

**RESPONSES OF TWO GRASS SPECIES TO PLANT GROWTH  
REGULATORS, FERTILIZER N, CHELATED Fe, SALINITY  
AND WATER STRESS**

by

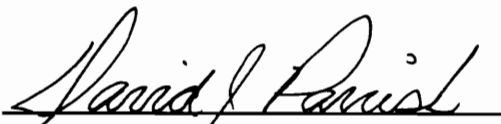
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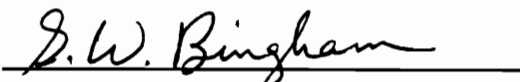
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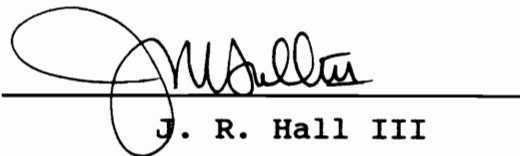
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**R. E. Schmidt, chairman**

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**(ABSTRACT)**

A series of studies were initiated to investigate growth responses of Kentucky bluegrass (Poa praetensis L.) and creeping bentgrass (Agrostis palustris Huds.) to foliar applications of two plant growth regulators (PGR) and/or chelated Fe (Na Fe diethylene triamine pentaacetate). Environmental variables considered were N levels, soil moisture regimes, and saline irrigations

The two materials investigated for PGR properties were a commercial product called Roots (a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine) and the systemic fungicide propiconazole, trade name: (Banner) and chemical name: [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2yl)methyl-1 H-1,2,4-triazole)]. Fortified seaweed extract (FSE) was applied at 9.3 L/ha, and propiconazole (PPC) was applied at 0.93 L a.i/ha. Each was applied alone or in conjunction with chelated Fe at 0.11 kg a.i/ha.

Kentucky bluegrass foliage height, root and shoot dry weight, and several foliar nutrients increased following PGR

treatments when grown under either limited soil moisture or saline irrigation.

Foliar applications of PGR and/or chelated Fe to creeping bentgrass reduced wilting and evapotranspiration, and increased leaf water status, root strength, and shoot dry matter at two levels of N during and after drought stress.

**DEDICATED TO  
MY MOM, MY BROTHERS AND SISTERS,  
AND THE LOVING MEMORY OF MY FATHER.**

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## **CHAPTER 1**

### **INTRODUCTION AND LITERATURE REVIEW**

The growth of plants involves genetic events and hormonal, physiological, and morphological factors. Since all these factors respond to environmental conditions, it is not surprising that environmental stresses can have a negative impact on plant growth and development (Parsons, 1982; Krizek et al., 1985). Water deficit and substrate salinity are major problems in agricultural production in many parts of the world; however, the mechanisms by which these stresses limit plant growth are not well understood (Munns and Termaat, 1986). Drought stress and toxic concentrations of ions such as Na and Cl can ultimately limit the supply of assimilate to the growing regions, resulting in low yields. Prolonged transpiration can concentrate salts in growing tissues, especially in older leaves, eventually killing them (Munns and Termaat, 1986). Drought stress and salinization can reduce plant survival in any habitat. These stresses can impair physiological and morphological determinants, photosynthesis, protein biosynthesis, hormonal function, ion movement, cell division, and differentiation of the plants.

#### **WATER DEFICITS**

Tissue water deficits occur when transpiration rate exceeds absorption (Kozlowski, 1968). Transpiration depletes water from the root zone, and the soil water potential drops, altering the relationship between water uptake and loss. Plants subjected to limited water grow slowly and experience alterations in metabolism and organelle activities that can result in further growth reductions and death (Hsiao, 1973). Turner (1979) noted that water stress produces physical changes such as cessation of cytoplasmic streaming and alterations in metabolic functions. Willey (1970) and Cocucci et al. (1976) reported that water deficits cause a rapid and severe wilting in the above ground parts of plants. Leaf expansion in particular and vegetative growth in general decline under moderate water stress (Turner and Kramer, 1980).

Sionit et al. (1973) observed that sunflower plants produce smaller leaves and less vegetative growth as soil moisture levels drop below  $-0.03$  MPa. Oak leaf and seedling growth slow as the soil water potential drops from  $-0.03$  to  $-0.8$  MPa (Larson and Palashev, 1973). Under water stress, roots elongate irregularly without root hairs, and they may die if the drought is prolonged (Turner and Kramer, 1980).

Cell growth and differentiation are related to cell turgor. Water stress or reduced osmotic potential at the

cellular level results in decreasing intercellular spaces and changes in membrane properties (Burstrom, 1975; Smart and Bibgham, 1974). Reduction in leaf water status can reduce photosynthesis due to stomatal closure. The failure of guard cells to maintain solute movement and generate sufficient osmotic potential leads to loss of turgor pressure for stomatal activity (Ehret and Boyer, 1979; Kech and Boyer, 1974). Turner and Kramer (1980) proposed that abscisic acid might behave as a cytokinin inhibitor in root and shoot tissues of water-stressed plants. Chloroplast activity, especially the enzymes associated with thylakoid formation, protein synthesis, and polyribosome formation decline as water potential decreases (Boyer, 1976; Ben-Zion et al., 1967; Cocucci et al., 1976). Deltour and Jacquard (1974) reported that the disturbance in polyribosome formation that is induced by water stress is preceded by a disturbance in the metabolism of DNA and RNA, which play a primary role in protein biosynthesis. Physiological disorders and biochemical imbalances under water stress reduce plant size and biomass production.

### **SALINITY**

Soluble salts, especially NaCl, either in soil or irrigation water, can be detrimental to agriculture. About 40

million hectares of the world's 220 million hectares of irrigated soils are adversely affected by excessive salt concentrations (Wignearajah, 1990). With a further 160 million hectares in desert regions, nearly 40% of the world's arable land can be classified as areas with potential salinity problems.

Substrate salinity, especially that produced by NaCl, has a negative impact on crop productivity. Sodium and Cl are commonly the most prevalent ions in a saline soil. Their specific toxicities can account for many of the adverse effects of salinity on growth and plant quality (Shannon, 1984). Plants subjected to a saline environment encounter three basic problems: a) a reduced soil water potential, making water less available, b) phytotoxic substances that interfere with physiological and biochemical processes, and c) essential nutrient ions must be obtained despite the predominance of other ions (Rains, 1979). Yield reductions on irrigated land that was once productive are often due to accumulation of salts. Such salinization reduces soil moisture availability and alters ionic balances within the soil particles (Hoffman et al., 1983).

Higher plants can be categorized in two general groups according to their ability to cope with salinity: a) halophytes, those that are capable of surviving at relatively high salt concentrations in their root zones, and b) non-

halophytes or glycophytes, those that are not able to adjust to substrate salinities in their root zones (Salisbury and Ross, 1978). Werker et al. (1983) proposed that growth reductions of non-halophyte plants in a saline environment are brought about by various physiological, morphological, and anatomical changes due to excessive amounts of soluble salts in their root zones. These changes can be related either to reduce water availability near the roots and an osmotic inhibition of water uptake or to toxic materials such as Na and Cl ions in the soil solution (Zekri and Lawrence, 1989). Plants in a saline soil have leaves that grow more slowly with smaller leaf-area expansion (Munns et al., 1982). Wignarajah (1990) concluded that salinity-induced reductions of leaf and whole plant size may be due to: a) reduced size and number of stomata per leaf, b) lowered chlorophyll content and rate of photosynthesis, c) reduced water availability, and d) increased transpiration. At low salinity, root growth may not be affected while vegetative growth is reduced.

Substrate salinity effects are seen at the subcellular levels. Salt concentrations impair membrane structure and integrity and cause organelle swelling and structural distortion in chloroplast, mitochondria, Golgi bodies, and the endoplasmic reticulum (Mass and Nieman, 1978; Udovenko et al., 1984). They also noted that specific enzymes, total proteins, and nucleic acids are significantly reduced in

quantity because the plants are smaller; however, specific activity and concentration often remain constant, in balance with a slower growth rate. Kuiper (1984) reported that, when plants are subjected to a saline soil, ATPase activity is suppressed, and long-term exposure of the plant to NaCl induces a conformational change of the K translocator of the ATPase complex. Jeschke (1984) also noted that salinity stress can stimulate production of abscisic acid (ABA) levels. In beet and barley roots, concentrations of ABA shifted ion uptake in favor of Na and inhibited K.

Since it is often not feasible to modify environments to suit plants, researchers are now being challenged to alter plants for survival in adverse environments (Staples and Toenniessen, 1984). In recent years, naturally occurring and synthetic plant growth regulators, especially cytokinins and triazole compounds, have been tested. Application of essential mineral nutrients, and breeding strategies are also becoming increasingly important tools in the field of plant growth regulation in order to integrate approaches for crop productivity under drought and saline environments.

#### **PLANT GROWTH REGULATORS**

Plant growth regulators (PGR) are naturally occurring or synthetic compounds that can modify plant growth and

development when applied in small quantities. The use of PGR offers unique possibilities in agricultural production. Some of these substances can modify plant characteristics that affect the ability to cope with environmental stress (Fletcher, 1985). One type of PGR currently being used for enhancing plant growth under harsh environments is fortified seaweed extract. Such extracts are marketed as containing naturally occurring phytohormone including cytokinins. Also having potential PGR properties are triazole compounds, such as propiconazole (Banner). These fungicides are known to be ergosterol biosynthesis inhibitors (Brain et al., 1973; Fletcher, 1985). Their growth regulatory abilities apparently stem from their alteration of biosynthesis of important endogenous plant growth substances like abscisic acid, gibberellin, and cytokinins.

Cytokinins were first identified in 1955, when kinetin (6-furfurylamino purine) was shown to promote mitosis and cell differentiation in tobacco callus (Moore, 1979). Ten years later, another cytokinin, zeatin, was extracted from developing corn seeds. This compound is also a major component of coconut milk (Letham, 1968). By 1967, cytokinins had been isolated and identified from almost 40 plant species (Moore, 1979). They are now thought to be of essentially universal occurrence in higher plants. Cytokinins are a unique class of plant hormones; they contain an adenine group, which also

occurs in nucleic acids. They are able to bind with tRNA during DNA translation (Moore, 1979; Letham, 1982).

Six-benzylamino purine, an adenine derivative, is a synthetic cytokinin. It has been used to fortify seaweed extracts, which contain naturally occurring cytokinins, and for agricultural production (Kende, 1971; Kane and Smiley, 1983). [Aqueous seaweed extracts are natural products. In addition to having cytokinin activity, they also contain inorganic compounds that serve as essential nutrients (Blunden and Wildgoose 1977; Nelson and Van Standen, 1984).

The triazole derivatives propiconazole and triadimefon, which are active in controlling several important plant diseases, are also included in plant growth regulators (Ragsdale, 1977; Ali et al., 1979; Brain et al., 1973). Booth (1966), Thomas (1974), and Gautam et al. (1984) discussed cytokinin-like activity and cytokinin-containing materials that have been suggested as active ingredients in these substances. Fletcher and Arnold (1986) found an enhancement of cytokinin production in triazole-treated cucumber, while a bioassay revealed no indication of cytokinin-like activity.

#### **PHYSIOLOGICAL GROWTH RESPONSES TO PLANT GROWTH REGULATORS**

It is now well established that plant roots are the sites of cytokinin biosynthesis (Itai and Vaadia, 1965). Plant roots

not only synthesize the cytokinins but transport them through the xylem to above-ground parts of the plant (Thimann et al., 1974). They also noted that cytokinins are capable of promoting cell division and differentiation, chlorophyll production, photosynthesis, DNA transcription, and enzyme activity. Michael and Beringer (1980) reported that wheat spikelet number, floret number, terminal spikelet differentiation, and seed formation and development can be influenced by endogenous cytokinins.

In an experiment with bean leaves soaked in solutions of kinetin, the cytokinin enhanced leaf size by cell division and expansion (Miller, 1956). Stomatal aperture, transpiration, and proteins associated with activity of guard cells were enhanced by a range of naturally occurring and synthetic cytokinins (Jewer and Incoll, 1980; Kurashi and Ishikawa, 1977). Foliar applications of cytokinins and cytokinin-like growth regulators to seedlings of Kentucky bluegrass stimulated lateral bud formation, leaf number, root elongation, and root and shoot biomass (Goatley, 1988; Goatley and Schmidt, 1990). Watermelon hypocotyl and cotyledons of cucumber are promoted by cytokinin-like solutions (Ross and Rayle, 1982). Application of cytokinin-containing materials has produced different effects on root formation. Kentucky bluegrass treated with benomyl produced a larger root mass and greater rhizome production (Smiley et al., 1985).

Benzyladenine enhanced cambial growth in spruce tree roots (Philipson and Coutts, 1980). Nelson and Van Standen (1984) observed that plants under nutrient stress treated with cytokinin-like substances showed a positive growth response.

Sexton and Woolhouse (1984) reported that cytokinin-like growth regulators stimulate chlorophyll content and protein biosynthesis. Proteins are usually degraded during the senescence process in leaves. Cytokinins inhibited the activity of free radical groups, hydrogen peroxide, and super oxide, which are the main elements for chlorophyll degradation during senescence. Cytokinin materials, in addition to acting as anti-senescence agents, stabilize polysome aggregates and chloroplast activity and direct the movement of certain essential ions and amino acids to portions of a plant that have been treated with these cytokinins (Marre et al., 1974). Cytokinin-like growth regulators can activate enzymes and the hormone-receptor complex within plant cells. Such regulation leads to an activation of RNA promoter and RNA polymerases in the cell nucleus. This results in activation of the whole system of protein biosynthesis and leads to higher photosynthetic rates (Chernyadcev et al., 1984).

