Alexandria outside plants had significantly ($t_{(cal)} > t_{(tab)}$) higher Na^+ content than inside plants. Outside plants also had greater main tiller height, more infertile spikelets per spike, fewer grains per spike and low average grain weight than inside plants in Alexandria (Tables 3.10 and 3.13).

3.4 DISCUSSION

3.4.1 Inter-varietal variation

Salt tolerance mechanisms vary from cultivar to cultivar within species (Greenway and Munns, 1980). The results of these studies indicate significant inter-varietal variation for leaf Na^+ and K^+ content between the three wheat varieties tested. The varieties were also found to differ significantly in K^+/Na^+ ratio. Similarly Ashraf and McNeilly (1988) reported significant differences under saline conditions between nine wheat cultivars for Na^+ and K^+ content. KRL1-4 which was found to be highly salt tolerant had lower Na^+, high K^+ and high K^+/Na^+ ratio than Alexandria and Kharchia-65. Therefore these results suggest that there is a possibility in wheat to select tolerant genotypes by selecting for these physiological traits. Similarly Shannon and Noble (1995) reported variability in salt tolerance among subterranean clover cultivars and

Table 3.8. Means, S.E of leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio of inside and outside plants in Kharchia-65 wheat under saline conditions.

Trait	Inside plants		Outside plants			
	Means	±S.E	Means	±S.E	t test	df
Na^+	160	9.5	164	11.6	-0.21NS	23
K^+	92	5.8	102	6.8	-1.19NS	24
K^+/Na^+	0.6	0.05	0.6	0.04	-0.70NS	24

NS = P > 0.05

Table 3.9. Means, S.E of leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio of inside and outside plants in KRL1-4 wheat under saline conditions.

Trait	Inside plants		Outside plants			
	Means	±S.E	Means	±S.E	t test	df
Na^+	146	10.9	126	4.7	1.70NS	18
K^+	158	16.8	146	6.6	0.64NS	18
K^+/Na^+	1.1	0.09	1.2	0.05	-0.47NS	21

NS = P > 0.05

Table 3.10. Means, S.E of leaf ion contents and K^+/Na^+ ratio of inside and outside plants in Alexandria wheat under saline conditions.

Trait	Inside plants		Outside plants			
	Means	±S.E	Means	±S.E	t test	df
Na^+	147	15.2	189	11.2	-2.22*	19
K^+	107	7.7	119	7.2	-1.13NS	23
K^+/Na^+	0.8	0.08	0.6	0.03	1.73NS	13

NS = P > 0.05

* = P < 0.05

Table 3.11. Means, S.E of grain weight per plant and various yield components of inside and outside plant in Kharchia-65 wheat.

Trait	Inside plants		Outside plants			
	Means	±S.E	Means	±S.E	t test	df
Grain weight per plant (g)	4.1	0.6	3.1	0.3	1.44NS	18
Main tiller height (cm)	72.8	1.7	81.9	0.9	-4.64*	19
Number of spikes per plant	4.2	0.4	3.5	0.3	1.50NS	23
Straw weight per plant (g)	3.1	0.4	2.9	0.3	0.47NS	21
Infertile spikelets per spike	0.7	0.2	0.5	0.1	1.36NS	20
Fertile spikelets per spike	12.4	0.3	12.5	0.2	-0.40NS	21
Number of grains per spike	24.4	1.3	26.3	0.6	-1.30NS	18
Average grain weight (mg)	36.0	2.0	33.0	1.0	1.20NS	24

NS = $P > 0.05$
* = $P < 0.05$

Table 3.12. Means, S.E of grain weight per plant and various yield components of inside and outside plant in KRL1-4 wheat.

Trait	Inside plants		Outside plants			
	Means	S.E	Means	S.E	t test	df
Grain weight per plant (g)	2.0	0.2	2.3	0.2	-0.73NS	24
Main tiller height (cm)	65.3	1.2	67.5	0.8	-1.55NS	23
Number of spikes per plant	1.9	0.2	2.0	0.0	--	--
Straw weight per plant (g)	2.0	0.2	2.2	0.0	-1.19NS	22
Infertile spikelets per spike	1.9	0.2	2.0	0.1	-0.20NS	23
Fertile spikelets per spike	14.6	0.4	14.9	0.3	-0.54NS	24
Number of grains per spike	35.8	1.9	36.9	1.4	-0.47NS	24
Average grain weight (mg)	29.0	2.0	30.0	2.0	-0.39NS	21

NS = $P > 0.05$

Table 3.13. Means, S.E of grain weight per plant and various yield components of inside and outside plant in Alexandria wheat.

Trait	Inside plants		Outside plants			
	Means	±S.E	Means	±S.E	t test	df
Grain weight per plant (g)	6.2	0.5	5.4	0.5	1.06NS	22
Main tiller height (cm)	89.0	0.7	92.0	0.9	-3.02*	24
Number of spikes per plant	3.3	0.2	3.0	0.2	0.96NS	18
Straw weight per plant (g)	5.7	0.5	4.8	0.4	1.60NS	22
Infertile spikelets per spike	0.6	0.2	1.3	0.2	-3.09*	24
Fertile spikelets per spike	20.5	0.3	19.7	0.3	1.83NS	24
Number of grains per spike	56.3	2.7	48.6	2.5	2.14*	23
Average grain weight (mg)	34.0	1.0	37.0	1.0	-2.13*	24

NS = $P > 0.05$

* = $P < 0.05$

concluded that improvement in salt tolerance is possible through selection. Salama *et al.* (1994) concluded that salt tolerance in wheat may be due to different capabilities of roots to exclude Na^+ and maintenance of internal K^+ and Mg^{2+} concentrations. However, in common with the results obtained here many other workers have reported that salt tolerance in wheat also depends on maintaining a high K^+/Na^+ ratio (Rana *et al.*, 1980; Rashid, 1986; Shah *et al.*, 1987; Gorham *et al.*, 1987).

There were also significant differences between varieties in yield and in its components. Alexandria was higher yielding than KRL1-4 and Kharchia-65 due to more fertile spikelets and grains per spike. Alexandria also had significantly greater average grain weight and straw weight per plant then KRL1-4.

Overall it was concluded from the results that KRL1-4 is salt tolerant but potentially lower yielding than Alexandria. However it should be borne in mind that comparisons for yield are not reliable for saline conditions because salinity was removed during the growth period.

3.4.2 Intra-varietal variation

There were intra-varietal variations in ion contents and K^+/Na^+ ratio within each variety. Differences within varieties were larger than differences between varieties. Comparison of inside and outside plants showed that this

variability was generally not due to sampling position. All varieties showed a wide range of values for ion uptake and K^+/Na^+ ratio.

However overall, variability in Na^+, K^+ content and K^+/Na^+ ratio was found to be similar in the 3 varieties. There was a lot of overlap between the varieties so that for example, although Alexandria had higher maximum Na^+ than maximum Na^+ for KRL1-4, its minimum Na^+ was lower than the minimum Na^+ revealed for KRL1-4 (Table 3.4). Yeo and Flowers (1984) reported higher variability in Na^+ than in K^+ levels in rice under saline conditions.

There was variability within Alexandria, KRL1-4 and Kharchia-65 for yield and its components except main tiller height (cm) and number of fertile spikelets per spike. Therefore these results suggest that there are intra-varietal variations for ion content, yield and yield components and there is possibility of selection within a variety for these traits. Such variability within wheat has been reported by many workers, in Blue Silver (Rashid, 1986; Shah, 1987; Salam, 1993) and in Kharchia (Joshi,1992). The results provided evidence that individual plants of the landrace Kharchia-65 were more variable for yield then those of the pure breeding lines. However variability in ion uptake was similar in the 3 varieties. It was expected that landraces should have more variability than pure genotypes. However there were no significant differences found in

variability in Na^+, K^+ uptake and K^+/Na^+ ratio between Alexandria, KRL1-4 (selection from within Kharchia-65) and Kharchia-65 (landrace).

However significantly higher variability was found for grain weight per plant, number of spikes per plant, main tiller height, number of infertile spikelets per spike and average grain weight in Kharchia-65 (landrace) than in Alexandria (pure variety) under non-saline conditions. Kharchia-65 also had significantly higher variability in main tiller height and number of infertile spikelets per spike than KRL1-4 (selection from within Kharchia-65) under non-saline conditions.

To determine if these observed differences between individual plants were due to real genetic differences individual plants were selected, multiplied and then tested in replicated randomised experiments. These are described in subsequent Chapters.

Subsequent experiments aimed to assess:

1) The possibility of making selections within varieties to increase yield (either by selecting for K^+/Na^+ ratio or yield). Greenway and Munns (1980) suggested values of ion content as a selection criteria in non-halophytes. Yield was positively correlated with K^+/Na^+ ratio in wheat (Salam *et al.,* 1992; Salam 1993). This correlation suggests the possibility of selection on the basis of high K^+/Na^+ ratio.

2) The potential for crossing salt-tolerant with high yielding lines, to determine the heritability of traits and the possibility of increasing yield by this method. Following this experiment two lines per variety were selected on bases of high and low K^+/Na^+ ratio. Two lines per variety were also selected on bases of high and low yield per plant (Table 3.14). Sebsequent experiments were intended to assess the relative benefits of selecting for either high yield or high K^+/Na^+ ratio.

Table 3.14. Means of K^+/Na^+ ratio and yield for three wheat varieties and selections with high (H) and low (L) values of individual traits from within three varieties.

----------Varieties----------			**---------Single plant selection--------**		
	K^+/Na^+ ratio	**Yield per plant (g)**	**Lines**	**K^+/Na^+ ratio**	**Yield per plant (g)**
Alexandria	0.70	5.72	Alex-1 (H K^+/Na^+ ratio)	0.96	7.87
			Alex-24 (L K^+/Na^+ ratio)	0.36	8.05
			Alex-3 (H yield)	0.74	9.72
			Alex-14 (L yield)	0.74	3.14
			Alex-9 (L K^+/Na^+ ratio)	0.44	8.13
KRL1-4	1.14	2.16	KRL-24 (H K^+/Na^+ ratio)	1.77	2.10
			KRL-21 (L K^+/Na^+ ratio)	0.71	1.41
			KRL-26 (H yield)	1.22	3.72
			KRL-3 (L yield)	0.98	1.03
			KRL-5 (H K^+/Na^+ ratio)	1.73	2.40
Kharchia-65	0.62	3.58	Khar-1 (H K^+/Na^+ ratio)	1.10	2.51
			Khar-5 (L K^+/Na^+ ratio)	0.44	2.05
			Khar-4 (H yield)	0.57	7.99
			Khar-17 (L yield)	0.40	1.28

CHAPTER 4

EFFECTS OF LEAF AGE ON ION CONTENT IN WHEAT UNDER SALINE CONDITIONS.

4.1 INTRODUCTION

It is considered by many workers that biological variation in Na^+ and K^+ contents is an important factor in the genetic basis of salt tolerance in wheat (Joshi *et al.*, 1979; Shah *et al.*, 1987; Singh *et al.*, 1988; Gorham, 1988; Salam, 1993). Hence it has been suggested that ion content can be used as a breeding tool for selecting salt-tolerant genotypes. Therefore it is very important to know the extent of variation in Na^+, K^+ and Cl^- contents in leaves. Many people have measured ion contents, usually by sampling at a single time from fully expanded fourth or flag leaves. However it is very important to know the pattern of ion uptake of genotypes, because genotypes initially with a low content might have a higher content at later growth stages. In breeding, differences in phenology are also important. When comparing early and late maturing varieties it is impossible to harvest the same leaf from all plants, at the same growth stage and on the same day. Such variation could give misleading information if differences in maturity are significantly large. Jones and Qualset (1984) suggested that precise and efficient analytical techniques are needed to confirm such biological variation in plants.

Differences in ion content due to leaf age must be identified because it is important in determining the ionic differences between tolerant and sensitive wheat genotypes. The experiments reported in this Chapter were done to examine if differences between genotypes in ion content and K^+/Na^+ ratio were consistent over a range of sampling dates. A later experiment (Chapter 5) looked at variations in ion content at different leaf positions.

4.2 MATERIALS AND METHODS

4.2.1 Experiment 2

Three wheat varieties Alexandria, Kharchia-65 and KRL1-4 were tested in this experiment, to see if differences between varieties in ion uptake and K^+/Na^+ ratio were consistent over a range of sampling dates. It was conducted in a glass-house at the University of Wales, College Farm, Aber, Bangor during the period October to December 1993. Temperature was not controlled and no supplementary lighting was used. Average temperature in the glass-house was 15.4±0.56°C.

4.2.1.1 Raising the seedlings

The seeds of the three varieties were germinated in a growth-room set at 20°C on capillary matting starting on 29-10-1993. The light intensity in the growth-room was 200-300 μ mol m^{-2} s^{-1} PAR at the leaf surface. Seedlings were transplanted into hydroponic culture in four plastic pots on 5-11-1993.

The pots were painted black on the outside with bitumenized paint to prevent light encouraging algal growth in the nutrient solution (Figure 4.1). The nutrient solution was aerated as described previously (section 3.2.1). Each polystyrene lid was painted black and bored with 16 holes using a 9 mm heated cork borer. The holes were spaced to give a plant-to-plant and row-to-row distance of 4 cm. There were 4 plants per variety grown in each of four replicates. A completely randomized design was used. Size of the pot was 21 × 21 × 23 cm. Salt stress (100 mol m^{-3} NaCl) was introduced in three increments over a period of five days starting on 16-11-1993. Macro- and micro-nutrients were added in the solution following the procedure described in Chapter 3.

4.2.1.2 Chemical analysis

The fourth leaf from a single plant per variety per replication was sampled on 7-12-1993 (28 days after transplanting), 14-12-1993 (35 days after transplanting), 21-12-1993 (42 days after transplanting) and 28-12-1993 (49 days after transplanting). Leaves were fully expanded on 14-12-1993. The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The samples were placed in Eppendorf tubes and stored in a freezer set at -10°C. Cell sap was extracted and cation concentrations determined as described in Chapter 3. Chlorides were measured withan ion selective electrode

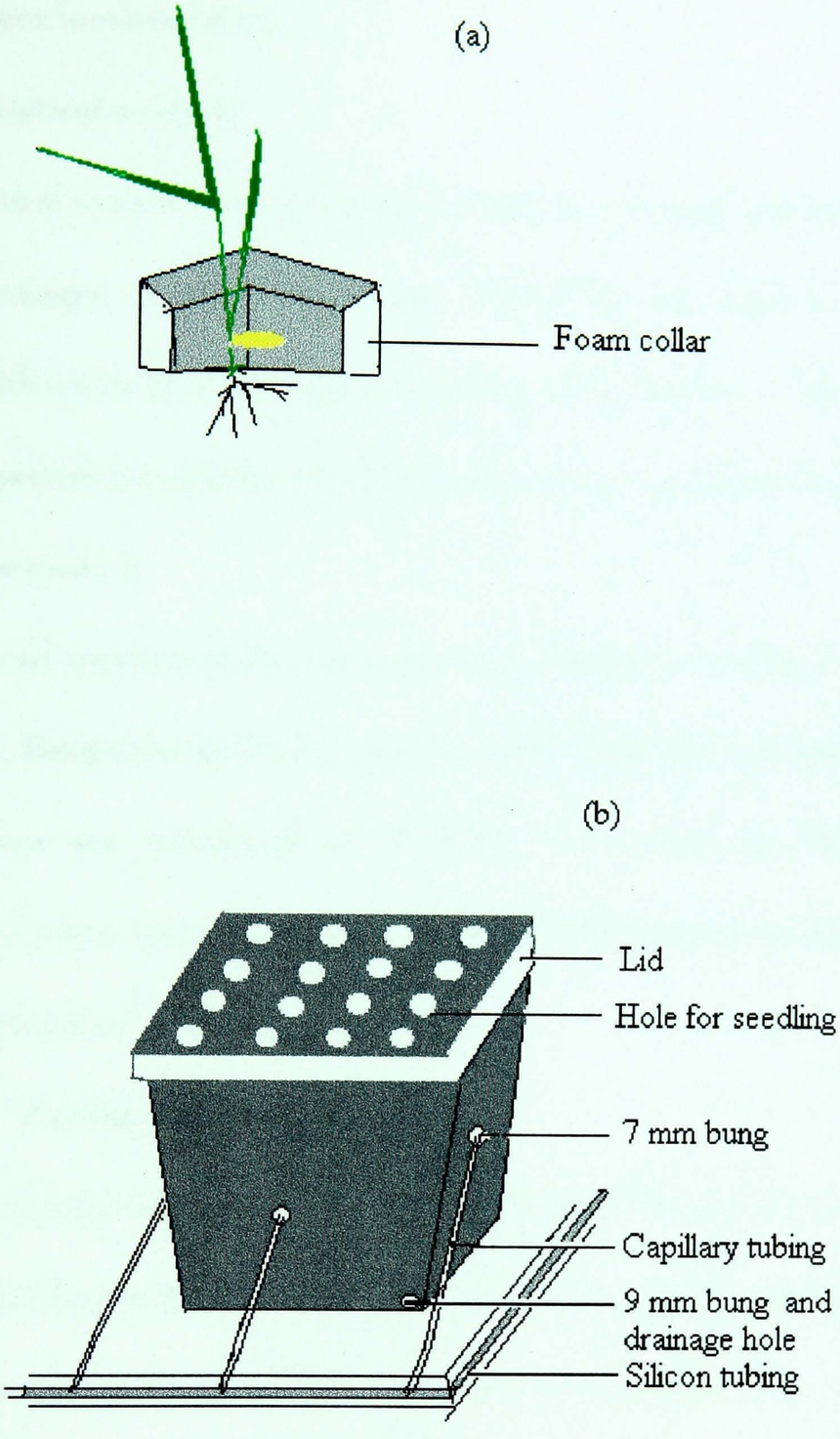


Figure 4.1. (a) Expanded diagram of supportive foam collar around seedling. (b) Growth container showing aeration lines, drainage hole and lid.

(Microprocessor Ionalyzer/901).

4.2.1.3 Statistical analysis

Statistical analysis were performed by using the Minitab and SYSTAT statistical packages. Analyses of variance (ANOVA) were used to assess significant differences ($P<0.05$) between means of the varieties. They were performed separately for each harvest and are presented in appendices 4.1-4.16.

4.2.2 Experiment 3

A second experiment was conducted in a glass-house at Pen Y Ffridd Field Station, Bangor during October to December 1994. The temperature of the glass-house was maintained at 18-20°C. The natural day light was supplemented when necessary by 400 W Son-T Sodium vapour lamps to provide a photoperiod of 16 hrs.

4.2.2.1 Raising the seedlings

Three S_1 selections Alex-1, KRL-24 and Khar-1 (their origin is detailed in Table 3.14, Chapter 3) were tested at 100 mol m^{-3} NaCl in this experiment. The seeds were sown in the glass-house at 18-20°C on capillary matting starting on 24-10-1994. Seedlings were transplanted into hydroponic culture on 3-11-1994 (replication 1 and 2) and 4-11-1994 (replication 3). A total of 60 plants (20 plants per selection per replication) were grown in three replications. The plant-to-plant and row-to-row distance was 3.5 cm and 6.0 cm respectively. A

completely randomized design was used. Size of the pot was 52 × 35 × 16 cm. Salt stress (100 mol m^{-3} NaCl) was introduced in three increments over a period of five days starting on 9-11-1994. Aeration was supplied as described in Chapter 3. Macro and micro nutrients were added in the solution following the procedure described in Chapter 3.

4.2.2.2 Chemical analysis

Fourth leaves from two plants per variety per replication were sampled on 26-11-1993 (23 days after transplanting), 3-12-1993 (30 days after transplanting), 10 -12-1993 (37 days after transplanting) and 17-12-1993 (44 days after transplanting). The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The samples were placed in Eppendorf tubes and stored in a freezer set at -10°C. Cell sap was extracted and cation concentrations determined as described in Chapter 3. Chlorides were measured with an ion selective electrode (Microprocessor Ionalyzer/901).

4.2.2.3 Statistical analysis

Statistical analyses were performed by using the Minitab, SYSTAT statistical packages. Analyses of variance (ANOVA) were used to assess significant differences ($P<0.05$) between means of the varieties. They were performed separately for each harvest and are presented in appendices 4.17-4.32. The original data and their standard errors are presented in appendices 1 and 2.

4.3 RESULTS

4.3.1 Experiment 2

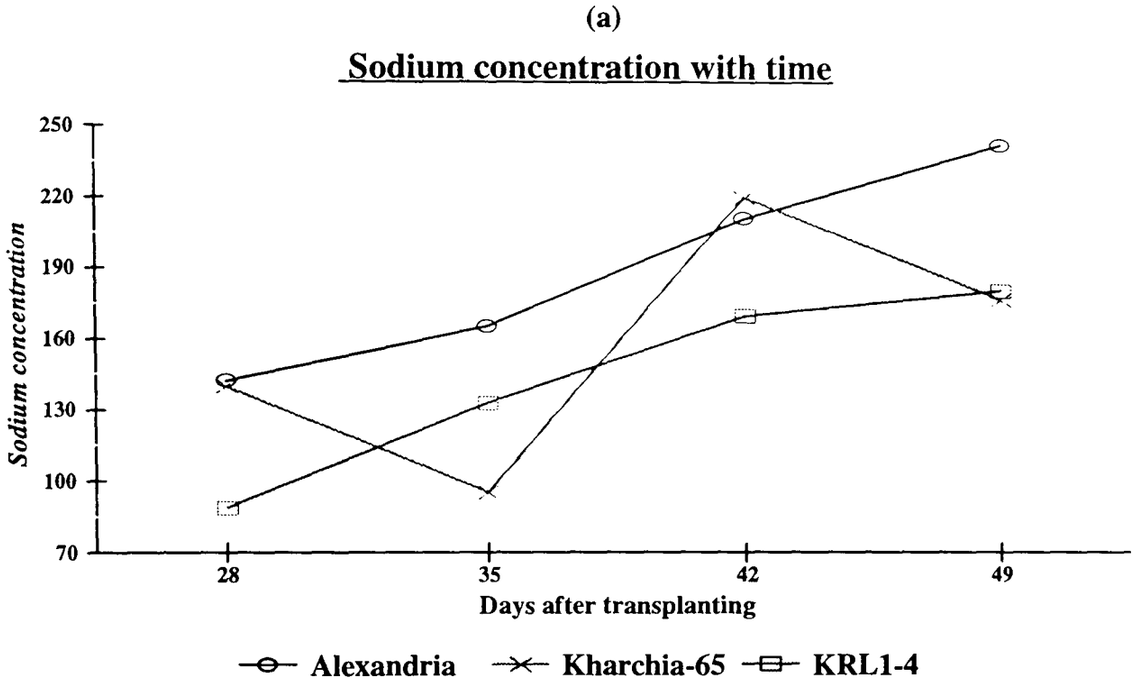
There were inconsistencies found in the increase in ion content from the first to the final harvest (Figures 4.2 and 4.3). This may be due to the limited number of plants tested or may be due to genetic variability within varieties as described in Chapter 3.

4.3.1.1 Harvest effects on ion uptake and K^+/Na^+ ratio

The general trend in all varieties was for Na^+ and Cl^- to increase, and K^+ and K^+/Na^+ ratio to decrease. However the trends were not consistent for all varieties throughout the sampling period (Figures 4.2 and 4.3).

4.3.1.2 Varietal effects on ion contents and K/Na ratio

There were no significant differences in Na^+ and K^+ content and K^+/Na^+ ratio between varieties at 28, 35, 42 and 49 days after transplanting. This is due to the large S.E's in relation to treatment means (appendix 1). However there were significant differences ($P<0.05$) in Cl^- content, where KRL1-4 had significantly lower Cl^- content than Alexandria and Kharchia-65. Although there were no significant differences between the varieties at any sampling dates, the differences were found to be consistent between Alexandria and KRL1-4 in Na^+ and Cl^+ content and K^+/Na^+ ratio (Figures 4.2, 4.3).



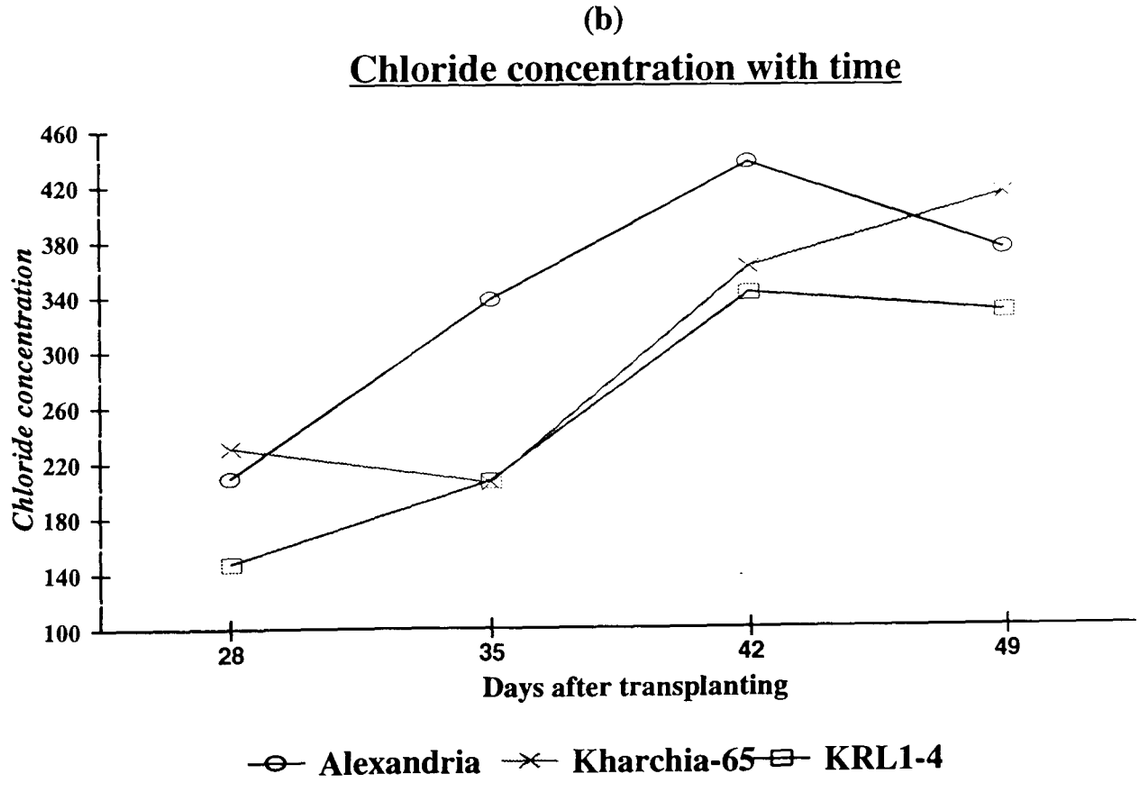
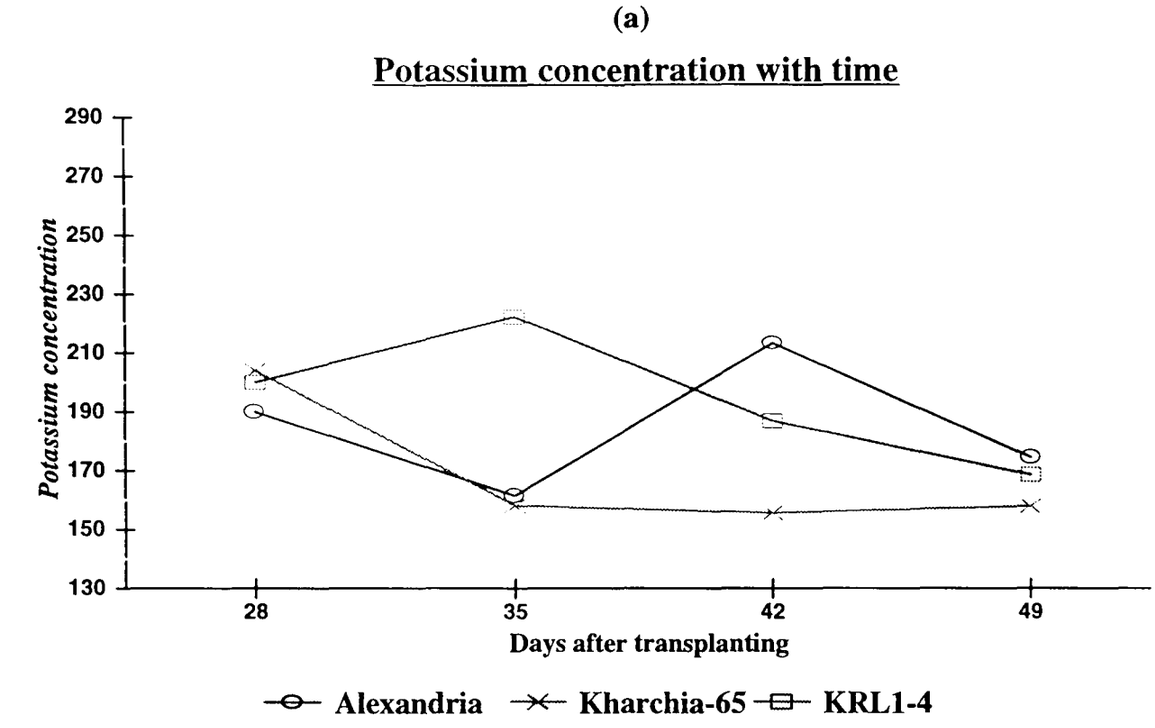


Figure 4.2. Effect of age on (a)- Na^+ (b)- Cl^- contents (mol m^{-3}) in three wheat varieties under saline conditions.



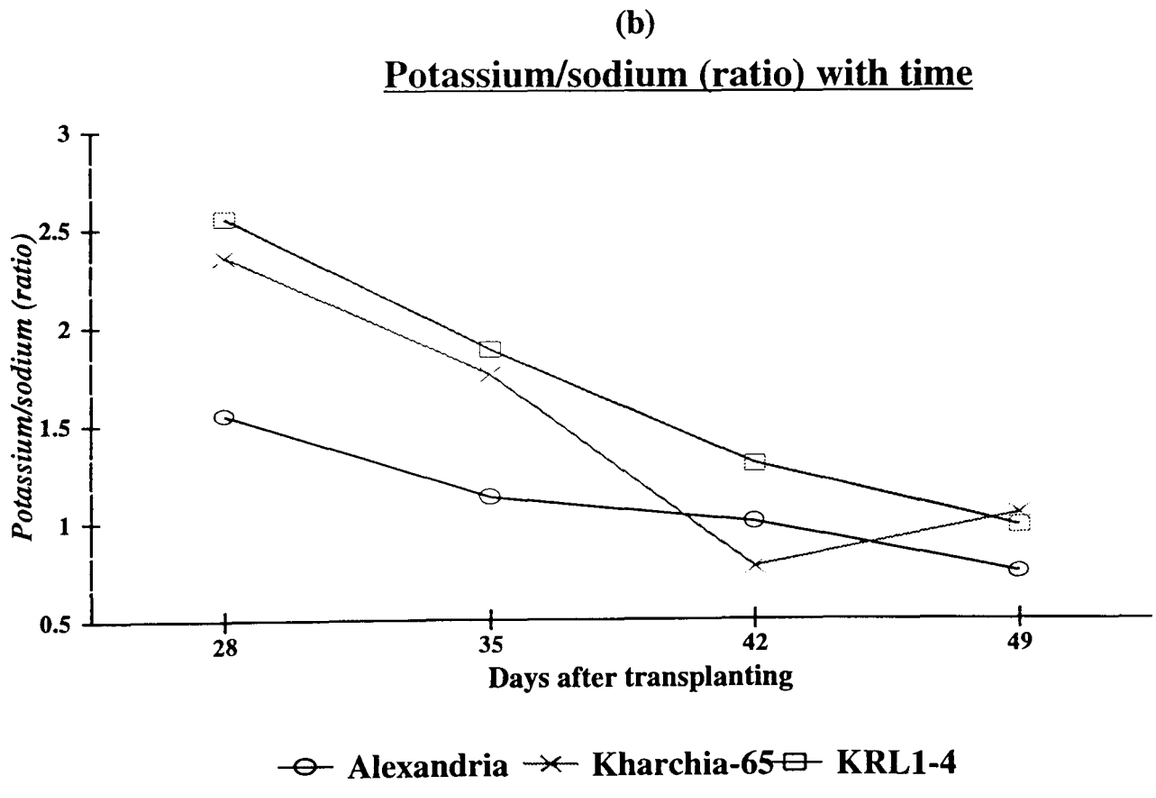


Figure 4.3. Effect of age on (a)- K^+ content (mol m^{-3}) (b)- K^+/Na^+ (ratio) in three wheat varieties under saline conditions.

4.3.2 Experiment 3

The trends in the results of this experiment were found to be more consistent than those of experiment 2. This is because double the number of plants were sampled in this experiment and as result the S.E's were smaller in relation to the means (appendix 2).

4.2.2.1 Harvest effects on ion uptake and K^+/Na^+ ratio

The trend in all S_1 lines was for Na^+ and Cl^- to increase and K^+ and K^+/Na^+ ratio to decrease. The trend in ion contents and K^+/Na^+ ratio were found to be consistent between Alex-1 and KRL-24. Khar-1 was found to be less consistent than these varieties (Figures 4.4 and 4.5). However the data obtained for this selection in experiment 3 was more consistent than the data obtained in experiment 2.

4.3.2.2 Varietal effects on ion contents and K^+/Na^+ ratio

There were no significant differences between varieties in Na^+, Cl^- and K^+ content and K^+/Na^+ ratio, except that KRL-24 had significantly ($P<0.05$) higher K^+/Na^+ ratio than at 30 days after transplanting. Alex-1 also had significantly ($P<0.05$) higher Cl^- content then KRL-24 at 23 and 30 days after transplanting.

Although most of the differences between the varieties in ion content and K^+/Na^+ ratio were found to be non significant as in experiment 2, differences

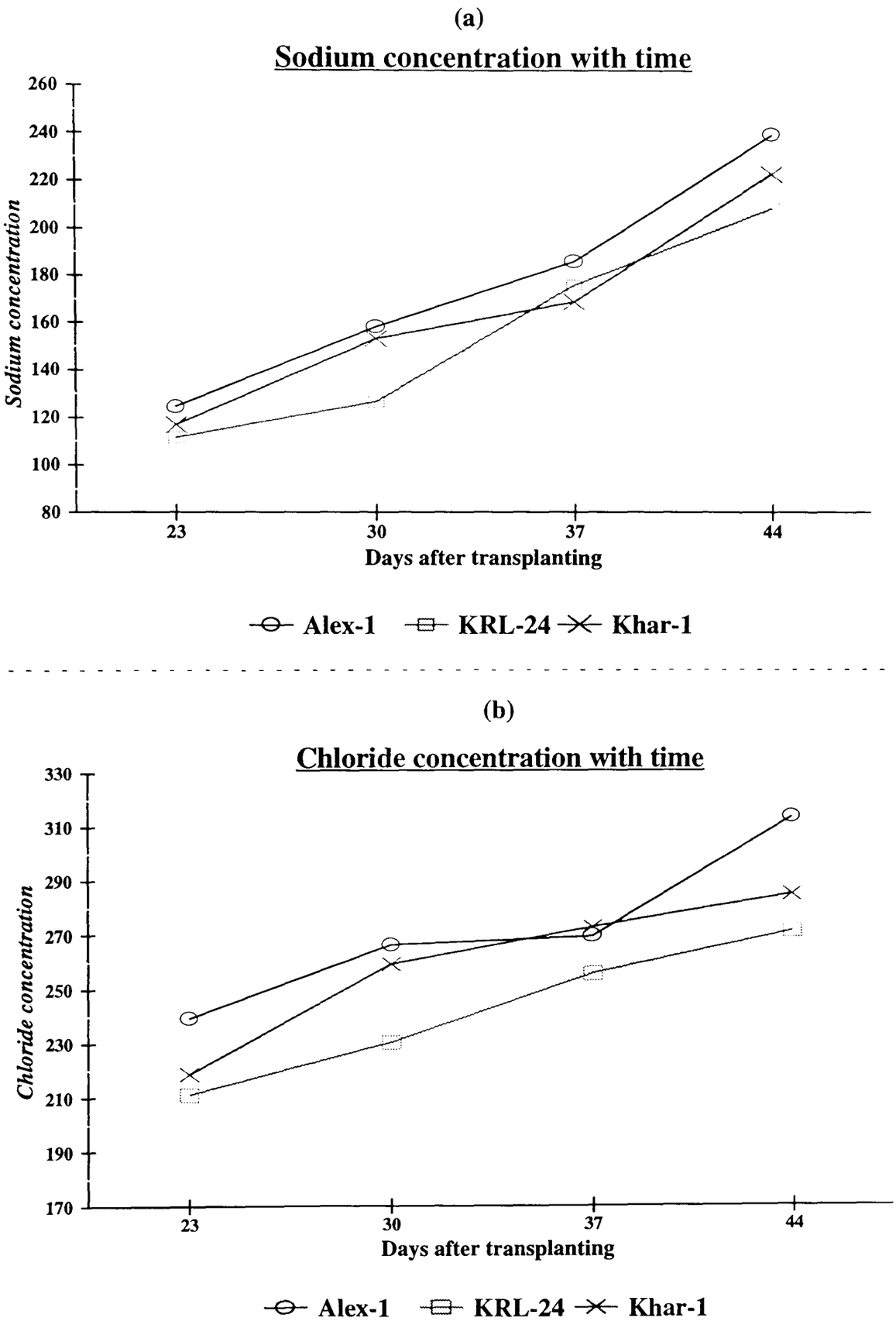
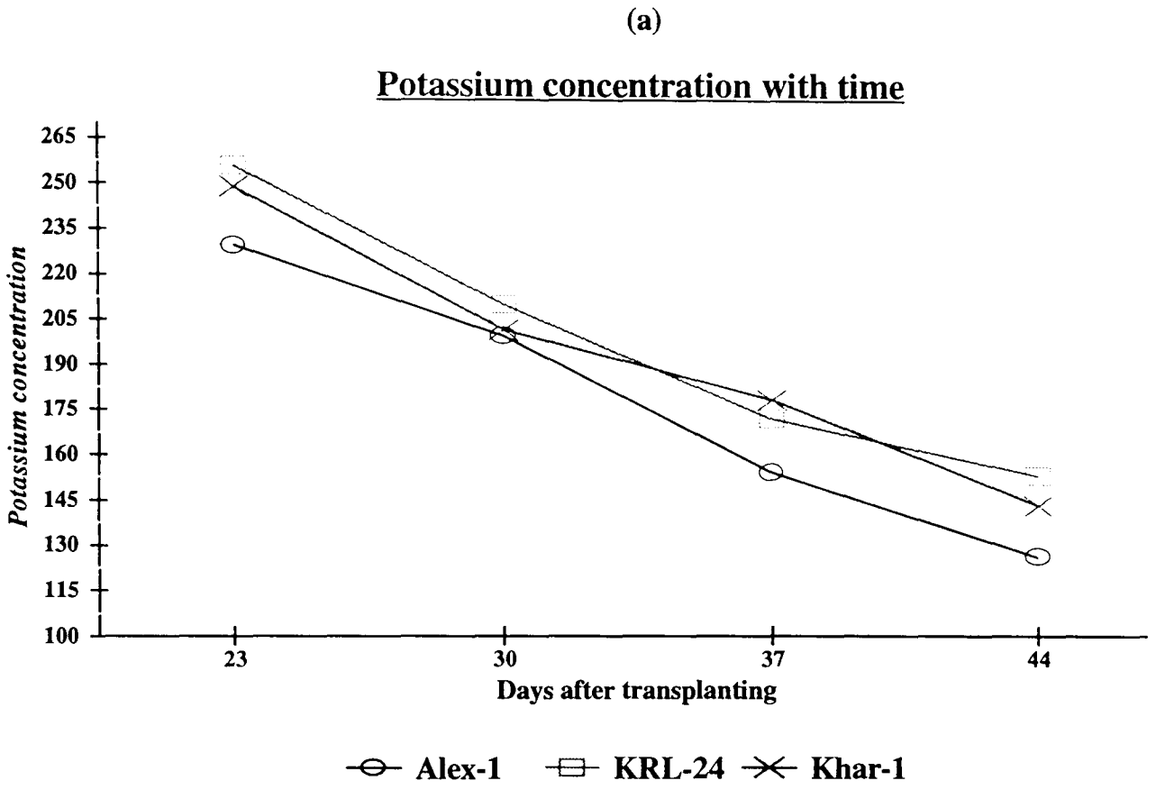


Figure 4.4. Effect of age on (a)- Na^+ (b)- Cl^- contents (mol m^{-3}) in selections from within three wheat varieties under saline conditions.



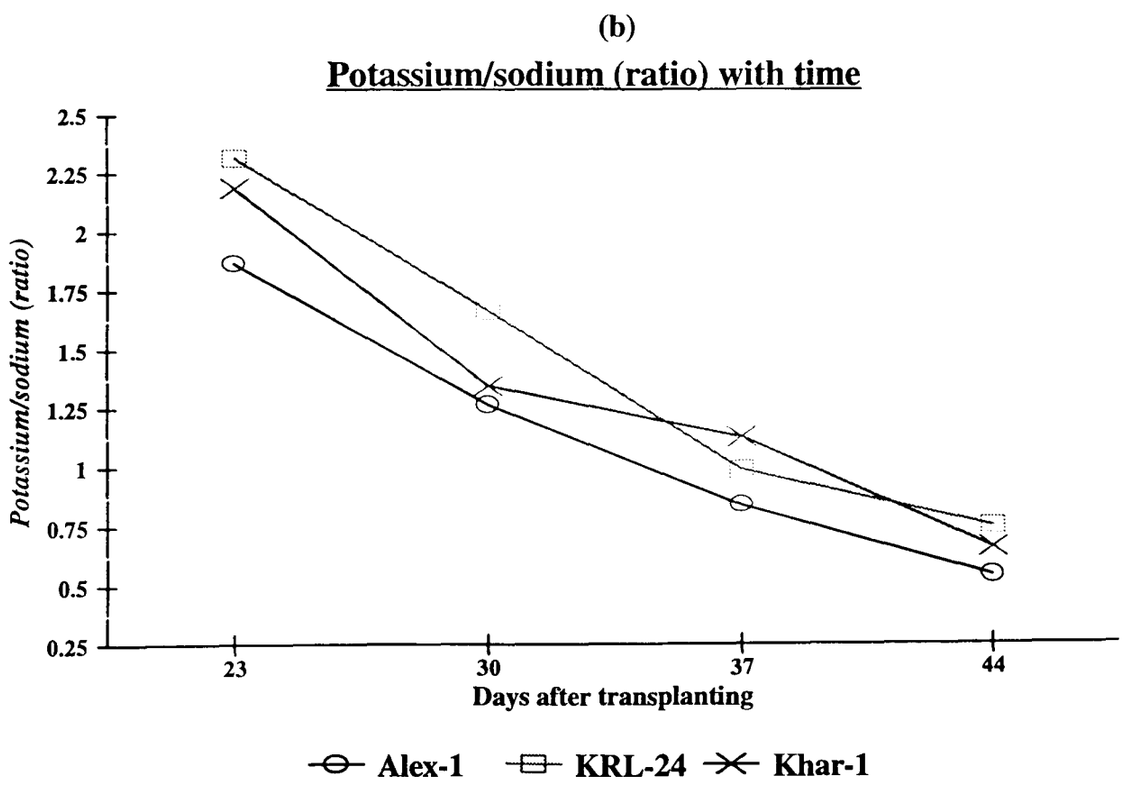


Figure 4.5. Effect of age on (a)- K^+ content (mol m^{-3}) (b)- K^+/Na^+ (ratio) in selections from within three wheat varieties under saline conditions.

between Alex-1 and KRL-24 in ion content and K^+/Na^+ ratio were found to be consistent (Figure. 4.4 and 4.5)

4.4 DISCUSSION

In experiment 2, there were no significant differences between these varietes in ion contents and K^+/Na^+ ratio at any sampling date. This might be due to the smaller number of plants tested in experiment 2 or might be due to the variability within varieties as identified in Chapter 3. The S.E's for all parameters were also larger in relation to the means in experiment 2. Therefore, differences were found to be non significant between the varieties.

A greater number of plants for S_1 lines were tested in experiment 3. Although the S.E.'s were smaller, again there were no significant differences between the genotypes, except for K^+/Na^+ ratio at 30 days and Cl^- at 23 and 30 days. It is possible that because all these lines were selected on the basis of high K^+/Na^+ ratio (Table 3.14) and the leaves were fully expanded at 30 days after transplanting in this experiment differences between varietes were smaller than in experiment 2.

The differences between genotypes in ion content and K^+/Na^+ ratio were found to be consistent in both studies except for Kharchia-65. Khar-1 was found to be intermediate between Alex-1 and KRL-24 at three out of four sampling dates. This variability between sampling dates suggests that these

physiological traits may be less useful when comparing genotypes. Generally Na^+ and Cl^- content were found to increase and K^+ and K^+/Na^+ ratio were found to decrease with leaf age in both studies. It is suggested from the results that sampling should be done when leaves are fully expanded and at least 6 leaves per genotype should also be sampled. These finding are considered elsewhere in this thesis. Differences in ion content between varieties were generally consistent over time.

CHAPTER 5

STUDY OF WHEAT VARIETAL BEHAVIOUR IN HYDROPONIC AND SOIL CULTURE

5.1 INTRODUCTION

The main purpose of this experiment was to study the performance of different wheat cultivars in soil versus performance in solution culture and the correlation in performance of the varieties in these systems under saline conditions. The majority of the research on salinity has been done in solution culture as it is easy to standardise and control salinity. It also avoids potential confounding effects due to effects of salinity on soil structure. The hydroponic medium tends to be acid to facilitate availability and uptake of trace elements especially iron, whereas most soils are more pH neutral and in many salt-affected areas in Pakistan they are alkaline as well. Electrical conductivity in solution culture is relatively constant whereas in soil it fluctuates in response to rainfall and irrigation. Therefore there is a need to show that performance in solution culture correlates with performance in soil culture if breeding and selection is to be done in solution culture. Storey (1995) reported that rates of net K^+, Na^+ and Cl^- uptake and transport of two genotypes of citrus grown in solution culture were substantially higher than those of plants grown in sand culture and that increase in solution culture was greater for a salt resistant lime

than salt a sensitive lime.

There is very little research published on this topic. Hence the present studies were conducted to study the performance of tolerant and susceptible varieties of wheat in soil versus performance in solution culture and the correlations in performance of the varieties in the two systems.

5.2 MATERIALS AND METHODS

In this experiment seven wheat varieties were tested (Table 5.1) It was conducted in a glass-house at the University of Wales, College Farm, Aber, Bangor during summer 1994. Temperature was not controlled and no supplementary lighting was used. Average temperature was 25.9°C (maximum 37.8°C and minimum 14.0°C).

Table 5.1. Varieties and their origin, response to NaCl, source and review in the literature.

Variety	Origin	Response	Source	Reference
1- SARC-III	Pakistan	Tolerant	Professor R.H.Qureshi, U.A.F., Pakistan	-
2- KRL1-4	India	Tolerant	Dr. S. Quarrie, Norwich, London	Chapter 3
3- Alexandria	Netherlands	Susceptible	Twyford Seeds, UK.	Chapter 3
4- LU26S	Pakistan	Tolerant	Dr. A. Salam, U.A.F., Pakistan	Ashraf & McNeilly (1988)
5- Bhawalpur-73	Pakistan	Unknown	U.A.F., Pakistan	-
6- Kharchia-65	India	Tolerant	Dr. S. Quarrie, Norwich, London	Prakash & Sastry (1992)
7- Blue Silver	Pakistan	Unknown	Dr. A. Salam, U.A.F., Pakistan	-

5.2.1 Raising the seedlings

Seeds of the seven wheat varieties were sown on capillary matting on 15-6-1994 in a growth-room set at 17.5°C. Seedlings were transplanted on 23-6-1994 into each system. Sixteen plants per variety per pot with plant-to-plant and row-to-row distance of 4 cm were grown in three replications in each system. A Completely Randomized Design was used. Pot size was 21 × 21 × 23 cm for both hydroponics and soil culture. Salt stress (100 mol m^{-3} NaCl) was commenced on 29-6-1994 and introduced in three increments over a period of five days in both systems.

5.2.1.1 Hydroponic culture solution

For plants grown in hydroponics the macro and micro nutrients were added in solution following the procedure described in section 3.2, Chapter 3. Seedlings were transplanted following the procedure described in section 4.2.1.1, Chapter 4.

5.2.1.2 Soil culture

Soil (clay loam) was taken from a cultivated field on the College Farm, that had been in a rotation of cereals and grass. It was sieved using a 2 mm sieve to remove the stones and placed in the pots.

To supply macro nutrients 0.5 g Phostrogen (see Chapter 3) per litre was added in two litres water. It was applied twice, at sowing and fifteen days later

to each pot. No micro nutrients were applied. Two litres 100 mol m^{-3} NaCl solution was applied twice a week to each pot. It was applied very carefully and slowly to avoid excessive leaching.

5.2.1.2.1 Electrical conductivity (dS/m)

Three extra soil pots without plants were included in the experiment. Samples were taken regularly from these pots, usually before and one day after applying the saline water. Soil samples were air dried and distilled water was added in the ratio 1:5. Samples were stirred for five minutes and then the solution was extracted using a funnel and filter paper. The EC of the extract was measured and then calculated as follows:

$EC_e = 6.4 \times EC_{1:5}$ (Talsma, 1968; Loveday *et al.*, 1972)

On occasions when the soil was dry and EC_e was higher than 12 dS/m one litre water per pot was applied in the soil to moisten the soil and decrease the EC_e. The maximum EC_e recorded during the growth period was 18.3 dS/m and the average EC_e was 11.1 dS/m whereas the EC in hydroponic culture was 10 dS/m.

5.2.2 Chemical analysis

Youngest fully-expanded leaves from two plants per variety per replication were sampled on 12-7-1994 (fourth leaf) and 01-8-1994 (flag leaf). The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The

samples were placed in Eppendorf tubes and stored in a freezer set at -10°C. Cell sap was extracted and ion contents were determined following the method of Gorham *et al.* (1984) as described previously (section 3.2.2). The cell sap was diluted with distilled water for the estimation of cations (Na^+, K^+, Ca^{2+}, Mg^{2+}) using an atomic absorption spectrophotometer-151 (Model 151, Instrumentation Laboratory). Chlorides (Cl^-) were measured with an ion selective electrode (Microprocessor Ionalyzer/901).

The experiment was harvested at maturity on 28-08-94 and data were recorded for main tiller height, spikes per plant, tillers per plant, straw dry weight, infertile spikelets per spike and fertile spikelets per spike. Tiller index was calculated by using the formula:

Tiller index = Spikes per plant × 100 /Tillers per plant

Threshing was done by hand and grain weight per plant, grain dry weight per spike, number of grains per plant, number of grains per spike and average grain weight were determined. Dry weight were determined following oven drying at 80°C for 48 hours.

5.2.3 Statistical analysis

Statistical analyses were performed by using the Minitab and SYSTAT statistical packages using GLM to assess significant differences ($P>0.05$) between the means of the varieties and systems (appendices 5.1-5.25). Where

differences between means were found to be significant ($P<0.05$) an LSD test was applied at the 5% level of significance.

LSD was calculated as $\sqrt{2} \times [$ S.E. of Means $\times t_{df\ 5\%}]$

Variety means for parameters studied were plotted to determine the relationships between these parameters in hydroponic and soil culture and values of the linear correlation coefficient (r) and the coefficient of determination (r^2) were computed.

5.3 RESULTS

In the following sections effects of variety are presented as means of two growing systems, and effects of growing systems as means of seven varieties. The performance of varieties in the two systems was studied using correlation analysis.

5.3.1 Effects of growing system

The effects of growing system under saline conditions on anion and cation uptake, number of grains per plant and various yield components (Tables 5.2 and 5.3). Plants grown in hydroponic culture had a yield significantly ($P\leq0.001$) lower and approximately 10% of those grown in soil. This was due to fewer grains per plant, fewer grains per spike. Average grain weight of plants grown in hydroponic culture was also very low ($P\leq0.000$). In comparison to these yield components, number of spikes per plant, number of tillers per plant,

Table 5.2. Effect of culture systems on yield per plant and yield components of wheat (data are the means of seven varieties) under saline conditions.

Trait	**Soil culture**		**Hydroponic culture**		
	Means	**±S.E**	**Means**	**±S.E**	**LSD**
Grain weight per plant (mg)	959.5	83.0	95.0	20.3	80.0***
Alive plants per pot	11.9	0.1	6.8	0.6	0.5***
Main tiller height (cm)	60.0	1.0	42.5	1.6	3.3***
Spikes per plant	1.5	0.1	1.3	0.1	NS
Straw weight per plant (mg)	947.1	43.7	822.5	87.7	NS
Infertile spikelets per spike	2.6	0.2	2.5	0.1	NS
Fertile spikelets per spike	11.2	0.3	8.4	0.3	0.6***
Tillers per plant	1.6	0.1	1.5	0.1	NS
Tiller index	96.0	1.1	87.9	2.0	3.5***
Number of grains per plant	23.8	1.6	9.2	1.3	3.5***
Number of grains per spike	16.1	1.1	6.9	0.9	2.1***
Grain weight per spike (mg)	659.2	65.2	69.7	12.1	80.0***
Average grain weight (mg)	41.0	3.5	9.0	0.8	7.0***

NS = $P > 0.05$

*** = $P < 0.001$

Note: Analyses of variance for these traits are presented in appendices 5.13-5.25.

Table 5.3. Effect of culture systems on ion contents (mol m^{-3}) and K^+/Na^+ ratio of wheat (data are the means of seven varieties) under saline conditions.

Trait	**Soil culture**		**Hydroponic culture**		
Fourth Leaf	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+	61	5.7	120	8.5	15.1***
K^+	195	6.5	138	7.0	18.2***
K^+/Na^+	3.9	0.5	1.3	0.1	0.8***
Cl^-	143	8.8	207	10.0	21.1***
Ca^{2+}	24	2.4	1.6	0.1	4.3***
Mg^{2+}	22	1.4	16	1.3	3.2**
Flag Leaf					
Na^+	64	5.1	138	6.7	12.7***
K^+	147	8.4	117	6.8	18.5**
K^+/Na^+	2.7	0.3	0.9	0.1	0.4***
Cl^-	204	9.5	330	17.9	40.4***
Ca^{2+}	14	1.1	1.1	0.04	2.4***
Mg^{2+}	17	0.8	13	0.5	1.4***

** = P > 0.01

*** = P > 0.001

Note: Analyses of variance for these traits are presented in appendices 5.1-5.12

straw weight per plant and number of infertile spikelets per spike were much less affected. The number of alive plants per pot was much less significantly lower ($P \leq 0.000$) in hydroponic than in soil culture.

Na^+ and Cl^- contents were significantly higher ($P \leq 0.001$) in plants grown in hydroponic culture than in plants grown in soil. K^+, Ca^{2+} and Mg^{2+} contents were significantly lower ($P \leq 0.01$) in plants grown in hydroponic culture than in plants grown in soil. K^+/Na^+ ratio was also significantly lower ($P \leq 0.001$) in plants grown in hydroponics than in plants grown in soil. The trends were the same in both the fourth and flag leaf.

5.3.2 Varietal effects

5.3.2.1 Yield and yield components

The effects of varieties on yield and yield components are shown in Table 5.4. The two salt sensitive varieties Alexandria and Bhawalpur-73 had significantly lower ($P \leq 0.001$) yield than the other varieties due to fewer grains per plant, grains per spike and lower average grain weight. LU26S was also lower yielding than SARC-III, KRL1-4, Kharchia-65 and Blue Silver due to decrease in grains per plant, grains per spike, grain weight per spike, average grain weight, but the differences were non significant. Alexandria and Bhawalpur-73 had significantly l ($P \leq 0.001$) fewer alive plants per pot than SARC-III and Kharchia-65. KRL1-4, LU26S and Blue Silver also had more

Table 5.4. Varietal effects on yield, and yield components of seven wheat varieties (data are the means of two growing systems) under saline conditions.

Trait	SARC-III		KRL1-4		Alexandria		LU26S		Bhawalpur-73		Kharchia-65		Blue Silver		
	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Grain weight per plant (mg)	678	262	642	223	247	98	540	259	220	72	640	212	700	286	160***
Alive plants per pot	10.5	0.7	9.5	1.1	8.2	1.7	9.7	1.8	7.5	2.0	10.7	0.7	9.3	1.3	1.1***
Main tiller height (cm)	51.4	3.5	54.3	2.9	50.9	6.1	49.9	5.6	45.4	4.6	55.6	5.0	51.2	3.1	6.8*
Spikes per plant	1.5	0.1	1.1	0.1	1.4	0.1	1.5	0.1	1.3	0.2	1.6	0.2	1.4	0.1	NS
Straw weight per plant (mg)	790	55	637	46	1028	55	1058	110	1277	144	818	97	625	62	259***
Infertile spikelets per spike	2.7	0.1	1.9	0.1	1.7	0.1	3.3	0.2	3.8	0.2	1.8	0.2	2.5	0.1	0.4***
Fertile spikelets per spike	10.0	0.6	11.0	0.6	10.9	0.9	9.2	1.3	9.4	0.5	10.2	0.8	8.0	0.4	1.2***
Tillers per plant	1.5	0.1	1.1	0.1	1.6	0.1	1.7	0.1	1.6	0.2	1.8	0.2	1.5	0.1	NS
Tiller index	98	1.3	100	0.0	89	3.7	88	3.5	86	4.5	93	3.3	89	1.7	7.2**
Grains per plant	19.7	4.3	19.9	3.1	12.2	3.0	15.2	5.6	9.6	1.9	22.9	5.0	16.2	4.1	7.1**
Grains per spike	13.3	2.9	17.9	2.8	8.3	1.9	9.7	3.3	7.2	1.2	13.4	2.6	10.8	2.1	4.3***
Grain weight per spike (mg)	461	186	590	212	170	66	350	167	160	53	380	132	430	165	150***
Average grain weight (mg)	27	7.4	32	12.9	15	4.6	24	9.9	20	3.8	24	7.0	31	10.3	NS

NS = $P > 0.05$
* = $P < 0.05$
** = $P < 0.01$
*** = $P < 0.001$
Note: Analyses of variance for these traits are presented in appendices 5.13-5.25.

alive plants per pot than Bhawalpur-73.

5.3.2.2 Anion and cation uptake

There were significant differences ($P<0.05$) between the varieties in contents of all ions in fourth leaf, and of all ion contents except Cl^- and Ca^{+2} in the flag leaf (Table 5.5). The general trends in ion content between varieties were the same in both leaves. Alexandria, LU26S and Bhawalpur-73 had higher ($P\leq0.001$) Na^+ than SARC-III, KRL1-4 and Kharchia-65. Alexandria, LU26S, Bhawalpur-73 and Blue Silver had lower ($P\leq0.01$) K^+/Na^+ ratio than SARC-III, KRL1-4 and Kharchia-65. Alexandria and Bhawalpur-73 had high Cl^- ($P\leq0.01$) but lower Ca^{2+} ($P<0.05$) and Mg^{2+} than SARC-III, KRL1-4, LU26S and Kharchia-65. Blue Silver had low Ca^{2+} and Mg^{2+} in fourth leaf and also high Mg^{2+} in flag leaf.

4.3.3 Correlation coefficients

In this section linear correlation were calculated using the data of the two systems (hydroponics and soil culture), separately and combined. The relationships between the valuse of parameters recorded in hydroponic and soil culture were also investigated.

5.3.3.1 Relations between yield and yield components in both systems and in combined data

In soil culture, yield per plant was significantly positively correlated with number of grains per plant, number of grains per spike, grain weight per spike, average grain weight and negatively correlated with straw weight per plant but

Table 5.5. Varietal effects of ion contents (mol m^{-3}) and K^+/Na^+ ratio of seven wheat varieties (data are the means of two growing systems) under saline conditions.

Trait	SARC-III		KRL1-4		Alexandria		LU26S		Bhawalpur-73		Kharchia-65		Blue Silver		
Fourth leaf	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+	63	9.2	65	14.7	118	11.4	99	11.6	100	16.4	71	13.5	121	27.6	30.6***
K^+	169	13.7	172	16.1	139	11.2	193	14.4	152	16.8	185	17.7	155	25.1	36.9*
K^+/Na^+	3.1	0.6	4.1	1.4	1.4	0.3	2.0	0.3	1.9	0.5	3.6	1.1	1.9	0.6	1.6**
Cl^-	150	13.6	147	15.1	198	13.8	152	28.7	228	22.7	171	22.5	177	28.2	42.6**
Ca^{2+}	17	7.3	18	8.0	7.6	2.8	17	7.2	10	4.5	10	4.5	10	4.6	8.6*
Mg^{2+}	24	3.6	20	1.3	13	1.5	21	1.2	17	2.7	23	3.4	13	1.6	6.5**
Flag leaf															
Na^+	69	12.9	85	16.1	109	23.2	114	12.1	123	19.6	85	17.2	124	23.8	25.8***
K^+	180	16.5	131	14.3	104	11.2	112	14.0	114	6.0	142	15.6	138	8.7	37.5**
K^+/Na^+	3.2	0.7	2.0	0.5	1.3	0.4	1.1	0.2	1.1	0.2	2.3	0.7	1.4	0.4	0.9***
Cl^-	272	29.4	245	33.5	307	45.2	253	8.7	270	35.4	255	45.8	309	52.5	NS
Ca^{2+}	8	3.3	9	3.7	6.4	2.7	8	3.2	6	2.2	8	3.4	9.4	4.0	NS
Mg^{2+}	16	1.1	17	1.8	13	1.1	14	1.3	11	0.8	15	0.7	16	0.6	2.9**

NS = $P > 0.05$
* = $P < 0.05$
** = $P < 0.01$
*** = $P < 0.001$
Note: Analyses of variance for these traits are presented in appendices 5.1-5.12.

there were no consistent relationships with other yield components (Table 5.6). Yield per plant was positively correlated with number of alive plants per pot, number of spikes per plant, number of fertile spikelets per spike and main tiller height, number of grains per spike, grain weight per spike and average grain weight in hydroponics and in the combined data from two systems. Tiller index was positively correlated with yield in the combined data only.

5.3.3.2 Relations among anion and cation concentrations

Na^+ and K^+ contents were significantly correlated with K^+/Na^+ ratio in the fourth and the flag leaf in the combined data from the two systems (Table 5.7) as well as in hydroponics (Table 5.8) but not in soil culture (Table 5.9) where K^+ was not significantly correlated with K^+/Na^+ ratio in the fourth leaf. Other correlations between anion and cation contents were generally significant in the combined data but not in individual systems. In soil culture Cl^- was significantly correlated with K^+ in the fourth leaf and Na^+ was significantly correlated with K^+ in the flag leaf. Mg^{2+} was significantly correlated with Na^+ and K^+/Na^+ ratio in the fourth leaf in the hydroponic culture system.

5.3.3.3 Relations between anion and cation contents and yield per plant

5.3.3.3.1 Fourth leaf

Yield per plant was significantly and positively correlated with K^+,

Table 5.6. Linear correlation coefficients between yield per plant and various yield components of 7 wheat varieties (data from hydroponics, soil culture and combined) under saline conditions.

Trait	**----------Yield per plant (g)----------**		
	Hydroponic culture	**Soil culture**	**Combined**
Alive plants per pot	0.463*	-0.085NS	0.734**
Main tiller height (cm)	0.737**	0.107NS	0.763**
Spikes per plant	0.603**	0.038NS	0.315*
Straw weight per plant (g)	0.014NS	-0.534*	0.048NS
Tillers per plant	0.429NS	-0.027NS	0.115NS
Tiller index	0.370NS	0.260NS	0.505**
Infertile spikelets per spike	-0.196NS	-0.252NS	-0.088NS
Fertile spikelets per spike	0.564**	-0.249	0.556**
Number of grains per plant	0.937**	0.626**	0.843**
Number of grains per spike	0.722**	0.550**	0.798**
Grain weight per spike (g)	0.853**	0.855**	0.953**
Average grain weight (g)	0.658**	0.688**	0.900**

NS = Non significant

* = Significant at 5% level of significance

** = Significant at 1% level of significance

Table 5.7. Linear correlation coefficients between wheat leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}) and K^+/Na^+ ratio (combined data of 7 varieties from the two systems) under saline conditions.

	Na^+	K^+	K^+/Na^+	Cl^-	Ca^{2+}
Fourth leaf					
K^+	-0.580**				
K^+/Na^+	-0.787**	0.650**			
Cl^-	0.601**	-0.588**	-0.595**		
Ca^{2+}	-0.616**	0.599**	0.571**	-0.590**	
Mg^{2+}	-0.589**	0.506**	0.558**	-0.462**	0.450**
Flag leaf					
K^+	-0.493**				
K^+/Na^+	-0.815**	0.783**			
Cl-	0.696**	-0.402**	-0.554**		
Ca^{2+}	-0.708**	0.390*	0.560**	-0.666**	
Mg^{2+}	-0.528**	0.421**	0.566**	-0.485**	0.545**

* = Significant at 5% level of significance
** = Significant at 1% level of significance

Table 5.8. Linear correlation coefficients between wheat leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}) and K^+/Na^+ ratio (data of 7 varieties from the hydroponic culture system) under saline conditions.

	Na^+	K^+	K^+/Na^+	Cl^-	Ca^{2+}
Fourth Leaf					
K^+	-0.238NS				
K^+/Na^+	-0.833**	0.663**			
Cl^-	0.303NS	-0.041NS	-0.187NS		
Ca^{2+}	-0.115NS	0.334NS	0.175NS	-0.398NS	
Mg^{2+}	-0.540*	0.371NS	0.637**	-0.402NS	0.066NS
Flag leaf					
K^+	-0.179NS				
K^+/Na^+	-0.719**	0.752**			
Cl^-	0.370NS	-0.205NS	-0.375NS		
Ca^{2+}	-0.320NS	0.011NS	0.195NS	0.121NS	
Mg^{2+}	-0.207NS	0.239NS	0.273NS	-0.230NS	-0.251NS

NS = Non significant

* = Significant at 5% level of significance

** = Significant at 1% level of significance

Table 5.9. Linear correlation coefficients between wheat leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}) and K^+/Na^+ ratio (data of 7 varieties from the soil culture system) under saline conditions.

	Na^+	K^+	K^+/Na^+	Cl^-	Ca^{2+}
Fourth leaf					
K^+	-0.180NS				
K^+/Na^+	-0.848**	0.380NS			
Cl^-	0.295NS	-0.545*	-0.419NS		
Ca^{2+}	-0.212NS	0.072NS	0.053NS	-0.181NS	
Mg^{2+}	-0.330NS	0.255NS	0.415NS	-0.117NS	0.222NS
Flag leaf					
K^+	-0.485*				
K^+/Na^+	-0.809**	0.835**			
Cl^-	0.139NS	-0.196NS	-0.157NS		
Ca^{2+}	0.110NS	0.110NS	-0.094NS	-0.248NS	
Mg^{2+}	-0.216NS	0.289NS	0.362NS	-0.189NS	0.225NS

NS = Non significant

* = Significant at 5% level of significance

** = Significant at 1% level of significance

K^+/Na^+ ratio, Ca^{2+} and Mg^{2+} but negatively correlated with Na^+ and Cl^- in the combined data from the two systems. In soil culture, yield per plant was significantly positively correlated with K^+ and K^+/Na^+ ratio and negatively with Na^+ and Cl^-. No significant correlations were found in hydroponics (Table 5.10).