#### **PLANT GROWTH REGULATORS AND ENVIRONMENTAL STRESSES**

Environmental stresses such as drought and salinity can

lead to changes and imbalances of endogenous plant growth regulators. Under some stress conditions, the production of abscisic acid (ABA) is enhanced. Among other effects, ABA inhibits H/K exchange across the plasmalemma of guard cells, synthesis of  $\alpha$ -amylase, and root geotropism (Sivakumaran and Hall, 1978; Rashke, 1977). Itai and Vaadia (1965) documented that environmental stresses might reduce transport of cytokinins from root systems to above ground parts of plants, lowering cytokinin concentrations in shoot tissues with consequent effects on shoot physiology. Plant roots are sites of cytokinin formation and are responsible for biosynthesis of some proteins in leaf tissues (Chibnall, 1939). Tomato root systems supply a factor that prevents leaf chlorosis (Went and Bonner, 1943). This is old work that we now know/believe is explained as root production of cytokinins and cytokinin prevention of senescence. (Chlorosis is one sign of senescence.) Induction of ABA synthesis in the stressed plant can lead to direct effects on root activity and availability of minerals (Sivakumaran and Hall, 1978).

Taylor (1980) and Bhan et al. (1973) proposed that those plants that develop larger root systems with a vigorous root tip are able to avoid yield losses from rapid wilting injuries. A larger root system is required, especially in dry environments where moisture is deeper in the soil. Humphries et al. (1967) reported that foliar application of plant growth

regulators to wheat under drought conditions stimulated root systems that enabled the plants to obtain more water and nutrients from deeper in the soil profile.

Cytokinin-like growth regulators applied to plants under drought stress might replace endogenous cytokinins whose production is reduced by stress conditions (Fletcher et al., 1986; Van Steveninck, 1972; Aharoni et al., 1977; Bengston et al., 1979). Increases of cytokinins enhance ion exchange across membranes of guard cells, tend to reduce abscisic acid levels, and improve leaf transpiration (Sivakumaran and Hall, 1978; Itai and Vaadia, 1965). Cooper and Digby (1972) showed that cytokinin-like substances can be used to suppress rapid wilt. The triazole compounds are known to protect plants from various stresses, including low and high temperatures, air pollutants, and drought (Fletcher et al., 1986). Application of gibberellic acid (GA) to celery plants treated with a NaCl solution to the roots resulted in substantial increases in leaf tissues and petiole elongation (Aloni and Pressman, 1980). Lettuce seeds soaked in fusicoccin or in a combination of three growth regulators, kinetin, ethephon, and gibberellic acid, germinated and emerged more readily in a saline soil (Joseph and Anwar, 1976). Flowers and Yeo (1983) reported that polyethylene glycol added to NaCl solution increased the rate of rice seedling survival.

**IRON**

Iron is the fourth most abundant plant nutrient, existing in the form of ferromagnesium silicate in the earth's lithosphere. During weathering, Fe precipitates as oxides or hydroxides, and only a small part of these oxides is released into secondary silicate form (Schwertmann and Taylor, 1977). Iron deficiencies have become a potential problem for agricultural production in many arid and semiarid regions. Plants that suffer from Fe deficiencies usually develop chlorotic interveinal areas in the new leaves, resulting in lower photosynthetic rates (Schwertmann and Taylor, 1977). Iron chlorosis is due to the inability of plant species to take up oxidized Fe(III), the form of Fe that is predominately in soil particles (Blaylock et al., 1989). Inorganic Fe, such as oxidized ferric hydroxides and Fe(III), have to be reduced to Fe(II) form in order to be absorbed by plant roots (Chen and Barak, 1988). Higher plants can be categorized into two general groups according to their ability to obtain Fe from the soil. Most dicot species are Fe-efficient; they are able to respond to Fe oxides by activating some biochemical processes in their root systems and to convert Fe oxides into the Fe(II) available form. Monocot species, including grasses, are generally unable to change Fe oxides into an available form of Fe. These plants are called Fe-inefficient and must

receive Fe fertilization during the growing season (Brown, 1978; Romheld and Marschner, 1986; Camp et al., 1987; Marschner et al., 1986).

#### **PHYSIOLOGICAL AND BIOCHEMICAL RESPONSES TO IRON**

Iron plays a major role in the establishment of chloroplast constituents, electron carriers, enzyme synthesis, and photosynthetic activity in higher plants. Many pathways of biosynthesis are highly dependent on the presence of Fe in the leaves (Terry, 1979; Blylock et al., 1989; Pushnik and Gene, 1989; Miller et al., 1982; Cherskin and Castelfranco, 1982). The use of chelated Fe has become an common in nutrient media, since Fe is the most difficult of all plant nutrients to maintain in available form (Brown, 1978). Pushnik and Gene (1989) observed that chlorophyll levels and photosynthetic rates of tobacco (Nicotiana tabacum) were increased by 96% within the first 10 days of foliar application of chelated Fe. Root growth development of Merion Kentucky bluegrass significantly improved when Fe was added to nutrient culture (Deal and Engel, 1956). Vegetative growth of bentgrass plants significantly improved when chelated Fe was applied in combination with N fertilizer (Snyder and Schmidt, 1973). Schmidt and Snyder (1984) also showed that application of chelated Fe in conjunction with high N caused better root

structures in creeping bentgrass under moisture stress. Goatley and Schmidt (1990) reported variable responses from Kentucky bluegrass seedlings when cytokinin-like growth regulators were applied with chelated Fe. Spiller and Terry (1980) reported that the chlorophyll-protein complex, the condensation of P700, cytochrome f, chloroplast membranes, and electron carriers sharply declined in Fe-stressed leaves. When chelating agents were used to supply Fe to nutrient culture of corn plants, root and shoot growth significantly increased over plants grown in solutions without added Fe (Majamder and Dunn, 1958). Heath and Clark (1956) showed that lack of Fe in nutrient solutions of grass species caused a significant reduction in photosynthetic rates and cell division and differentiation of the root tissues.

**THE OBJECTIVES OF THESE STUDIES WERE TO :**

i) evaluate the influences of soil moisture, plant growth regulators, and chelated Fe on Kentucky bluegrass growth and development.

ii) examine the effect of plant growth regulators, chelated Fe, and nitrogen fertility on creeping bentgrass subjected to drought stress.

iii) determine if salinity stress on Kentucky bluegrass can be alleviated by plant growth regulators and chelated Fe.

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## CHAPTER 2

### INFLUENCE OF SOIL MOISTURE, PLANT GROWTH REGULATORS, AND CHELATED Fe ON KENTUCKY BLUEGRASS PERFORMANCE

#### ABSTRACT

Kentucky bluegrass (*Poa praetensis* cv. 'plush') seeds were sown at 16 kg/ha in plastic flats filled with Groseclose silt loam soil. Two weeks after germination, when the plants were at the two-leaf stage, seedlings were treated with two plant growth regulators (PGR) and chelated Fe. One PGR was Roots, a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine (FSE). It was applied at 9.3 L/ha. The other PGR was the systemic fungicide propiconazole (trade name: Banner, chemical name: [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl)methyl-1H-1,2,4-triazole] (PPC). It was applied at 0.93 L active ingredient (a.i)/ha. Each PRG was applied alone or in conjunction with Na-diethylene-triamine-pentaactate chelated Fe at 0.11 kg a.i/ha.

Moisture release curves were used to develop soil water potentials of -0.5, -0.3, and -0.03 MPa, respectively. Terrarium-like boxes were constructed, and into each of these boxes was placed soil adjusted to one of the three moisture levels. Each terrarium received transplanted plugs that had received each of the six chemical treatments. Terraria were

placed in an air-conditioned greenhouse in which temperature was maintained at 21°C in the day and 18°C at night.

As soil water potential increased, foliage growth of treated plants increased. Both PGR either alone or with chelated Fe stimulated plant height under all three soil moisture levels. Plants maintained a better leaf water status (LWS) when grown under the higher than the lower soil moisture tension. All chemical treatments improved LWS as compared to the control plants.

Root development of chemically treated plants were greater under two moderately low soil moisture tensions. This resulted in higher root dry matter in plants that grown under the lower (-0.5 MPa) than the higher (-0.03 MPa) soil water potential. Propiconazole plus Fe and Fe alone at -0.5 MPa, Fe alone at -0.03 MPa, and PPC at -0.3 MPa were among plant treatments that increased shoot dry matter.

When averaged across all chemical treatments, total leaf protein content of plants increased at the highest soil moisture level. Bluegrass seedlings treated with PGR and/or chelated Fe exhibited improved uptake of several minerals. Treated leaf tissues were higher in P, K, and Fe when compared to seedlings that received no PGR or chelated Fe treatments under three different soil moisture tensions. Chelated Fe was equally or more beneficial than (FSE) or PPC, but these did not caused synergistic or additive effects.

## INTRODUCTION

Moisture stress affects plant growth and development in several ways. Plants subjected to low soil moisture grow slowly; the relative growth rate of various organs is inhibited (Nus and Hodges, 1986). Schultz and Matthews (1988) showed that growth of grape (Vitis vinifera L.) is substantially inhibited at a soil water potential of  $-0.065$  MPa and ceases completely at  $-0.54$  MPa. Lateral bud initiation and rhizome formation in grass species are reduced drastically during periods of soil water deficits (Beard, 1973). Water stress during vegetative growth affects leaf expansion and total leaf number, resulting in less dense plant canopies (Shaw, 1977; Arkin et al., 1983). Moisture stress not only reduces final leaf size but also decreases the rate of leaf initiation, causing decreases in plant size (Krieg and Sung, 1979). Reducing or halting vegetative growth and leaf emergence limits carbohydrate production (Jones et al., 1981; Steinberg et al., 1990). Limited soil moisture also can reduce water content and net photosynthesis (Frank et al., 1973).

Mayaki et al. (1976) and Shaw (1978) documented that above-ground vegetative growth may be restricted where soil water is limited for a short period, but deeper and more prolific root growth may result. Taylor (1980) found that plants grown under relatively dry soil conditions have deeper

and larger root systems. Such roots can absorb water stored deep in the soil and provide for plant survival during periods of moisture stress. Plant roots not only transport water and nutrients, but they also provide shoots with phytohormones, such as cytokinins, that are essential for growth and development (Blackman and Davies, 1985; Turner, 1986). Moisture stress can interrupt cytokinin production in roots, which results in reduction of nutrient uptake and restriction of stomatal activity (Blackman and Davies, 1985; Davies et al., 1986). Bengston et al. (1979) reported that plants subjected to water stress accumulate abscisic acid in roots, and that might cause an inhibition of cytokinin biosynthesis in the roots.

Moisture stress affects stomatal activity, leaf water potential, hormonal activities, and the availability of nutrients, especially K and P (Duniway, 1971; Ehret and Boyer, 1979; Mouat and Ness, 1986; Jupp and Newman, 1987). A reduction of K concentration in leaf tissue can impair the activity of guard cells. Chlorophyll contents activation of enzymes associated with light reactions, levels of free amino acids, and protein synthesis are reduced significantly in response to soil water deficits (Kemble and MacPherson, 1954; Kech and Boyer, 1974; Ehret and Boyer, 1979; Mouate and Ness, 1986).

Naturally occurring and synthetic plant growth regulators

Limit water potential

kg m<sup>-2</sup>

29

applied in conjunction with mineral nutrients have drawn considerable attention for crop production in adverse environments (Majamder and Dunn, 1958; Cothren and Stutte, 1973). Plant growth regulators have been shown to modify responses to drought stress. Lovett and Campbell (1973) found that foliar applications of cycocel (CCC) to sunflower (Helianthus annuus L.) significantly improved physiological and morphological characteristics of moisture-stressed plants. Two anti-gibberellin plant growth regulators applied to corn (Zea mays L.) under limited soil moisture stimulated vegetative growth and promoted yield about 16% relative to untreated plants (Shanahan and Nielsen, 1987). Mepequat chloride enhanced leaf water potential and the Ca, P, and Mg concentrations in leaves of cotton (Gossypium hirsutum L.) under limited irrigation (Cothren et al., 1983). Naturally occurring and synthetic cytokinins are known to induce plant growth and development under adverse environments (Nelson and Van Standen, 1984). Cytokinin responses include enhanced cotyledon and leaf expansion, leaf number, root initiation and elongation, lateral bud formation, and rhizome production (Miller, 1956; Philipson and Coutts, 1980; Nelson and Van Standen, 1984; Smiley et al., 1985; Goatley and Schmidt, 1990). Foliar application of cytokinins to drought-stressed plants increased stomatal activity, transpiration (Jewer and Incoll, 1980), chlorophyll content, and protein biosynthesis

(Kuraishi and Ishikawa, 1977; Sexton and Woolhouse, 1984).

Schmidt and Snyder (1984) reported that chelated Fe promoted growth of bentgrass (Agrostis palustris L.) under low soil moisture. However, foliar application of seaweed extract or propiconazole in conjunction with chelated Fe to well-irrigated seedling Kentucky bluegrass (Poa pratensis L.) produced variable growth responses (Goatley and Schmidt, 1990). Chelated Fe increased root growth of 'Merion' Kentucky bluegrass in nutrient culture (Deal and Engel, 1965). Majamder and Dunn (1958) noted that chelating agents significantly increased root and shoot growth of corn plants grown in solution.