5.3.3.3.2 Flag leaf

Yield per plant was significantly positively correlated with K^+, K^+/Na^+ ratio, Ca^{2+}, Mg^+ and negatively correlated with Na^+ and Cl^- in the combined data from the two systems. No significant correlations were found in hydroponics and soil culture except for Mg^{2+} content in hydroponics and K^+, Ca^{2+} and Mg^{2+} contents in soil culture, which were significantly positively correlated with yield per plant (Table 5.10).

5.3.3.4 Relationships between fourth and flag leaf ion contents

Most of correlations between fourth leaf ion contents and flag leaf ion contents were found to be significant in the combined data (Table 5.11), except K^+ and Mg^{2+} which were non significant. There were considerably fewer significant correlations in hydroponic culture (Table 5.12) where fourth leaf Na^+ was significantly correlated with flag leaf Na^+, Cl^- and K^+/Na^+ ratio. Fourth leaf Mg^{2+} was also significantly correlated with flag leaf Na^+ and K^+/Na^+ ratio. Other correlations between fourth and flag leaf anion and cation

Table 5.10. Linear correlation coefficients between wheat leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}), K^+/Na^+ ratio and yield per plant (data of 7 varieties from the soil culture, hydroponics and combined data from two the systems) under saline conditions.

Trait	Yield per plant (g)		
	Hydroponics	**Soil culture**	**Combined**
Fourth leaf			
Na^+	-0.319NS	-0.514*	-0.701**
K^+	0.230NS	0.504*	0.735**
K^+/Na^+	0.229NS	0.502*	0.751**
Cl^-	-0.281NS	-0.717**	-0.755**
Ca^{2+}	-0.109NS	0.367NS	0.809**
Mg^{2+}	0.365NS	0.039NS	0.411**
Flag leaf			
Na^+	-0.412NS	-0.149NS	-0.733**
K^+	-0.032NS	0.444*	0.489**
K^+/Na^+	0.177NS	0.392NS	0.707**
Cl^-	0.104NS	-0.075NS	-0.607**
Ca^{2+}	0.093NS	0.503*	0.872**
Mg^{2+}	0.528*	0.690**	0.734**

NS = Non significant

* = Significant at 5% level of significance

** = Significant at 1% level of significance

Table 5.11. Linear correlation coefficients between fourth leaf and flag leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}) and K^+/Na^+ ratio (Combined data of 7 varieties from the two systems) under saline conditions.

Flag leaf	**----------Fourth leaf----------**					
	Na^+	K^+	K^+/Na^+	Cl^-	Ca^{2+}	Mg^{2+}
Na^+	0.796**	-0.669**	-0.693**	0.675**	-0.692**	-0.678**
K^+	-0.425**	0.231NS	0.531**	-0.346*	0.382*	0.373*
K^+/Na^+	-0.670**	0.479**	0.726**	-0.546**	0.616**	0.595**
Cl^-	0.698**	-0.552**	-0.549**	0.583**	-0.563**	-0.485**
Ca^{2+}	-0.662**	0.605**	0.678**	-0.652**	0.784**	0.308*
Mg^{2+}	-0.521**	0.548**	0.530**	-0.584**	0.555**	0.253NS

NS = Non significant
* = Significant at 5% level of significance
** = Significant at 1% level of significance

Table 5.12. Linear correlation coefficients between fourth leaf and flag leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}) and K^+/Na^+ ratio (data of 7 varieties from the hydroponic culture system) under saline conditions.

Flag leaf	**----------Fourth leaf----------**					
	Na^+	K^+	K^+/Na^+	Cl^-	Ca^{2+}	Mg^{2+}
Na^+	0.650**	-0.424NS	-0.694**	0.420NS	-0.247NS	-0.840**
K^+	-0.076NS	-0.258NS	0.039NS	-0.031NS	-0.027NS	0.109NS
K^+/Na^+	-0.462*	-0.037NS	0.416NS	-0.300NS	0.092NS	0.622**
Cl^-	0.493*	-0.060NS	-0.380NS	0.269NS	-0.391NS	-0.428NS
Ca^{2+}	-0.124NS	-0.264NS	-0.049NS	-0.070NS	-0.198NS	0.220NS
Mg^{2+}	-0.164NS	0.242NS	0.341NS	-0.237NS	0.230NS	0.261NS

NS = Non significant
* = Significant at 5% level of significance
** = Significant at 1% level of significance

Table 5.13. Linear correlation coefficients between fourth leaf and flag leaf Na^+, K^+, Cl^-, Ca^{2+}, Mg^{2+} contents (mol m^{-3}) and K^+/Na^+ ratio (data of 7 varieties from the soil culture system) under saline conditions.

Flag leaf	**----------Fourth leaf----------**					
	Na^+	K^+	K^+/Na^+	Cl^-	Ca^{2+}	Mg^{2+}
Na^+	0.421NS	-0.036NS	-0.402NS	0.245NS	-0.061NS	-0.352NS
K^+	-0.445*	0.131NS	0.510*	-0.224NS	0.129NS	0.339NS
K^+/Na^+	-0.550**	0.080NS	0.524*	-0.285NS	0.158NS	0.468*
Cl^-	0.222NS	-0.283NS	-0.179NS	0.204NS	0.183NS	-0.049NS
Ca^{2+}	-0.298NS	-0.027NS	0.279NS	-0.352NS	0.154NS	-0.298NS
Mg^{2+}	-0.396NS	0.355NS	0.292NS	-0.501*	0.273NS	-0.106NS

NS = Non significant

* = Significant at 5% level of significance

** = Significant at 1% level of significance

contents were found to be non significant. In soil culture (Table 5.13) most of the correlations between fourth and flag leaf anion and cation contents were also found to be non significant. Fourth leaf Na^+ and K^+/Na^+ ratio were significantly correlated with flag leaf K^+ and K^+/Na^+ ratio. Fourth leaf Mg^{2+} was significantly correlated with flag leaf K^+/Na^+ ratio. There was also significant correlations between fourth leaf Cl^- and flag leaf Mg^{2+}.

5.3.3.5 Relationships between the values of certain traits in hydroponics and soil culture

The relationships between values of certain agronomic traits, ion uptake and K^+/Na^+ ratio in hydroponics and soil culture are shown in Figures 5.1-5.7. The correlations between the values of yield and most of its components in hydroponic culture and soil culture were non significant, except for straw weight per plant and infertile spikelets per spike.

Also all of the relationships between ion contents in hydroponics and soil culture were found to be non significant, except for fourth leaf Ca^{2+} and flag leaf K^+/Na^+ ratio.

5.4 DISCUSSION

5.4.1 Performance in hydroponics versus soil culture

The varieties tested had higher values for yield and most yield components in soil than in hydroponic culture. In soil culture harvest index was 51%, whereas it was only 10% in hydroponic culture. Although average EC was

Figure 5.1. Relationships between values of certain parameters in hydroponic and soil culture under saline conditions.

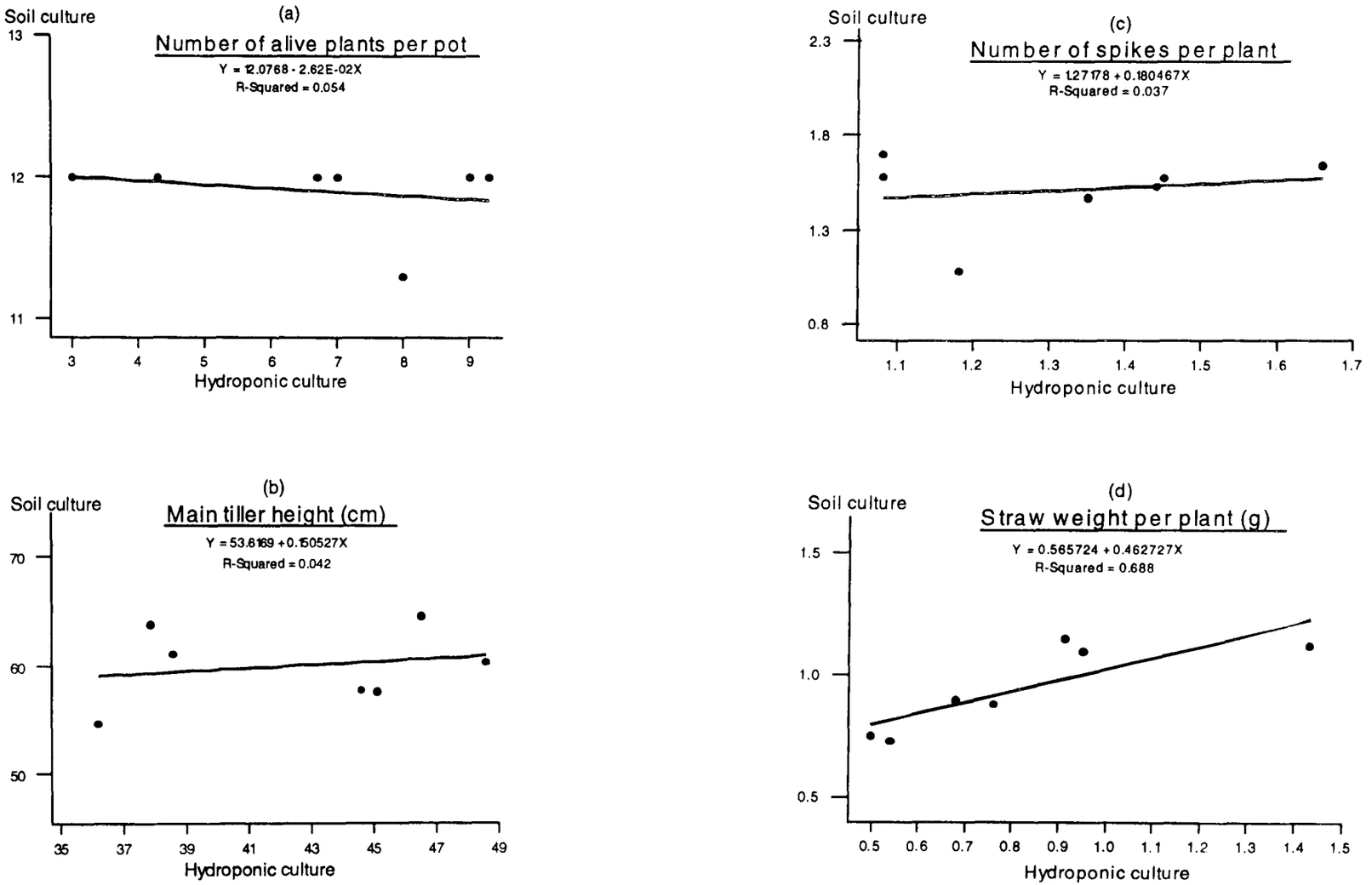
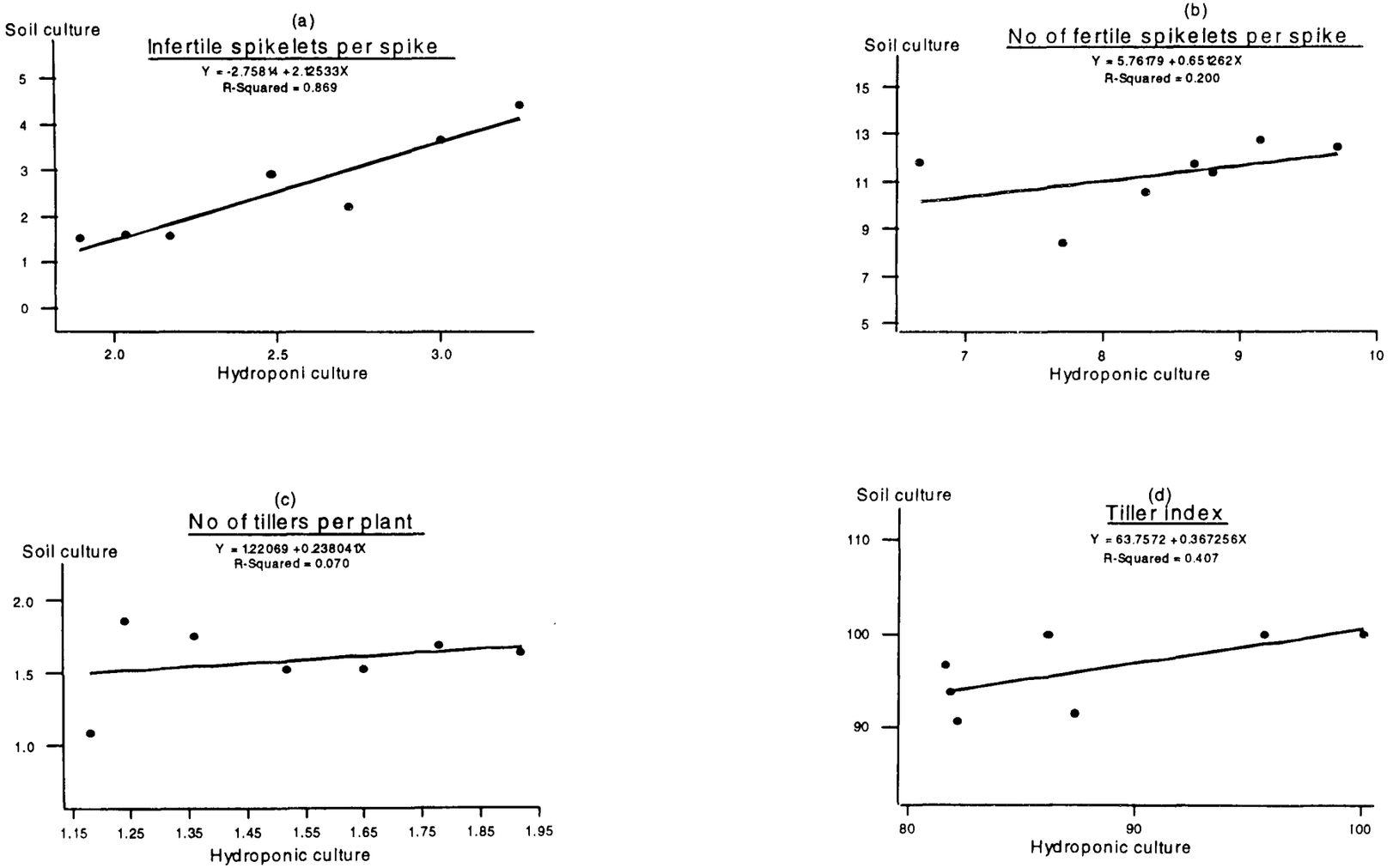


Figure 5.2. Relationships between values of certain parameters in hydroponic and soil culture under saline conditions.



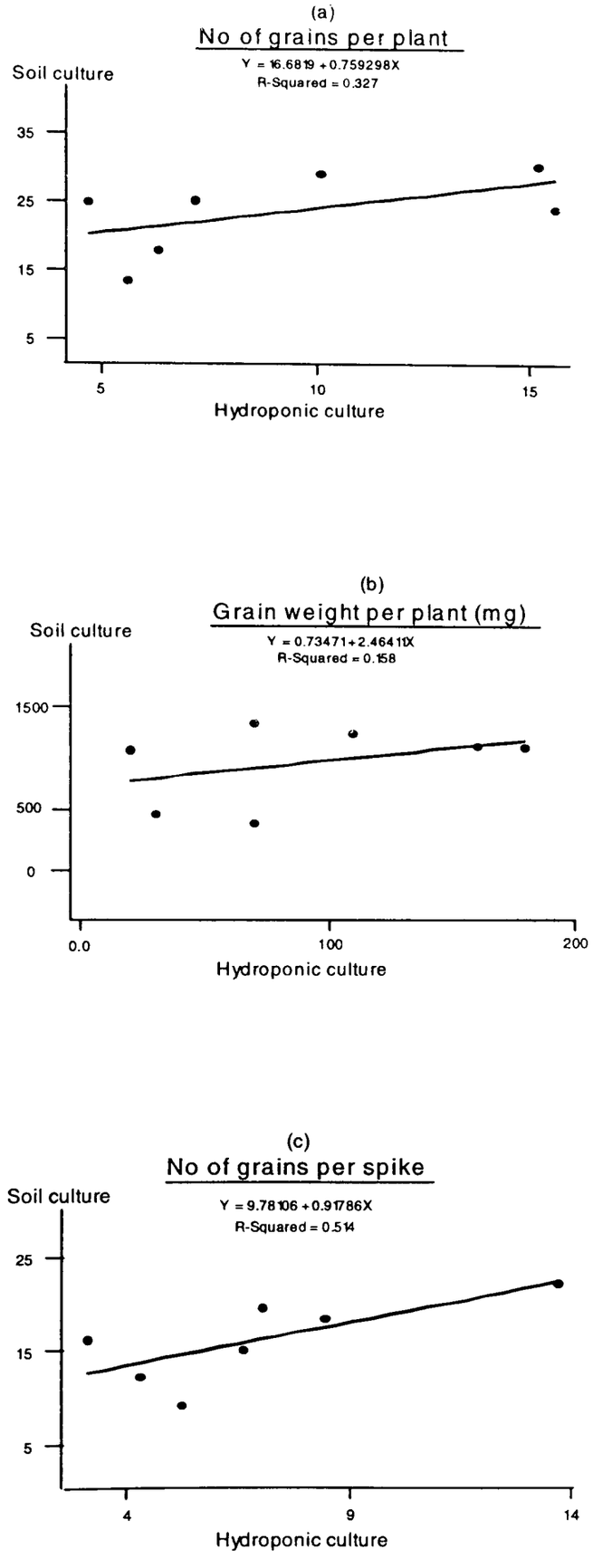


Figure 5.3. Relationships between values of certain parameters in soil and hydroponic culture under saline conditions.

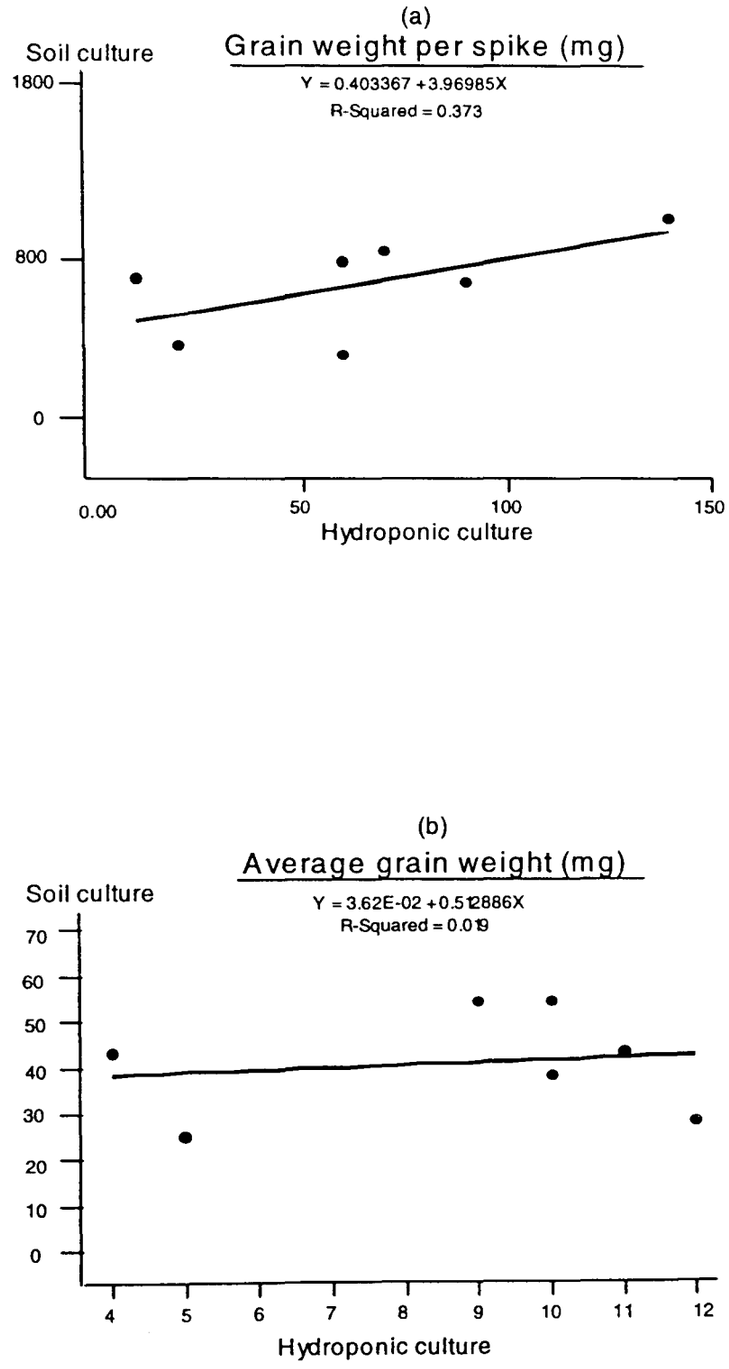


Figure 5.4. Relationships between values of certain parameters in soil and hydroponic culture under saline conditions.

Figure 5.5. Relationships between ion contents (mol m^{-3}) in hydroponic and soil culture under saline conditions.

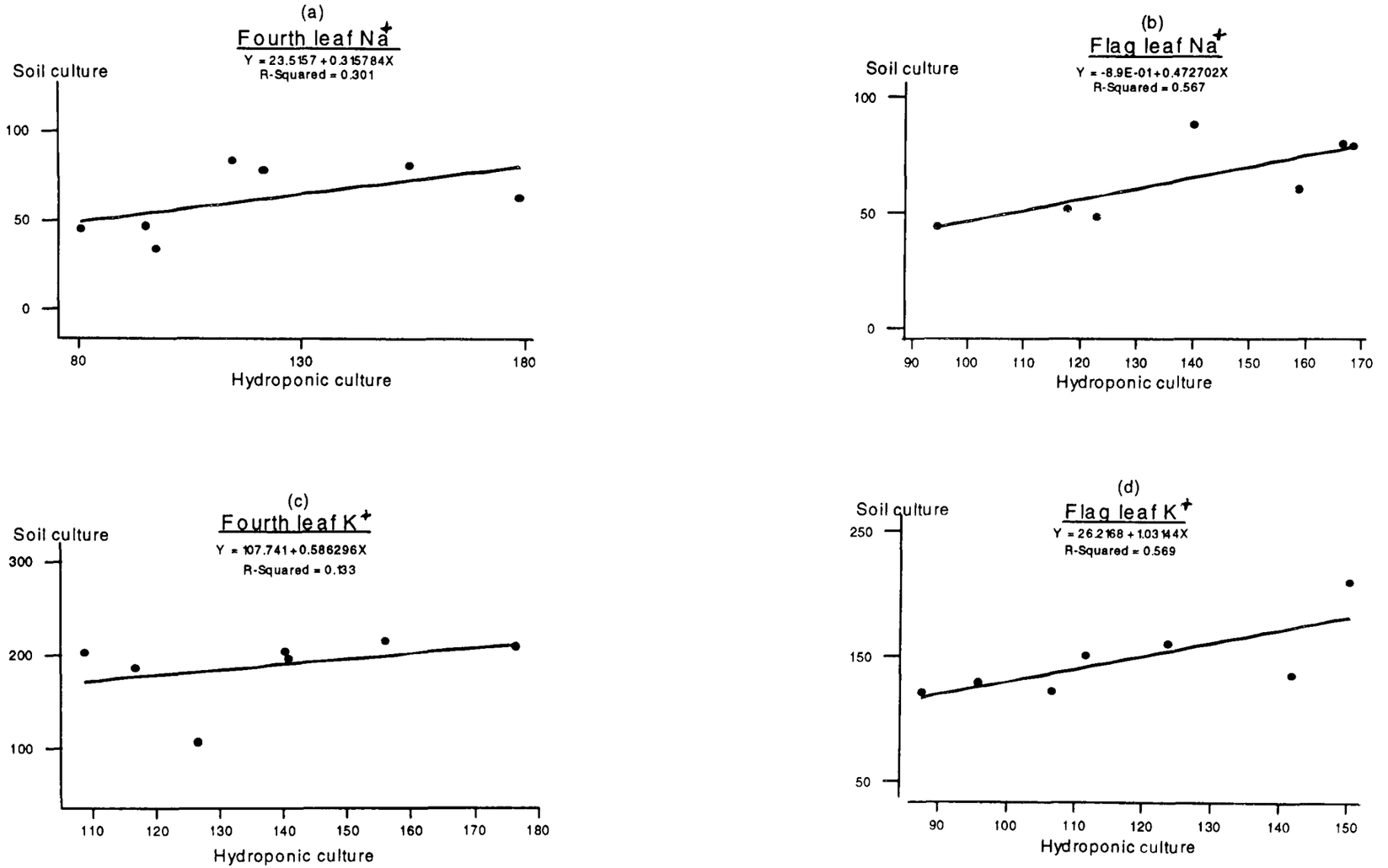


Figure 5.6. Relationships between ion contents (mol m^{-3}) in hydroponic and soil culture under saline conditions.

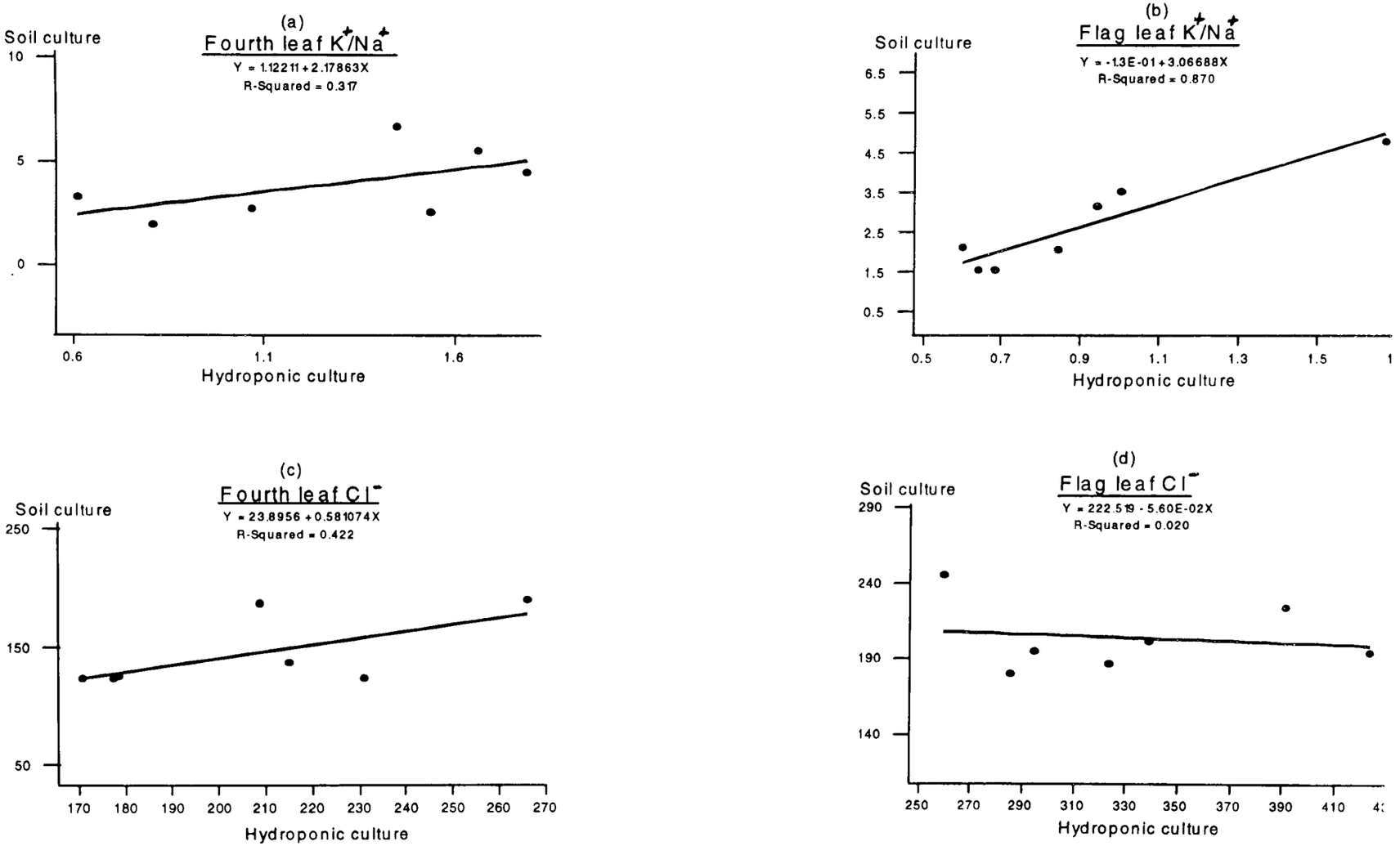
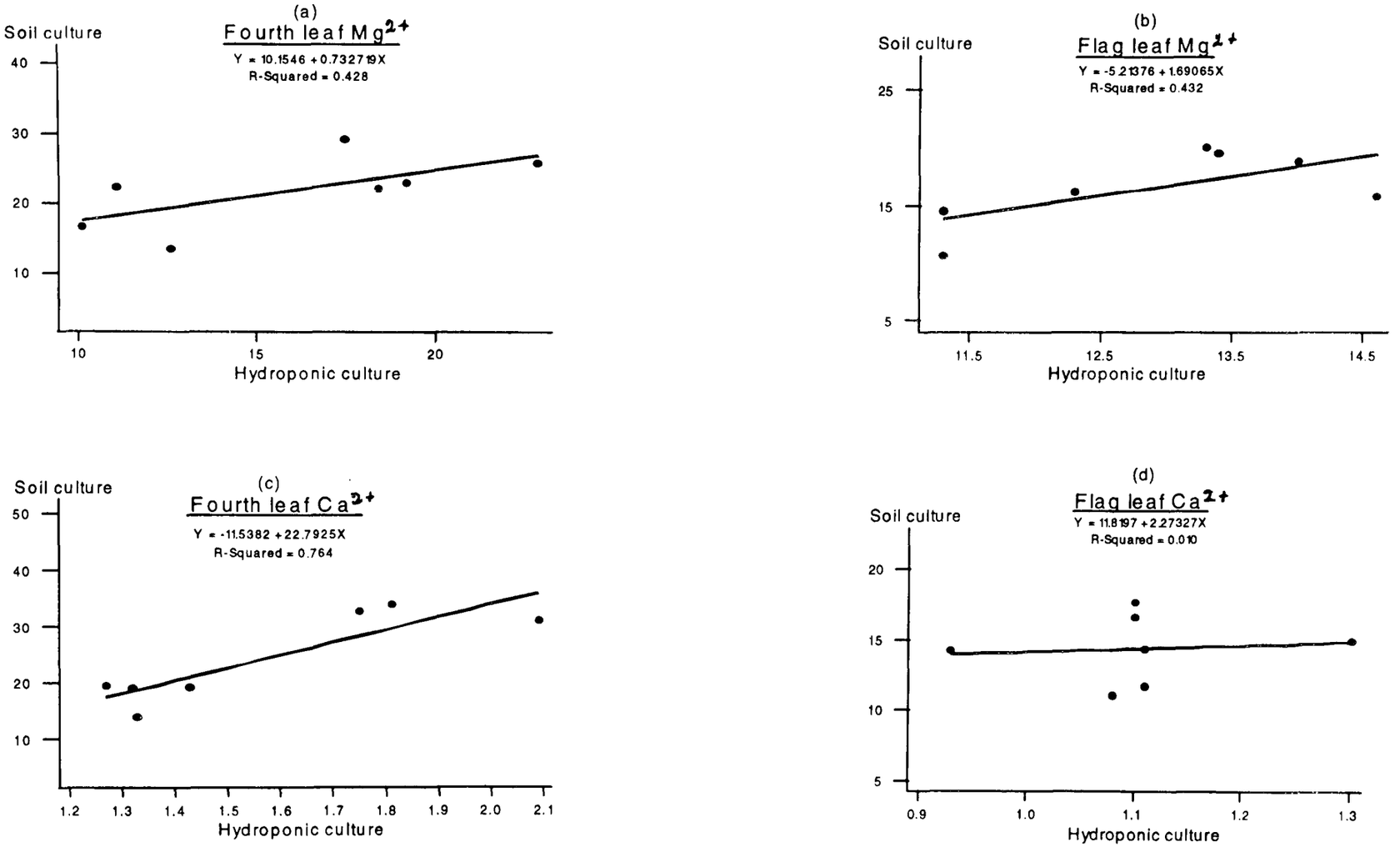


Figure 5.7. Relationships between ion contents (mol m^{-3}) in hydroponic and soil culture under saline conditions.



11.1 dS/m in soil culture, it was 10 dS/m in hydroponic culture. Therefore the better performance in soil is not due to lower EC. The observed differences in performance might be due to differences in the uptake of ions. Uptake of Na^+ and Cl^- were significantly higher in hydroponics than soil culture.

K^+, Ca^{2+} ,Mg^{2+} contents uptake were lower in plants grown in hydroponics than in soil culture. The uptake of these ions is likely to be higher as fertile field soil was used. K^+/Na^+ ratio was also found to be lower in hydroponic culture. Therefore the results suggest that amount of ion uptake in hydroponics is greater than the amount of ion uptake in soil culture. Similarly Storey (1995) reported in lime that ion uptake of plants grown in solution culture was higher than that of plants grown in sand culture.

However in hydroponic culture Ca^{2+} uptake was found to be low. Ehret *et al.* (1990) reported a greater reduction in growth and a higher incidence of foliar Ca^{2+} deficiency symptoms in wheat under hydroponic salinity. This might be responsible for the pronounced reduction in number of grains per plant and grain weight per plant in hydroponic culture which suggests post-anthesis effects. It is apparent from the literature that Ca^{2+} sometimes reduces the effects of salt (LaHaye and Epstein, 1969), supplementary Ca^{2+} improves plant growth (Hyder and Greenway, 1965; Alberico and Cramer, 1993; Cramer *et al.*, 1994a) and increases Na^+ exclusion of plant roots exposed to NaCl stress

(LaHaye and Epstein, 1971). Further roots supplied with elevated levels of Ca^{2+} are often able to maintain their K^{+} concentrations under saline conditions, whereas roots supplied with lower Ca^{2+} frequently cannot (Lauchli, 1990).

In general the relationships between yield and yield components were found to be different in hydroponic culture, soil culture and the combined data from the two systems. However number of grains per plant, number of grains per spike, grain weight per spike and average grain weight were correlated with yield in both systems. Number of grains and average grain weight were significantly and positively correlated with yield per plant. It suggested that varieties that have high values of these components under saline conditions have high yield. Similarly Sharma and Sastry (1992) also observed from their studies that tillers per meter, 100-grain weight followed by grains per ear are the most important yield determinants in wheat grown under salinity. The results of studies of yield correlations in wheat under saline conditions suggested to Matveev and Vakulenko, 1990 that high grain number per ear appeared more desirable. 1000-grain weight was positively correlated with yield in pearl millet hybrids (Dua and Bhattacharyya, 1988).

However most of the relationships between ion contents in the fourth and flag leaf were non significant in hydroponics and soil culture, but were

significant when the data from both systems was combined. This is partly due to the different range in values found in the two systems. The observation that contents of ions within leaves are not correlated suggest that they are taken up independently (except Na^+ versus K^+). However Won *et al.* (1992) reported a relatively high correlation between Na^+ and K^+ contents in rice. No significant correlations were found between yield and ion contents in hydroponic culture, except Mg^{2+}. But all correlations were found to be significant in the combined data from the two systems. There were no correlations between yield per plant and Na^+, K^+, K^+/Na^+ ratio and Cl^- except in the case of the fourth leaf in soil culture. Similar results in wheat have been reported by Ashraf and McNeilly (1988) and they proposed that whole plant performance be used for assessment of salt tolerance but in contrast Salam *et al.* (1992) reported highly significant negative correlations between Na^+, Cl^- and yield in wheat. They also reported high positive correlations between youngest leaf K^+/Na^+ ratio and yield. Further experiments are required to establish the reasons why these apparently contrasting results have been found.

Similarly most of the correlations between fourth and flag leaf ion contents were found to be non significant in hydroponic culture and soil culture. But there were significant correlations between fourth and flag leaf anion and cation contents in the combined data from the two systems. Ion

contents in the fourth leaf were not correlated with ion contents in the flag leaf. Hence although susceptible varieties have more Na^+, Cl^- and less K^+ content is not a good predictor of yield and uptake by one leaf is not a good predictor of uptake by other leaves.

5.4.2 Varietal differences

The varieties tested differed significantly in overall performance under saline conditions. SARC-III, KRL1-4 and Kharchia-65 were found to be more salt tolerant and high yielding out of seven genotypes tested. This might be due to low Na^+, Cl^-, and high K^+, Ca^{2+}, Mg^{2+} contents and high K^+/Na^+ ratio. Thus the overall performance of these varieties would seem to support the suggestion (Wyn Jones 1981) that at least to some extent, salinity tolerance may be related to an ability to restrict or control ion accumulation in shoot tissue. Sastry and Prakash (1993) reported significant differences between 8 selected wheat genotypes for Na^+ and K^+ content and increasing Na^+ over K^+ in these genotypes. Joshi and Bhoite (1988) reported all ions in decreasing order: $Cl^->Na^+>Mg^{2+}>Ca^{2+}>K^+$ in soil and in vegetative parts of the halophyte (*Aeluropus lagopoides* L.), but in contrast Albert and Popp (1977) found more K^+ uptake than Na^+ in monocotyledonous halophytes.

Although, Blue Silver was also high yielding it had higher Na^+ and K^+ contents and low K^+/Na^+ ratio. Blue Silver also had low Ca^{2+} and Mg^{2+}

contents. For screening or selection different workers (Roy, 1991; Kuiper *et al.*, 1988; Weimberg and Shannon, 1988; Falconer, 1960; Cramer *et al.*, 1994a; Matveev and Vakulenko, 1990; Greenway and Munns 1980; Sastry and Prakash, 1993) have suggested use of different traits responsible for salt tolerance, but the results of this study indicate that no single trait is enough.

5.4.3 Associations between performance in hydroponics and soil culture

Most of the relationships between the agronomic traits of the seven wheat varieties studied in hydroponic and soil culture were found non significant except straw weight per plant and number of infertile spikelets per spike. A similar trend was noted in the case of ion contents. Values in hydroponic and soil culture were found to be significantly correlated only in the cases of fourth leaf Ca^{2+} and flag leaf K^+/Na^+ ratio.

Therefore it is concluded that genotypes tested or evaluated under hydroponic salinity can behave differently under soil salinity. However it is suggested that genotypes must be tested under soil salinity before recommending for saline cultivation. Values in hydroponic culture were not correlated with values in soil culture. However hydroponic and soil culture found to be two independent systems.

It is concluded from the results that:

1- Ion content in one leaf is not a good indicator of ion content in another.

2- Ion content is not consistently correlated with grain yield per plant.

3- Good performance of variety in hydroponic culture does not imply good performance in soil. Hence breeding and evaluation of varieties for saline areas should be done under saline field conditions.

CHAPTER 6

STUDY OF VARIABILITY WITHIN THREE WHEAT VARIETIES FOR ION UPTAKE, YIELD AND YIELD COMPONENTS UNDER SALINE CONDITIONS

6.1 INTRODUCTION

Generally it is assumed that commercial wheat varieties are true breeding. However this depends on the method by which a particular variety has been developed and also on the conditions under which it has been tested. If a variety has been developed from a pure-line (by selecting a single plant) it should be true to type. However, if a variety is a multi-line and has been developed by selecting phenotypically alike plants, it may not be.

Intra-varietal variation in wheat has been reported by several workers (Joshi, 1992; Rashid, 1986; Salam, 1993; Shah ,1987; Leonard and Martin, 1963) and in rice (Flowers and Yeo, 1981).

In the present studies selections from within three wheat varieties: Alexandria (salt sensitive), Kharchia-65 (salt tolerant) and KRL1-4 (salt tolerant); were tested to estimate the effects of selfing and selection from within agronomically desirable varieties. Lines selected for high and low yield and K^+/Na^+ ratio were compared to determine the effects of selecting for these traits. Selected lines from within wheat varieties with increased salt tolerance

and high yield could be used as cultivars or as salt tolerant parents in breeding programmes. The effects of leaf detachment on yield and its components were also determined in the present studies. Determination of K^+/Na^+ ratio involves extracting sap from a detached leaf. This technique could not be used in the early stages of a breeding and selection programme if it has adverse effects on yield. However if leaf detachment has no adverse effects on the relative yields of varieties then this technique can be used without the need to discard the sampled plants from the breeding programme. It could ultimately be useful in saving time and resources.

6.2 MATERIALS AND METHODS

6.2.1 Experiment 5

Twelve single plant selections obtained from the material originally screened in Experiment 1, (Chapter 3) S_0 were tested in this experiment. Selections from within a variety were made on the basis of yield per plant and K^+/Na^+ ratio. Four lines per variety were selected within Alexandria, KRL1-4 and Kharchia-65. The actual values of yield and K^+/Na^+ ratio for these units are given in Table 3.14, Chapter 3.

Source	Selections	Selection criteria
Alexandria	(a) Alex-1	High K^+/Na^+ ratio
	(b) Alex-24	Low K^+/Na^+ ratio

	(c) Alex-3	High yield per plant
	(d) Alex-14	Low yield per plant
KRL1-4	(a) KRL-24	High K^+/Na^+ ratio
	(b) KRL-21	Low K^+/Na^+ ratio
	(c) KRL-26	High yield per plant
	(d) KRL-3	Low yield per plant
Kharchia-65	(a) Khar-1	High K^+/Na^+ ratio
	(b) Khar-5	Low K^+/Na^+ ratio
	(c) Khar-4	High yield per plant
	(d) Khar-17	Low yield per plant

The experiment was conducted in a glass-house at the University of Wales, College Farm, Aber, Bangor during the period September to January 1993. Temperature was not controlled and no supplementary lighting was used. Some panes of the glass-house were broken on 23-12-1993 due to high wind. The pots were transferred to a growth-room. A sixteen hour photoperiod was used. Average temperature during growth period was 18.3±0.40°C.

6.2.1.1 Raising the seedlings

The seeds of the twelve selections were germinated in a growth-room set at 20°C on capillary matting starting on 16-9-1993. Seedlings were transplanted into hydroponic culture on 24-9-1993. In each replicate there

were 10 plants per selection grown in a row with plant-to-plant and row-to-row distances of 3.5 cm and 6.0 cm respectively. A completely randomized design was used with three replicates. Plants were grown in pots 52 × 35 × 16 cm. Aeration was applied as mentioned in Chapter 3. Salt stress (130 mol m^{-3} NaCl) was introduced in three increments over a period of five day starting on 4-10-1993. Macro- and micro-nutrients were added to the solution following the procedure described in Chapter 3. The solution was changed in the pots was changed every 15 days.

6.2.1.2 Chemical analysis

Youngest fully-expanded leaves from two plants per selection per replication were sampled on 27-10-1993 (fourth leaf) and 10-11-1993 (sixth leaf). The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The samples were placed in Eppendorf tubes and stored in a freezer set at -10°C. Cell sap was extracted and K^+, Na^+ and Cl^- concentrations were determined as described in Chapter 5.

6.2.1.3 Final harvest

The remaining plants (6 per replicate) were harvested at maturity, on 24-01-1994 and main tiller height and number of spikes per plant were recorded. The ears were detached and straw weight per plant, infertile spikelets per spike and fertile spikelets per spike were recorded. Threshing was done by hand and

grain weight per plant and number of grains per plant were determined.

6.2.1.4 Statistical analysis

Statistical analyses were performed by using the Minitab, SYSTAT and Genstat statistical packages. Analysis of variance (ANOVA) was used to assess significant differences between the means of the selections (appendices 6.1-6.45). Where differences between means were significant ($P<0.05$) an LSD test was applied at the 5% level of significance.

6.2.2 Experiment 6

Seeds of S_0 lines harvested from Experiment 5 were multiplied and selfed by sowing in soil in pots in a green-house on 12-6-1994. Each pot was 21 × 21 × 23 cm. A solution containing macro- and micro-nutrients was applied to the pots twice during the whole period. Seeds of the second selfed generation (S_1) were harvested at maturity on 31-8-94.

Twelve S_1 selections and their parents (Alexandria, Kharchia-65 and KRL1-4 as described in experiment 2) were tested in this experiment. It was conducted in a glass-house at the University of Wales, College Farm, Aber, Bangor during the period January to May 1995. Temperature was not controlled and natural day light was supplemented by mercury vapour bulbs (model 3808 MP) to give a photoperiod of 16 hrs. Average temperature in the glass-house was 16.4±0.44°C.

6.2.2.1 Raising the seedlings

The seeds of the twelve selections and their parents were germinated on capillary matting in a growth-room set at 20°C starting on 13-1-1995. Seedlings were transplanted into hydroponic culture on 22-1-1995. There were 10 plants (1 row) per selection and 20 plants (2 rows) per parent in each of three replicates. Plant-to-plant and row-to-row distances of 3.5 cm and 6.0 cm respectively were used. A Completely Randomized Design was used. Plants were grown in 6 pots 52 × 35 × 16 cm. Salt stress (100 mol m^{-3} NaCl) was introduced in three increments over a period of five day starting on 28-1-1995. The solution in the pots was kept well aerated and changed as mentioned in Chapter 3. Macro- and micro-nutrients were added in the solution following the procedure described in Chapter 3.

6.2.2.2 Chemical analysis

Youngest fully-expanded fourth leaves from three plants per selection and five plants per parent per replication were sampled on 16-02-1995 (replication 1) and 17-02-1995 (replication 2 & 3). The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The samples were placed in Eppendorf tubes and stored in freezer set at -10°C. Cell sap was extracted and K^+, Na^+ and Cl^- concentrations were determined as described in Chapter 5.

6.2.2.3 Final harvest

All plants (those with the fourth leaf intact and fourth leaf detached) were separately harvested at maturity on 15-5-1995 (replication 2 and 3) and on 16-5-1995 (replication 1). Main tiller height and number of spikes per plant were recorded. The ears were detached and straw weight per plant, infertile spikelets per spike and fertile spikelets per spike were recorded.

Threshing was done by hand and grain weight per plant, grain weight per spike, number of grains per plant, grains per spike and average grain weight were determined.

6.2.2.4 Statistical analysis

Analysis of variance (ANOVA) was used to assess significant differences between the means of the selections (appendices 6.46-6.87). Where differences between means were significant ($P<0.05$) an LSD test was applied at the 5% level of significance. The means of plants with the fourth leaf either intact or detached were also compared using Students t test.

6.3 RESULTS

6.3.1 Experiment 5

This experiment evaluated the performance of the original S_0 selections. Overall there were very few significant differences between the

selected lines (Tables 6.1-6.6). This may be due to the limited number of plants tested.

6.3.1.1 Ion contents

There were no significant differences ($P \geq 0.05$) for Na^+, K^+, Cl^- concentrations and K^+/Na^+ ratio (fourth and sixth leaf) between Alex-1, Alex-24, Alex-3 and Alex-14 (Table 6.1). There were also no significant differences ($P \geq 0.05$) for ion contents and K^+/Na^+ ratio (fourth and sixth leaf) between Khar-1, Khar-5, Khar-4 and Khar-17 (Table 6.2). Similar results were found between KRL-24, KRL-21, KRL-26 and KRL-3 except that in the sixth leaf KRL-21 had significantly higher ($P < 0.05$) Cl^- concentrations than the KRL-24, KRL-3 and KRL-26 (Table 6.3).

Even though there were no significant differences ($P \geq 0.05$) between selections for ion concentrations the behaviour of most of the selected lines was true to selection, expect Khar-1 and Khar-5. Lines selected for high K^+/Na^+ ratio had high K^+/Na^+ ratio and lines selected for low K^+/Na^+ ratio had low K^+/Na^+ ratio.

6.3.1.2 Yield and yield components

Alex-3 had higher yield than Alex-14 but Alex-1 had significantly higher ($P < 0.05$) yield then all selections (Table 6.4). There were no significant differences ($P \geq 0.05$) for all other yield components. Differences in yield

between selections were mainly due to differences in number of grains per plant.

There were no significant differences ($P \geq 0.05$) for yield and yield components between the four selected Kharchia-65 lines (Table 6.5).

KRL-26 had significantly higher ($P < 0.05$) yield than KRL-24, and KRL-21. This is because KRL-26 had more fertile spikelets per spike, fewer infertile spikelets per spike and more grains per plant. Main tiller height was higher and straw weight per plant was greater in KRL-26 than in KRL-3. (Table 6.6). KRL-24 also had significantly higher ($P < 0.05$) yield than KRL-21 and KRL-3.

6.3.2 Experiment 6

This experiment evaluated the performance of the S_1 lines, obtained by selfing the orignial selections.

6.3.2.1 Comparison between plants with fourth leaf detached and fourth leaf undetached

Yield and yield components of plants with and without the fourth leaf were compared. No significant differences were found in Alexandria (Table 6.7). In KRL1-4 there were no significant differences except in straw weight per plant which was significantly greater in plants with the fourth leaf (Table 6.8). There were no significant differences (in Kharchia-65 (Table 6.9).

Table 6.1. Means and S.E of leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio under saline conditions of four inbred lines selected from Alexandria wheat.

Trait	Alex-3 High yield		Alex-14 Low yield		Alex-1 High K^+/Na^+		Alex-24 Low K^+/Na^+		
Fourth leaf	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+	88	15.3	96	7.6	122	42.9	106	16.2	NS
K^+	206	28.3	167	1.0	229	34.4	135	5.6	NS
K^+/Na^+	2.4	0.3	1.8	0.1	2.2	0.4	1.3	0.2	NS
Cl^-	172	26.5	189	24.0	193	13.5	244	48.3	NS
Sixth leaf									
Na^+	108	19.2	104	4.1	127	13.0	111	34.5	NS
K^+	172	10.9	165	8.9	204	21.0	177	1.5	NS
K^+/Na^+	1.7	0.5	1.6	0.1	1.6	0.02	1.8	0.5	NS
Cl^-	192	14.5	204	14.7	197	6.5	230	18.5	NS

NS = $P > 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.1-6.8.

Table 6.2. Means and S.E of leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio under saline conditions of four inbred lines selected from Kharchia-65 wheat.

Trait	Khar-4 High yield		Khar-17 Low yield		Khar-1 High K^+/Na^+		Khar-5 Low K^+/Na^+		
Fourth leaf	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+	141	22.8	169	39.9	97	17.9	85	16.0	NS
K^+	173	6.4	165	13.3	171	11.8	172	12.7	NS
K^+/Na^+	1.3	0.2	1.1	0.3	1.8	0.2	2.2	0.5	NS
Cl^-	212	10.9	241	8.7	202	22.9	220	17.2	NS
Sixth leaf									
Na^+	108	2.5	132	13.6	149	18.3	150	37.3	NS
K^+	204	41.5	155	15.3	168	24.7	162	9.9	NS
K^+/Na^+	1.9	0.4	1.2	0.1	1.2	0.2	1.2	0.3	NS
Cl^-	226	8.0	201	22.9	204	13.3	251	15.6	NS

NS = $P > 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.16-6.23.

Table 6.3. Means and S.E of leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio under saline conditions of four inbred lines selected from KRL1-4 wheat.

Trait	**KRL-26 High yield**		**KRL-3 Low yield**		**KRL-24 High K^+/Na^+**		**KRL-21 Low K^+/Na^+**		
Fourth leaf	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+	88	21.9	116	3.3	78	26.5	97	22.3	NS
K^+	192	10.7	162	11.3	163	11.0	164	21.5	NS
K^+/Na^+	2.5	0.7	1.4	0.1	2.6	0.7	1.9	0.4	NS
Cl^-	212	22.0	250	41.9	192	36.4	227	20.7	NS
Sixth leaf									
Na^+	79	15.4	129	10.7	118	22.5	197	48.1	NS
K^+	172	8.7	171	5.9	179	8.9	184	8.2	NS
K^+/Na^+	2.4	0.6	1.3	0.1	1.6	0.3	1.1	0.31	NS
Cl^-	192	3.3	203	5.8	221	11.6	282	19.2	33.0*

NS = $P > 0.05$

* = $P < 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.31-6.38

Table 6.4. Means and S.E of yield per plant and various yield components of four inbred lines selected from Alexandria wheat variety.

Trait	**Alex-3 High yield**		**Alex-14 Low yield**		**Alex-1 High K^+/Na^+**		**Alex-24 Low K^+/Na^+**		
	Means	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Grain weight per plant (mg)	39	1.8	12	3.9	71	1.0	14	5.8	29.7*
Main tiller height (cm)	66.0	1.4	58.4	1.5	69.7	0.7	60.2	4.6	NS
No of spikes per plant	1.2	0.2	1.2	0.1	1.3	0.04	1.1	0.1	NS
Straw weight per plant (g)	2.2	0.5	2.0	0.5	2.2	0.3	1.9	0.2	NS
Infertile spikelets per spike	2.1	0.4	1.8	0.2	0.7	0.1	1.7	0.4	NS
Fertile spikelets per spike	18.1	0.6	17.5	0.3	19.0	1.1	17.7	1.4	NS
No of grains per plant	13.0	4.8	5.7	3.1	17.8	1.5	7.6	3.0	NS

NS = $P > 0.05$

* = $P < 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.9-6.15.

Table 6.5. Means and S.E of yield per plant and various yield components of four inbred lines selected from Kharchia-65 wheat variety.

Trait	Khar-4 High yield		Khar-17 Low yield		Khar-1 High K^+/Na^+		Khar-5 Low K^+/Na^+		
	Means	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Grain weight per plant (mg)	58	28.0	53	19.0	89	22.0	68	54.0	NS
Main tiller height (cm)	57.5	1.8	63.5	4.0	64.1	1.3	63.2	2.7	NS
No of spikes per plant	1.3	0.2	1.4	0.4	1.7	0.3	1.6	0.4	NS
Straw weight per plant (g)	0.9	0.1	0.9	0.1	1.0	0.2	1.3	0.3	NS
Infertile spikelets per spike	1.2	0.04	1.0	0.2	1.4	0.4	1.3	0.2	NS
Fertile spikelets per spike	10.5	0.1	10.8	0.2	11.0	0.5	11.3	0.3	NS
No of grains per plant	9.3	4.2	8.2	2.3	12.0	4.3	8.8	4.9	NS

NS = $P > 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.24-6.30.

Table 6.6. Means and S.E of yield per plant and various yield components of four inbred lines selected from KRL1-4 wheat variety.

Trait	KRL-26 High yield		KRL-3 Low yield		KRL-24 High K^+/Na^+		KRL-21 Low K^+/Na^+		
	Means	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Grain weight per plant (mg)	389	40.0	147	55.0	287	33.0	214	40.0	119.6*
Main tiller height (cm)	69.8	0.5	55.7	5.1	67.8	1.2	64.1	2.4	8.2*
No of spikes per plant	1.2	0.1	1.1	0.1	1.0	0.0	1.0	0.0	NS
Straw weight per plant (g)	0.9	0.1	0.4	0.0	0.7	0.1	0.6	0.1	0.2*
Infertile spikelets per spike	1.7	0.2	2.8	0.1	1.9	0.3	2.4	0.3	0.7*
Fertile spikelets per spike	13.1	0.04	9.3	0.9	12.3	0.3	10.8	0.8	1.7*
No of grains per plant	39.1	3.7	17.0	4.3	30.6	1.5	22.3	5.7	11.6*

NS = $P > 0.05$

* = $P < 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.39-6.45.

Table 6.7. Means, S.E. for yield and yield components of plant with and without fourth leaf in Alexandria wheat (combined data from parents and selections).

Trait	Detached		Undetached			
	Means	±S.E	Means	±S.E	t test	df
Grain weight per plant (mg)	119.7	22.8	114.9	9.0	0.19NS	43
Main tiller height (cm)	48.2	2.1	52.8	1.1	-1.95NS	52
No of spikes per plant	1.1	0.09	1.1	0.03	0.07NS	42
Straw weight per plant (g)	0.6	0.01	0.7	0.03	-0.34NS	40
Infertile spikelets per spike	2.2	0.3	2.2	0.1	-0.14NS	52
Fertile spikelets per spike	11.8	0.5	11.7	0.3	0.11NS	64
No of grains per plant	13.3	2.3	12.5	1.0	0.31NS	44
No of grains per spike	11.4	1.6	11.6	0.9	-0.15NS	55
Grain weight per spike (mg)	101.0	16.1	106.2	8.7	-0.29NS	53
Average grain weight (mg)	6.4	0.7	7.4	0.5	-1.16NS	62

NS = $P > 0.05$.

Table 6.8. Means, S.E. for yield and yield components of plants with and without fourth leaf in KRL1-4 wheat (combined data from parents and selections).

Trait	Detached		Undetached			
	Means	±S.E	Means	±S.E	t test	df
Grain weight per spike (mg)	350.8	23.2	343.0	17.1	0.27NS	94
Main tiller height (cm)	65.4	1.2	66.6	0.9	-0.82NS	97
No of spikes per plant	1.1	0.03	1.2	0.04	-0.83NS	77
Straw weight per plant (g)	0.6	0.03	0.7	0.02	-2.02*	115
Infertile spikelets per spike	2.8	0.2	2.5	0.1	1.85NS	82
Fertile spikelets per spike	11.1	0.3	11.0	0.2	0.13NS	102
No of grains per plant	25.5	1.5	26.7	1.2	-0.65NS	102
Grain weight per plant (mg)	386.2	26.1	399.1	20.4	-0.39NS	99
No of grains per spike	23.8	1.6	23.1	1.1	0.34NS	88
Average grain weight (mg)	15.7	0.8	15.0	0.4	0.70NS	40

NS = $P > 0.05$
* = $P < 0.05$

Table 6.9. Means, S.E. for yield and yield components of plants with and without fourth leaf in Kharchia-65 wheat (combined data from parents and selections).

Trait	Detached		Undetached			
	Means	±S.E	Means	±S.E	t test	df
Grain weight per plant (mg)	310.0	34.6	297.1	26.9	0.29NS	107
Main tiller height (cm)	65.8	1.8	66.8	1.2	-0.49NS	92
No of spikes per plant	1.7	0.1	1.8	0.1	-0.63NS	94
Straw weight per plant (g)	0.8	0.1	0.8	0.1	-0.32NS	98
Infertile spikelets per spike	1.1	0.1	1.2	0.1	-0.81NS	116
Fertile spikelets per spike	9.5	0.2	9.4	0.2	0.08NS	138
No of grains per plant	23.9	2.4	24.8	1.7	-0.31NS	96
No of grains per spike	13.5	0.9	13.4	0.5	0.04NS	82
Grain weight per spike (mg)	173.0	14.1	152.9	9.8	1.16NS	95
Average grain weight (mg)	12.6	0.8	10.8	0.5	1.91NS	97

NS = $P > 0.05$

6.3.2.2 Selections within Alexandria (Table 6.10 and 6.11)

There was a significant difference ($P<0.05$) in K^+/Na^+ ratio between Alex-1 (high K^+/Na^+ ratio) and Alex-24 (low K^+/Na^+ ratio). This was due to lower Na^+ and higher K^+ uptake by Alex-1. There were no significant differences ($P\geq0.05$) in Cl^- uptake between Alex-1 and Alex-24. There were also no significant differences ($P\geq0.05$) in Na^+, K^+, Cl^- uptake and K^+/Na^+ ratio between the Alexandria parent and selections Alex-1 and Alex-24.

No significant differences ($P\geq0.05$) in Na^+, K^+, Cl^- ion contents and K^+/Na^+ ratio were found between the Alexandria parent, Alex-3 (high yield) and Alex-14 (low yield).

There were no significant differences ($P\geq0.05$) in yield per plant between the Alexandria parent, Alex-3 (high yield) and Alex-14 (low yield) and also no significant differences ($P\geq0.05$) for any other parameter. Grain weight per plant was low due to low average grain weight and number of grains per plant.

Although Alex-1 (high K^+/Na^+ ratio) had higher yield per plant and greater number of grains per plant than Alex-24 (low K^+/Na^+ ratio) and the parent, but the differences were not significant ($P\geq0.05$) for yield and any of its components. Alex-1 (high K^+/Na^+ ratio) had a significantly greater ($P\leq0.01$) number of fertile spikelets per spike than Alex-24.

Table 6.10. Means, S.E. of fourth leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio under saline conditions of Alexandria and selections within Alexandria variety.

Trait	Parent		Selections								
	Alexandria		Alex-3 High yield		Alex-14 Low yield		Alex-1 High K^+/Na^+		Alex-24 Low K^+/Na^+		
	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Na^+	248	10.4	254	10.9	249	11.7	205	8.9	254	13.7	NS
K^+	125	7.6	122	5.2	112	5.4	139	7.4	113	8.6	NS
K^+/Na^+	0.5	0.04	0.5	0.03	0.5	0.03	0.7	0.06	0.4	0.03	0.1*
Cl^-	380	7.9	360	10.8	379	9.8	327	12.8	366	7.5	NS

NS = $P > 0.05$

* = $P < 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.46-6.49.

Table 6.11. Means, S.E. of yield per plant and various yield components under saline conditions of Alexandria and selections within Alexandria variety.

Trait	Parent		Selections								
	Alexandria		Alex-3 High yield		Alex-14 Low yield		Alex-1 High K^+/Na^+		Alex.24 Low K^+/Na^+		
	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Grain weight per plant (mg)	89	38.2	90	6.5	80	7.9	202	48.4	67	18.8	NS
Main tiller height (cm)	48.2	5.0	48.4	3.6	49.4	2.5	59.5	1.7	42.1	4.6	NS
No of spikes per plant	1.0	0.02	1.0	0.03	1.0	0.01	1.3	0.2	1.1	0.1	NS
Straw weight per plant (g)	0.5	0.1	0.6	0.1	0.5	0.1	0.9	0.1	0.5	0.1	NS
Infertile spikelets per spike	2.9	0.9	1.8	0.4	1.9	0.1	1.7	0.4	3.2	0.3	NS
Fertile spikelets per spike	10.6	0.7	11.7	0.4	12.7	0.4	14.2	0.2	9.2	1.2	1.7**
No of grains per plant	9.5	4.2	11.7	0.9	10.5	1.8	22.0	5.6	6.8	2.1	NS
No of grain per spike	9.4	4.3	11.4	1.1	10.5	1.8	16.6	1.5	6.6	2.3	NS
Grain weight per spike (mg)	88	38.7	87	5.8	80	7.9	153	11.3	64	21.5	NS
Average grain weight (mg)	9.4	0.3	7.8	0.6	7.7	0.5	9.2	0.1	10.4	1.3	NS

NS = $P > 0.05$
** = $P < 0.01$

6.3.2.3 Selection within Kharchia-65 (Table 6.12 and 6.13)

Khar-1 (high K^+/Na^+ ratio) had significantly higher ($P<0.05$) K^+ uptake and K^+/Na^+ ratio than Khar-5 (low K^+/Na^+ ratio). However there were also significant differences ($P<0.05$) in K^+/Na^+ ratio between the Kharchia-65 parent and these selections. There were also no significant ($P\geq0.05$) differences in Na^+, K^+, Cl^- uptake and K^+/Na^+ ratio between the Kharchia-65 parent and selections Khar-4 (high yield).

Khar-4 (high yield) had a higher grain weight per plant than Khar-17 (low yield) and the parent but differences were not significant ($P\geq0.05$). There were also no significant differences ($P\geq0.05$) for other yield components between the Kharchia-65 parent and Khar-4 (high yield) and Khar-17 (low yield). Yield per plant and other yield components were not significantly different ($P\geq0.05$) between parent and selections Khar-1 (high K^+/Na^+ ratio) and Khar-5 (low K^+/Na^+ ratio).

6.3.2.4 Selection within KRL1-4 (Table 6.14 and 6.15)

There were no significant differences ($P\geq0.05$) in K^+/Na^+ ratio and ion contents between the KRL1-4 parent and selections KRL-24 (high K^+/Na^+ ratio) and KRL-21 (low K^+/Na^+ ratio). Similarly Na^+, K^+, Cl^- uptake and K^+/Na^+ ratio were not significantly different ($P\geq0.05$) between the KRL1-4 parent and selections KRL-26 (high yield) and KRL-3 (low yield) but KRL-26

Table 6.12. Means, S.E. of fourth leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio under saline conditions of Kharchia-65 and selections within Kharchia-65 variety.

Trait	Parent		Selections								
	Kharchia-65		Khar-4 High yield		Khar-17 Low yield		Khar-1 High K^+/Na^+		Khar-5 Low K^+/Na^+		
	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Na^+	197	9.4	186	13.8	179	8.1	181	6.0	217	11.6	NS
K^+	139	8.0	151	6.9	155	10.1	157	11.9	128	4.1	13.8*
K^+/Na^+	0.7	0.06	0.8	0.07	0.9	0.06	0.9	0.07	0.6	0.03	0.1**
Cl^-	282	9.3	277	18.2	271	14.6	270	11.2	288	8.6	NS

NS = $P > 0.05$

* = $P < 0.05$

** = $P < 0.01$

Note: Analyses of variance for these traits are presented in appendices 6.60-6.63.

Table 6.13. Means, S.E. of yield and various yield components under saline conditions of Kharchia-65 and selections within Kharchia-65 variety.

Trait	Parent		Selections								
	Kharchia-65		Khar-4 High yield		Khar-17 Low yield		Khar-1 High K^+/Na^+		Khar-5 Low K^+/Na^+		
	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Grain weight per plant (mg)	241	86	386	135	337	162	363	125	233	99.3	NS
Main tiller height (cm)	63.7	2.0	71.2	7.1	67.9	5.6	67.7	4.9	64.5	5.6	NS
No of spikes per plant	1.8	0.3	1.9	0.3	1.7	0.5	1.6	0.2	1.7	0.3	NS
Straw weight per plant (g)	0.8	0.2	0.9	0.2	0.8	0.3	0.8	0.2	0.7	0.2	NS
Infertile spikelets per spike	1.4	0.3	0.9	0.1	1.1	0.1	1.0	0.1	1.3	0.3	NS
Fertile spikelets per spike	9.5	0.2	9.3	0.3	9.7	0.5	9.6	0.4	8.8	0.2	NS
No of grains per plant	22.9	7.6	29.6	7.8	25.6	9.7	24.4	8.1	21.7	8.3	NS
No of grain per spike	12.1	2.3	15.4	2.0	14.4	1.2	14.5	3.0	11.6	2.4	NS
Grain weight per spike (mg)	125	29.3	194	48.6	180	45.3	215	56.1	122	32.6	NS
Average grain weight (mg)	10.1	0.8	12.4	2.6	12.3	2.6	14.5	2.1	10.2	0.9	NS

NS = $P > 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.64-6.73.