The data on PGR and Fe effects on relief of drought stress are inconsistent, especially in studies with grass species. The need still exists for conclusive studies of growth performance of grass species under a wide range of soil moisture conditions. The objective of this study was to determine the growth responses of seedling Kentucky bluegrass to foliar applications of two PGR materials and chelated Fe under different soil moisture regimes.

#### **MATERIALS AND METHODS**

Kentucky bluegrass (Poa praetensis cv. 'Plush') seeds were sown on 17, June 1989 at 16 kg/ha in plastic flats filled

with Groseclose silt loam soil (Typic Hapludult, pH 6.4). Soil P and K were 30 and 57  $\mu\text{g/g}$ , respectively. Seeded flats received 50 to 60 ml of water twice a day. Two weeks after germination, when the grass was at the two-leaf stage, seedlings were treated with two PGR materials and Fe. One PGR was Roots, a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine marketed by Roots Inc. It was applied at 9.3 L/ha. The systemic fungicide propiconazole (trade name: Banner, chemical name: [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2yl)methyl-1 H-1,2,4-triazole] (PPC), produced by Ciba Geigy, was also tested for its PGR-like properties. It was applied at 0.93 L a.i/ha. Each PGR was applied alone or in conjunction with Na-diethylene-triamine-pentaactate chelated Fe (Ciba Geigy 330Fe) at 0.11 kg a.i/ha. All treatments were applied with a compressed-air sprayer delivering 784 L/ha at 290 kPa. The treated seedlings were kept in the greenhouse and irrigated daily. Mean temperature in the greenhouse during the experiment was 24°C in the day and 20°C at night.

Groseclose soil was air-dried in a greenhouse for 2 weeks and then screened with a 2-mm mesh sieve. Moisture release curves, determined according to the methods of Richards (1965), showed the soil required 11.6, 14.4, and 20.1 g of water per 100 g of soil to develop soil water potentials of -0.5, -0.3, and -0.03 MPa, respectively. Aliquots of 22 kg of

this soil with appropriate water and a 20-20-20 soluble fertilizer (to supply 50 kg N/ha) were mixed thoroughly before being placed into plastic containers, sealed, and allowed to equilibrate for 72 h.

Terrarium-like boxes (30 cm high × 40 cm wide × 75 cm long) were constructed with 0.15-mm clear plastic sides and tops. Into each of these boxes was placed 22 kg of soil adjusted to one of the three moisture levels. This provided a 7.5-cm soil depth. Plugs, 3.8 cm in diameter, of the treated Kentucky bluegrass seedlings were removed from the flats 2 weeks after chemical treatments. The soil was gently washed from the roots, and the plugs were transplanted to the terraria containing each of the three moisture levels. Each terrarium received 24 plugs (four replicates of transplanted plugs that had received each of the six chemical treatments). Two terraria were set up for each moisture regime.

Tape was used to seal the seams of each terrarium. Straws were placed in two, 0.5-cm diameter holes in each top to partially ventilate the terrarium and minimize moisture losses and condensation. The terraria were placed in an air-conditioned greenhouse in which temperature was maintained at 21°C in the day and 18°C at night. Thermocouples were used to record soil and air temperatures inside the terraria twice a day, at 9 a.m and at 7 p.m. Mean air temperatures were 24, 22, and 21°C in the morning and 21, 20, and 22°C in the evening for

-0.5, -0.3, and -0.03 MPa soil water potentials, respectively. Mean soil temperatures were 18, 20, and 22°C in the morning and 20, 23, and 22°C in the evening for -0.5, -0.3, and -0.03 MPa soil water potentials, respectively. Air and soil temperatures of the terraria dropped into cooler ranges at night. Irradiance inside the terraria ranged from 455 to 485  $\mu\text{E}/\text{m}^2/\text{s}$  during early to late morning, and 512 to 475  $\mu\text{E}/\text{m}^2/\text{s}$  during early to late afternoon.

The tops of the terraria were removed weekly to check for diseases or wilting. The seedlings received no additional PGR, fertilizer, or water treatments for the 6-week duration of this experiment. Three weeks after transplanting, the seedlings were clipped at 5 cm. Plant heights were measured twice, 3 weeks after transplanting (before being clipping to 5 cm) and again at 6 weeks. Plant height, leaf water status, percent leaf protein, root length, shoot and root biomass, and mineral composition of leaf tissues were determined at the end of 6 weeks.

Leaf water status (LWS) was determined by removing the last fully emerged leaf from a tiller and placing it in a Campbell-Brewster leaf press chamber. The chamber was activated by a hydraulic pump that was capable of compressing the leaf as described by Jones (1980). The pressure that caused water to be exudated uniformly from tip, bottom, and edges of leaf was recorded. The greater the pressure required

to exude water, the lower the leaf water status. The mean of three measurements from each treated plug were taken for leaf water status.

Six weeks after transplanting, the plants were removed, the soil was washed from the roots, and root length was recorded. Tillers were separated from roots, and biomass was determined by weighing after drying the plant tissues in an oven at 60°C for 24 h.

Mineral composition of leaf tissues was determined following perchloric acid digestion according to the methods of Lim and Jackson (1958) and Jones and Steyn (1973). Phosphorus, K, Ca, Mg, and Fe were analyzed by an inductively coupled plasma spectrograph. Total leaf protein was determined by using a Kjeltac Auto 1030 analyzer according to the methods described by MacKenzie and Wallace (1954). The means of two measurements were calculated for the total leaf protein in each treated plant.

Treatments were analyzed as split plot design. The main plots were soil moisture levels and subplots were chemical treatments. All treatments were replicated four times. For mineral and protein analysis, additional plant tissue was needed. Samples were pooled from two duplicate terraria of four replicates each to provide sufficient tissues. Analyses of Variance (ANOVA) were used to analyze the data, and means were separated by Duncan's multiple range test at 10%

probability.

## RESULTS

Some treatments improved leaf elongation under each soil moisture condition tested (Tables 2.1 and 2.2). Greater plant heights developed when plants were grown under the higher (-0.03 MPa) soil water potential as compared to low (-0.5 MPa) and moderate (-0.3 MPa) soil moisture 3 and 6 weeks after transplanting. Plant growth regulators and soil moisture levels interacted for plant height in the third and sixth week after transplanting.

Three weeks after transplanting, only plants treated with Fe had increased leaf elongation under -0.5 MPa soil moisture when compared to the control (Table 2.1). Propiconazole and PPC plus chelated Fe-treated plants had less leaf elongation than the control plants at the moderate soil moisture. Fortified seaweed extract reduced foliage elongation than relative to control at the -0.03 MPa soil water potential 3 weeks after transplanting. Chelated Fe alone produced higher leaf growth than other chemical treatments or the control plants at the highest soil moisture. Addition of PPC or FSE eliminated the effect of chelated Fe. Propiconazole alone also promoted growth relative to the control plants.

At the end of the sixth week after transplanting, Fe

**TABLE 2.1.** Plant height of Kentucky bluegrass 5 weeks after chemical treatment and 3 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and soil moisture interacted at the  $p=0.1$  level.

Treatment	Amount		Soil Moisture Level (MPa)		
	PGR	Fe	-0.5	-0.3	-0.03
	L/ha	kg/ha	-----cm-----		
PPC	0.93	0.0	6.5ab*	5.0b	11.7b
FSE	9.3	0.0	6.7ab	6.7a	7.7e
Fe	0.0	0.11	7.7a	5.7ab	12.5a
PPC+Fe	0.93	0.11	5.2bc	4.5b	8.7d
FSE+Fe	9.3	0.11	5.7b	6.2a	9.7c
Control	0.0	0.0	6.0b	6.2a	9.2cd
Mean			6.3y*	5.7y	10.0x

\*Means within a column (a,b,c,d) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

alone was again the only chemical treatment that stimulated plant height under the lowest soil moisture (Table 2.2). Addition of either PGR to chelated Fe treatments reduced leaf elongation to control levels. All chemical treatments, except FSE plus Fe, promoted leaf elongation at the moderate soil moisture. Under  $-0.03$  MPa soil moisture, PPC and chelated Fe enhanced foliage development when compared to all other treatments including the control. Propiconazole plus chelated Fe at the high soil moisture was the only chemical treatment that did not enhance leaf elongation as compared to the control plants.

Transplants grown under  $-0.5$  and  $-0.3$  MPa soil moisture regimes had increased root length as compared to the plants grown under  $-0.03$  MPa soil water level (Table 2.3). Plant growth regulators and soil moisture levels interacted for root length. All chemical treatments caused greater root length as compared to the control plants when grown under the lowest soil moisture level. Similar results were obtained at  $-0.3$  MPa soil water potential, except PPC plus Fe-treated plants were not different from the control. Root lengths of chemically treated plants were not different than the control when grown under the highest soil moisture.

Kentucky bluegrass transplants had higher shoot dry weights under  $-0.03$  MPa than at the two other soil moisture levels (Table 2.4). Plant growth regulators and soil moisture

**TABLE 2.2.** Plant height of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Plants had been clipped to 5 cm at 3 weeks after transplanting Chemical treatment and soil moisture interacted at the p=0.1 level.

Treatment	Amount		Soil Moisture Level (MPa)		
	PGR	Fe	-0.5	-0.3	-0.03
	L/ha	kg/ha	-----cm-----		
PPC	0.93	0.0	15.2b*	19.1a	25.3a
FSE	9.3	0.0	15.0b	19.2a	23.0b
Fe	0.0	0.11	18.8a	19.4a	26.2a
PPC+Fe	0.93	0.11	14.9b	18.2a	19.4c
FSE+Fe	9.3	0.11	12.1c	14.2c	23.0b
Control	0.0	0.0	13.4bc	16.6b	20.7c
Mean			15.0z*	17.8y	23.0x

\*Means within a column (a,b,c) or row (x,y,z) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 2.3.** Root length of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and soil moisture interacted at the  $p=0.1$  level.

Material	Amount		Soil Moisture Level (MPa)		
	PGR	Fe	-0.5	-0.3	-0.03
	L/ha	kg/ha	-----cm-----		
PPC	0.93	0.0	14.5b*	15.5b	12.7a
FSE	9.3	0.0	16.2ab	13.7c	12.2a
Fe	0.0	0.11	15.5b	15.0b	13.5a
PPC+Fe	0.93	0.11	15.2b	12.5d	10.7a
FSE+Fe	9.3	0.11	17.2a	17.7a	13.0a
Control	0.0	0.00	11.7c	11.0d	10.2a
Mean			15.0x*	14.0x	12.0y

\*Means within a column (a,b,c,d) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according Duncan multiple range test.

**TABLE 2.4.** Shoot Dry matter of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and soil moisture interacted at the  $p=0.1$  level.

Treatment	Amount		Soil Moisture Level (MPa)		
	PGR	Fe	-0.5	-0.3	-0.03
	L/ha	kg/ha	-----mg dry wt/cm <sup>2</sup> -----		
PPC	0.93	0.0	19.2b*	24.0b	36.8a
FSE	9.3	0.0	20.8b	22.4b	30.4b
Fe	0.0	0.11	29.6a	32.8a	30.4b
PPC+Fe	0.93	0.11	28.8a	12.8c	23.2bc
FSE+Fe	9.3	0.11	19.2b	23.2b	29.6b
Control	0.0	0.0	18.4b	21.6b	24.0c
Mean			22.4y*	22.8y	28.8x

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

regimes interacted on shoot dry weight. Plants treated with chelated Fe and PPC plus chelated Fe under  $-0.5$  MPa soil moisture regime had higher shoot biomass production as compared to the other chemical treatments and control plants. Addition of the FSE to chelated Fe decreased the effect of the FSE. Iron was the only chemical treatment that enhanced shoot dry weight at the moderate soil moisture, and PPC plus Fe had reduced dry matter when compared to the control plants. In the highest moisture regime, all chemical treatments promoted shoot weight, except PPC plus chelated Fe.

Root dry weight was greater under the lowest soil moisture regimes than under the other two moisture levels (Table 2.5). All chemical treatments caused an increase in root development when compared to the control plants at all three moisture levels. Iron and FSE plus chelated Fe were the most active materials in enhancing root growth. Propiconazole again seemed to reduce the chelated Fe effect.

As soil water potential decreased, leaf water status (LWS) of treated plants changed (Table 2.6). Greater LWS values indicate greater likelihood of leaf stress. The highest LWS was developed under plants grown at the lowest moisture level. Plants were able to maintain the best LWS under the  $-0.03$  MPa soil water potential, and LWS was better in the  $-0.3$  MPa regime than at  $-0.5$  MPa. All chemical treatments improved LWS plant under all soil moisture regimes. The PPC plus

**TABLE 2.5.** Root dry weight of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			
	PGR	Fe	-0.5	-0.3	-0.03	Mean
	L/ha	kg/ha	-----mg dry wt/cm <sup>2</sup> -----			
PPC	0.93	0.0	16.8	20.8	16.0	17.6b*
FSE	9.3	0.0	18.4	15.2	16.8	16.8b
Fe	0.0	0.11	26.4	19.2	20.0	21.6a
PPC+Fe	0.93	0.11	21.6	16.8	14.4	17.6b
FSE+Fe	9.3	0.11	20.0	22.4	18.4	20.0a
Control	0.0	0.0	9.6	13.6	13.6	12.0c
Mean			18.4y*	17.6x	16.8x	

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 2.6.** Leaf water status of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			Mean
	PGR	Fe	-0.5	-0.3	-0.03	
	L/ha	kg/ha	-----kPa-----			
PPC	0.93	0.0	195	210	200	201c*
FSE	9.3	0.0	220	200	165	195c
Fe	0.0	0.11	220	200	180	200c
PPC+Fe	0.93	0.11	230	210	195	212b
FSE+Fe	9.3	0.11	220	200	180	200c
Control	0.0	0.0	265	245	230	247a
Mean			225z*	211x	192y	

\*Means within a column (a,b,c) or row (x,y,z) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

chelated Fe treatment was less effective overall than the other four chemical applications.

When averaged across all chemical treatments, total leaf protein content of plants was greater at the highest soil moisture level (Table 2.7). At all three soil moisture regimes, foliar applications of PGR either alone or in combination with chelated Fe had no effect on leaf protein content.

Bluegrass seedlings treated with PGR and/or chelated Fe had improved uptake of several minerals (Tables 2.8 to 2.12). In several cases, P, K, and Fe of chemically treated plants were higher than in seedlings that received no PGR plus chelated Fe treatments 6 weeks after transplanting under three different moisture regimes.

The P concentrations in seedlings were lower at  $-0.3$  MPa as compared to the two other soil water potentials (Table 2.8). Plants that received PPC had higher P content in leaf tissues than the control plants across all three soil moisture levels 6 weeks after transplanting. All other chemically treated plants had higher P, but not significantly, than the control plants.

Propiconazole alone and chelated Fe alone were the only treatments that enhanced leaf K (Table 2.9), but the PPC plus chelated Fe treatment was not promotive. Other chemical treatments tended to increase K uptake, but K was not

**TABLE 2.7.** Leaf protein of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			
	PGR	Fe	-0.5	-0.3	-0.03	Mean
	L/ha	kg/ha	-----%			
PPC	0.93	0.0	22	24	22	23a*
FSE	9.3	0.0	22	22	24	23a
Fe	0.0	0.11	21	22	25	23a
PPC+Fe	0.93	0.11	21	25	24	23a
FSE+Fe	9.3	0.11	21	22	28	24a
Control	0.0	0.0	20	22	21	21a
Mean			21y*	23y	24x	

\*Means within a column (a) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 2.8.** Phosphorus concentration of leaf tissue of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			Mean
	PGR	Fe	-0.5	-0.3	-0.03	
	L/ha	kg/ha	-----%-----			
PPC	0.93	0.0	0.31	0.27	0.30	0.30a*
FSE	9.3	0.0	0.29	0.25	0.30	0.28ab
Fe	0.0	0.11	0.30	0.26	0.27	0.28ab
PPC+Fe	0.93	0.11	0.25	0.25	0.31	0.27ab
FSE+Fe	9.3	0.11	0.27	0.27	0.28	0.27ab
Control	0.0	0.0	0.25	0.26	0.24	0.25b
Mean			0.28x*	0.26y	0.28x	

\*Means within a column (a,b) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 2.9.** Potassium concentration of leaf tissue of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			
	PGR	Fe	-0.5	-0.3	-0.03	Mean
	L/ha	kg/ha	-----%-----			
PPC	0.93	0.0	2.7	3.1	3.1	2.9a*
FSE	9.3	0.0	2.9	2.9	2.7	2.8ab
Fe	0.0	0.11	2.9	2.9	2.9	2.9a
PPC+Fe	0.93	0.11	2.6	2.8	2.6	2.6ab
FSE+Fe	9.3	0.11	2.7	2.9	2.6	2.7ab
Control	0.0	0.0	2.4	2.7	2.5	2.5b
Mean			2.7y*	2.9x	2.7y	

\*Means within a column (a,b) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

significantly greater than in control plants.

Foliar applications of PGR in conjunction with chelated Fe on bluegrass seedlings had no effect on leaf Mg content under three different soil moisture levels 6 weeks after transplanting into the terraria (Table 2.10). Propiconazole caused significantly higher Mg than the control, but PPC plus Fe was equivalent to control.

Plant growth regulators with or without chelated Fe had no positive effect on leaf Ca uptake regardless of soil water potential 6 weeks after transplanting into terraria (Table 2.11). The FSE treatment caused lower Ca than the control.

Plant foliage showed differences in Fe content in response to applications of PGR with or without chelated Fe under all three soil moisture regimes (Table 2.12). There was a significant interaction between treatments and soil moisture levels. Plants grown under -0.3 MPa soil moisture regime had the lower Fe concentrations in their leaves when compared to the two other soil moisture regimes. Under the -0.5 MPa soil water potential, addition of chelated Fe either alone or with PGR resulted in approximately two- to three-fold increases in the Fe concentration in the leaves as compared to the control. Those plants that received PPC or FSE without chelated Fe under the -0.5 MPa soil moisture regime were also higher in Fe than the control plants. Under -0.3 MPa soil moisture level, leaf tissues had greater Fe contents after treatment only with

**TABLE 2.10.** Magnesium concentration of leaf tissue of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			
	PGR	Fe	-0.5	-0.3	-0.03	Mean
	L/ha	kg/ha	-----%-----			
PPC	0.93	0.0	0.27	0.28	0.27	0.27a*
FSE	9.3	0.0	0.24	0.25	0.26	0.25b
Fe	0.0	0.11	0.27	0.25	0.25	0.26ab
PPC+Fe	0.93	0.11	0.27	0.26	0.23	0.25b
FSE+Fe	9.3	0.11	0.25	0.26	0.26	0.26ab
Control	0.0	0.0	0.24	0.26	0.25	0.25b
Mean			0.26x*	0.26x	0.25x	

\*Means within a column (a,b) or row (x) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 2.11.** Calcium concentration of leaf tissue of Kentucky bluegrass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Soil Moisture Level (MPa)			
	PGR	Fe	-0.5	-0.3	-0.03	Mean
	L/ha	kg/ha	-----%-----			
PPC	0.93	0.0	0.54	0.53	0.57	0.54a*
FSE	9.3	0.0	0.47	0.47	0.54	0.49b
Fe	0.0	0.11	0.55	0.54	0.52	0.53a
PPC+Fe	0.93	0.11	0.53	0.55	0.57	0.54a
FSE+Fe	9.3	0.11	0.50	0.50	0.54	0.51ab
Control	0.0	0.0	0.50	0.53	0.54	0.52a
Mean			0.51x*	0.52x	0.54x	

\*Means within a column (a,b) or row (x) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 2.12.** Iron concentration of leaf tissue of Kentucky blue grass 8 weeks after chemical treatment and 6 weeks after transplanting into various soil moistures as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and soil moisture interacted at the p=0.1 level.

Treatment	Amount		Soil Moisture Level (MPa)		
	PGR	Fe	-0.5	-0.3	-0.03
	L/ha	kg/ha	-----%-----		
PPC	0.93	0.0	0.078c*	0.041d	0.056c
FSE	9.3	0.0	0.078c	0.064cd	0.062c
Fe	0.0	0.11	0.105ab	0.094b	0.170b
PPC+Fe	0.93	0.11	0.120a	0.150a	0.200a
FSE+Fe	9.3	0.11	0.120a	0.074bc	0.064c
Control	0.0	0.0	0.045d	0.064cd	0.065c
Mean			0.092y*	0.072x	0.100y

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

PPC plus chelated Fe or chelated Fe alone. Addition of PPC increased the effect over chelated Fe alone. Similar results were obtained from PPC plus chelated Fe and chelated Fe alone when plants were grown under the  $-0.03$  MPa soil water potential; addition of PPC increased the effect of chelated Fe.

## DISCUSSION

Exogenous applications of FSE and PPC either alone or in combination with chelated Fe enhanced several aspects of transplanted seedling Kentucky bluegrass growth under different soil moisture regimes. Chelated Fe alone was also very beneficial. These substances caused different growth responses under different soil moisture conditions, as has also been observed by others (Schmidt and Snyder 1984; Walster et al., 1987); but growth of treated plants was regularly increased over untreated plants.

Moisture treatments affected on plant growth as would be expected. Plant height and shoot biomass were greater at the higher than under the other two soil moisture treatments. Conversely, root length was significantly greater in low and moderate soil moisture regimes when compared to plants grown under the higher soil moisture. Root dry matter was also greater under the more limited soil moisture environments than

under well-watered conditions. This confirms earlier research reports that plants grown under low to moderate soil moisture develop more extensive root systems than when well-watered (Mayaki et al., 1976; Shaw, 1978).

Growth-promoting activity of FSE, PPC, and chelated Fe increased root elongation and dry weight at the lower soil moisture. These findings support previous research that cytokinin-like materials are capable of promoting root and development (Philipson and Coutts, 1980; Nelson and Van Standen 1984; Goatley and Schmidt 1990). Plants with larger and deeper roots are naturally capable of absorbing water that is stored in the deeper soil zones during periods of moisture stress.

Water stress during vegetative growth inhibits activity of stomata, carbohydrate partitioning, and relative water content of leaf tissues (Frank et al., 1973; Steinberg et al., 1990). Other workers have suggested that limited soil moisture tends to reduce lateral bud initiation, rhizome production, and growth rate of other organs, resulting in a smaller plant size (Beard, 1973; Krieg and Sung, 1979; Arkin and Rosenthal, 1983; Nus and Hodges, 1986). Bengston et al. (1979) concluded that leaf water deficits result in the production of abscisic acid in the roots and shoots and consequently the suppression of cytokinin synthesis. The present study provides evidence that FSE and PPC either alone or in conjunction with chelated

Fe stimulated leaf activity, perhaps via cytokinins.

The application of these chemical substances substantially improved leaf water status and plant biomass production, particularly at the low and moderate moisture regimes. Kane and Smiley (1983) reported that foliar applications of systemic triazole compounds to Kentucky bluegrass suppressed root and shoot development under a good soil moisture regime. However, Goatley and Schmidt (1990) suggested the possibility of some type of interaction between cytokinin-like materials and the addition of chelated Fe on seedlings of Kentucky bluegrass grown under good soil moisture conditions.

This investigation indicated that PPC is capable of enhancing plant growth. The addition of the chelated Fe at times increased the effects of PPC resulting in greater growth responses. In general, PPC either alone or in combination with chelated Fe promoted seedling growth as compared to the control under different soil moisture levels, in particular at the limited soil water regime. Similar results were obtained from plants that received FSE with or without chelated Fe at the different soil moisture regimes, but some antagonisms may have occurred also.

Shanahan and Nielsen (1987) showed that application of two anti-gibberellin plant growth regulators to corn plants increased the plant population and improved grains per kernel

in dry soil conditions. Plant roots are not thought to synthesize cytokinin during severe water deficits (Nelson and Van Standen, 1984). Foliar applications of FSE and PPC may result in cytokinin-like properties and inhibit abscisic acid, which is a primary factor in slowed growth during water stress (Bengston et al., 1979). These PGR have been reported to accelerate transport of endogenous cytokinins along with certain amino acids to areas where these chemicals will be utilized (Nelson and Van Standen, 1984). The increased cytokinin concentrations in leaf tissues retarded transpiration rate, and prevented rapid wilting of plants because of low water potential in soils (Bengston et al., 1979).

The enhancement of root and shoot dry matter production had a great effect on some of the minerals and on total protein in leaf tissues. Seedlings treated with PGR and chelated Fe under different soil moisture levels had increases in several nutrients especially Fe, P, and K. Increases in nutrient concentrations, primarily K, possibly promoted stomatal activity and osmotic water potential of plant leaves in order to generate solute movement and  $K^+/H^+$  exchanges across the guard cells (Ehret and Boyer, 1979). Similar observations regarding exogenous applications of several PGR to moisture-stressed plants were noted by Cothren et al. (1983), Blackman and Davies (1985), and Davies et al. (1986). Foliar

application of PGR in conjunction with chelated Fe also induced increases in Ca, Mg, and Fe, which play significant roles in enzyme activation and photosynthesis (Cothren et al., 1983; Mouate and Ness, 1986).

Investigation in this experiment indicated that these ions also concentrated in leaf tissues of treated plants when grown under different soil moisture regimes. Leaf tissue analyses indicated mineral nutrients, particularly K, and P in the seedlings treated with PGR either alone or in combination with chelated Fe may be responsible for stimulating the activity of leaf tissues, which might allow plants to maintain a better LWS during periods when soil water is limited. Cothren et al. (1983), Duniway (1971), and Mouate and Ness (1986) reported a stimulation of Ca, Mg, and P in the leaf tissues when treated with PGR was important for energy reserves, leaf permeability, enzyme biosynthesis, and possibly plant quality and dry weight under limited soil moisture regimes.

Kemble and MacPherson (1954) and Ehret and Boyer (1979) reported that water content and protein biosynthesis were drastically reduced as soil water potential decreased. Findings in this experiment indicate the application of PGR with or without chelated Fe can improve leaf water status and the total of leaf proteins under different soil moisture levels as compared to control plants. Similar results were

reported by Rao (1973) that suggested that protein quality and biomass production of crops can be improved when additional mineral nutrients are added into growth media under different soil irrigation systems.