Table 6.14. Means, S.E. of fourth leaf ion contents (mol m^{-3}) and K^+/Na^+ ratio under saline conditions of KRL1-4 and selections within KRL1-4 variety.

Trait	**Parent**		**----------Selections----------**								
	KRL1-4		**KRL-26 High yield**		**KRL-3 Low yield**		**KRL-24 High K^+/Na^+**		**KRL-21 Low K^+/Na^+**		
	Means	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+	204	9.5	184	8.4	218	10.4	189	9.6	177	12.2	NS
K^+	149	9.2	167	11.9	122	6.5	152	10.6	145	7.9	NS
K^+/Na^+	0.7	0.05	0.9	0.09	0.6	0.06	0.8	0.09	0.9	0.09	NS
Cl^-	300	8.6	273	10.1	314	12.7	275	10.9	284	6.9	NS

NS = $P > 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.74-6.77.

Table 6.15. Means, S.E. of yield and various yield components under saline conditions of KRL1-4 and selections within KRL1-4 variety.

Trait	Parent		Selections								
	KRL1-4		KRL-26 High yield		KRL-3 Low yield		KRL-24 High K^+/Na^+		KRL-21 Low K^+/Na^+		
	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	Means	±S.E	LSD
Grain weight per plant (mg)	373	37.2	439	14.1	278	76.7	422	37.0	481	65.5	NS
Main tiller height (cm)	67.8	4.4	66.9	3.2	63.0	4.1	65.5	5.5	64.9	3.0	NS
No of spikes per plant	1.2	0.08	1.1	0.03	1.2	0.09	1.3	0.2	1.2	0.1	NS
Straw weight per plant (g)	0.7	0.1	0.6	0.1	0.5	0.1	0.8	0.1	0.7	0.1	NS
Infertile spikelets per spike	2.6	0.5	2.4	0.2	2.8	0.5	2.1	0.5	2.9	0.2	NS
Fertile spikelets per spike	10.8	1.0	10.7	0.6	10.2	0.7	12.1	0.5	11.3	0.8	NS
No of grains per plant	26.3	4.6	26.4	2.9	21.2	5.6	28.8	5.0	28.5	7.0	NS
No of grain per spike	23.1	4.5	24.7	3.1	18.2	4.6	24.7	7.2	24.6	6.8	NS
Grain weight per spike (mg)	322	15.8	413	20.1	235	58.7	355	70.4	407	56.9	NS
Average grain weight (mg)	14.8	2.0	16.9	1.4	13.0	1.1	15.1	1.3	17.8	2.2	NS

NS = $P > 0.05$

Note: Analyses of variance for these traits are presented in appendices 6.78-6.87.

had higher yield than KRL-3.

There were no significant differences ($P \geq 0.05$) in yield per plant and yield components between KRL1-4 parent and selections KRL-26 (high yield) and KRL-3 (low yield).There were also no significant differences ($P \geq 0.05$) in yield per plant and other yield components between KRL1-4 Parent and KRL-24 (high K^+/Na^+ ratio) and KRL-21(low K^+/Na^+ ratio).

6.3.3 Average grain weight

In both experiments average grain weight was low. This was due to increased temperatures during the grain filling period and plants were tested under complete salt stress. The fact that no supplementary Ca was added to the nutrient solution as mentioned in Chapter 3 may have aggravated the salinity effect.

Maximum temperature exceeded 30°C in experiment 5 (Figure 6.1 a) and approached 40°C in experiment 6 (Figure 6.1 b). Salt sensitivity in plants increases with temperature due to enhanced uptake of ions and decreased plant growth (Oertli, 1960). Gale (1975) also found that plant growth under salt stress was sensitive to air temperature. Na^+-K^+ imbalance also adversely affects grain yield (Devitt *et al.*1981). The decreased average grain weight ultimately resulted in plants having lower yield. Harvest index in cereals is often around 50%, but can be decreased by increasing salinity (Iqbal, 1992; Torres and Binghum, 1973). On the basis of this, and using a straw weight of 800 mg per plant, a grain weight around 400 mg per plant might have expected (Table 5.2, Chapter 5).

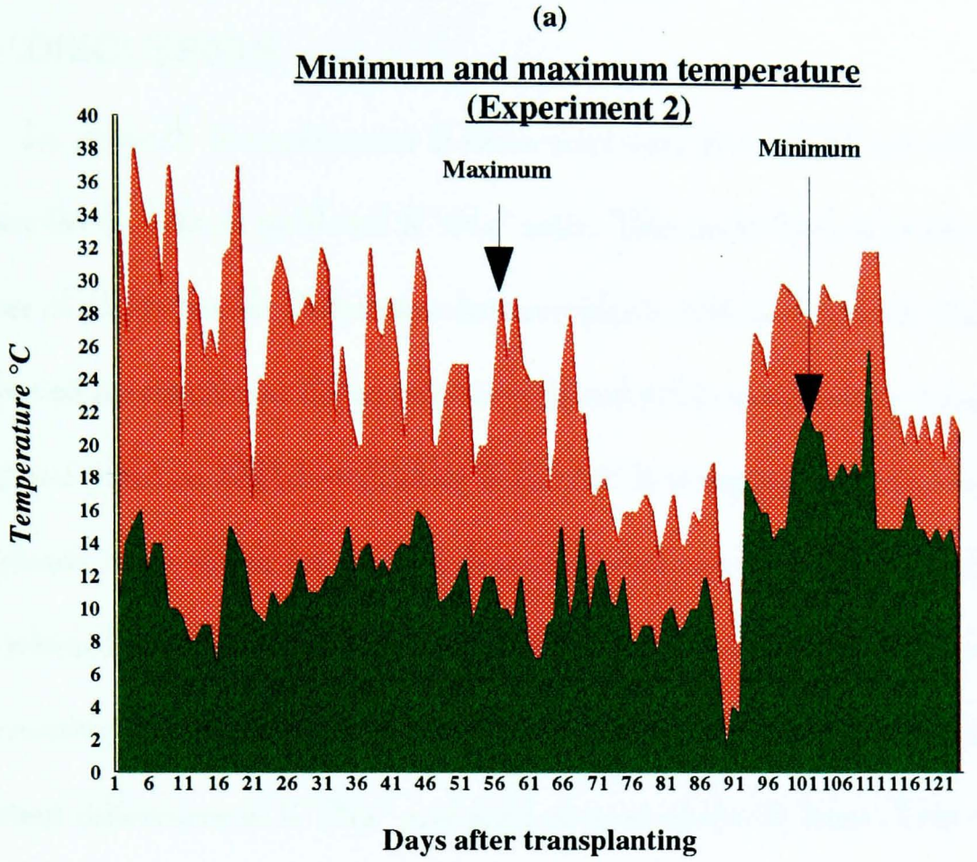


Figure 6.1. Minimum and maximum temperature during growth period of wheat under saline conditions.

6.4 DISCUSSION

In general, in experiment 5 there were very few significant differences between the S_0 lines in yield and K^+/Na^+ ratio. This might be due to the limited number of plants tested. Differences between plants with and without the fourth leaf showed no significant differences in yield and yield components except straw weight per plant in KRL1-4 (Tables 6.7-6.9). It is suggested from the results that plants from which leaves have been sampled can be included with plants from which leaves have not been sampled for yield comparison. In experiment 6 increasing the number of plants tested resulted in more pronounced and consistent differences in K^+/Na^+ and yield of most of the S_1 lines. This suggests that a greater number of plants needed to be tested during such studies. Increasing the number of plants, reduced the experimental error. Selection and selfing also increased the yield and K^+/Na^+ ratio in most of the selected lines.

6.4.1 Effects of selecting and selfing for K^+/Na^+ ratio on K^+/Na^+ ratio

In Alexandria wheat selecting S_0 lines for high and low K^+/Na^+ ratio resulted in plants with differing K^+/Na^+ ratios, but the differences were not significant for any leaf and the trends were not consistent in the sixth leaf (Table 6.1). Selfing S_0 lines resulted in a pronounced and consistent increase in K^+/Na^+ ratio in Alexandria (Table 6.10).

In Kharchia-65 K^+/Na^+ ratio did not follow the expected trend in the selected S_0 lines (Table 6.2). However after selfing, Khar-1 (high K^+/Na^+ ratio) and Khar-5 (low K^+/Na^+ ratio) lines trends in K^+/Na^+ ratio and Na^+ uptake

followed the expected trend (Table 6.12).

In KRL1-4 K^+/Na^+ ratios followed the expected trend in selected S_0 lines but not after selfing (Table 6.3 and 6.14). Yeo *et al.* (1988) reported similar inconsistencies in Na^+ uptake in rice varieties. They isolated and selfed lines with high and low Na^+ transport rate and reported that lines selected for low and high Na^+ concentrations did not show consistency from the S_1 to S_2 generation. In later generations from S_4 to S_5, they found clear and consistent trends showing that 90% of the progeny of plants with low Na^+ parents had low Na^+ contents and plants selected for high Na^+ produced progeny with high Na^+ concentrations. Therefore the lines tested in these experiments should be selfed to determine their clear response to selection and selfing in later generations.

6.4.2 Effects of selecting and selfing for yield on yield

The varieties differed in their response to selection and selfing. The effects of variety type on responses to selection will be discussed in the general discussion (Chapter 8). In Alexandria selecting plants for high yield produced progeny with high yield while plants selected for low yield produced progeny having low yield (Tables 6.4). However trends in yield between lines were not consistent from S_0 to S_1, so that differences between S_1 lines were not significant (Tables 6.4 and 6.11).

A similar trends were evident in Kharchia-65 and the differences were smaller and not significant (Table 6.5 and 6.13). Similar trend was found in KRL1-4. Plants selected for high yield produced progeny with high yield and plants selected for low yield had low yielding progeny. Differences in yield

between the lines were significant in S_0 but not in S_1 (Tables 6.6 and 6.15). Different workers have reported different responses to selection from within varieties, Joshi (1992) reported highly significant differences in grain yield and its attributes under saline conditions in Kharchia collections. However Weltzien and Fischbeck (1990) tested homozygous lines of barley under drought and dry land salinity stress and reported greater variation among yield components between than within populations.

6.4.3 Relative increases in yield as a result of selecting for yield or K^+/Na^+ ratio

The results gave no clear indication as to whether it is better to select for yield or K^+/Na^+ ratio. The Alexandria S_0 lines selected for (Alex-1) high K^+/Na^+ ratio had higher yield than lines selected with high yield and this trend was consistent from S_0 to S_1 generation (Table 6.4 and 6.11).

In Kharchia-65 the S_0 line selected with high K^+/Na^+ ratio (Khar-1) produced higher yield than the line selected with high yield (Khar-4). This trend was not clear and not consistent from S_0 to S_1 generation (Table 6.5 and 6.13).

In KRL1-4 the S_0 line selected for high yield (KRL-26) produced relatively higher yielding progeny than the line selected for high K^+/Na^+ ratio (KRL-24). In S_1 the low K^+/Na^+ ratio selection (KRL-21) gave higher yield (Tables 6.6 and 6.15). It is suggested that further selfing to later generations is required to find out whether it is best to select for yield or K^+/Na^+ ratio under saline conditions.

6.4.4 CONCLUSIONS

It is generally concluded from the performance of these two selfed generations of Alexandria, Kharchia-65 and KRL1-4 that there is genetic variation in K^+/Na^+ ratio and grain yield within these three wheat varieties under saline conditions.

Therefore, there is a possibility to select lines from within these varieties with high K^+/Na^+ ratio and or high yield per plant. The selected lines could be produced with high K^+/Na^+ ratio and / or high yield by continuous selection and selfing in successive generations. These lines could be utilised for cultivation on salt affected soils. They could also be used in a breeding programme to improve yield and enhance K^+/Na^+ ratio and side by side to produce genetic information of some physiological and agronomic aspects, which are very important for plant breeding strategies to evolve varieties with increased salt-tolerance.

There are also two possibilities suggested from the results which could be tested in further experiments involving a large number of plants.

1) KRL1-4 is already salt-tolerant. Can greatest improvement be achieved by selecting for yield?

2) Alexandria and Kharchia are less salt-tolerant. Can greatest improvement be achieved by selecting for K^+/Na^+ ratio or another character associated with increased salt-tolerance?

CHAPTER 7

GENETICAL ANALYSIS OF SALT TOLERANCE IN SPRING WHEAT (*Triticum aestivum* L.)

7.1 INTRODUCTION

Crop plant responses to salt stress including aspects of growth, development, and yield have been well documented as described in Chapter 2. For successful increases in plant salt tolerance, breeding and selection techniques can be used (Epstein *et al.,* 1980). For this to be achieved the traits associated with salt tolerance should be genetically controlled and potentially heritable (Shannon, 1984). In addition patterns of inheritance (qualitative and or quantitative), the number of genes contributing to salt tolerance and the nature of gene action should be known.

Salt tolerance in wheat is associated with accumulation of inorganic ions (Na^+, K^+ and Cl^-). Salam *et al.* (1992) found a highly significant negative correlation between Na^+ and Cl^- contents and yield. Youngest leaf K^+/Na^+ ratio showed a very high positive correlation with yield and its components. They concluded that salt tolerance was under genetic control. Gorham and Wyn Jones (1990) reported that high leaf K^+/Na^+ ratio has been associated with salt tolerance and this character is genetically controlled in durum wheat and they also reported development of most promising lines from Chinese Spring ×

Agropyron junceum [*Elymus farctus* spp. *bessarabicus*] hybrid.

Gregorio and Senadhira (1993) reported salt tolerance in a nine-parent complete diallel including reciprocals in rice. They found that salt tolerance was associated with Na^+ exclusion and absorption of K^+ to maintain a good Na^+-K^+ balance in the shoot. These workers also found that Na^+-K^+ ratio is controlled by both additive and dominance gene effects. The trait exhibited overdominance. Heritability of the trait was low because environmental effects were large. They concluded that selection must be done in later generations and under controlled conditions so as to minimize environmental effects.

Asins *et al.* (1993) reported heritability estimates of 53% for total fruit weight (TW) and 73% for number (FN) in 206 progeny derived from an interspecific hybrid (*L. esculentum* x *L. pimpinellifolium*) by self pollination under saline conditions. Non additive gene effects were detected for TW, FN and for average fruit weight (FW). Different types of gene action were found depending on the presence and absence of high NaCl concentrations in the nutrient solution. A different set of genes, or genes, differently regulated, must be involved in the expression of TW, FN and other fruit related characters depending on environmental conditions.

Ashraf (1994) reported broad-sense heritability estimates calculated at different salinity levels in two F_2 wheat populations. One was derived from a

cross between LU26S (from Pakistan) and Kharchia (from India) varieties. The second F_2 population was derived from a cross between LU26S and Candeal (from CIMMYT) parents. Broad-sense heritability for number of tillers per plant ranged from 49 to 60%; for 1000-seed weight from 57 to 80%; for number grains per spike from 64 to 78%; and for seed yield from 60 to 91%. Yadav (1993) found high genetic variability under saline conditions for number of tillers per plant, spike length and 1000-grain weight in barley (*Hordeum vulgare*). Heritability was lower under saline than in non-saline conditions for all the traits expect 1000-grain weight. Genetic correlations were modified under saline conditions.

Phung *et al.* (1992) reported heritability estimates under saline conditions in F_2 generation of 4 crosses in rice. Heritability estimates were high for number of grains per panicle for all crosses. Path analysis revealed that number of panicles per plant had the highest direct effect on yield in all crosses.

Although these studies provide some information on the inheritance of ion exclusion, yield and its components, additional studies especially for wheat are needed to determine effective selection procedures. In this section the results of experiments involving the parents, F_1, F_2, BC_1 and BC_2 populations of a cross between Alexandria (high yielding) and KRL1-4 (salt tolerant) are presented to provide information about the nature of genetic effects and

heritability estimates of leaf ion contents, yield and its components. Phenotypic and genotypic correlations for these traits are also presented.

7.2 MATERIALS AND METHODS

7.2.1 Raising of inbred parents

Single plants were selected from within Alexandria (high yielding, salt sensitive) and KRL1-4 (low yielding, salt tolerant) and they were used as parents of crosses. The generations used in these studies were:

Population	Pedigree
P_1(♀)	Alex-9
P_2(♂)	KRL-5
F_1	$P_1 \times P_2$
F_2	Selfed F_1
BC_1	$P_1 \times F_1$
BC_2	$P_2 \times F_1$

The experiment was conducted in a glass-house at the University of Wales, College Farm, Aber, Bangor during September 1993. The seeds of the parents were sown starting on 25-9-1993. Four seeds per pot per parent were sown at 4 different times to help synchronization of flowering and permit crossing because KRL1-4 was an early variety and Alexandria was late. The plants were grown in 36 pots using soil. The pot size was as described in section

5.2.1, Chapter 5. A solution containing macro- and micro-nutrients was applied to pots at twenty day intervals during early growth stages. Average temperature in the glass-house was 17.2±0.45°C

Some panes of the glass-house were broken on 23-12-1993 due to high wind. The pots were transferred to a glass-house at Pen Y Ffridd field station. The temperature of the glass-house was 16-18°C and natural day length was supplemented to a photoperiod of 16 hrs.

7.2.1.1 Emasculation and pollination

To produce F_1 seeds, florets of each spikelet were hand emasculated by using pointed forceps, and were pollinated using a small hair brush.

Anthesis in wheat generally starts in the middle of the spike and progresses upwards and downwards. The terminal and basal florets usually have functionless flowers. Depending on the size of the ear, 3-5 upper and basal spikelets were removed with the help of pointed forceps (Fehr, 1987). All tertiary florets were also removed by gently pulling these florets downward and upward with pointed forceps. The upper third of top lemma and palea was removed using a pair of scissors. In the female parent, three immature anthers were very carefully removed with pointed forceps from each floret to avoid injuring the stigma. The emasculated spikes were bagged immediately with 7.5" × 2.5" glassine bags. Spikes of the male parent were also bagged separately to

avoid foreign pollen contamination. In the morning after bagging, pollen was collected from the male parent in the bag by gently shaking the spike. The pollen was transferred to a petri dish and then dusted onto the feathery stigmas of the emasculated florets of the female parent, which were again covered by bags. The F_1 crosses were labelled as ♀ x ♂. Pollination was done two to three times to increase seed setting. Hand and all equipment used were sterilized with absolute alcohol before proceeding to next pollination. 80 crosses were made and 50 seeds from single and 41 seeds from reciprocal crosses were obtained. F_1 and parental seeds were harvested at maturity on 10-2-1994.

7.2.2 Producing the F_1, F_2, and backcross (BC_1 and BC_2) generations

These generations were produced under glass-house conditions at the University of Wales, College Farm, Aber, Bangor during summer 1994. The seeds of the parents and F_1 were sown on 12-5-1994. A single seed per pot per parent was sown at 4 different sowing dates to control the synchronization of flowering problem. There were 24 pots per generation. The plants were grown in pots using soil. Pot size was 15 cm diameter. A solution containing macro- and micro-nutrients was applied at twenty day intervals during early growth stages.

7.2.2.1 Emasculation and pollination

Emasculation and pollination were done as described above in section

7.2.1.1. At maturity, 400 seeds from each parent, 500 from F_2, 54 from F_1, 32 from BC_1 and 42 BC_2 respectively were harvested on 21-8-1994.

7.2.3 Growing the parents, F_1, F_2, BC_1 and BC_2 in NaCl solution

The experiment was conducted in a glass-house at the University of Wales, College Farm, Aber, Bangor starting in January 1995. Temperature was not controlled and a 16 hrs photoperiod consisting of natural day light was supplemented by bulbs used as described in experiment 6, Chapter 6. Average temperature in the glass-house was 16.4±0.44°C.

7.2.3.1 Raising the seedlings of basic generations

The parents and progenies (F_1, F_2, BC_1 and BC_2) were tested at 100 mol m^{-3} NaCl. The seeds were germinated in a growth-room at 20°C on capillary matting starting on 13-01-1995. Seedlings were transplanted into hydroponics in 6 pots on 22-01-1995. The total number of plants were 60 for each parent, 52 for F_1, 270 for F_2, 30 for BC_1 and 42 BC_2. The plants were grown in three replicates to facilitate leaf sampling and final harvesting, with up to 10 plants per row. The plant-to-plant and row-to-row distance was 3.5 cm and 6.0 cm, respectively. A Randomized Complete Block Nested Design was used. Size of the pot was 52 × 35 × 16 cm. Salt stress was introduced in three increments over a period of five day starting on 28-1-1995. Macro and micro nutrients were added in the solution following the procedure described in Chapter 3.

7.2.3.2 Chemical analysis

Youngest fully-expanded fourth leaves were used for chemical analyses. They were sampled from 15 random plants per parent, 14 from F_1, 89 for F_2, 11 from BC_1 and 16 from BC_2. Replication 1 was sampled on 16-02-1995, and replications 2 and 3 on 17-2-1995. The leaves were rinsed quickly in distilled water and blotted dry with tissue paper. The samples were placed in Eppendorf tubes and stored in a freezer at -10°C. Cell sap was extracted and ions were determined as described in Chapter 5.

7.2.3.3 Final harvest

The experiment was harvested at maturity on 15-5-1995 (replication 2 and 3) and on 16-5-1995 (replication 1) and main tiller height and the number of spikes per plant were recorded. The total number of plants harvested were 40 from P_1, 46 from P_2, 27 from F_1, 230 from F_2, 23 from BC_1 and 36 from BC_2. The ears were detached and straw weight per plant, number of infertile spikelets per spike and number of fertile spikelets per spike were recorded.

Threshing was done by hand and grain weight per plant (g), grain weight per spike (g) , number of grains per plant, number of grains per spike and average grain weight (g) were determined .

7.2.3.4 Statistical and biometrical analysis

Statistical analyses were performed using the Minitab for Windows

Package. ANOVA was used to assess significant differences between the means of generations and pairwise comparison where appropriate were made by Fisher's test at 5%.

Standard errors (S.E) of the mean of each generation (P_1, P_2, BC_1, BC_2, F_1 and F_2) (Snedecor and Cochran 1989) were estimated by constructing the following ANOVA to know the rows and replication effects.

Source	df	EMS
Between replications	(b-1)	$\sigma^2w+k\sigma^2r+kb\sigma^2b$
Between rows within replications	b(r-1)	$\sigma^2w+k\sigma^2r$
Between plants within rows within replications	br(k-1)	σ^2w
Total	brk-1	

The SS due to replications is an orthogonal and linear estimate of block effect. It does not contribute to the variance of mean. However, the between rows within replicates SS contributes to the variance of mean. It contains the interaction of rows within replicate blocks.

If the MS due to differences between rows within replications was significant then the generation variance was obtained as:

$$V\bar{x} = \sigma^2w+k\sigma^2r / brk$$

If the MS due to differences between rows within replications was non-significant its SS was pooled with the between plants within rows within replication SS to obtain the pooled mean square which was divided by (brk) to

get $V\bar{x}$. Pooled $\sigma^2 w$ over rows and replications was used for the analysis of second degree statistics.

7.2.3.5 Generation means analysis

A generation means analysis was performed as described by Mather and Jinks (1982). A computer programme supplied by Dr. H.S. Pooni, School of Biological Sciences, University of Birmingham, was used. The analysis was performed for ion contents associated with salt tolerance, and yield and its components under saline conditions.

The coefficients of the genetic components of generation means are presented in Table 7.1. Weighted least squares analysis (Mather and Jinks, 1982) was performed on the generation means. A simple one-parameter model was tried first and tested for goodness of fit. If the one-parameter model, [m] did not fit then a two-parameter model, [m] and [d], was fitted and tested for goodness of fit. If the two-parameter model did not fit then a dominance parameter was included in the model. If any parameter was non significant then it was dropped and then next one parameter tried, although $\chi 2$ was non significant. The higher value parent was always taken as P_1 in the model fitting for each trait (For instance Alexandria having higher Na^+ content was taken as P_1 for the analysis of Na^+ content while KRL-5 bearing high K^+ content was taken as P_1 for analysis of K^+ content). The model was selected when

Table 7.1. Coefficients for the genetic effects for the weighted least squares analysis of generation means (Mather and Jinks, 1982).

Generation	**----------Components of genetic effects----------**				
	m	**[d]**	**[h]**	**[i]**	**[j]**	**[l]**
P_1	1	1	0	1	0	0
P_2	1	-1	0	1	0	0
F_1	1	0	1	0	0	1
F_2	1	0	0.5	0	0	0.25
BC_1	1	0.5	0.5	0.25	0.25	0.25
BC_2	1	-0.5	0.5	0.25	-0.25	0.25

m = Mean
[d] = Additive
[h] = Dominance
[i] = Additive x additive
[J] = Additive x dominance
[l] = Dominance x dominance

Table 7.2. Coefficients for the genetic variance components for the weighted least squares analysis of generation variances (Mather and Jinks, 1982).

Generations	**----------Genetic components----------**			
	D	**H**	**F**	**E**
P_1	0	0	0	1
P_2	0	0	0	1
F_1	0	0	0	1
F_2	0.5	0.25	0	1
BC_1	0.25	0.25	-0.5	1
BC_2	0.25	0.25	0.5	1

D = Additive component
H = Dominance component
F = Cross product between additive and dominance
E = Environmental component

parameters tested were significant at infinity and $\chi 2$ value was non-significant at 5%.

7.2.3.6 Analysis of components of genetic variances

A weighted least squares analysis of variances was performed as described by Mather and Jinks (1982). The data of the experiment containing six generations (parents, F_1, F_2, BC_1 and BC_2) was analyzed using a computer programme supplied by Dr. H.S. Pooni, University of Birmingham. The coefficients of genetic components of the generation variance are presented in Table 7.2, Models incorporating E, (D and E), (D, H and E), (D, F and E) and (D, H, F and E) were tried. The best fit model was selected, when $\chi 2$ was non significant with all significant parameters.

7.2.3.7 Heritability estimates

Narrow sense heritability for F_2 and F infinity generation was calculated from the components of variance from the best fit model of the weighted least squares analysis using the formulae:

$h^2(F_2)$

a) = 0.5D/(0.5D+E)

(when the simple DE model fitted the data)

b) = 0.5D/(0.5D+0.25H+E)

(when the DHE model fitted the data)

$h^2(F_\infty) = D/(D+E)$

7.2.3.8 Correlations

7.2.3.8.1 Phenotypic correlations

The phenotypic (r_P) correlations between two traits, x and y, were calculated using Minitab for Windows. The correlations between ion contents, K^+/Na^+ ratio, yield and its components were computed from the 89 plants of the F_2 population as followed:

$r_P = Cov_P(x, y)/\sqrt{V_P(x).\ V_P(y)}$

Where:

$Cov_P(x, y)$ = Mean product of xyth traits in F_2 generation.

$V_P(x)$ and $V_P(y)$ = Mean squares for xth and yth traits respectively in F_2 generation.

7.2.3.8.2 Genotypic correlations

The genetic correlations (r_G) between two characters, x and y, were calculated by the formula:

$r_G = Cov_g\ (x,y)/\sqrt{V_g(x).\ V_g(y)}$

Where:

$Cov_g\ (x,y) = Cov(x,y)F_2\text{-}Cov(x,y)E$

$Cov\ (x,y)E = (¼)[Cov(x,y)P_1 + Cov(x,y)P_2 + 2Cov(x,y)F_1]$

$Cov_g(x,y)$, $Cov(x,y)E$, $Cov(x,y)P_1$, $Cov(x,y)P_2$, $Cov\ (x,y)F_1$ and $Cov(x,y)F_2$ are covariances of x and y associated with genetic effects, non-genetic effects, P_1, P_2, F_1 and F_2 generations, respectively

$V_g(x) = V(x)F_2\text{-}V(x)E$

$V_g(y) = V(y)F_2 - V(y)E$

$V(x)E = (¼)[V(x)P_1 + V(x)P_2 + 2V(x)F_1]$

$V(y)E = (¼)[V(y)P_1 + V(y)P_2 + 2V(y)F_1]$

$V_g(x)$ and $V_g(y)$ are genetic variances of x and y respectively.

7.3 RESULTS

Overall differences between the generations were found to be significant for all physiological and agronomic traits studied (Tables 7.3 and 7.4).

7.3.1 Response of Parents and F1 to NaCl

7.3.1.1 Ion contents and K^+/Na^+ ratio (Table 7.3)

There was a significant decrease ($P \leq 0.001$) in Na^+, Cl^- uptake and an increase ($P \leq 0.001$) in K^+ uptake in the F_1 hybrid compared to both parents. K^+/Na^+ ratio was also higher ($P \leq 0.001$) in the F_1 than in the parents. KRL1-4 also had significantly less Na^+, Cl^- uptake, but increased K^+ and K^+/Na^+ ratio than Alexandria.

7.3.1.2 Yield and yield components (Table 7.4)

Grain weight per plant was significantly greater ($P \leq 0.001$) in F_1 than in Alexandria. This was due to more grains per plant, more grains per spike, higher average grain weight, greater grain weight per spike and more fertile spikelets per spike. Main tiller height was also significantly higher ($P \leq 0.01$) in the F_1 than in Alexandria.

Table 7.3. Generation means and S.E of ion contents (mol m^{-3}) and K^+/Na^+ ratio in a cross between Alexandria (P_1) and KRL1-4 (P_2) wheat under saline conditions.

Trait	P_1		P_2		F_1		F_2		BC_1		BC_2		
	Means	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Probability**
Na^+	238^{abcd}	18.30	188^{a}	12.98	137^{aef}	4.95	185^{be}	6.98	191^{cf}	17.10	171^{d}	12.02	0.001**
K^+	112^{abcd}	5.40	167^{a}	8.23	224^{aef}	9.21	172^{be}	6.37	161^{cf}	16.86	190^{d}	13.28	0.000***
K^+/Na^+	0.5^{abc}	0.06	1.0^{a}	0.10	1.7^{aef}	0.12	1.1^{be}	0.08	1.0^{f}	0.19	1.2^{c}	0.14	0.000***
Cl^-	355^{abcd}	6.68	274^{a}	5.00	225^{aef}	4.24	271^{be}	6.76	272^{cf}	18.27	260^{d}	12.55	0.000***

** = $P < 0.01$

*** = $P < 0.001$

Note : Values with same letters had significant differences at Fisher's test at 5%.

Table 7.4. Generation means and S.E of yield and yield components in a cross between Alexandria (P_1) and KRL1-4 (P_2) wheat under saline conditions.

Trait	P_1		P_2		F_1		F_2		BC_1		BC_2		
	Means	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**Probability**
Main tiller height (cm)	54.7^{abcde}	1.13	61.0^{a}	1.69	67.3^{b}	1.64	62.6^{c}	0.96	62.5^{d}	2.93	62.9^{e}	2.31	0.007**
Number of spikes per plant	1.5^{a}	0.12	1.0^{abcd}	0.03	1.2	0.09	1.3^{b}	0.04	1.4^{c}	0.10	1.4^{d}	0.08	0.014*
Straw weight per plant (g)	0.7	0.05	0.6^{abc}	0.03	0.7	0.05	0.8^{a}	0.04	0.8^{b}	0.12	0.8^{c}	0.08	0.035*
Infertile spikelets per spike	1.8^{ab}	0.11	2.4^{acd}	0.15	1.7^{ce}	0.24	2.3^{be}	0.08	1.8^{d}	0.21	2.0	0.17	0.005**
Fertile spikelets per spike	11.0^{ab}	0.44	9.7^{abcd}	0.22	12.4^{bf}	0.40	12.0^{ae}	0.18	11.0^{c}	0.60	11.7^{def}	0.44	0.000***
Number of grains per plant	10.2^{abcd}	1.22	22.9^{a}	1.13	33.6^{aef}	1.70	24.6^{be}	1.13	23.4^{cf}	3.10	29.4^{d}	2.34	0.000***
Grain weight per plant (mg)	134.0^{abc}	14.57	377.0^{a}	20.03	458.0^{b}	25.73	397.0^{c}	16.35	334.0^{bd}	43.98	464.0^{d}	32.30	0.000***
Number of grains per spike	8.0^{abcd}	0.97	22.3^{a}	1.16	30.4^{aef}	1.80	19.3^{be}	0.73	16.7^{ac}	2.01	21.3^{df}	1.27	0.000***
Grain weight per spike (mg)	104.0^{ab}	11.34	366.0^{bc}	19.46	419.0^{ad}	28.80	317.0^{de}	11.53	236.0^{ace}	31.41	339.0^{a}	16.71	0.000***
Average grain weight (mg)	10.7^{abcde}	0.63	16.9^{a}	0.55	14.5^{b}	1.07	16.4^{c}	0.51	14.4^{d}	1.32	16.7^{e}	0.77	0.000***

* = $P < 0.05$

** = $P < 0.01$

*** = $P < 0.001$

Note : Values with same letters had significant differences at Fisher's test at 5%.

The F_1 also had significantly ($P \leq 0.001$) more grains per plant, grains per spike, fertile spikelets per spike and fewer ($P \leq 0.01$)) infertile spikelets per spike than KRL1-4, but there were no significant differences in grain weight per plant, grain weight per spike and average grain weight.

KRL1-4 had higher ($P \leq 0.001$) yield than Alexandria. This was due to more grains per plant, more grains per spike, higher average grain weight and greater grain weight per spike. However KRL1-4 had significantly ($P \leq 0.001$) fewer fertile spikelets per spike, fewer spikes per plant, more infertile spikelets per spike and higher main tiller height than Alexandria.

7.3.2 Generation means analysis (Tables 7.5 and 7.7)

The three-parameter (mdh) model provided the best fit of the observed to the expected generation means for ion uptake, K^+/Na^+ ratio, main tiller height, straw weight per plant, fertile spikelets per spike, number of grains per plant, grain weight per plant and grain weight per spike.