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### CHAPTER 3

#### EFFECT OF PLANT GROWTH REGULATORS, CHELATED Fe, AND NITROGEN FERTILITY ON CREEPING BENTGRASS SUBJECTED TO DROUGHT STRESS

##### ABSTRACT

Creeping bentgrass (Agrostis palustris Huds.) that was established in 1985 on a Groseclose silt loam received 25 and 50 kg/ha of urea on 15 May 1989 for low and high N regimes respectively. Ten days after fertilization, plots were treated with two plant growth regulators (PGR) and chelated Fe. One PGR was Roots, a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine (FSE). The other PGR was the systemic fungicide propiconazole (trade name: Banner, chemical name: [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl)methyl-1H-1,2,4-triazole)] (PPC). The FSE was applied at 9.3 L/ha and PPC was applied at 0.93 L active ingredient (a.i)/ha. Each PRG was applied alone or in conjunction with Na-diethylene-triamine-pentaactate chelated Fe at 0.11 kg a.i/ha.

Two weeks after spray treatments a plug was taken from each field plot and transplanted into a drained container. Two weeks after transplanting, all of the containers were placed

under a rain shelter for 3 weeks and water was withheld, during which several parameters were monitored. After 3 weeks of withholding water, plants were irrigated to field capacity three times per week. Additional plant responses were measured over the next 3 weeks.

Foliar applications of FSE or PPC either alone or in combination with chelated Fe to creeping bentgrass increased tolerance to drought under different N fertility levels.

Cumulative evapotranspiration (CET) of treated plants was greater under the higher N fertilization. The increased CET caused bentgrass to be wilted after 3 weeks of withholding water. Plant growth regulator treatments with or without chelated Fe, except for PPC, resulted in significantly less wilting than in the control plants with either low or high N fertility. Plant growth regulators plus Fe treatments resulted in significantly less stress to leaves than in untreated control plants. Leaves were more drought stressed under high than low N fertility.

Foliage recovery following a drought was significantly greater under the lower than the higher N fertility 3 weeks after rewatering. All PGR with or without chelated Fe significantly improved root growth with either low or high N fertility levels 3 weeks after irrigation resumed. Chelated Fe was equally or more beneficial than FSE or PPC, but Fe and PGR did not cause synergistic or additive effects.

**INTRODUCTION**

Drought is the occurrence of a substantial water deficit in the soil and consequently in plant tissues. If drought persists, significant injuries to plants result (Taylor et al., 1983). Willey (1970) and Cocucci and Trecani (1976) documented that the most visible plant response to drought is a rapid wilt. Water deficits accompanying rapid wilting in the above ground parts of plants cause a change in the protoplasm of leaf tissues and impair permeability of the cell membrane to water (Levitt et al., 1936).

Low soil water potential can limit the allocation of carbon resources to the root system and reduced intermediary metabolism, water absorption, and mineral uptake (Willey, 1970; Malik et al., 1979). Mitchell and Russell (1971) proposed that, in order to avoid yield losses associated with drought, one should select plant cultivars that have deep, large root systems and low transpiration rates.

Growth, quality, and yield sometimes have been improved under stressful environments by supplying mineral nutrients and plant growth regulators (PGR) (Clarkson and Hanson 1980; Kuiper and Staal, 1987). Cycocel (CCC) is a PGR that increases plant resistance to drought (Halevy and Kessler, 1963). Fletcher (1985) and Nelson and Van Standen (1984) documented that compounds with cytokinin-like activity not only increase

crop yields but also protected plants against various environmental stresses. A report by Davies et al. (1988) indicated that plants receiving cytokinin-like materials appeared to survive longer than untreated plants during drought stress.

Exogenous applications of cytokinin-like PGR to plants grown in N-deficient soils have induced positive plant growth responses (Kuiper and Staal, 1987; Mardanov, 1985). Emerson et al. (1986) noted that winter wheat treated with an anti-gibberellin PGR produced higher yields under a low rate of N as compared to a heavy rate of N. Cytokinins added to a growth medium without additional N enhanced birch seedling growth (Horgan and Wareing, 1980). Foliar application of chelated Fe to Kentucky bluegrass grown at a low level of mineral nutrition significantly improved plant growth and development (Deal and Engle, 1965).

Kentucky bluegrass grown in nutrient culture with low N levels was more heat tolerant than plants under high N levels (Pellet and Roberts, 1963). Banpna and Khuspe (1980) and Agarwal and Sinha (1983) reported that plant water use and dry matter accumulation are highly dependent upon the presence of N in the soil and plant tissue. Information is limited concerning the interaction of PGR and N fertilization for crops growing under arid conditions. Applications of heavy rates of N to plants grown in dry soil have a negative impact

on plant water use efficiency and dry matter distribution (Kuiper and Staal, 1987; Heitholt, 1990). Reduction of root growth, increase of certain diseases, and high water use have been associated with excessive use of N fertilizer (Yust et al., 1984; Heitholt, 1990).

The objectives of this study were : a) to evaluate growth responses of creeping bentgrass under drought stress when treated with PGR, Fe, and two N fertility regimes, and b) to evaluate the effects of the above treatments on recovery from drought stress of creeping bentgrass.

#### **MATERIALS AND METHODS**

Creeping bentgrass (Agrostis palustris Huds.) was established in 1985 on a Groseclose silt loam (clayey, kaolinitic, mesic Typic Hapludult). Soil P and K at the time of this experiment were 21 and 37  $\mu\text{g/g}$ , respectively. Soil pH was 6.2. On 15 May 1989, urea was applied at 25 and 50 kg N/ha for low and high N regimes, respectively. This experiment also received 22 kg N/ha in fall 1988.

Ten days after N fertilization, plots were treated with two PGR materials and chelated Fe. One PGR was Roots (FSE), a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine marketed by ROOTS INC. The systemic fungicide propiconazole (trade name: Banner,

chemical name: [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2yl)methyl-1 H-1,2,4-triazole] produced by Ciba Geigy, was also tested for its PGR-like properties. Propiconazole and FSE were applied individually and in conjunction with chelated Fe (Ciba Geigy 330Fe). A compressed-air sprayer that delivered 784 l/ha of solution at a pressure of 290 kPa was used for foliar application. All treatments were applied to 0.9 m by 1.8 m plots arranged in a randomized complete block design and replicated six times.

Two weeks after spray treatments plots were clipped to 5 cm, and a 14-cm diam by 14-cm deep plug was taken from each field plot and transplanted into a drained metal container that was 15-cm in diam and 15-cm high. Approximate field capacity of each plug was established by thoroughly watering the container, allowing it to drain for 24 h, and then weighing.

Another 72 metal containers of the size described above were filled with 4.4 kg of Groseclose silt loam with pH of 6.2. Root rings were made from 10-cm diam PVC pipe cut to a 5-cm depth. Two steel wires 3 mm in diam were inserted at right angles through holes drilled 6.2 mm from the angles of the rings. Hardware cloth with 6-mm mesh, cut to fit the inside of the ring, was placed into the ring and rested on the steel wires (Schmidt et al., 1986). The root rings were placed on the soil surface in the metal containers. A 10-cm diam by 10-

cm deep plug was taken from each field plot and transplanted into the root rings, allowing the plant roots to grow into the soil through the hardware cloth. Two weeks after transplanting plugs into root rings, approximate field capacity of the soils were determined as described above.

Two weeks after transplanting, all of the metal containers were placed under a rain shelter for 3 weeks and water was withheld, during which several parameters were monitored (see below). After 3 weeks of withholding water, plants were removed from the rain shelter and irrigated to field capacity three times per week. Additional plant responses were measured over the next 3 weeks.

#### **Measurements during drought**

Cumulative evapotranspiration (CET) was determined after 1 and 3 weeks of withholding water by weighing the metal containers and subtracting those values from weights of the containers when at the original approximate field capacity. Percent wilting was determined by placing a 4-cm diam ring on the plugs, counting all leaves, and counting wilted leaves, using indicators such as bluish color and leaf rolling. The means of two ratings per plug were recorded for percent wilting.

Leaf water status (LWS) was determined by removing the

last fully emerged leaf from a tiller and placing it in a Campbell-Brewster leaf press chamber. The chamber was activated by a hydraulic pump that was capable of compressing the leaf as described by Jones and Carably (1980). The pressure that caused water to be exuded uniformly from tips, bottom, and edges of the leaves was recorded. The greater the pressure required to exude water, the lower the LWS. Means of three LWS measurements from each treated plug were recorded. Shoot dry weight accumulation was approximated by harvesting creeping bentgrass at heights of 5 cm (after withholding water for 3 weeks) and oven drying at 60°C for 24 h.

#### **Post-drought measurements**

Plant recovery was evaluated by using a 4-cm diam ring placed over the transplanted plugs, and the number of leaves that were green were counted to determine green leaf density. The means of two measurements per plug were recorded for plant recovery 2 and 3 weeks after rewatering. Color of plants was rated based upon a visual score of 9 for dark green color, and 6 for acceptable color, and 1 for no green color. Leaf water status was determined as described previously.

Root development was determined by the vertical root lift method (Schmidt et al., 1984). A hand-held pulling gauge scale (Model DPPH-100, John Chatillion and Sons, Inc., 83-30 Kew

Gardens Road, Kew Gardens, NY) with two metal hooks extending from the base was attached to the rooting rings and vertically lifted. The amount of force required to lift the roots free from the soils correlated to the amount of root mass. Post-drought shoot dry weight accumulation was determined 3 weeks after rewatering by harvesting foliage at a height of 5 cm and oven drying at 60°C for 24 h.

A randomized complete block design with two N fertility regimes and six chemical treatment were used. Treatments were replicated six times. An analysis of variance (ANOVA) was used to analyze the data, and means of treatment effects were compared according to Duncan's multiple range test at 10% probability.

## **RESULTS**

Foliar applications of PGR and/or chelated Fe to bentgrass grown under different N fertility regimes caused substantial differences in water use during and following a simulated drought. Cumulative evapotranspiration was affected by the amount of N applied. Plants at the lower fertility level and treated with PGR maintained a higher leaf water status during the drought. Plants transpired relatively large amounts of water during the first week; but as the drought was prolonged, the plants lost less water regardless of N

fertility. Cumulative evapotranspiration was generally 10 to 15% greater with the higher rate of N (Tables 3.1, and 3.2).

None of the foliar treatments affected CET during the first week water was withheld (Table 3.1). The PPC treatments tended to reduce CET of bentgrass plants, but not significantly. Nitrogen level did affect CET. Plants grown under the lower N rate had significantly less CET than plants grown under the higher N.

Chemical treatments and N fertility interacted for CET after 3 weeks without watering (Table 3.2). No differences between any chemical treatment and the control were observed under the lower N regime. At the higher N fertility, FSE was active in lowering CET, while PPC+Fe-treated plants had significantly higher CET. Cumulative evapotranspiration continued to be greater at the higher N rate.

Wilting was evident in some plants 2 weeks after water was withheld (Table 3.3). Foliar PGR applications with or without chelated Fe, except for PPC alone, reduced wilting when compared to the control after 2 weeks of induced drought. This was true at both N fertility levels. Propiconazole plus Fe was the most active treatment in protecting plants against wilt regardless of the N applications. However, PPC alone caused more wilting than in the control. Plant growth regulators and N fertility regimes interacted for plant wilting, but there was no difference between the two N levels

**TABLE 3.1.** Cumulative evapotranspiration of creeping bentgrass 3 weeks after chemical treatment and after 1 week without watering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----kg/m <sup>2</sup> -----		
PPC	0.93	0.0	14.8	23.5	19.1a*
FSE	9.3	0.0	19.7	23.4	21.5a
Fe	0.0	0.11	19.7	21.5	20.6a
PPC+Fe	0.93	0.11	14.8	23.7	19.2a
FSE+Fe	9.3	0.11	17.3	21.7	19.5a
Control	0.0	0.0	18.5	24.0	20.3a
Mean			17.4y*	23.0x	

\*Means within a column (a) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 3.2.** Cumulative evapotranspiration of creeping bentgrass 5 weeks after chemical treatment and after 3 weeks without watering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes. Chemical treatments and nitrogen levels interacted at  $p=0.1$  level.

Treatment	Amount		N rates (kg/ha)	
	PGR	Fe	25	50
	L/ha	kg/ha	-----kg/m <sup>2</sup> -----	
PPC	0.93	0.0	22.0b*	33.0b
FSE	9.3	0.0	25.5ab	25.5c
Fe	0.0	0.11	28.0a	33.0b
PPC+Fe	0.93	0.11	26.3ab	38.3a
FSE+Fe	9.3	0.11	28.0a	29.8b
Control	0.0	0.0	27.1ab	33.0b
Mean			26.1y*	32.0x

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at the  $p=0.1$  according to Duncan multiple range test.

**TABLE 3.3.** Wilting of creeping bentgrass 4 weeks after chemical treatment and after 2 weeks without watering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes. Chemical treatment and nitrogen level interacted at  $p=0.1$  level.

Treatment	Amount		N rates (kg/ha)	
	PGR	Fe	25	50
	L/ha	kg/ha	-----% Wilted-----	
PPC	0.93	0.0	41a*	39a
FSE	9.3	0.0	22c	24cd
Fe	0.0	0.11	20d	26c
PPC+Fe	0.93	0.11	17e	21d
FSE+Fe	9.3	0.11	25c	27c
Control	0.0	0.0	30b	33b
Mean			26x	28x

\*Means within a column (a,b,c,d) or row (x) followed by the same letter are not significantly different at the  $p=0.1$  according to Duncan multiple range test.

when averaged across chemical treatments.