In the case of number of spikes per plant and average grain weight per plant a four-parameter (mdhl) model provided the best fit of the observed to the expected generation means. In the case of number of infertile spikelets per spike and number of grains per spike a four-parameter (mdhi) model provided a best fit of the observed to the expected generation means.

The additive genetic effects were found to be smaller than the dominance

effects. This can arise if there is overdominance or unidirectional dominance or dispersion of genes in the parents leading to reduced estimation of the [d] component in relation to [h] component. The dominance effects were negative for Na^+, Cl^- uptake and number of infertile spikelets per spike showing thereby that decreases for these traits were dominant of the non-allelic interactions, [i] and [l] components were only important. The negative [i] for infertile spikelet number shows that it is possible to obtained less infertility in the F_∞ generation. The positive [i] for number of grains shows that it is possible to fix additive x additive interactions for increased number of grains per spike. The comparison of [h] and [l] for number of spikes per plant and average grain weight shows that there exist duplicate gene interactions for these traits are likely to be very difficult to exploit in the improvement of recombinant inbred lines.

The consistently significant [d] component for all traits undoubtedly reveals that the additive variation is pronounced for all traits in this cross. Clearly, there exists a scope for the genetic improvement for all traits.

7.3.3 Generation variances analysis (Tables 7.6 and 7.8)

In the generation variances analyses, the model incorporating DE (additive and environmental) components gave the best fit for all ion contents and K^+/Na^+ ratio. The generation variances analysis also provide the best fit for DE (additive and environmental) for almost all agronomic traits except number

Table 7.5. Best model fit estimates for generation mean parameters by weighted least squares analysis of ion contents (m mol^{-3}) and K^+/Na^+ ratio in cross between Alexandria and KRL1-4 wheat under saline conditions.

	----------Parameters----------						
Trait	**m**	**±S.E**	**[d]**	**±S.E**	**[h]**	**±S.E**	**χ2 (3df)**
Na^+	220.7	8.38	25.5***	9.71	-82.0***	10.68	1.32
K^+	137.6	4.63	26.8***	4.75	79.8***	9.65	1.44
K^+/Na^+	0.7	0.05	0.2***	0.06	0.9***	0.12	1.88
Cl^-	314.5	3.96	39.6***	4.09	-89.2***	5.95	1.64

m = Mean

[d] = Additive effects

[h] = Dominance effects

*** = $P < 0.005$

Table 7.6. Components of variation, D (additive) and E (environmental) and narrow sense heritability estimates for ion contents and K^+/Na^+ ratio in cross between Alexandria and KRL1-4 wheat under saline conditions.

	----------Variance components----------					Narrow sense	
Trait	**(D)**	**±S.E**	**(E)**	**±S.E**	**χ2 (4df)**	**h^2 (F_2)**	**h^2 (F_∞)**
Na^+	7214.1***	1307.58	905.0***	189.49	0.43	79.9	88.8
K^+	5295.6***	1116.15	1005.1***	209.47	2.15	72.5	84.0
K^+/Na^+	0.76***	0.16	0.15***	0.03	0.74	71.7	83.5
Cl^-	8175.7***	1212.59	387.0***	81.47	4.74	91.3	95.5

*** = $P < 0.005$

Table 7.7. Estimates of parameters of best fit model on means of basic generations of the cross Alexandria × KRL1-4 in wheat under saline conditions.

	Parameters										
Trait	**m**	**±S.E**	**[d]**	**±S.E**	**[h]**	**±S.E**	**[i]**	**±S.E**	**[l]**	**±S.E**	**χ2 (df)**
Grain weight per plant (mg)	263.7	11.71	125.2***	11.97	229.0***	25.57					4.19(3)
Main tiller height (cm)	59.8	0.92	2.9***	0.96	9.4***	1.81					0.51(3)
Number of spikes per plant	1.2	0.06	0.2***	0.05	0.5*	0.24			-0.5*	0.26	5.46(2)
Straw weight per plant (g)	0.7	0.03	0.1*	0.03	0.1*	0.06					7.43(3)
Infertile spikelets per spike	2.9	0.28	0.3***	0.09	-1.4***	0.48	-0.8***	0.29			1.36(2)
Fertile spikelets per spike	10.6	0.22	0.8***	0.23	2.3***	0.43					5.90(3)
Number of grains per plant	16.5	0.79	6.3***	0.81	17.1***	1.71					0.64(3)
Number of grains per spike	8.6	2.31	6.7***	0.71	20.5***	3.79	6.2**	2.41			5.14(2)
Grain weight per spike (mg)	227.7	10.46	122.5***	10.37	161.5***	24.19					5.70(3)
Average grain weight (mg)	13.8	0.42	3.0***	0.40	8.4***	2.30			-7.6***	2.80	1.76(2)

m = Mean, [d] =Additive effects, [h] = Dominance effects, [i] = Additive x edditive effects, [l] = Dominance x dominance effects

* = $P < 0.05$

** = $P < 0.01$

*** = $P < 0.005$

Table 7.8. Components of variance, D (additive), E (environmental) and narrow sense heritability estimates for yield and yield components under saline conditions in a cross between Alexandria and KRL1-4 wheat.

	----------Variance components------					Narrow sense	
Trait	**(D)**	**±S.E**	**(E)**	**±S.E**	**χ2 (df)**	**h^2 (F_2)**	**h^2 (F_∞)**
Grain weight per plant (mg)	93932.11***	11717.46	15117.79***	2067.44	0.80 (4)	75.6	86.1
Main tiller height (cm)	304.41***	42.78	65.15***	8.88	2.22 (4)	70.0	82.3
Number of spikes per plant			0.35***	0.03	7.54 (5)		
Straw weight per plant (g)	0.68***	0.07	0.07***	0.01	1.06 (4)	82.9	90.7
Infertile spikelets per spike			1.15***	0.08	8.40 (5)		
Fertile spikelets per spike			6.62***	0.47	9.69 (5)		
Number of grains per plant	443.93***	56.47	75.16***	10.27	1.26 (4)	74.7	85.5
Number of grains per spike	1 08.92***	27.97	63.35***	8.48	1.76 (4)	46.2	63.2
Grain weight per spike (mg)	24893.53***	7125.15	16741.22***	2234.45	6.82 (4)	43.6	59.8
Average grain weight (mg)	68.17***	11.90	22.09***	2.99	4.00 (4)	60.7	75.5

*** = $P < 0.005$

of spikes per plant, number of infertile spikelets per spike and fertile spikelets per spike where model E (environmental components) gave the best fit.

7.3.4 Heritability (Tables 7.6 and 7.8)

The infinity generation heritability estimates were consistently higher than those for F_2 generation. This means that the proportion of genetic component of variance that can be fixed among inbred lines is very high. There is thus a possibility of improvement of all traits except number of spikes per plant, number of infertile spikelets per spike and number of fertile spikelets per spike.

7.3.5 Frequency distribution of F_2 population

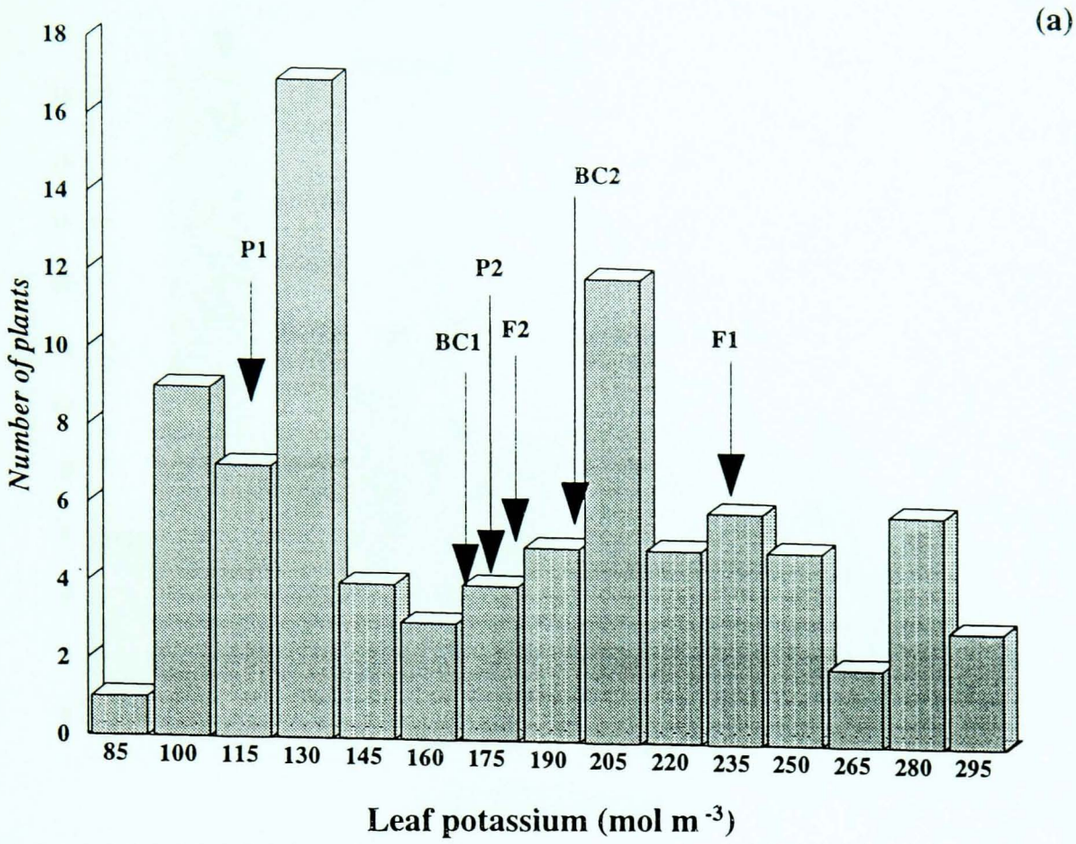
The frequency distributions of physiological and agronomic traits for the F_2 populations are given in Figures 7.1-7.7. The graphs for all traits show near-normal distributions in the F_2 which also exhibit transgressive segregation. The F_1 means fall outside the parental range for all traits except number of spikes per plant, except average grain weight. Thus heterosis in F_1 was greatly pronounced. This can arise from any one of the following individually or in combination:

i) Overdominance.

ii) Unidirectional dominance with gene dispersion.

iii) Non-allelic interactions.

iv) Maternal effects.



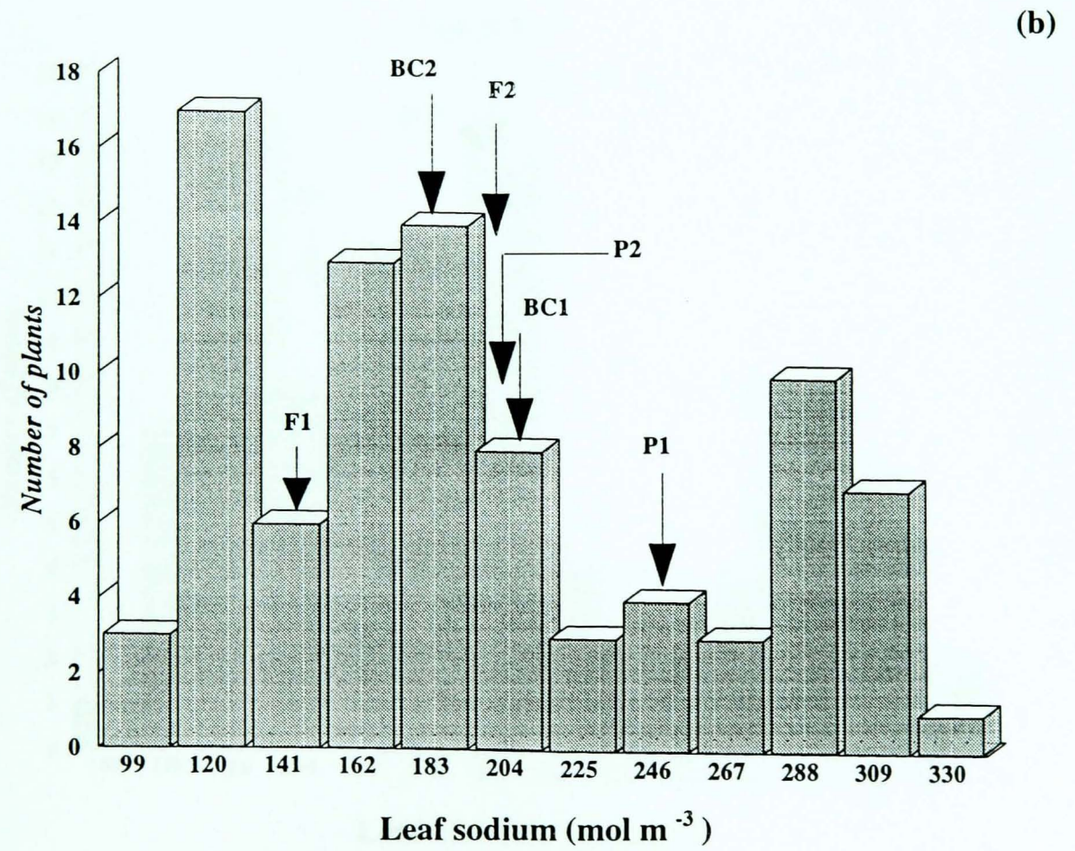
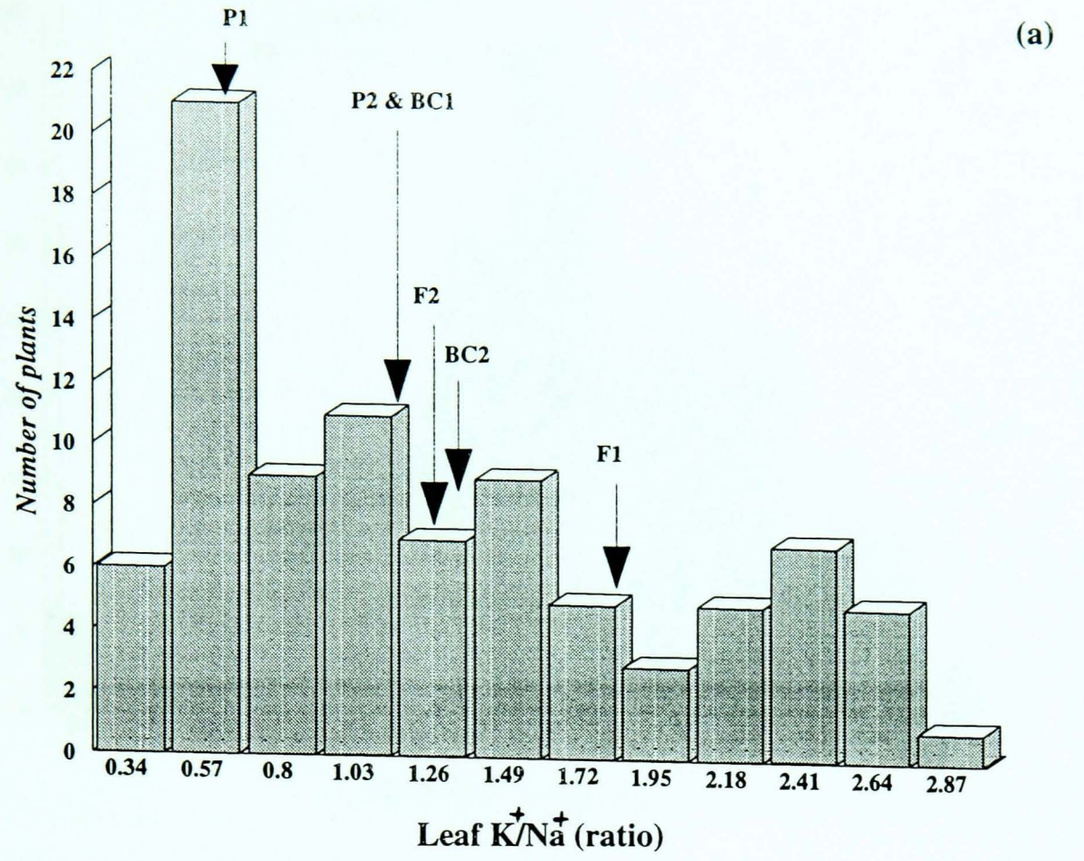


Figure 7.1. Frequency distribution of the F2 for leaf (a)sodium and (b) potassium contents under saline conditions.



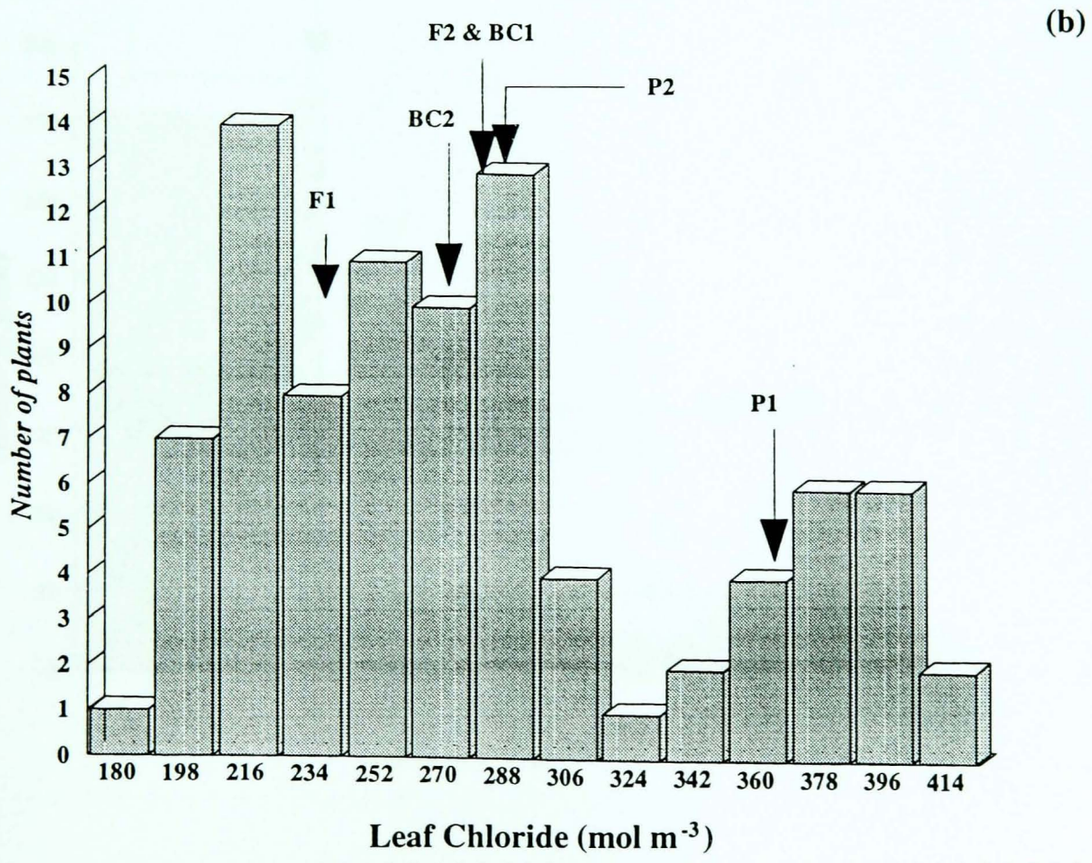


Figure 7.2 Frequency distribution of the F2 for leaf (a) chloride content and (b) K^+/Na^+ ratio under saline conditions.

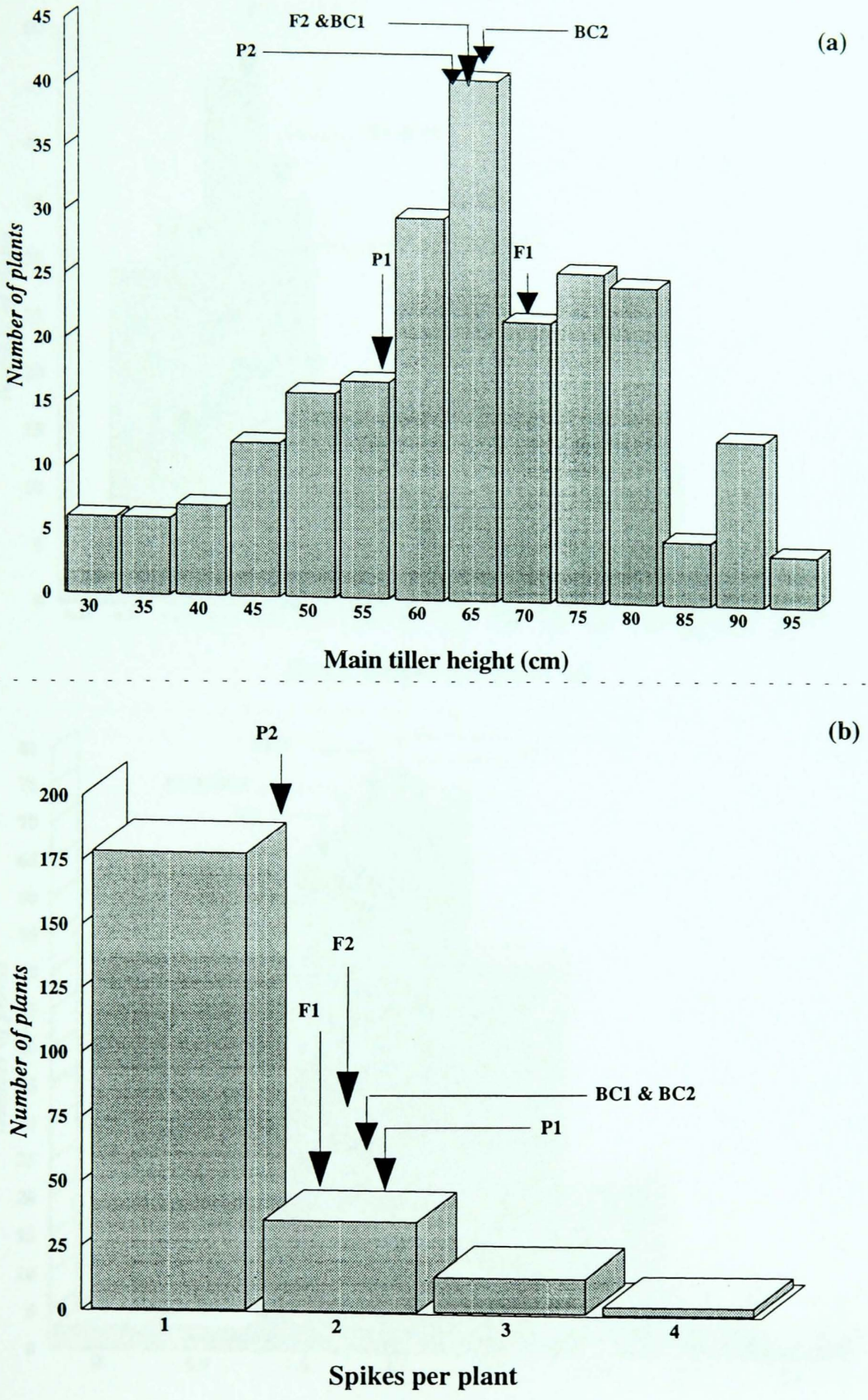


Figure 7.3. Frequency distribution of the F2 for (a) main tiller height and (b) spikes per plant under saline conditions.

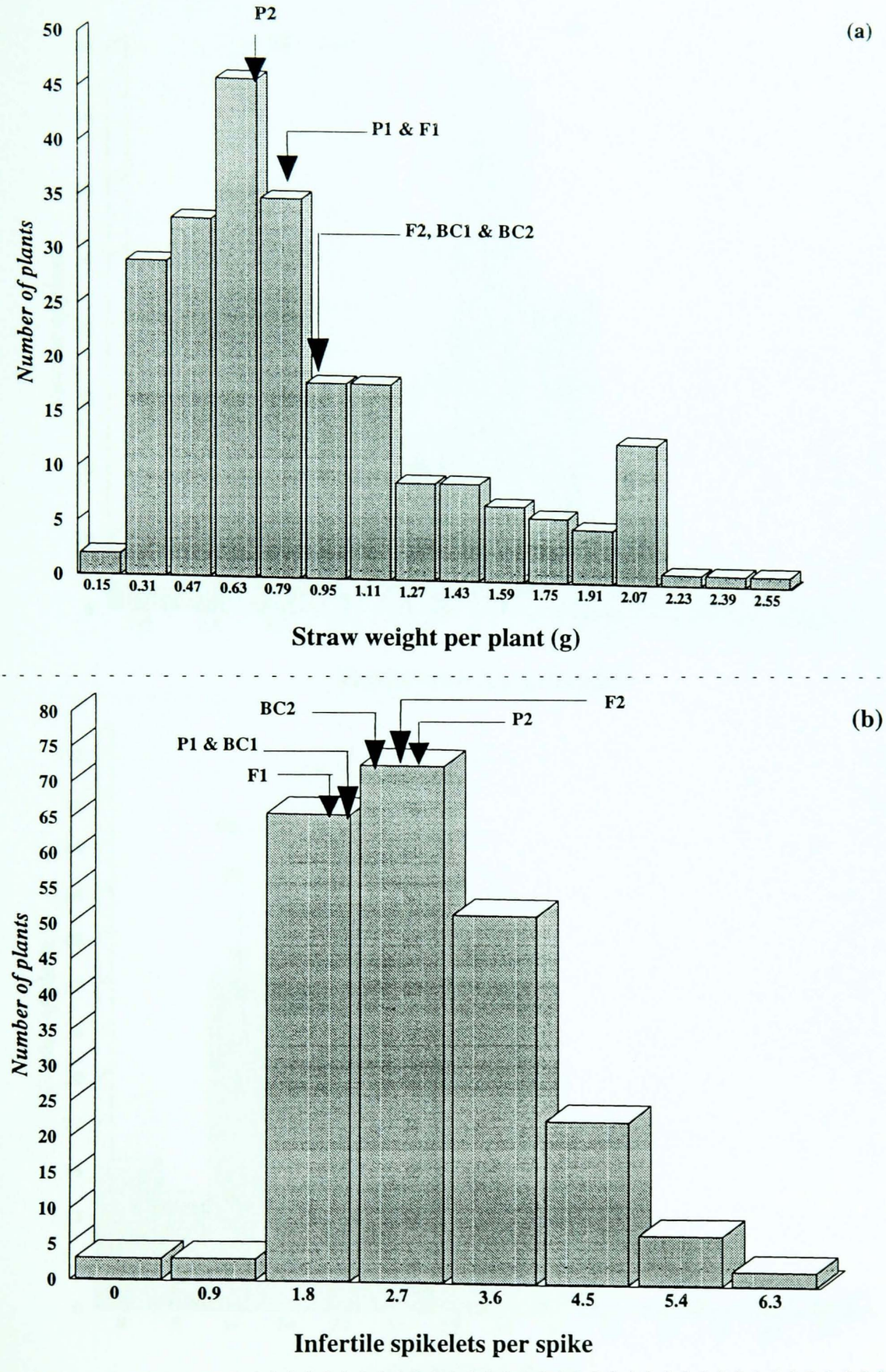


Figure 7.4. Frequncy distribution of the F2 for (a) straw weight per plant and (b) infertile spikelets per spike under saline conditions

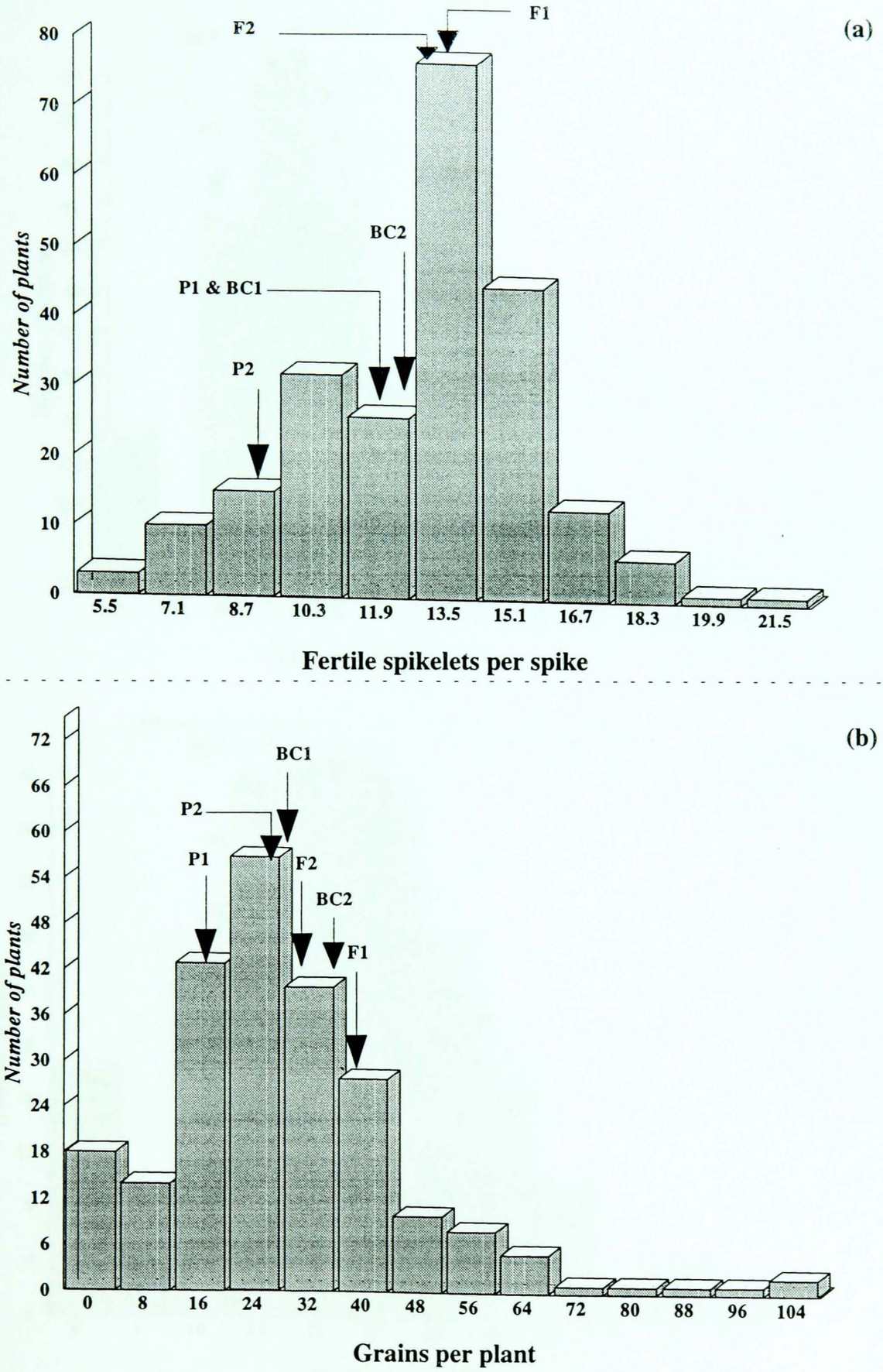
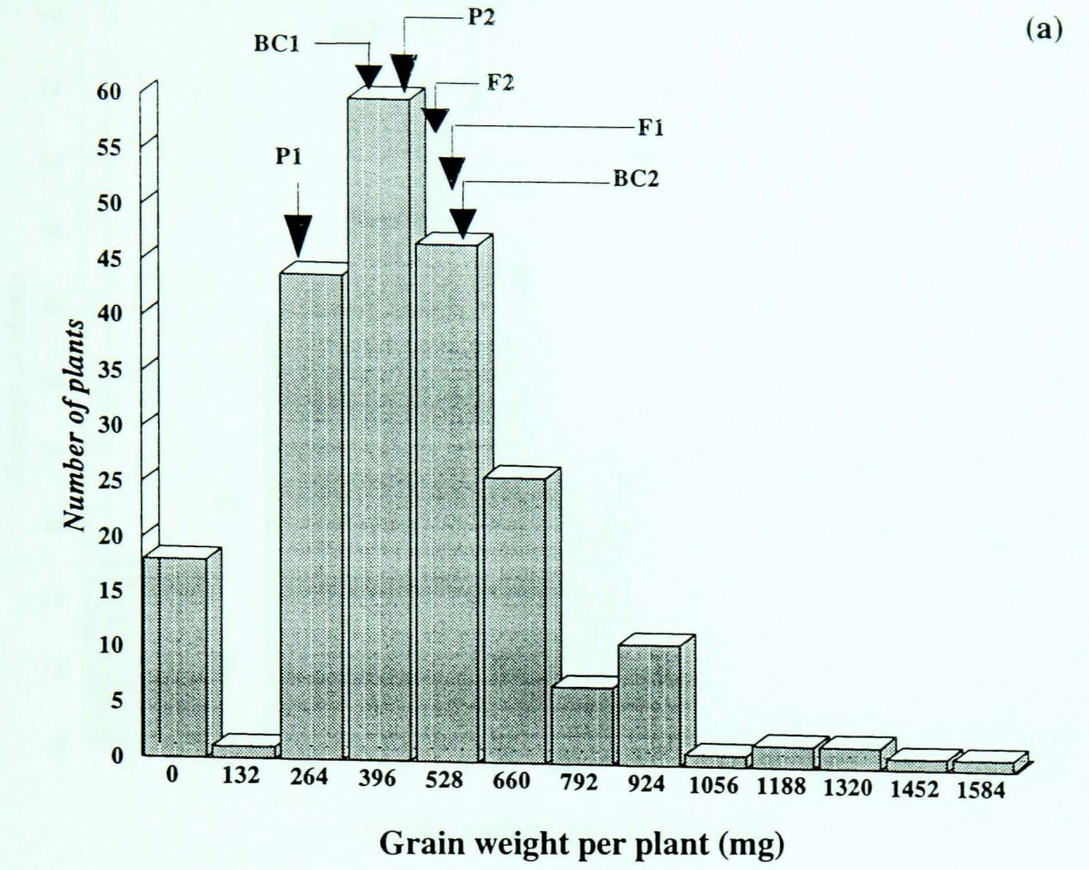


Figure 7.5. Frequency distribution of the F2 for (a) fertile spikelets per spike and (b) grains per plant under saline conditions.



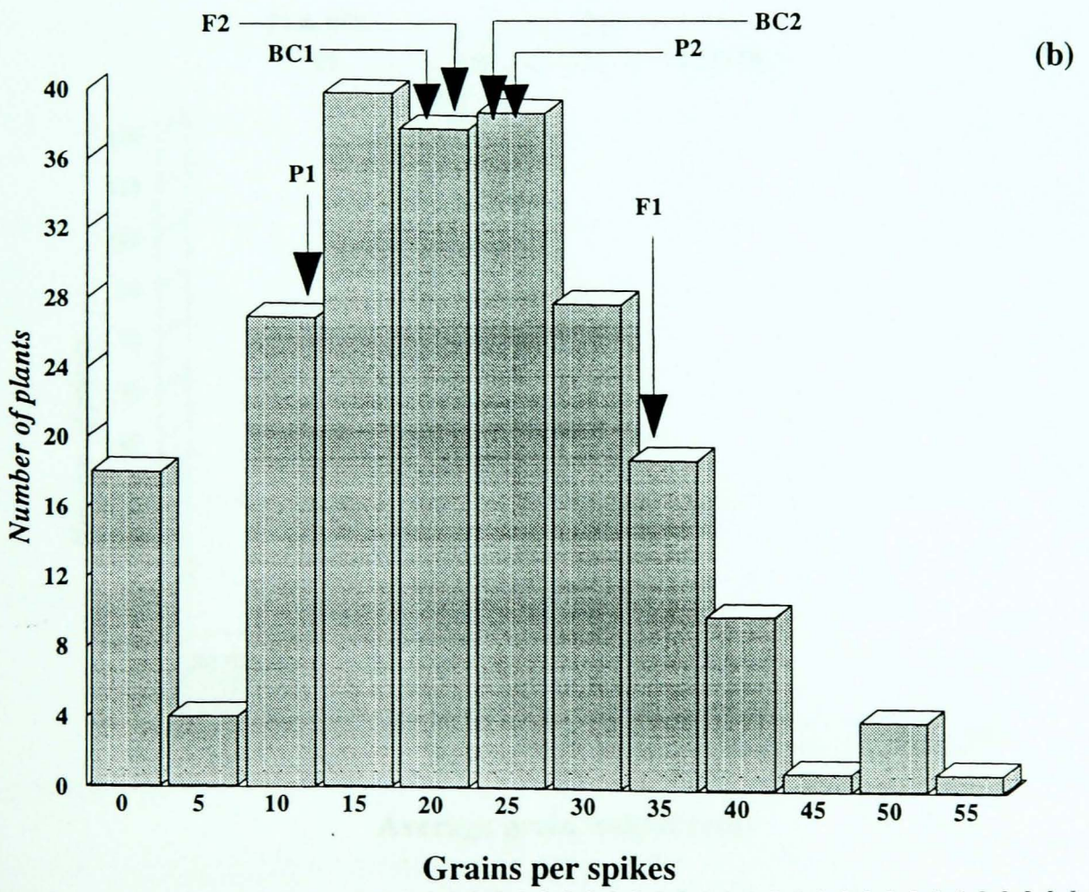
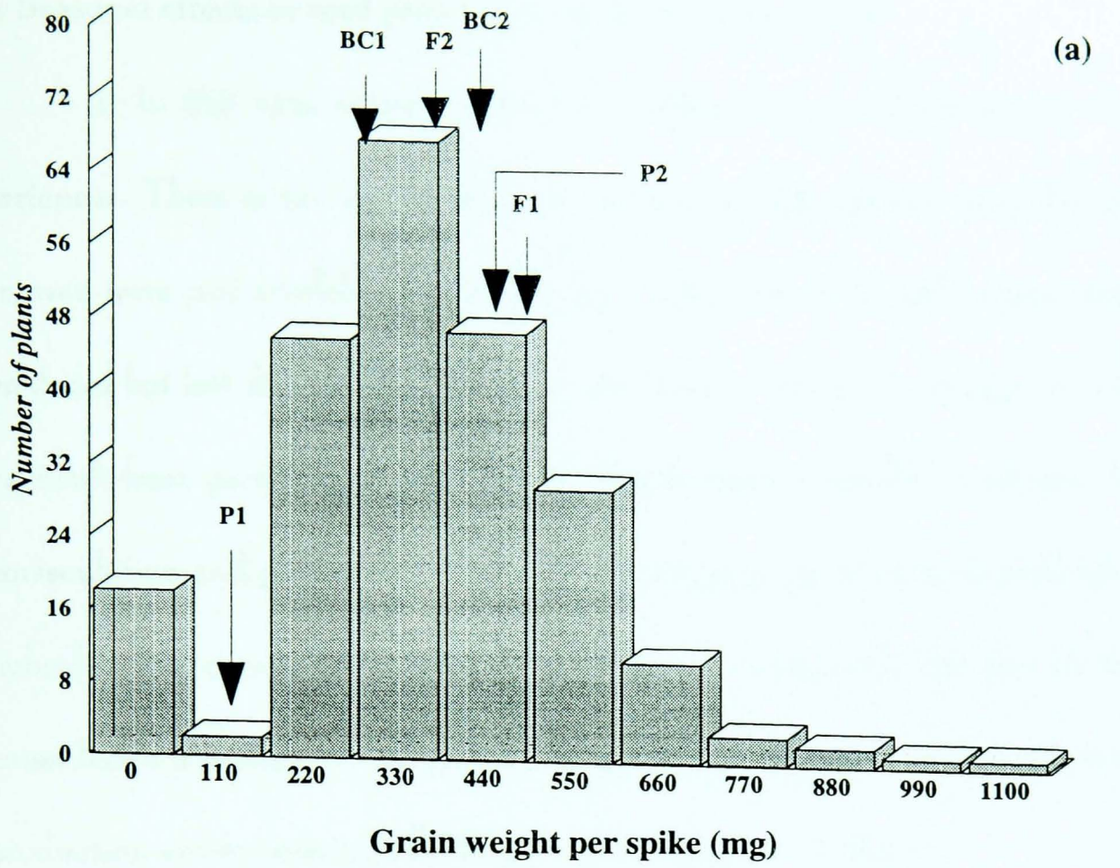


Figure 7.6. Frequency distribution of the F2 for (a) grain weight per plant and (b) grains per spike under saline coditions.



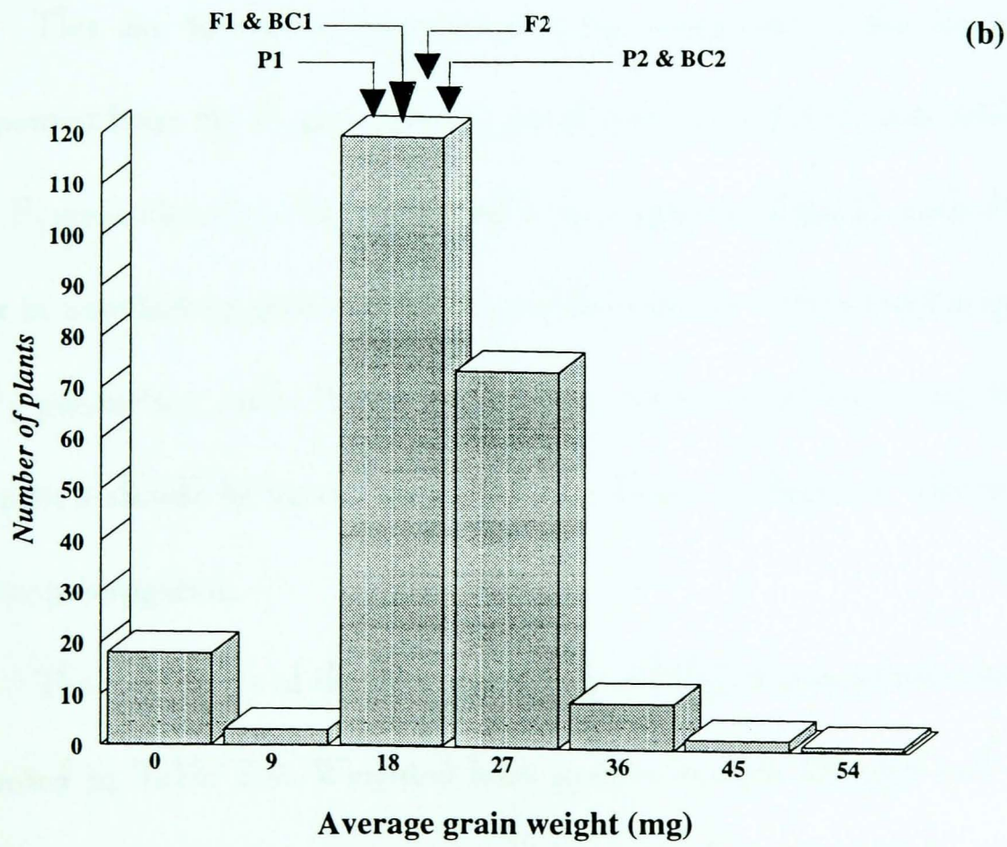


Figure 7.7. Frequncy distribution of the F2 for (a) grain weight per spike and (b) average grain weight under saline conditions.

v) Seasonal effects or seed production environmental effects.

(i) to (iii) were examined by model fitting on generation means and variances. There is no way to verify (iv) in the present material as reciprocal crosses were not available for the analysis, although reciprocal crosses were produced but lost due to a failure in the glass-house ventilation system. (v) can be result from greater seed size of F_1 produced under controlled conditions by emasculation and pollination. Only a few seeds were borne on each head after hybridisation compared to several by selfing. Consequently the size of the crossed-seed is usually greater. As a result it is very common to confuse the seed production environmental effects with spurious overdominance.

This can be verified by estimating the magnitude of the dominance component from the F_1 generation $[h_1]$ and comparing it with that estimated from F_2 generation $[h_2]$. The two h's will be homogeneous if the F_1 seeds did not differ in manifesting greater initial capital because the environment is specific to F_1 generation only. If $[h_1] \neq [h_2]$, the estimates of $[h_1]$ using the F_1 generation should be viewed very carefully. Thus, the heterotic effects need further investigation.

The coefficients of the dominance $[h_1]$ and $[h_2]$ of generation means are presented in Table 7.9. Weighted least squares analysis (Mather and Jinks, 1982) was computed on the generation means, while other effects such as

additive × additive, additive × dominance and dominance × dominance were ignored. The $[h_1]$ and $[h_2]$ were compared applying t test at 5% (Tables 7.10 and 7.11). $[h_1]$ was found to be significantly higher than $[h_2]$ for Na^+, K^+, K^+/Na^+ ratio. It was also found to be significantly higher for number of grains per spike and grain weight per spike. However $[h_1]$ was significantly less then $[h_2]$ for straw

Table 7.9 Coefficients for the dominance effects for the weighted least squares analysis of generation means.

Generations	Parameters			
	m	**[d]**	**$[h_1]$**	**$[h_2]$**
P_1	1	1	0	0
P_2	1	-1	0	0
F_1	1	0	1	0
F_2	1	0	0	0.5
BC_1	1	0.5	0	0
BC_2	1	-0.5	0	0

m = Mean
[d] = Additive
$[h_1]$ = Dominance due to F_1
$[h_2]$ = Dominance due to F_2

weight per plant, number of infertile spikelets, number of fertile spikelets and average grain weight. In general, the magnitude of $[h_2]$ was smaller than $[h_1]$ even when the coefficient of dominance is smaller in the F_2 generation which usually results in larger estimates of dominance components having larger standard errors. This means that $[h_2]$ is closer to the real dominance effects.

Table 7.10. Estimated dominance [h_1] from F_1 and dominance [h_2] from F_2 for ion content (m mol^{-3}) and K^+/Na^+ ratio in Alexandria and KRL1-4 wheat under saline conditions.

Trait	----------Parameters----------				
	[h_1]	±S.E	[h_2]	±S.E	t.test
Na^+	-58.4	9.06	-20.9	20.63	2.03*
K^+	77.7	10.20	51.5	15.50	2.25*
K^+/Na^+	0.9	0.13	0.5	0.19	2.38*
Cl^-	-83.4	5.76	-74.9	15.61	0.59NS

NS = P > 0.05
* = P < 0.05

Table 7.11. Estimated dominance [h_1] from F_1 and dominance [h_2] from F_2 generations for yield per plant and yield components in Alexandria and KRL1-4 wheat under saline conditions.

Trait	----------Parameters----------				
	[h_1]	±S.E	[h_2]	±S.E	t.test
Grain weight per spike (mg)	168.2	30.03	132.5	28.69	4.02*
Main tiller height (cm)	8.4	1.86	7.4	2.59	0.55NS
Number of spikes per plant	-0.1	0.10	-0.0	0.12	1.51NS
Straw weight per plant (g)	0.1	0.06	0.4	0.10	-3.62*
Infertile spikelets per spike	-0.4	0.25	0.4	0.21	-6.05*
Fertile spikelets per spike	1.8	0.45	2.7	0.54	-3.22*
Number of grains per plant	15.5	1.86	12.9	2.72	1.30NS
Grain weight per plant (mg)	175.9	28.02	228.0	39.50	-1.87NS
Number of grains per spike	14.2	1.90	6.2	1.91	41.30*
Average grain weight (mg)	0.3	1.13	4.4	1.24	-8.10*

NS = P > 0.05
* = P < 0.05

7.3.6 Phenotypic and genotypic correlations (Tables 7.12 and 7.13)

Phenotypic correlations between grain weight per plant and its components were generally significant and positive, except infertile spikelets per spike which was negatively correlated with yield. Yield was significantly negatively correlated with Na^+ and Cl^- contents but positively correlated with K^+ content and K^+/Na^+ ratio. Number of spikes per plant was positively correlated with K^+ content and K^+/Na^+ ratio. Number of grains per plant was also negatively correlated with Na content.

Genetic correlations between Na^+ content, K^+, K^+/Na^+, yield, and most of the yield components were significant but negative. Cl^- content and number of infertile spikelets per spike were positively correlated with Na^+ content. Yield and most of its components were significantly and positively correlated with K^+ and K^+/Na^+ ratio, and negatively correlated with leaf Cl^-. Number of infertile spikelets were positively correlated with Cl^-.

7.4 DISCUSSION

There were significant differences in ion uptake, K/Na ratio, yield and yield components between parents and the F_1. All traits showed heterosis, except average grain weight and number of spikes per plant. Akbar and Yabuno (1975) reported genetically controlled salt tolerance in rice after studying salt-tolerant and salt-sensitive varieties and their F_1 hybrid, although they used salinity

Table 7.12. Phenotypic correlations (r_p) for ion contents, K/Na ratio, grain weight per plant and yield components in a cross between Alexandria (P_1) and KRL1-4 (P_2) wheat (data were from 89 F_2 plants) under saline conditions.

	Na^+	K^+	K^+/Na^+	Cl^-	Grain weight per plant (mg)
K^+	-0.830**				
K^+/Na^+	-0.879**	0.956**			
Cl^-	0.460**	-0.366**	-0.381**		
Grain weight per plant (mg)	-0.238*	0.218*	0.221*	-0.246*	
Main tiller height (cm)	-0.117NS	0.145NS	0.167NS	-0.113NS	0.494**
Number of spikes per plant	-0.208NS	0.232*	0.247*	-0.143NS	0.488**
Straw weight per plant (g)	-0.050NS	0.089NS	0.081NS	-0.099NS	0.437**
Infertile spikelets per spike	0.185NS	-0.135NS	-0.184NS	0.076NS	-0.407**
Fertile spikelets per spike	-0.171NS	0.122NS	0.111NS	-0.209NS	0.492**
Number of grains per plant	-0.227*	0.206NS	0.202NS	-0.203NS	0.881**
Number of grains per spike	-0.205NS	0.131NS	0.126NS	-0.186NS	0.633**
Grain weight per spike (mg)	-0.181NS	0.123NS	0.114NS	-0.199NS	0.673**
Average grain weight (mg)	-0.063NS	0.030NS	0.028NS	-0.055NS	0.384**

NS = Non significant
* = Significant at 5% level of significance
**= Significant at 1% level of significance

Table 7.13. Genetic correlations (rG) for ion contents, K^+/Na^+ ratio, grain weight per plant and yield components in a cross between Alexandria (P_1) and KRL1-4 (P_2) wheat (data were from 89 F_2 plants) under saline conditions.

	Na^+	K^+	K^+/Na^+	Cl^-	Grain weight per plant (mg)
K^+	-0.984				
K^+/Na^+	-0.941	0.975			
Cl^-	0.605	-0.441	-0.457		
Grain weight per plant (mg)	-0.351	0.300	0.329	-0.331	
Main tiller height (cm)	-0.243	0.348	0.362	-0.146	0.490
Number of spikes per plant	-0.304	0.322	0.332	-0.211	0.780
Straw weight per plant (g)	-0.017	0.117	0.072	-0.135	0.343
Infertile spikelets per spike	0.461	-0.439	-0.509	0.219	-0.559
Fertile spikelets per spike	-0.379	0.373	0.360	-0.286	0.693
Number of grains per plant	-0.355	0.346	0.357	-0.230	0.975
Number of grains per spike	-0.556	0.485	0.523	-0.297	0.772
Grain weight per spike (mg)	-0.415	0.298	0.324	-0.361	0.544
Average grain weight (mg)	-0.157	-0.027	-0.017	-0.129	0.295

induced panicle sterility as the criterion for salt tolerance. Singh *et al.* (1988) reported better performance of elite wheat lines developed from crosses between Kharchia and commercial varieties and reported that salt tolerance is transferable from tolerant to sensitive genotypes.

A comprehensive knowledge of associations, gene action and heritability for a trait is a prerequisite for its manipulation in a breeding programme. It was clear from an examination of the F_2 population frequency distributions for all traits that they were quantitatively inherited. Gene dispersion or non-allelic interactions or involvement of modifiers are suggested from the transgressive segregation in the F_2 populations. The results of these studies clearly provide evidence that traits responsible for salt tolerance, such as Na^+, K^+, Cl^- uptake and K^+/Na^+ ratio are heritable and significantly correlated with yield under saline conditions.

The significantly different estimates of the dominance component in F_2 for Na^+, K^+, K^+/Na^+ ratio, number of grain per spike and grain weight per spike than that obtained from F_1 indicate spurious overdominance exhibited by the F_1 generation for these traits.

7.4.1 Genetical effects

7.4.1.1 Gene effects for ion uptake and K^+/Na^+ ratio

In the generation means analysis the observation that the three-

parameter model provided the best fit to the data for Na^+, K^+, Cl^- uptake and K^+/Na^+ ratio suggests that the inheritance of these traits is relatively simple. Both additive and dominance genetic effects were found to be pronounced for all these traits. Similarly Gregorio and Senadhira (1993) reported in rice that good Na^+-K^+ balance was maintained by Na^+ exclusion and increased absorption of K^+ which were responsible for salinity tolerance. They also reported that low Na^+-K^+ ratio is governed by both additive and dominance gene effects. The trait exhibited overdominance, and two groups of genes were detected.

7.4.1.2 Gene effects for yield and its components

The results of generation means analysis for main tiller height, straw weight per plant, number of fertile spikelets per spike, number of grains per plant, yield per plant and yield per spike showed significant additive and dominance genetic effects. This means that the inheritance of these traits is relatively simple and it is assumed that the genes involved are independent of each other in producing their effects. In the case of number of spikes per plant and average grain weight additive, dominance and dominance x dominance genetic effects were detected. The inheritance of these traits is polygenic and not found to be so simple. For number of infertile spikelets per spike and number of grains per spike additive, dominance and additive x additive interactions were

involved in the inheritance.. This generally suggests that inheritance of all these traits is polygenic. Narayanan and Rangasamy (1991) reported similar significant additive and dominance effects for number of days to flowering, height, tiller number, panicle length, number of spikelets per panicle, 1000-grain weight and dry mater accumulation under normal and saline conditions. However, they found significant additive effects for grain yield only under saline conditions. They suggested that varieties with more additive gene effects for grain yield would perform better in saline soils. Salam (1993) reported intermediate responses for most of the traits such as Na^+, K^+, Cl^- uptake, K^+/Na^+ ratio, osmotic pressure, plant height, spikes per plant, 100 grain weight, harvest index and grain yield per plant and suggested partial dominance and additive gene action for these traits.

However, in generation variance analysis only additive genetic effects were involved in the inheritance of ion uptake and K^+/Na^+ ratio, yield and most of its components. But the generation means analysis show that both additive and dominance components were involved in the inheritance of all these traits. These inconsistencies may be due to the estimation precision of the two analyses. Although the generation means analysis found more integral and informative than that of generation variances.

7.4.2 Heritability estimates

Only narrow sense heritability estimates were computed, because in the least squares analysis of generation variances the simple DE (additive and environmental) model gave the best fit which suggests that additive variance comprised the significant part of total genetic variance.

F infinity heritabilities were high for ion uptake and K^+/Na^+ ratio which suggests that high genetic gain is possible. A high heritability estimate suggests that genetic improvement is possible for these traits in wheat from selection in segregating populations. Gregorio and Senadhira (1993) reported in rice low heritability for Na^+-K^+ ratio, but they found large environmental effects and suggested that selections must be done in later generations and under controlled conditions in order to minimize environmental effects.

Heritability estimates were also high for grain weight per plant and most of its components, but in some parameters such as number of grains per spike and grain weight per spike they were found to be comparatively low. These high heritability estimates suggest that yield can be improved using selection during successive generations. There is very little information available on heritability estimates of all these traits in wheat. In this experiment, heritability estimates were not computed for number of spikes per plant, number of infertile spikelets per plant and number of fertile spikelets per plant as there were no additive and

dominance components involved in the inheritance of these traits. Yadav (1993) reported in barley that heritability estimates were lower for tillers per plant and spike length under saline conditions than non saline conditions except 1000-grain weight. Narayanan and Rangasamy (1991) reported in rice high heritability estimates for dry matter accumulation, 1000-grain weight and spikelet number and concluded that selection on the basis of such traits would be effective in producing salt-tolerance varieties.

In these studies heritability estimates were found to be high for most of the traits. It is thought that this is because this experiment was conducted in hydroponic culture with controlled salinity stress. This minimised the effects of experimental error. Physiological and agronomic traits were measured more accurately. This also reduced the experimental error, as reported by Fehr (1987) that any precautions which may reduce experimental error will improve the estimate of heritability of a character.

7.4.3 Phenotypic and genotypic correlations

Phenotypic correlations (r_p) between ion uptake, K^+/Na^+ ratio and yield per plant were highly significant. Yield was also highly significantly correlated with all yield components. This suggests that there might be linkages between the genes which control yield and genes responsible for ion uptake. In the case of yield components only number of spikes per plant was significantly positively

correlated with K^+ uptake and K^+/Na^+ ratio and there was a significant negative correlation between number of grains per plant and Na^+ uptake (Table 6.9). Genetic correlations (r_G) were also derived between these traits to find any suitable marker closely linked with these traits. Salam (1993) concluded from the results that salinity markers for Na^+, K^+, Cl^-, and osmotic pressure are under genetic control. He also suggested that the K^+/Na^+ ratio of the youngest leaf and Cl^- contents of the mature leaves could be used as reliable criteria for screening salt tolerant wheat.

The magnitude of almost all genetic correlations (r_G) were higher between Na^+, K^+, Cl^- uptake, K^+/Na^+ ratio, yield and yield components, except straw weight per plant and average grain weight. K^+, K^+/Na^+ ratio, yield and its components were negatively correlated with Na^+ and Cl^-, except number of infertile spikelets which had positive correlation with Na^+ and Cl^-, and negative with K^+, K^+/Na^+ ratio and yield. These interrelations indicate that these traits might be controlled by common genes. Rana (1985) reported negative correlations between Na^+ contents and yield components. Salam *et al.* (1992) reported significant correlations between Na^+ and Cl^- and yield in wheat. They also found that K^+/Na^+ ratio, particularly for the youngest leaf had very high correlation with yield and yield components.

It is generally concluded from the results that traits are genetically

controlled and transferable from tolerant to sensitive genotypes. The genes controlling physiological traits are linked with the genes controlling yield and its components. High K^+/Na^+ ratio or high yield can be used as selection criteria for screening wheat under saline conditions. The results suggest that promising recombinant can be obtained by screening during later generations for saline conditions. There was no significant relationships between certain parameter in hydroponic culture and soil culture as described in section 5.3.3.4, Chapter 5. Therefore it is generally suggested that the later generations should be tested for genetic effects and heritability estimates under saline field conditions.

CHAPTER 8

GENERAL DISCUSSION

The problems of salt affected soil need more attention from plant breeders to evaluate promising plant cultivars which can grow better and also give desired grain yield to feed the burgeoning human population. This can be achieved by developing salt tolerant crops plant (Epstein *et al.,* 1980; Shannon, 1990).

Wheat is staple food for most of the human beings in the world as well as in Pakistan. It is grown on 8.1 million hectares in Pakistan (FAO, 1994). Pakistan has extensive salt affected areas (Rafique, 1975; Muhammad, 1978, 1983) and the area of saline arable land is growing at a rate of 250 acres per day (Rozema *et al.,* 1990). Therefore in these studies wheat was selected to be improved for saline cultivation. To achieve such a goal, wheat varieties were studied for their physiological mechanisms of salt-tolerance and their genetic basis. Some workers had already reported the presence of considerable genetic variation in salt-tolerance between rice varieties (Akbar *et al.*, 1972; Akbar and Yabuno, 1975). Some other plant breeders reported that salinity tolerance is governed by polygene in rice (Akbar and Yabuno, 1975, 1977; Akbar *et al.*, 1985).

The experiments reported in this thesis were planned to study inter-varietal variation (Greenway and Munns, 1980; Rana *et al.*, 1980; Shah , 1987; Shah *et al.*, 1987; Salam 1993) and intra-varietal variation (Joshi *et al.*, 1979; Qureshi *et al.*, 1980; Rashid, 1986; Shah, 1987; Salam, 1993) in salt-tolerance of wheat. In experiment 1 (Chapter 3) it was found that there were inter- and intra-varietal variations in ion contents under saline conditions and in yield and yield components under non-saline conditions. Although environmental conditions were uniform, variability within varieties was found to be higher than the variability between varieties. The variety KRL1-4 was found to be salt-tolerant under saline conditions but low yielding under non-saline conditions as compared to other varieties. These inter- and intra-varietal variations suggested that improvement might be achieved through selection from within varieties or by crossing tolerant and sensitive genotypes.

It was expected that landraces should have more variability than pure genotypes. But there was no difference in variability in Na^+ and K^+ uptake and K^+/Na^+ ratio between Alexandria (pure genotype), KRL1-4 (selection from within Kharchia-65) and Kharchia-65 (landrace). Surprisingly Alexandria (pure genotype) was found to be slightly more variable in Na^+ uptake than Kharchia-65 (landrace) under saline conditions experiment 1 (Chapter 3).

However more variability was found for grain weight per plant, number

of spikes per plant, main tiller height, straw weight per plant and average grain weight in Kharchia-65 (landrace) than in Alexandria (pure variety). Kharchia-65 also had more variability for main tiller height and number of infertile spikelets per spike than KRL1-4, experiment 1 (Chapter 3). These results may not be found under saline conditions. Further research is necessary to identify the extent of genetic variation in ion uptake and yield of other landraces. Such research should be done initially under hydroponic saline conditions where the environment can be controlled. However selections should subsequently be examined under saline field conditions.

Effects of leaf age, leaf position and location of plants in the pot on growth, yield and ion uptake were also considered in this study. Higher Na^+ and Cl^- concentrations were found in the older leaves and high K^+ concentrations in the younger leaves. However the considerable variations with leaf age in experiment 2 and 3 (Chapter 4), and leaf position in experiment 4, (Chapter 5), suggest that physiological traits are less useful as selection criteria. Most of the correlations between ion concentrations in the fourth and flag leaf in experiment 4 (Chapter 5), were found to be non-significant. This also supports the idea that physiological traits are less useful while as selection criteria. However the absence of differences in ion uptake, yield and yield components between inside and outside plants in experiment 1 (Chapter 3), suggest that the random

sampling including inside and outside plants in a pot can be used to identify intra-varietal variation.

Richards (1983) argued that because in saline fields most of the yield comes from the areas with lowest salinity, then it is better to select for high yield under non-saline conditions. The results of these studies do not support this hypothesis. Alexandria was found to be higher yielding than KRL1-4 and Kharchia-65 under non-saline conditions in experiment 1 (Chapter 3), but it was found to be lower in yield than these varieties under saline conditions in experiment 4 (Chapter 5). Therefore it is suggested from the results that selection under non-saline conditions cannot be useful to predict performance under saline conditions. There were no significant differences in Na^+, K^+ contents and K^+/Na^+ ratio between Alexandria and Kharchia-65 in experiment 1 and experiment 2, (Chapter 3 and Chapter 4 respectively) but there were significant differences for ion contents and K^+/Na^+ ratio between these varieties in experiment 4, (Chapter 5).

Therefore these variations for physiological traits between experiments also suggest that these traits might be less useful selection criteria.

In experiment 4 (Chapter 4), Alexandria was found to be lower yielding than KRL1-4 and it was also found to be lower in yield in experiment 7, (Chapter 7). It is suggested from these results that yield under saline conditions

is the most useful selection criteria for salt tolerance.

There were two breeding techniques used in this study to increase the salt-tolerance and improve the yield of spring wheat under saline conditions. The first involved selecting tolerant and sensitive lines from within already existing cultivars and selfing these lines their behaviour was then studied in the second selfed generation. The second involved crossing, a salt-sensitive genotype with a salt-tolerant genotype. Biometrical genetic analysis were done to establish the genetic basis of salt-tolerance and to determine the likelihood of achieving increases in salt-tolerance by this approach.

In experiment 5 (Chapter 6) selections from within varieties were found to be true to selection in most of the S_0 lines, but some inconsistencies were also found. These results gave no clear indication as to whether it is better to select for yield or K^+/Na^+ ratio. KRL1-4 was found to be more salt tolerant than Alexandria and Kharchia-65. Alexandria S_0 lines selected with high K^+/Na^+ ratio had higher yield than lines selected with high yield and this trend was consistent from S_0 to S_1 generation. In Kharchia-65 (S_0) lines selected with high K^+/Na^+ ratio produced relatively higher yielding progeny than lines selected with high yield, but the trend was not clear and not consistent from S_0 to S_1 generation. In KRL1-4 (S_0) lines selected for high yield produced relatively higher yielding progeny than the lines selected with high K^+/Na^+ ratio.

Therefore it is concluded from the results that the usefulness of K^+/Na^+ ratio as a selection criteria varies from genotype to genotype. It cannot be useful as a selection criteria for all genotypes.

In experiment 6 (Chapter 6) S_1 lines with high K^+/Na^+ ratio gave higher yield. This trend was consistent from S_0 to S_1 generation. Selfing increased the salt-tolerance and yield of some lines. Most of the results suggest that selection and selfing of successive generations might be useful to improve the salt-tolerance and yield of existing salt-sensitive cultivars. Yeo *et al.* (1988) reported in later generations of rice from S_4 to S_5 clear and consistent trends showing that plants with low Na^+ parents had low Na^+ contents and plants selected for high Na^+ produced progeny with high Na^+ concentrations. Therefore, it suggested that more selfing should be done in later generations to find out whether yield or K^+/Na^+ ratio under saline conditions can be used as selection criteria. However it should be noted that these lines were selected on the basis of high and low yield under non-saline conditions, and high and low K^+/Na^+ ratio under saline conditions. High yield is associated with low Na^+, low Cl^-, and high K^+/Na^+ ratio in wheat (Salam *et al.,* 1992) and this suggests the possibility, as reported by Falconer (1960), that the relative efficiency of selection in the moderately saline environments can also be approached using the concept of genetic correlation.

Phenotypic and genotypic associations between high yield and Low Na^+, low Cl^-, high K^+, and high K^+/Na^+ ratio found in experiment 6 (Chapter 7) suggest that ion contents can be used as selection criteria for salt-tolerance. However due to the variations for ion contents as discussed earlier these traits are not reliable. Additive and dominant genetic effects were found to be involved in the inheritance of ion content, K^+/Na^+ ratio, yield and most of the yield components, which suggested similar gene action for all these traits. These inter-relationships and similar gene action indicate that these traits might be controlled by some common genes. The high heritability estimates for Na^+, Cl^-, K^+, K^+/Na^+ ratio, yield and most of the yield components indicates that these traits will have good response to selection and considerable progress may be expected from selection in segregating generations. The results of this study show that no single agronomic or physiological trait was highly correlated with yield. However, bearing in mind the high heritabilities of some of these traits, the results suggest that it may be possible to develop salt-tolerant varieties by combing these in a single variety. This concept put forward by Yeo and Flowers (1986) is termed "pyramiding". Similar results in wheat under drought stress were also reported by Malik (1995). Although high heritability is potentially useful it can be less useful for physiological traits due to variation for these traits which were found in experiments 2, 3, 4 (Chapter 4 and Chapter 5). There was

high heterosis in the F_1. The dominance effects were compared between F_1 and F_2 generations. The results indicate that overdominance is not trustworthy. This might be due to the greater seed size of F_1.