As drought continued, wilting increased at both N levels (Table 3.4). After 3 weeks of drought, all treated plants, except the PPC-treated plants, had a lower percentage of wilted leaves as compared to the control plants under both N regimes. Chelated Fe plus PPC was most effective at reducing wilting at both levels of N treatments. There was a difference in wilting between N application after 3 weeks without added water; the higher rate of N resulted in more wilt.

Plant growth regulator treatments, chelated Fe, and low N treatments improved LWS during drought stress. Foliar treatments and N fertility regimes interacted for LWS. There was a better LWS under 25 kg N/ha than under the higher N level (Table 3.5). The LWS was more favorable in plants treated with PPC, FSE, and chelated Fe after 1 week without watering. Addition of Fe to the PGR tended to reduce the effect of PPC or FSE, but these treatments still were superior to the control under both N levels.

All chemical treatments, except FSE and FSE+Fe, stimulated shoot growth above clipping height (5 cm) as compared to the control plants (Table 3.6). Propiconazole plus Fe was the most active material in enhancing growth above 5 cm after 3 weeks of induced drought. Nitrogen treatments were without effect on clipping mass, and there was no interaction of N and PGR treatments.

**TABLE 3.4.** Wilting of creeping bentgrass 5 weeks after chemical treatment and after 3 weeks without watering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes. Chemical treatments and nitrogen level interacted at p=0.1 level.

Treatment	Amount		N rates (kg/ha)	
	PGR	Fe	25	50
	L/ha	kg/ha	-----% wilting-----	
PPC	0.93	0.0	57a*	57b
FSE	9.3	0.0	44b	49c
Fe	0.0	0.11	39b	48c
PPC+Fe	0.93	0.11	27c	35d
FS+Fe	9.3	0.11	44b	53c
Control	0.0	0.0	56a	65a
Mean			44y*	51x

\*Means within a column (a,b,c,d) or row (x,y) followed by the same letter are not significantly different at the p=0.1 according to Duncan multiple range test.

**TABLE 3.5.** Leaf water status of creeping bentgrass 3 weeks after chemical treatment and after 1 week without watering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----kPa-----		
PPC	0.93	0.0	150	180	165d*
FSE	9.3	0.0	165	185	175cd
Fe	0.0	0.11	175	180	175cd
PPC+Fe	0.93	0.11	175	190	180bc
FSE+Fe	9.3	0.11	180	195	187b
Control	0.0	0.0	195	225	210a
Mean			175y*	194x	

\*Means within a column (a,b,c,d) or row (x,y) followed by the same letter are not significantly different at the p=0.1 according to Duncan multiple range test.

**TABLE 3.6.** Dry matter of creeping bentgrass clipping 5 weeks after chemical treatment and after 3 weeks without watering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----g/m <sup>2</sup> -----		
PPC	0.93	0.0	1.00	0.87	0.90b*
FSE	9.3	0.0	0.90	0.83	0.87bc
Fe	0.0	0.11	0.97	0.98	0.97ab
PPC+Fe	0.93	0.11	1.07	1.00	1.02a
FSE+Fe	9.3	0.11	0.87	0.90	0.91bc
Control	0.0	0.0	0.80	0.87	0.83c
Mean			0.90x	0.91x	

\*Means within a column (a,b,c,) or row (x) followed by the same letter are not significantly different at the p=0.1 according to Duncan multiple range test.

Green leaf density was significantly greater for plants grown under lower than higher N regimes after rewatering (Tables 3.7 and 3.8). Propiconazole plus Fe and chelated Fe alone were the only chemical treatments that increased bentgrass recovery 2 weeks after rewatering when compared to the control plants. Addition of chelated Fe increased the effect of PPC 2 weeks after rewatering. Plant growth regulator treatments with or without chelated Fe resulted in more green leaves under lower than higher levels of N 3 weeks after rewatering. Propiconazole plus Fe and chelated Fe alone caused an increase in regreening when compared to the control plants, while PPC treatments caused less regreening growth than in untreated plants.

Foliar applications, of PGR and/or chelated Fe produced a darker green leaf than in the control plants (Table 3.9). Leaf color was not affected by N levels. Plants that received PPC with or without chelated Fe had the darkest green color 3 weeks after watering resumed.

Plants fertilized with 25 kg N/ha had a better LWS than plants fertilized with 50 kg N/ha after 3 weeks of rewatering (Table 3.10). The PGR treatments and N levels interacted. All foliar treatments, except FSE+Fe, had a better LWS than the control plants under the lower N regime. The PPC- and FSE-treated plants at the lower N level and PPC+Fe and chelated Fe treated plants at the higher N level were among treatments

**TABLE 3.7.** Green-leaf density of creeping bentgrass 7 weeks after chemical treatment and 2 weeks after rewatering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----no./cm <sup>2</sup> -----		
PPC	0.93	0.0	4.12	3.17	3.65c
FSE	9.3	0.0	4.20	3.09	3.65c
Fe	0.0	0.11	4.36	4.04	4.20b
PPC+Fe	0.93	0.11	5.13	4.12	4.60a
FSE+Fe	9.3	0.11	4.20	3.73	3.96c
Control	0.0	0.0	3.96	3.73	3.80c
Mean			4.36x*	3.70y	

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at the p=0.1 according to Duncan multiple range test.

**TABLE 3.8.** Green-leaf density of creeping bentgrass 8 weeks after chemical treatment and 3 weeks after rewatering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----no./cm <sup>2</sup> -----		
PPC	0.93	0.0	5.55	4.68	5.07c*
FSE	9.3	0.0	6.26	5.63	6.03b
Fe	0.0	0.11	6.34	6.50	6.26a
PPC+Fe	0.93	0.11	6.82	5.87	6.34a
FSE+Fe	9.3	0.11	5.87	5.55	5.71b
Control	0.0	0.0	5.55	5.95	5.71b
Mean			6.1y*	5.9x	

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at the p=0.1 according to Duncan multiple range test.

**TABLE 3.9.** Leaf color of creeping bentgrass 5 weeks after chemical treatment and 3 weeks after rewatering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----Rating#-----		
PPC	0.93	0.0	8.7	8.7	8.7a*
FSE	9.3	0.0	8.2	8.7	8.4b
Fe	0.0	0.11	8.2	8.8	8.5b
PPC+Fe	0.93	0.11	8.6	8.6	8.6a
FSE+Fe	9.3	0.11	8.4	8.2	8.3b
Control	0.0	0.0	7.2	8.0	7.6c
Mean			8.2x	8.5x	

\*Means within a column (a,b,c) or row (x) followed by the same letter are not significantly different at the p=0.1 according to Duncan multiple range test.

#Plant color was ranked from 1 to 9, where 1 is no green color, 6 is acceptable, and 9 is the best.

**TABLE 3.10.** Leaf water status of creeping bentgrass 8 weeks after chemical treatment and 3 weeks after rewatering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes. Chemical treatment and nitrogen level interacted at  $p=0.1$  level.

Treatment	Amount		N rates (kg/ha)	
	PGR	Fe	25	50
	L/ha	kg/ha	-----kPa-----	
PPC	0.93	0.0	150d*	194bc
FSE	9.3	0.0	170c	197bc
Fe	0.0	0.11	175bc	190c
PPC+Fe	0.93	0.11	180b	186c
FSE+Fe	9.3	0.11	190a	202b
Control	0.0	0.0	195a	220a
Mean			177y*	198x

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at the  $p=0.1$  according to Duncan multiple range test.

with the most leaf moisture. Addition of chelated Fe reduced the effect of PPC and FSE at the lower N level and of FSE at the higher N.

After 3 weeks recovery from drought, root-pull strength of PGR-treated plants was significantly enhanced at both N levels (Table 3.11). Root-pull strength was greater at the higher than at the lower N level. All foliar treatments enabled plants to establish more root strength than the control plants under both fertility levels. Addition of chelated Fe reduced the effect of PPC at the lower N, while addition of chelated Fe increased the effect of PPC and reduced the effect of FSE at high N fertility.

Plant growth regulators with or without chelated Fe caused few significant increases in dry weight of bentgrass clippings compared to the control plants with either low or high N regimes 3 weeks after rewatering (Table 3.12). Propiconazole plus chelated Fe was the only chemical treatment that increased clipping dry weight when compared to the control plants.

## **DISCUSSION**

Many studies have shown that water deficits reduce growth rates of various plant organs and cause wilting (Taylor et al., 1983; Willey, 1970; Cocucci and Trencani, 1976; Wilson,

**TABLE 3.11.** Root-pull strength of creeping bentgrass 5 weeks after chemical treatment and 3 weeks after rewatering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes. Chemical treatment and nitrogen level interacted at  $p=0.1$  level.

Treatment	Amount		N rates (kg/ha)	
	PGR	Fe	25	50
	L/ha	kg/ha	-----kg/m <sup>2</sup> -----	
PPC	0.93	0.0	29a*	30b
FSE	9.3	0.0	29a	30b
Fe	0.0	0.11	24b	34a
PPC+Fe	0.93	0.11	27ab	34a
FSE+Fe	9.3	0.11	29a	27c
Control	0.0	0.0	20c	24d
Mean			26y*	28x

\*Means within a column (a,b,c,d) or row (x,y) followed by the same letter are not significantly different at the  $p=0.1$  according to Duncan multiple range test.

**TABLE 3.12.** Dry matter of creeping bentgrass clippings 8 weeks after chemical treatment and 3 weeks after rewatering as affected by propiconazole (PPC), fortified seaweed extract (FSE), Fe, and two nitrogen fertility regimes.

Treatment	Amount		N rates (kg/ha)		Mean
	PGR	Fe	25	50	
	L/ha	kg/ha	-----g/m <sup>2</sup> -----		
PPC	0.93	0.0	0.93	0.73	0.83ab*
FSE	9.3	0.0	0.90	0.80	0.86ab
Fe	0.0	0.11	0.86	1.00	0.93ab
PPC+Fe	0.93	0.11	1.13	1.06	1.10a
FSE+Fe	9.3	0.11	0.93	0.93	0.93ab
Control	0.0	0.0	0.67	0.86	0.77b
Mean			0.90x	0.90x	

\*Means within a column (a,b) or row (x) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

1983; Pellet and Roberts, 1963). Others have shown that applications of cytokinins to plants grown under low fertility levels can protect plants against various types of environmental stresses, in particular heat and drought (Fletcher, 1985; Nelson and Van Standen, 1984).

Each of the foliar treatments tested appeared to be more active in increasing drought resistance of creeping bentgrass under the lower N fertility. Dexter (1937) reported that high rates of N fertilization reduced drought tolerance of temperate grass species.

Results of this study indicated that some PGR plus Fe treatments lowered evapotranspiration and therefore reduced wilting of bentgrass under limited water availability. Root growth, but not foliar growth of bentgrass was enhanced with application of PGR and/or Fe under either N level during drought stress. Goatley and Schmidt (1990) noted that certain PGR compounds when combined with chelated Fe showed variable growth responses.

This experiment also showed that 50 kg N/ha stimulated CET and wilting and lowered LWS. Even with higher CET and moderate LWS, PGR and Fe treatments lowered wilted leaves relative to the control. This was associated with improvement of root strength, which presumably reflects root development. Improved root development would allow the plants to obtain deeper moisture when water was limited near the soil surface

during the drought period.

Application of N at 50 kg/ha increased clipping dry matter as compared to 25 kg N/ha. The higher N rate caused increased vegetative growth, and that resulted in higher CET. Loss of greater amounts of water from pots with limited water available caused wilting and lower growth recovery following a drought. Others have shown that increased top growth associated with application of N accelerates transpiration and rapid assimilation of soluble carbohydrates to leaves and is correlated with reduced root growth (Green and Beard, 1969; Snyder and Schmidt, 1974). This investigation showed that foliar applications of PGR and/or chelated Fe enhanced bentgrass root growth and root strength under drought with two different N treatments. Banpna and Khuspe (1980) proposed that soil water potential and the availability of N in the soil play a significant role in transpiration rate and crop production.

Higher CET, more wilted leaves, and lower LWS in the plants grown at the higher N was followed by slower growth recovery from drought. Yust et al. (1984), Heitholt (1990), Malik et al. (1979), and Willey (1970) also reported increased incidence of certain diseases, lowered water use efficiency, and yield reductions associated with excessive use of N fertilization.

Lower CET, higher LWS, and less wilted leaves due the low

N level accounted for better growth recovery from drought stress. Emerson et al. (1986) found similar responses for winter wheat under low irrigation and different rates of N fertility following foliar applications of cycocel, a PGR.

Applications of PGR and/or Fe in this study increased dry matter in clippings under both N fertility regimes following a drought. An increase in LWS also was associated with foliar application of PGR and/or chelated Fe with each rate of N application. However, LWS of plants that had been chemically treated followed by drought had a better apparent LWS under the lower than the higher rate of N fertility. Plants with lower LWS had greater growth under both N regimes during the subsequent drought stress. These findings support previous work by Walton (1980), Pierce and Raschke (1980), and Blomquist et al. (1973) who noted that foliar applications of PGR in particular cytokinins, might increase the activity of enzymes or endogenous hormones in leaves. An accumulation of these chemicals in plant tissues can suppress abscisic acid, which is the primary element for reduction of cell turgidity and solute movement in leaves during drought stress.