The cross breeding technique seemed to be relatively more effective for the improvement of salt-tolerance of wheat as compared to selection from within a variety. Alexandria is a late maturing variety and awnless, two characters which are undesirable in the Pakistani wheat growing environment. Therefore in later generations deleterious combinations can be expected. Hybrids as expected involving Alexandria were intermediate in maturity and awnless. The awnless character might not be useful under saline conditions whereas it is very useful under drought conditions. Late maturing salt-tolerant varieties might not be acceptable to farmers. To produce and evaluate new salt-tolerant genotypes by cross breeding needs more time and resources than to improve salt-tolerance by selection from within already existing salt-tolerant genotypes. Therefore improvement by making selections from within already existing salt-tolerant varieties can be done with less time and resources.

Performance of varieties in soil and hydroponic culture were found to be independent of each another. The relationships between most of the parameters studied were found to be non significant. Yield per plant was very low under hydroponic salinity compared to that under soil salinity. The average grain

weight was also very low. This might be due to increasing temperature during grain filling stage. There was also no supplementary Ca^{2+} added in these experiments. It was noted in the literature review that Ca^{2+} decreases the damaging effects of Na^{+} ((LaHaye and Epstein, 1969; Hyder and Greenway, 1965; Alberico and Cramer, 1993; Cramer *et al.*, 1994a). Therefore it is concluded that addition of supplementary Ca^{2+} while comparing and evaluating genotypes can give misleading results. It is also suggested from the results that F_2 population and selections from within a variety need to be evaluated under saline field conditions during their later generations.

It is generally concluded from these results that high yield is the most useful selection criteria for salt tolerance under saline conditions. High yield under non-saline conditions was found to be ineffective. In experiments 5 and 6 (Chapter 6) there are also two possibilities suggested from the results which can be tested in further experiments involving larger numbers of plants. Single plants can be selected for yield from within already salt-tolerant but low yielding varieties to improve yield and single plants can also be selected for high K^{+}/Na^{+} ratio from within less salt-tolerant but high yielding varieties to improve their salt tolerance. Thus a substantial programme is needed to study such a complex phenomenon of salt stress.

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Appendix 1. Means and S.E. of ion contents (mol m^{-3}) and K^+/Na^+ ratio for four harvests in three wheat varieties under saline conditions.

	Varieties						
Days after	**Alexandria**		**KRL1-4**		**Kharchia-65**		
transplanting	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+							
28	142	30.6	89	22.8	140	52.9	NS
35	165	30.7	133	53.0	95	14.9	NS
42	211	27.4	170	35.6	220	40.9	NS
49	243	21.8	181	28.1	177	38.5	NS
K							
28	190	11.7	200	29.6	204	17.5	NS
35	161	8.0	222	49.5	158	12.7	NS
42	213	31.9	187	24.3	156	20.2	NS
49	175	24.6	169	9.7	158	24.7	NS
K^+/Na^+							
28	1.5	0.3	2.6	0.6	2.4	0.8	NS
35	1.1	0.3	1.9	0.3	1.8	0.3	NS
42	1.0	0.0	1.3	0.5	0.8	0.1	NS
49	0.7	0.1	0.9	0.1	1.0	0.3	NS
Cl^-							
28	209	12.0	147	12.7	231	23.0	49.9*
35	338	50.8	207	29.0	206	23.5	NS
42	439	50.2	343	7.0	362	41.0	NS
49	374	49.2	329	22.6	415	80.6	NS

NS = $P > 0.05$
* = $P < 0.05$

Appendix 2. Means and S.E. of ion contents (mol m^{-3}) and K^+/Na^+ ratio for four harvests in selections from within three wheat varieties under saline conditions.

	Varieties						
Days after	**Alex-1**		**KRL-24**		**Khar-1**		
transplanting	**Means**	**±S.E**	**Means**	**±S.E**	**Means**	**±S.E**	**LSD**
Na^+							
23	124	4.6	112	4.7	117	8.3	NS
30	158	9.4	127	5.4	153	11.2	NS
37	186	9.7	176	9.5	169	13.9	NS
44	240	11.3	209	11.2	223	9.0	NS
K^+							
23	230	7.8	256	7.3	249	10.1	NS
30	200	11.1	210	9.8	202	13.7	NS
37	154	11.9	172	9.4	178	13.3	NS
44	126	10.5	153	12.3	144	11.5	NS
K^+/Na^+							
23	1.9	0.1	2.3	0.1	2.2	0.2	NS
30	1.3	0.1	1.7	0.1	1.3	0.1	0.4*
37	0.8	0.1	1.0	0.1	1.1	0.2	NS
44	0.5	0.1	0.7	0.1	0.7	0.1	NS
Cl^-							
23	239	7.5	211	5.4	219	6.0	22.2*
30	266	8.4	230	10.8	253	6.3	30.6*
37	269	6.1	256	9.4	273	8.8	NS
44	314	14.4	271	13.1	285	7.9	NS

NS = $P > 0.05$
* = $P < 0.05$

Appendix 3.1 Analysis of variance for leaf Na^+ concentration (m mol^{-3}) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	17321	8660	5.00	0.009
Error	78	135113	1732		

Appendix 3.2 Analysis of variance for leaf K^+ concentration (m mol^{-3}) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	43127	21563	16.90	0.000
Error	78	99552	1276		

Appendix 3.3 Analysis of variance for leaf K^+/Na^+ (ratio) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	4.2455	2.1227	44.00	0.000
Error	78	3.7631	0.0482		

Appendix 3.4 Analysis of variance for number of spike per plant under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	48.914	24.457	28.46	0.000
Error	78	67.037	0.859		

Appendix 3.5 Analysis of variance for main tiller height (cm) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	8631.7	4315.2	171.59	0.000
Error	78	1961.8	25.2		

Appendix 3.6 Analysis of variance for straw weight per plant (g) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	138.292	69.146	48.40	0.000
Error	78	111.440	1.429		

Appendix 3.7 Analysis of variance for number of infertile spikelets per spike under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	26.397	13.198	29.82	0.000
Error	78	34.521	0.443		

Appendix 3.8 Analysis of variance for number of fertile spikelets per spike under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	825.17	412.59	305.82	0.000
Error	78	105.23	1.35		

Appendix 3.9 Analysis of variance for number of grains per spike under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	9519.9	4760.0	91.10	0.000
Error	78	4075.4	52.2		

Appendix 3.10 Analysis of variance for grain weight per plant (g) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	173.763	86.882	35.16	0.000
Error	78	192.762	2.471		

Appendix 3.11 Analysis of variance for average grain weight (mg) under saline conditions of three wheat varieties. (Experiment 1)

Source	D.F	S.S	M.S	F	P
Genotype	2	0.000504	0.000252	7.04	0.002
Error	78	0.002794	0.000036		

Appendix 4.1 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 28 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	7325	3663	0.65
Error	9	50562	5618	

Appendix 4.2 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 28 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	416	208	0.12
Error	9	15818	1758	

Appendix 4.3 Analysis of variance for leaf K^+/Na^+ (ratio), 28 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	2.308	1.154	0.78
Error	9	13.368	1.485	

Appendix 4.4 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 28 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	15011	7506	6.76
Error	9	10000	1111	

Appendix 4.5 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 35 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	9817	4908	1.27
Error	8	30828	3854	

Appendix 4.6 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 35 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	10469	5234	2.41
Error	8	17364	2170	

Appendix 4.7 Analysis of variance for leaf K^+/Na^+ (ratio), 35 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	1.3370	0.6685	2.27
Error	8	2.3602	0.2950	

Appendix 4.8 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 35 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	46086	23043	4.32
Error	8	42648	5331	

Appendix 4.9 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 42 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	5744	2872	0.63
Error	8	36718	4590	

Appendix 4.10 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 42 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	6690	3345	1.29
Error	8	20668	2584	

Appendix 4.11 Analysis of variance for leaf K^+/Na^+ (ratio), 42 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	0.5504	0.2752	1.32
Error	8	1.6704	0.2088	

Appendix 4.12 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 42 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	20102	10051	1.59
Error	8	50637	6330	

Appendix 4.13 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 49 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	10823	5412	1.48
Error	9	32942	3660	

Appendix 4.14 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 49 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	576	288	0.16
Error	9	15724	1747	

Appendix 4.15 Analysis of variance for leaf K^+/Na^+ (ratio), 49 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	0.2143	0.1071	0.71
Error	9	1.3602	0.1511	

Appendix 4.16 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 49 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	V.R
Genotype	2	14806	7403	0.59
Error	9	113121	12569	

Appendix 4.17 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 23 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	512.4	256.2	1.13	0.348
Error	15	3387.7	225.8		

Appendix 4.18 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 23 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	2219.4	1109.7	2.58	0.109
Error	15	6462.6	430.8		

Appendix 4.19 Analysis of variance for leaf K^+/Na^+ (ratio), 23 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	0.6635	0.3317	2.29	0.135
Error	15	2.1694	0.1446		

Appendix 4.20 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 23 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	2577.3	1288.7	5.34	0.018
Error	15	3617.2	241.1		

Appendix 4.21 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 30 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	3451.8	1725.9	3.56	0.054
Error	15	7262.3	484.2		

Appendix 4.22 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 30 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	381.9	190.9	0.23	0.794
Error	15	12203.1	813.5		

Appendix 4.23 Analysis of variance for leaf K^+/Na^+ (ratio), 30 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	0.53639	0.26820	4.02	0.040
Error	15	1.00091	0.06673		

Appendix 4.24 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 30 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	3957.4	1978.7	4.33	0.033
Error	15	6850.3	456.7		

Appendix 4.25 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 37 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	900.6	450.3	0.60	0.564
Error	15	11343.7	756.2		

Appendix 4.26 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 37 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	1946.0	973.0	1.18	0.335
Error	15	12391.8	826.1		

Appendix 4.27 Analysis of variance for leaf K^+/Na^+ (ratio), 37 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	0.24692	0.12346	1.26	0.312
Error	15	1.47129	0.09809		

Appendix 4.28 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 37 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	994.3	497.2	1.23	0.321
Error	15	6085.7	405.7		

Appendix 4.29 Analysis of variance for leaf Na^+ concentration (m mol^{-3}), 44 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	2846.3	1423.1	2.12	0.154
Error	15	10060.9	670.7		

Appendix 4.30 Analysis of variance for leaf K^+ concentration (m mol^{-3}), 44 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	2288.7	1144.3	1.45	0.266
Error	15	11867.5	791.2		

Appendix 4.31 Analysis of variance for leaf K^+/Na^+ (ratio), 44 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	0.13506	0.06753	2.04	0.164
Error	15	0.49538	0.03303		

Appendix 4.32 Analysis of variance for leaf Cl^- concentration (m mol^{-3}), 44 days after transplanting under saline conditions of three wheat varieties.

Source	D.F	S.S	M.S	F	P
Genotype	2	5611.4	2805.7	3.18	0.071
Error	15	13238.3	882.6		

Appendix 5.1 Analysis of variance for fourth leaf Na^+ concentrations (m mol^{-3}) of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	21413.7	21613.8	3602.3	6.74	0.000
System	1	36247.3	35044.8	35044.8	65.53	0.000
Genotype x System	6	7482.0	7482.0	1247.0	2.33	0.061
Error	27	14438.8	14438.8	534.8		

Appendix 5.2 Analysis of variance for fouth leaf K^+ concentration (m mol^{-3}) of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	13441.0	12347.9	2058.0	2.65	0.037
System	1	34132.8	33098.5	33098.5	42.67	0.000
Genotype x System	6	4611.1	4611.1	768.5	0.99	0.451
Error	27	20943.9	20943.9	775.7		

Appendix 5.3 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	32649	31176	5196	5.00	0.001
System	1	42357	41680	41680	40.14	0.000
Genotype x System	6	7406	7406	1234	1.19	0.342
Error	27	28033	28033	1038		

Appendix 5.4 Analysis of variance for fourth leaf K^+/Na^+ (ratio) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	36.176	36.633	6.105	4.31	0.004
System	1	72.695	69.730	69.730	49.18	0.000
Genotype x System	6	20396	20.396	3.399	2.40	0.055
Error	27	38.281	38.281	1.418		

Appendix 5.5 Analysis of variance for fourth leaf Ca^{2+} concentration (m mol^{-3}) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	778.77	626.30	104.38	2.47	0.049
System	1	5171.94	5227.59	5227.59	123.64	0.000
Genotype x System	6	549.80	549.80	91.63	2.17	0.078
Error	27	1141.61	1141.61	42.28		

Appendix 5.6 Analysis of variance for fouth leaf Mg^{2+} concentration (m mol^{-3}) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	767.21	757.25	126.21	5.15	0.001
System	1	362.36	352.65	352.65	14.40	0.001
Genotype x System	6	160.72	160.72	26.79	1.09	0.391
Error	27	661.31	661.31	24.49		

Appendix 5.7 Analysis of variance for flag leaf Na^{+} concentration (m mol^{-3}) of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	15878.0	16395.0	2732.5	7.23	0.000
System	1	56673.4	55354.3	55354.3	146.45	0.000
Genotype x System	6	3034.6	3034.6	505.8	1.34	0.275
Error	27	10205.5	10205.5	378.0		

Appendix 5.8 Analysis of variance for flag leaf K^{+} concentration (m mol^{-3}) of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	22626.3	23149.5	3858.3	4.81	0.002
System	1	9053.6	9050.7	9050.7	11.29	0.002
Genotype x System	6	3858.8	3858.8	643.1	0.80	0.577
Error	27	21645.2	21645.2	801.7		

Appendix 5.9 Analysis of variance for flag leaf Cl^- concentration (m mol^{-3}) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	32388	32134	5356	1.41	0.247
System	1	172042	162352	162352	42.77	0.000
Genotype x System	6	35411	35411	5902	1.55	0.199
Error	27	102493	102493	3796		

Appendix 5.10 Analysis of variance for flag leaf K^+/Na^+ (ratio) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	20.9837	21.4447	3.5741	7.70	0.000
System	1	32.3864	31.2299	31.2299	67.29	0.000
Genotype x System	6	6.6969	6.6969	1.1162	2.40	0.054
Error	27	12.5314	12.5314	0.4641		

Appendix 5.11 Analysis of variance for flag leaf Ca^{2+} concentration (m mol^{-3}) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	58.32	52.41	8.73	0.65	0.693
System	1	1783.99	1773.71	1773.71	131.07	0.000
Genotype x System	6	51.50	51.50	8.58	0.63	0.702
Error	27	365.38	365.38	13.53		

Appendix 5.12 Analysis of variance for flag leaf Mg^{2+} concentration (m mol^{-3}) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	163.181	164.161	27.360	5.57	0.001
System	1	138.744	138.717	138.717	28.23	0.000
Genotype x System	6	63.579	63.579	10.596	2.16	0.079
Error	27	132.659	132.659	4.913		

Appendix 5.13 Analysis of variance for number of alive plants per pot of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	49.512	47.863	7.977	11.54	0.000
System	1	275.392	268.138	268.138	387.84	0.000
Genotype x System	6	51.941	51.941	8.657	12.52	0.000
Error	27	18.667	18.667	0.691		

Appendix 5.14 Analysis of variance for main tiller height (cm) of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	378.13	387.28	64.55	2.47	0.049
System	1	3073.31	3106.35	3106.35	118.70	0.000
Genotype x System	6	255.56	255.56	42.59	1.63	0.178
Error	27	706.61	706.61	26.17		

Appendix 5.15 Analysis of variance for number of spikes per plant of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	0.95419	0.93958	0.15660	1.64	0.173
System	1	0.37586	0.36598	0.36598	3.84	0.060
Genotype x System	6	0.62856	0.62856	0.10476	1.10	0.388
Error	27	2.57100	2.57100	0.09522		

Appendix 5.16 Analysis of variance for straw weight per plant (g) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	2.04969	2.00670	0.33445	8.72	0.000
System	1	0.13907	0.14657	0.14657	3.82	0.061
Genotype x System	6	0.33368	0.33368	0.05561	1.45	0.232
Error	27	1.03512	1.03512	0.03834		

Appendix 5.17 Analysis of variance for number of infertile spikelets per spike of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	24.2133	23.4958	3.9160	35.40	0.000
System	1	0.0178	0.0365	0.0365	0.33	0.570
Genotype x System	6	4.4085	4.4085	0.7347	6.64	0.000
Error	27	2.9870	2.9870	0.1106		

Appendix 5.18 Analysis of variance for number of fertile spikelets per spike of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	37.2261	39.3060	6.5510	7.47	0.000
System	1	77.2586	80.5392	80.5392	91.86	0.000
Genotype x System	6	14.7535	14.7535	2.4589	2.80	0.030
Error	27	23.6714	23.6714	0.8767		

Appendix 5.19 Analysis of variance for number of tillers per plant of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	1.5474	1.5563	0.2594	1.93	0.112
System	1	0.0475	0.0413	0.0413	0.31	0.594
Genotype x System	6	0.9288	0.9288	0.1548	1.15	0.360
Error	27	3.6265	3.6265	0.1343		

Appendix 5.20 Analysis of variance for tiller index of seven wheat genotypes in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	921.25	961.80	160.30	5.47	0.001
System	1	660.87	674.80	674.80	23.01	0.000
Genotype x System	6	281.60	281.60	46.93	1.60	0.185
Error	27	791.67	791.67	29.32		

Appendix 5.21 Analysis of variance for number of grains per plant of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	774.94	784.55	130.76	4.57	0.003
System	1	2106.44	2149.32	2149.32	75.10	0.000
Genotype x System	6	230.53	230.53	38.42	1.34	0.273
Error	27	772.73	772.73	28.62		

Appendix 5.22 Analysis of variance for grain weight per plant (g) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	1.56557	1.49609	0.24935	17.21	0.000
System	1	7.59216	7.63984	7.63984	527.29	0.000
Genotype x System	6	1.15858	1.15858	0.19310	13.33	0.000
Error	27	0.39120	0.39120	0.01449		

Appendix 5.23 Analysis of variance for number of grains per spike of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	462.44	478.86	79.81	7.68	0.000
System	1	846.99	862.92	862.92	83.00	0.000
Genotype x System	6	82.34	82.34	13.72	1.32	0.282
Error	27	280.70	280.70	10.40		

Appendix 5.24 Analysis of variance for grain weight per spike (g) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	0.88776	0.87587	0.14598	10.61	0.000
System	1	3.54667	3.55461	3.55461	258.27	0.000
Genotype x System	6	0.59307	0.59307	0.09885	7.18	0.000
Error	27	0.37160	0.37160	0.01376		

Appendix 5.25 Analysis of variance for average grain weight (g) of seven wheat varieties in two growing systems (hydroponic and soil culture) under saline conditions. (Experiment 4)

Source	D.F	Seq.S.S	Adj.S.S	Adj.M.S	F	P
Genotype	6	0.0013071	0.0012865	0.0002144	1.91	0.115
System	1	0.0102813	0.0103526	0.0103526	92.41	0.000
Genotype x System	6	0.0011015	0.0011015	0.0001836	1.64	0.175
Error	27	0.0030247	0.0030247	0.0001120		

Appendix 6.1 Analysis of variance for fourth leaf Na^+ concentration (m mol^{-3}) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	1926	642	0.36
Error	8	14337	1792	

Appendix 6.2 Analysis of variance for fourth leaf K^+ concentration (m mol^{-3}) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	15652	5217	3.45
Error	8	12101	1513	

Appendix 6.3 Analysis of variance fourth for leaf K^+/Na^+ (ratio) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	1.9677	0.6559	2.71
Error	8	1.9385	0.2423	

Appendix 6.4 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	8680	2893	1.20
Error	8	19264	2408	

Appendix 6.5 Analysis of variance for sixth leaf Na^+ concentration (m mol^{-3}) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	919.7	306.6	0.37
Error	6	5037.8	839.6	

Appendix 6.6 Analysis of variance for sixth leaf K^+ concentration (m mol^{-3}) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2612.1	870.7	0.94
Error	6	5586.5	931.1	

Appendix 6.7 Analysis of variance sixth for leaf K^+/Na^+ (ratio) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.0598	0.0199	0.06
Error	6	2.1256	0.3543	

Appendix 6.8 Analysis of variance for sixth leaf Cl^- concentration (m mol^{-3}) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2642.2	880.7	1.59
Error	6	3326.3	554.4	

Appendix 6.9 Analysis of variance for main tiller height (cm) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	244.17	81.39	3.65
Error	7	156.02	22.29	

Appendix 6.10 Analysis of variance for number of spikes per plant of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.05923	0.01974	0.38
Error	7	0.36747	0.05250	

Appendix 6.11 Analysis of variance straw weight per plant (g) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.1830	0.0610	0.14
Error	7	3.1210	0.4459	

Appendix 6.12 Analysis of variance number of infertile spikelets per spike of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	3.3365	1.1122	3.06
Error	7	2.5475	0.3639	

Appendix 6.13 Analysis of variance for number of fertile spikelets per spike of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	3.948	1.316	0.54
Error	7	16.923	2.418	

Appendix 6.14 Analysis of variance for number of grains per plant of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	268.31	89.44	2.43
Error	7	257.33	36.76	

Appendix 6.15 Analysis of variance grain weight per plant (g) of four selections from within Alexandria wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.00676	0.002253	7.15
Error	7	0.00221	0.000315	

Appendix 6.16 Analysis of variance for fourth leaf Na^+ concentration (m mol^{-3}) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	13793	4598	2.27
Error	8	16181	2023	

Appendix 6.17 Analysis of variance for fourth leaf K^+ concentration (m mol^{-3}) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	118.9	39.6	0.10
Error	8	3115.3	389.4	

Appendix 6.18 Analysis of variance for fourth leaf K^+/Na^+ (ratio) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2.2739	0.7580	2.33
Error	8	2.6018	0.3252	

Appendix 6.19 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2413.7	804.6	1.06
Error	8	6094.0	761.7	

Appendix 6.20 Analysis of variance for sixth leaf Na^+ concentration (m mol^{-3}) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	3401	1134	0.69
Error	7	11495	1642	

Appendix 6.21 Analysis of variance for sixth leaf K^+ concentration (m mol^{-3}) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	4369	1456	1.12
Error	7	9088	1298	

Appendix 6.22 Analysis of variance for sixth leaf K^+/Na^+ (ratio) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	1.0926	0.3642	1.71
Error	7	1.4888	0.2127	

Appendix 6.23 Analysis of variance for sixth leaf Cl^- concentration (m mol^{-3}) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	4800.2	1600.1	1.93
Error	7	5810.7	830.1	

Appendix 6.24 Analysis of variance for main tiller height (cm) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	84.71	28.24	1.33
Error	8	170.39	21.30	

Appendix 6.25 Analysis of variance for number of spikes per plant of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.2083	0.0694	0.20
Error	8	2.7917	0.3490	

Appendix 6.26 Analysis of variance straw weight per plant (g) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.2795	0.0932	0.88
Error	8	0.8506	0.1063	

Appendix 6.27 Analysis of variance number of infertile spikelets per spike of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.2756	0.0919	0.53
Error	8	1.3819	0.1727	

Appendix 6.28 Analysis of variance for number of fertile spikelets per spike of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	1.0332	0.3444	1.21
Error	8	2.2705	0.2838	

Appendix 6.29 Analysis of variance for number of grains per plant of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	25.03	8.34	0.17
Error	8	395.07	49.38	

Appendix 6.30 Analysis of variance grain weight per plant (g) of four selections from within Kharchia-65 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.002330	0.000777	0.22
Error	8	0.027641	0.003455	

Appendix 6.31 Analysis of variance for fourth leaf Na^+ concentration (m mol^{-3}) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2278	759	0.60
Error	8	10138	1267	

Appendix 6.32 Analysis of variance for fourth leaf K^+ concentration (m mol^{-3}) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	1871.6	623.9	1.01
Error	8	4933.3	616.7	

Appendix 6.33 Analysis of variance fourth for leaf K^+/Na^+ (ratio) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2.8712	0.9571	1.02
Error	8	7.4704	0.9338	

Appendix 6.34 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	5362	1787	0.60
Error	8	23964	2996	

Appendix 6.35 Analysis of variance for sixth leaf Na^+ concentration (m mol^{-3}) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	21781	7260	3.05
Error	8	19056	2382	

Appendix 6.36 Analysis of variance for sixth leaf K^+ concentration (m mol^{-3}) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	335.3	111.8	0.58
Error	8	1547.3	193.4	

Appendix 6.37 Analysis of variance sixth for leaf K^+/Na^+ (ratio) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	3.1171	1.0390	2.23
Error	8	3.7289	0.4661	

Appendix 6.38 Analysis of variance for sixth leaf Cl^- concentration (m mol^{-3}) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	14684.9	4895.0	11.96
Error	8	3274.0	409.2	

Appendix 6.39 Analysis of variance for main tiller height (cm) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	352.39	117.46	4.68
Error	8	200.62	25.08	

Appendix 6.40 Analysis of variance for number of spikes per plant of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.05729	0.01910	1.83
Error	8	0.08333	0.01042	

Appendix 6.41 Analysis of variance straw weight per plant (g) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.28047	0.09349	6.48
Error	8	0.11540	0.01443	

Appendix 6.42 Analysis of variance number of infertile spikelets per spike of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	2.3318	0.7773	4.56
Error	8	1.3622	0.1703	

Appendix 6.43 Analysis of variance for number of fertile spikelets per spike of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	25.354	8.451	7.77
Error	8	8.701	1.088	

Appendix 6.44 Analysis of variance for number of grains per plant of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	845.05	281.68	5.60
Error	8	402.69	50.34	

Appendix 6.45 Analysis of variance grain weight per plant (g) of four selections from within KRL1-4 wheat variety under saline conditions. (Experiment 5)

Source	D.F	S.S	M.S	V.R
Genotype	3	0.096479	0.032160	5.98
Error	8	0.043049	0.005381	

Appendix 6.46 Analysis of variance for fourth leaf Na^+ concentration (m mol^{-3}) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	5166	1291	3.46	0.051
Error	10	3729	373		

Appendix 6.47 Analysis of variance for fourth leaf K^+ concentration (m mol^{-3}) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	1456	364	2.50	0.110
Error	10	1458	146		

Appendix 6.48 Analysis of variance for fourth leaf K^+/Na^+ (ratio) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.09593	0.02398	4.05	0.033
Error	10	0.05920	0.00592		

Appendix 6.49 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	5629	1407	3.53	0.048
Error	10	3985	398		

Appendix 6.50 Analysis of variance for main tiller height (cm) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	471.5	117.9	2.88	0.080
Error	10	409.0	40.9		

Appendix 6.51 Analysis of variance for number of spike per plant of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.1718	0.0430	1.17	0.380
Error	10	0.3669	0.0367		

Appendix 6.52 Analysis of variance for straw weight per plant (g) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.3716	0.0929	2.39	0.120
Error	10	0.3879	0.0388		

Appendix 6.53 Analysis of variance for number of infertile spikelets per spike of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	5.729	1.432	1.95	0.179
Error	10	7.354	0.735		

Appendix 6.54 Analysis of variance for number of fertile spikelets per spike of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	44.12	11.03	7.82	0.004
Error	10	14.10	1.41		

Appendix 6.55 Analysis of variance for number of grains per plant of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	407.5	101.9	2.95	0.076
Error	10	345.9	34.6		

Appendix 6.56 Analysis of variance for grain weight per plant (g) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.03592	0.00898	3.50	0.049
Error	10	0.02566	0.00257		

Appendix 6.57 Analysis of variance for number of grains per spike of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	161.0	40.3	2.22	0.140
Error	10	181.2	18.1		

Appendix 6.58 Analysis of variance for grain weight per spike (g) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.01397	0.00349	2.67	0.095
Error	10	0.01309	0.00131		

Appendix 6.59 Analysis of variance for average grain weight (g) of four wheat selections and their parent Alexandria under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.0000156	0.0000039	2.58	0.102
Error	10	0.0000151	0.0000015		

Appendix 6.60 Analysis of variance for fourth leaf Na^+ concentration (m mol^{-3}) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	2948	737	1.89	0.189
Error	10	3902	390		

Appendix 6.61 Analysis of variance for fourth leaf K^+ concentration (m mol^{-3}) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	1805.3	451.3	4.69	0.022
Error	10	962.7	96.3		

Appendix 6.62 Analysis of variance for fourth leaf K^+/Na^+ (ratio) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.16191	0.04048	6.62	0.007
Error	10	0.06113	0.00611		

Appendix 6.63 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	674	169	0.46	0.765
Error	10	3674	367		

Appendix 6.64 Analysis of variance for main tiller height (cm) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	111.94	27.99	0.33	0.852
Error	10	847.91	84.79		

Appendix 6.65 Analysis of variance for number of spike per plant of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.1176	0.0294	0.08	0.986
Error	10	3.5852	0.3585		

Appendix 6.66 Analysis of variance for straw weight per plant (g) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.1159	0.0290	0.21	0.925
Error	10	1.3569	0.1357		

Appendix 6.67 Analysis of variance for number of infertile spikelets per spike of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.6162	0.1540	1.39	0.306
Error	10	1.1086	0.1109		

Appendix 6.68 Analysis of variance for number of fertile spikelets per spike of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	1.1493	0.2873	0.74	0.583
Error	10	3.8593	0.3859		

Appendix 6.69 Analysis of variance for number of grains per plant of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	112.0	28.0	0.13	0.966
Error	10	2100.0	210.0		

Appendix 6.70 Analysis of variance for grain weight per plant (g) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.05950	0.01488	0.32	0.858
Error	10	0.46370	0.04637		

Appendix 6.71 Analysis of variance for number of grains per spike of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	33.58	8.39	0.55	0.705
Error	10	153.23	15.32		

Appendix 6.72 Analysis of variance for grain weight per spike (g) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.021085	0.005271	0.92	0.487
Error	10	0.057006	0.005701		

Appendix 6.73 Analysis of variance for average grain weight (g) of four wheat selections and their parent Kharchia-65 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.00003934	0.00000983	0.85	0.525
Error	10	0.00011555	0.00001155		

Appendix 6.74 Analysis of variance for fourth leaf Na^+ concentration (m mol^{-3}) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	3337	834	2.17	0.146
Error	10	3843	384		

Appendix 6.75 Analysis of variance for fourth leaf K^+ concentration (m mol^{-3}) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	3186	796	2.84	0.082
Error	10	2802	280		

Appendix 6.76 Analysis of variance for fourth leaf K^+/Na^+ (ratio) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.2154	0.0538	2.07	0.160
Error	10	0.2598	0.0260		

Appendix 6.77 Analysis of variance for fourth leaf Cl^- concentration (m mol^{-3}) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	2	3757	939	2.40	0.119
Error	10	3911	391		

Appendix 6.78 Analysis of variance for main tiller height (cm) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	40.61	10.15	0.20	0.935
Error	10	516.38	51.64		

Appendix 6.79 Analysis of variance for number of spike per plant of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.05429	0.01357	0.32	0.858
Error	10	0.42300	0.04230		

Appendix 6.80 Analysis of variance for straw weight per plant (g) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.11317	0.02829	1.60	0.249
Error	10	0.17700	0.01770		

Appendix 6.81 Analysis of variance for number of infertile spikelets per spike of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	1.2156	0.3039	0.62	0.661
Error	10	4.9356	0.4336		

Appendix 6.82 Analysis of variance for number of fertile spikelets per spike of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	6.487	1.622	0.95	0.477
Error	10	17.154	1.715		

Appendix 6.83 Analysis of variance for number of grains per plant of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	110.08	27.52	0.34	0.845
Error	10	808.63	80.86		

Appendix 6.84 Analysis of variance for grain weight per plant (g) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.072461	0.018115	2.30	0.130
Error	10	0.078811	0.007881		

Appendix 6.85 Analysis of variance for number of grains per spike of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	94.50	23.62	0.26	0.896
Error	10	902.72	90.27		

Appendix 6.86 Analysis of variance for grain weight per spike (g) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.063643	0.015911	2.16	0.148
Error	10	0.073829	0.007383		

Appendix 6.87 Analysis of variance for average grain weight (g) of four wheat selections and their parent KRL1-4 under saline conditions. (Experiment 6)

Source	D.F	S.S	M.S	F	P
Genotype	4	0.000042205	0.00001055	1.27	0.344
Error	10	0.000083040	0.00000830		

APPENDIX 7

EQUIPMENT USED

Aerators: 'Supa' Aquatic Supplies Ltd., 'Conway' Hawthorne Close, Barlborough, Chesterfield, Great Britain.

Air Compressor: Compair-Brown Wade, High Waycomb, England.

Balances: Sartorius, West Germany.

Bungs: Grey Neoprene Bungs, Scientific Services, High Street Tattenhall, Chseter, England.

Centrifuge: Clandon MLW T52.1, Centrifuge, England.

Conductivity Meter: Model p335, Portland Electronics Ltd., 18 Greenacres Road, Oldham, England.

Fridge: Vindon Scientific Ltd, Diggle, Oldham, England.

Large Drying Oven: Unitherm, Drying Oven, Russell-Lindsey Light Engineering Ltd., 60-62 Constitution Hill, Birmingham, England.

Needles: Terumo needles (236 x 1.25), Fiscorns/MSE, MSE Scientific Instruments, Manor Royals, Crawley West Sussex, England.

Phostrogen: Photrogen Ltd., Corwen, Clwyd, UK.

Pipettes: Eppendorf Varipipette (4720) and Multipipette (4780). Eppendorf Geratenbau, Netherlert, Hirz, Gmbh, Postfach 65, 0670, 2000, Hamburg 65, West Germany.

Plantpak Plug Trays P180: Cookson Plantpak, Mundon, Maldon, Essex, England.

Pots: WCB Container, Cookson Plantpak, Mundon, Maldon, Essex, England.

Seed Counter: Numigral-Tecator, Box 70, 5-26301, Hoganas, Sweden.

Tubing: Silicon Tubing and non-sterill polythene tubing, Portex Ltd., Hythe, Kent, England

Vortex Stirrer: Gallenkamph Spinmix, England.