Growth responses of bentgrass plants to PGR and/or Fe after 3 weeks recovery from drought showed that the regrowth generally was less with the higher N rate. Apparently drought conditions produced several effects, especially at the higher N fertility, which caused a decrease in the regrowth capacity

of the bentgrass plants. Kramer (1950) noted that, when wilted plants received water, the leaves regained turgidity within a few hours; but the effects of wilting on internal processes were evident for much longer. Levitt et al. (1936) and Loustalot (1945) reported that leaf wilting produced changes in the protoplasm and permeability of cell membranes. The drought experience resulted in lower photosynthetic rates and growth inhibition for several days following rewatering.

This study also showed that bentgrass plants wilted to a greater extent and had lower LWS at higher N fertility and that was correlated with slowed recovery from drought. These findings also confirm that plant regrowth following drought is promoted by foliar application of PGR and/or Fe.

Chemical substances containing cytokinin-like materials affect carbohydrate partitioning in plant tissues that can potentially induce growth under stressed environments (Nelson and Van Standen 1984; Kuiper and Staal, 1987; Mardanov, 1985). These cytokinins may protect plants in soils with mineral deficiencies. Halevy and Kessler (1963), Mardanov (1985), Kuiper and Staal (1987), and Davies et al. (1988) documented that cytokinins can modify intermediary metabolism in plant organelles and induce positive growth responses under harsh environments.

Leaf water status of bentgrass plants significantly differed in relation to N applications. Plants grown under low

N fertility had a better LWS. This was correlated with less CET from plant treatments. Plant growth regulators plus Fe and chelated Fe treatments played a significant role in minimizing the drought effects on bentgrass. Foliar applications of PGR and/or chelated Fe improved LWS when water became limited to plants. This improvement may correspond to activation of endogenous hormones that protect cell membranes of leaf tissues during drought (Pierce and Raschke, 1980; Walton, 1980; Blomquist et al., 1973). Hubick et al. (1986) also noted that enhancement of cytokinins in plant tissues caused an increase in cell wall permeability that may result in reduction of abscisic acid biosynthesis, which is a primary cause of stomatal closure during periods of drought stress.

Color comparisons of creeping bentgrass treated with PGR and/or chelated Fe with either level of N levels indicated no effect after rewatering. Plants treated with PGR and chelated Fe developed more green tissues than the untreated control. The appearance of a dark green color is presumably an indication of chlorophyll development following the stressful conditions. Hsiao (1973) indicated that drought is a primary cause for increased leaf senescence and chlorophyll destruction. Our findings also supported other workers who believe that cytokinins may deactivate free radical groups, superoxide, and hydrogen peroxide, which accelerate leaf senescence and chlorophyll break down in plant species during

water deficiency (Upadhyaya et al., 1985). Leaf rolling, decrease in leaf area, and the reduction in cytokinin supply from the roots in arid environments induces stomatal closure resulting in yield reductions (Blackman and Davies, 1983, 1985).

Earlier works showed that cytokinins in conjunction with N, P, and Fe can potentially increase plant resistance to drought stress and stimulate biomass production (Kuiper and Staal, 1987; Clarkson and Hanson 1980; Horgan and Wareing, 1980; Deal and Engle, 1965). Results of the present investigation also showed application of PGR and/or Fe can reduce bentgrass drought stress. Application of these chemicals appeared to be more beneficial to plants when grown under lower rather than higher N fertility.

Kramer (1950) found that severe leaf wilt and loss of leaf turgidity resulted in injury and cessation of root growth. Itai and Vaadia (1971) reported that cytokinin concentrations in leaves of sunflower plants were reduced by 53%, which resulted in root growth cessation during 1 week of drought stress. In the present study, PGR-treated plants were able to develop a better root system during and after a drought.

Plants with large and deep root systems (Taylor et al., 1980) are able to reach water when soil moisture is limited during dry conditions. Exogenous application of cytokinins-

like materials to plants may result in stimulation of endogenous hormones, possibly cytokinins, which are known to induce root elongation and protect cell organelles when plants are subjected to limited water (Blackman and Davies, 1985).

Findings in this study suggest that further research is needed to evaluate responses of PGR plus Fe with different grass species in relation to N fertility regimes.

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## CHAPTER 4

### ALLEVIATION OF SALINITY STRESS ON KENTUCKY BLUEGRASS BY PLANT GROWTH REGULATORS AND CHELATED Fe

#### ABSTRACT

Four-month old, well-established Kentucky bluegrass (Poa praetensis cv. 'Plush') was selected for this investigation. Plots were treated on 18 August 1990 with various combinations of PGR and chelated Fe. One PGR was Roots, a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine (FSE). It was applied at 9.3 L/ha. The other PGR was the systemic fungicide propiconazole (trade name: Banner, chemical name: [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2yl)methyl-1 H-1,2,4-triazole] (PPC). It was applied at 0.93 L active ingredient (a.i)/ha. Each PRG was applied alone or in conjunction with Na-diethylene-triamine-pentaactate chelated Fe at 0.11 kg a.i/ha. Two weeks after spray treatments a 10-cm diam plug was taken from each of the treated plots and transplanted into a drained container. The transplanted plugs were irrigated for 1 week with potable water, and plants were then clipped at 5 cm height. Containers were placed under a rain shelter, and 220 mL of water containing 0, 0.15, and 0.3 S m<sup>-1</sup> salt concentrations were supplied to separate containers three times a week for 7 weeks.

Leaf water status (LWS), shoot dry weight, and root mass decreased as salinity increased. All chemical treatments, except chelated Fe, enhanced growth under low salinity water. Under high salinity, PPC enhanced foliage weight, and all treatments, except Fe, stimulated rooting. Propiconazole-treated plants had greater root mass under high salinity, but the effect was reduced with addition of Fe.

Mineral composition of leaves was examined after 7 weeks of salinized irrigation. Potassium concentration decreased under both salinity water, and Mg content under high salinity was equivalent to potable water; but Mg content decreased under the lower salinity. Sodium, Ca, and Fe content of leaves increased as salinity level increased.

Plant Na content increased with highest salinity water under all PGR and/or Fe treatments. These results suggest that high concentrations of Na in leaves are not always phytotoxic. Possibly, the increase in Ca reduced Na phytotoxicity. Chelated Fe was equally or more beneficial than FSE or PPC, but these did not cause synergistic or additive effects.

## **INTRODUCTION**

Soluble salts in the soil or irrigation water, are detrimental to seedling development and plant establishment in many agriculture settings. It is estimated that nearly 40% of

the world's arable land has salinity problems (Wignarajah, 1990). Substrate salinity, in particular that created by NaCl, produces substantial adverse effects on crop quality and yields (Mass and Grieve, 1977). Slow growth due to dissolved salts in the root zone is attributed to reduced uptake of water and essential ions (Bernstein and Hayward, 1958; Jeschke et al., 1986). Chang and Dregne (1955) proposed that excess Na ions can increase soil water retention, reduce soil aeration, and cause growth reduction in plants. Harmer and Benne (1945), Cooper et al. (1953), and Pessoa da Costa and Smucker (1981) reported that Na in small quantities has positive influences on plant development; but Munns and Termaat (1986) and Glenn (1987) noted that Na toxicity can cause reductions in leaf water status and inhibit growth of plants in saline environments.

At high salinity levels, NaCl concentrations have been observed to rise in bundle sheath and mesophyll cells, and that was accompanied by a reduction of K content in leaf tissues (Stelze, 1981). The addition of NaCl to a nutrient culture without additive Ca significantly increased Na and Cl concentrations and decreased Ca, Mg, and K concentrations in citrus leaves; but when CaSO<sub>4</sub> and CaCl<sub>2</sub> were added to the growth media, Ca increased and Na decreased (Zekeri and Pearson, 1990).

Salinized plants may grow more slowly because of shoot

injury, decreased leaf expansion, and a lower rate of photosynthesis (Rawson and Munns, 1984; Mass and Grieve, 1987). Several researchers have demonstrated that Na and Cl ions are largely accumulated within the vacuoles; that can reduce water content and cause nutrient imbalances (Hellbust, 1976). Phospholipid levels in plant roots and shoots decrease when irrigated with saline water (Kuiper, 1984). Hsiao et al. (1976), Turner and Kramer (1980), and Caldwell (1974) indicated the ability of plants to survive in a saline environment might be dependent upon the availability of specific ions that aid in regulating leaf turgidity. Van Standen and Davey (1979) and Walker and Dumbroff (1981) noted that plant roots not only absorb mineral nutrients but also are sites of cytokinin biosynthesis. Cytokinins are plant growth regulators (PGR) that can be involved in ion movement and growth regulation during salinity stress.

Mineral nutrients, such as Ca, and PGR such as kinetin, ethephon, and gibberellin, and proline are known to alleviate salinity stress of plants (Joseph and Khan 1976; Hyder and Greenway, 1965; Shaddad, 1990; Zekri and Pearson, 1990). The addition of Ca to soils tends to decrease the uptake of Na ion and enhance K concentrations, which help maintain the permeability of leaf cell membranes (Waisel, 1962; Hansen and Munns, 1988; Zekri and Pearson, 1990). Seeds of lettuce plants soaked in fusicoccin or a combination of three PGR (kinetin,

ethephon, and gibberellic acid) had substantially enhanced germination and seedling development in a saline environment (Joseph and Khan, 1976). Aloni et al. (1980) reported that seedlings of celery grown with a continuous supply of NaCl increased substantially in leaf tissue and petiole elongation when gibberellic acid was applied to the foliage.

The objectives of this study were to evaluate key plant responses as influenced by two and/or chelated Fe on Kentucky bluegrass grown under various salinity regimes

#### **MATERIALS AND METHODS**

Four-month old, well-fertilized (N, P, and K were applied to provide 49 Kg N/ha at seeding with a soluble 10-10-10 fertilizer) Kentucky bluegrass (Poa praetensis cv. 'Plush') was established in the field. A Groseclose silt loam (a clayey, kaolinitic, mesic Typic Hapludult) was selected for this investigation. Soil pH was 6.2, and P, and K were 24 and 36  $\mu\text{g/g}$  respectively.

Plots were treated on 18 August 1990 with various combinations of PGR and chelated Fe. One PGR was Roots, a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine (FSE). It was applied at 9.3 L/ha. The other PGR was the systemic fungicide propiconazole (trade name: Banner, chemical name: [1-((2-(2,4-

dichlorophenyl)-4-propyl-1,3-dioxolan-2yl)methyl-1 H-1,2,4-triazole] (PPC). It was applied at 0.93 L active ingredient (a.i)/ha. Each PRG was applied alone or in conjunction with Na-diethylene-triamine-pentaactate chelated Fe at 0.11 kg a.i/ha. All treatments were applied to 0.9 m by 1.8 m plots and were replicated four times. A compressed-air sprayer that delivered 784 L/ha of solution at a pressure of 290 kPa was used for foliar applications. All treatments were applied to 0.9 m by 1.8 m plots and were replicated four times. Treated plants were allowed to grow for two weeks and irrigated as needed.

Root rings were made from plastic 10-cm-diam PVC pipe and cut to provide 5 cm depth. Two steel wires 3-mm in diameter were inserted at right angles through holes drilled 6 mm from the bottom edge of ring. Hardware cloth with 6 mm mesh, cut to fit the inside of the ring, was placed into the ring and rested on the steel wires (Schmidt et al., 1986). A root ring was placed in each of 72 metal containers 15 cm in diam and 15 cm high filled with 4.4 kg of the same Groseclose silt loam. Two weeks after spray treatments a 10-cm x 5-cm deep diam plug was taken from each of the treated plots and transplanted into a root ring in one of the containers. Plant roots were allowed to grow through the hardware cloth into the soil.

Prior to beginning salinized irrigations, the transplanted plugs irrigated for 1 week with potable water,

and plants were then clipped at 5 cm. Approximate field capacity of the soil was determined by saturating each container and permitting it to drain for 24 h. The weight of each container was then recorded.

Containers were placed under a rain shelter, and 220 mL of water containing 0, 0.15, and 0.3 S m<sup>-1</sup> salt concentrations were supplied to separate containers three times a week for 7 weeks. Saline irrigation treatments were made by dissolving 0.96 g NaCl/L or 1.92 g NaCl/L, in potable water.

Plant height, leaf water status (LWS), and dry weight of clippings were measured 3 weeks after initiation of salt treatments (AIST). Leaf water status was determined by removing the last fully emerge leaf from a tiller and placing it in a Campbell-Brewster leaf press chamber. The chamber was activated by a hydraulic pump that was capable of compressing the leaf as described by Jones and Corably (1980). The pressure that caused water to be exuded uniformly from tip, bottom, and the edges of the leaf was recorded. The greater the pressure required to exude water, the lower the LWS. The means for LWS were determined from three leaf measurements per treatment. Leaves were clipped at 5 cm 3 weeks AIST and dried at 60°C for 24 h.

The salinity regimes were continued for 4 more weeks. At 7 weeks AIST, plant heights, leaf color, LWS, root-pull strength, and clipping yields were ascertained for each

treatment. Root-pull strength of the transplanted plugs was measured by vertically lifting the root rings from the soil in the metal containers. The force required was measured with a hand-held pull-gauge (model DPPH-100, John Chatillion & Sons, Inc., 83-30 Kew Gardens Road, Kew Gardens., New York). Two metal hooks extended from the gauge and attached to the root rings (Schmidt et al. 1986). Leaf water status was determined as previously described above.

Leaf color was ranked from 1 to 9, where 9 was the darkest green and anything ranked below 6 was a lighter green than was considered acceptable. Foliage was again clipped 7 weeks AIST at 5 cm and was dried at 60°C for 24 h to determine dry weight. Mineral composition of leaf tissues was determined by using perchloric acid methods described by Lim and Jackson (1958) and Jones et al. (1973). Sodium, P, K, Ca, Mg, and Fe concentrations of leaves were analyzed by inductively coupled plasma spectroscopy.

A randomized complete block design with three salinity levels and six chemical treatments was replicated four times. Analysis of Variance (ANOVA) was applied to analyze the data, and means were separated by the Duncan's multiple range test at 10% probability when the ANOVA indicated significant treatment effects.

## **RESULTS**

Two different irrigation water salinities showed no significant effects on Kentucky bluegrass plant height when compared to plants grown under potable water (Table 4.1). All PGR and/or chelated Fe treatments, when averaged over the salinity regimes, stimulated leaf elongation relative to the control. Fortified seaweed extract, although not significantly different from PPC, was the most active chemical treatment in promoting foliage growth 3 weeks AIST.

There was no significant effect of salinity treatments on clipping dry weight 3 weeks AIST (Table 4.2). Propiconazole and FSE plus Fe caused significantly higher dry matter accumulation when average across salinities. Other treatments tended to increase plant dry weight but were not significantly different from the control.

Plants grown under the lower salinity were not different in LWS from plants under potable water 3 weeks AIST (Table 4.3). Irrigation at the higher salinity decreased LWS when compared to the potable water or low salinity treatments. All the PGR and/or chelated Fe treatments resulted in improved LWS as compared to the control 3 weeks AIST. Propiconazole was the most active chemical treatment in increasing LWS 3 weeks AIST. Addition of the chelated Fe tended to reduce the effect of FSE and PPC.

Bluegrass was shorter under both salinization treatments

**TABLE 4.1.** Height of Kentucky bluegrass 6 weeks after chemical treatment, 4 weeks after transplanting and after growing 3 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and chelated Fe.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----cm-----			
PPC	0.93	0.0	27.4	27.0	26.4	26.9ab*
FSE	9.3	0.0	29.2	27.0	27.0	27.7a
Fe	0.0	0.11	24.5	26.0	26.0	25.5b
PPC+Fe	0.93	0.11	26.3	25.0	24.5	25.2b
FSE+Fe	9.3	0.11	28.3	25.0	25.0	25.9b
Control	0.0	0.0	22.5	22.0	21.5	21.9c
Mean			26.4x*	25.3x	25.0x	

\*Means within a column (a,b,c) or row (x) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

**TABLE 4.2.** Shoot dry weight of Kentucky bluegrass 6 weeks after chemical treatment, 4 weeks after transplanting and after growing 3 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and chelated Fe.

Treatment	Amount		Irrigation Salinity (S m <sup>-1</sup> )			
	PGR	Fe	0	0.15	0.3	Meam
	L/ha	kg/ha	-----g dry wt/m <sup>2</sup> -----			
PPC	0.93	0.0	2.1	2.3	2.2	2.2a*
FSE	9.3	0.0	2.0	1.9	2.0	1.9ab
Fe	0.0	0.11	2.0	1.9	1.9	1.9ab
PPC+Fe	0.93	0.11	2.0	2.2	1.9	1.9ab
FSE+Fe	9.3	0.11	2.1	2.4	2.0	2.0a
Control	0.0	0.0	1.7	1.7	1.7	1.7b
Mean			2.0x*	2.1x	1.9x	

\*Means within a column (a,b) or row (x) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 4.3.** Leaf water status of Kentucky bluegrass 6 weeks after chemical treatment, 4 weeks after transplanting and after growing 3 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----kPa-----			
PPC	0.93	0.0	155	165	175	165a*
FSE	9.3	0.0	160	170	180	170ab
Fe	0.0	0.11	180	180	185	182c
PPC+Fe	0.93	0.11	165	170	180	172b
FSE+Fe	9.3	0.11	175	180	190	182c
Control	0.0	0.0	195	200	210	202d
Mean			172y*	177y	187x	

\*Means within a column (a,b,c,d) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

than plants grown under potable water for 7 weeks AIST (Table 4.4). The higher salinity suppressed leaf elongation most. The PGR with or without chelated Fe treatments stimulated foliage growth across all salinities.

Lighter green leaves were observed in plants grown under the higher salinity as compared to the lower salinity, and both were lighter than leaves on plants grown with potable water (Table 4.5). Propiconazole and chelated Fe alone improved leaf coloration when compared to the control plants 7 weeks AIST, but the PPC plus Fe treatment was not different from control.

Seven weeks AIST, as salinity increased, LWS of bluegrass leaves also decreased (Table 4.6). (Larger pressure values means lower LWS). Plant growth regulators and/or Fe enabled plants to maintain a better LWS in their leaf tissues as compared to the control plants. Propiconazole was the most active material in increasing bluegrass tolerance to salinity 7 weeks AIST. Addition of chelated Fe reduced the effect of PPC.

Bluegrass plants grown under the higher salinity irrigation had significantly lower clipping dry matter accumulation than plants grown under potable water or the lower salinity irrigation 7 weeks AIST (Table 4.7). Plant growth regulators and/or chelated Fe increased dry weight of bluegrass clippings when compared to the control.

**TABLE 4.4.** Height of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and chelated Fe.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----cm-----			
PPC	0.93	0.0	29.3	26.0	22.5	25.9a*
FSE	9.3	0.0	30.2	25.4	20.5	25.3a
Fe	0.0	0.11	27.4	25.5	22.5	25.1ab
PPC+Fe	0.93	0.11	27.5	24.5	20.5	24.1b
FSE+Fe	9.3	0.11	28.4	25.3	24.0	25.9a
Control	0.0	0.0	24.0	22.5	20.0	22.1c
Mean			28.0x*	25.0y	22.0z	

\*Means within a column (a,b,c) or row (x,y,z) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test

**TABLE 4.5.** Leaf color of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Irrigation Salinity (S m <sup>-1</sup> )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----Rating#-----			
PPC	0.93	0.0	9.0	8.5	6.0	8.0ab*
FSE	9.3	0.0	9.0	8.0	6.0	7.6bc
Fe	0.0	0.11	9.0	8.5	7.0	8.3a
PPC+Fe	0.93	0.11	8.5	7.5	6.0	7.3c
FSE+Fe	9.3	0.11	9.0	8.5	7.0	8.1a
Control	0.0	0.0	8.5	7.5	6.0	7.3c
Mean			9.0x*	8.0y	6.0z	

\*Means within a column (a,b,c) or row (x,y,z) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

#Color was ranked from 9 to 1, where 9 is the best, 6 acceptable, and 1 is the poorest.

**TABLE 4.6.** Leaf water status of Kentucky bluegrass leaf 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and chelated Fe.

Treatment	Amount		Irrigation Salinities ( $S\ m^{-1}$ )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----kPa-----			
PPC	0.93	0.0	170	180	205	185a*
FSE	9.3	0.0	170	195	220	195b
Fe	0.0	0.11	190	215	225	210c
PPC+Fe	0.93	0.11	185	195	225	202b
FSE+Fe	9.3	0.11	185	190	220	198b
Control	0.0	0.0	215	230	250	232d
Mean			186x*	201y	224z	

\*Means within a column (a,b,c,d) or row (x,y,z) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test

**TABLE 4.7.** Dry weight of clippings of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and chelated Fe.

Treatment	Amount		Irrigation Salinity (S m <sup>-1</sup> )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----g dry wt/m <sup>2</sup> -----			
PPC	0.93	0.0	2.0	2.1	1.9	2.0a*
FSE	9.3	0.0	2.0	1.9	1.6	1.8ab
Fe	0.0	0.11	1.8	1.5	1.5	1.7b
PPC+Fe	0.93	0.11	2.0	1.8	1.6	1.8ab
FSE+Fe	9.3	0.11	2.3	2.0	1.5	1.9ab
Control	0.0	0.0	1.6	1.5	1.3	1.5c
Mean			1.9x*	1.7y	1.6y	

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

As salt concentration increased in the irrigation water, root-pull strength decreased (Table 4.8). Plants grown under either low or high salinity irrigation had about 40% lower root-pull strength than plants irrigated with potable water. Chemical treatments and salinity regimes interacted for this parameter. Except at the intermediate salinity, all PGR with or without Fe caused an increase in root growth strength. Iron alone caused a decrease under the 0.15 S m<sup>-1</sup> treatment and no change under the higher salinity. Propiconazole alone and FSE plus Fe increased root strength when compared to other chemical treatments under 0.15 S m<sup>-1</sup> salinity. Chelated Fe plus FSE increased root strength, but neither was effective when applied alone at 0.15 S m<sup>-1</sup>. At the higher salinity, root strength was greatest with the FSE and PPC treatments, while addition of the chelated Fe to PPC significantly reduced the root effect. All PGR-treated bluegrass grown under the higher salinity developed stronger root systems than the control plants.

Mineral composition of leaves at 7 weeks AIST changed in response to salinity regimes (Tables 4.9 to 4.14). There was a chemical treatment by salinity interaction for all elements, except K. As salinity increased, the Na concentration in leaf tissue also increased (Table 4.9). Seven weeks AIST leaf tissue of plants irrigated with saline water had two to three fold Na increases when compared to plants irrigated with

**TABLE 4.8.** Root-pull strength of Kentucky bluegrass 10 weeks after chemical treatment, 7 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and water salinity interacted at the  $p=0.1$  level.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )		
	PGR	Fe	0	0.15	0.3
	L/ha	kg/ha	-----kg/m <sup>2</sup> -----		
PPC	0.93	0.0	26b*	23a	27a
FSE	9.3	0.0	29b	16c	21ab
Fe	0.0	0.11	23c	13d	10d
PPC+Fe	0.93	0.11	35a	18b	13c
FSE+Fe	9.3	0.11	26b	23a	17b
Control	0.0	0.0	18d	16c	11d
Mean			28x*	18y	16y

\*Means within a column (a,b,c,d ) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

**TABLE 4.9.** Sodium concentration of leaf tissue of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified extract (FSE), and Fe. Chemical treatment and water salinity interacted at the  $p=0.1$  level.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )		
	PGR	Fe	0	0.15	0.3
	L/ha	kg/ha	-----§-----		
PPC	0.93	0.0	0.12b*	0.29a	0.42b
FSE	9.3	0.0	0.14ab	0.28a	0.41bc
Fe	0.0	0.11	0.14ab	0.18c	0.48a
PPC+Fe	0.93	0.11	0.16a	0.28a	0.41bc
FSE+Fe	9.3	0.11	0.12b	0.23b	0.36c
Control	0.0	0.0	0.13b	0.22b	0.27d
Mean			0.13z*	0.24y	0.39x

\*Means within a column (a,b,c,d) or row (x,y,z) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

potable water. Under saline irrigation, several PGR and/or chelated Fe treatments resulted in higher Na content in leaf tissues. Addition of Fe tended to reduce the effect of FSE. Foliar application of PGR and/or chelated Fe to bluegrass grown under the higher salinity resulted in higher Na concentrations than in the control plants.

Kentucky bluegrass plants that received either level of added salinity had significantly higher Ca content in their leaf tissues than the plants receiving potable water (Table 4.10). Under potable water, all plant treatments, except chelated Fe alone and PPC+Fe, caused significantly less Ca uptake than the control. Under low salinity, all foliar treatments, except FSE plus Fe significantly increased leaf Ca content when compared to the control. Under high salinity, PPC either alone or in combination with Fe and Fe alone caused an increase in Ca content. Fortified seaweed extract with chelated Fe caused a decrease in Ca content. Addition of the chelated Fe tended to reduce the effect of PPC.

Potassium concentration of leaves was reduced by saline irrigation (Table 4.11). Plants grown under the higher salinity had less K content than those grown under potable water but more than those grown in low salinity. Most chemical treatments did not significantly affect K. Propiconazole plus chelated Fe decreased K content compared to the control plants. Addition of the Fe tended to reduce the K uptake

**TABLE 4.10.** Calcium concentration of leaf tissue of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and water salinity interacted at the  $p=0.1$  level.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )		
	PGR	Fe	0	0.15	0.3
	L/ha	kg/ha	-----%-----		
PPC	0.93	0.0	0.098bc*	0.142a	0.189a
FSE	9.3	0.0	0.102bc	0.140a	0.161bc
Fe	0.0	0.11	0.121a	0.140a	0.185a
PPC+Fe	0.93	0.11	0.112ab	0.140a	0.168b
FSE+Fe	9.3	0.11	0.097c	0.120b	0.147d
Control	0.0	0.0	0.127a	0.123b	0.154c
Mean			0.110z*	0.134y	0.167x

\*Means within a column (a,b,c,d) or row (x,y,z) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

**TABLE 4.11.** Potassium concentration of leaf tissue of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Irrigation Salinity (S m <sup>-1</sup> )			
	PGR	Fe	0	0.15	0.3	Mean
	L/ha	kg/ha	-----%-----			
PPC	0.93	0.0	2.0	1.6	2.4	2.0a*
FSE	9.3	0.0	2.3	1.3	1.6	1.7bc
Fe	0.0	0.11	2.2	1.2	1.6	1.7bc
PPC+Fe	0.93	0.11	1.8	1.4	1.7	1.6c
FSE+Fe	9.3	0.11	2.1	1.7	1.7	1.8abc
Control	0.0	0.0	2.2	1.7	1.8	1.9ab
Mean			2.1x*	1.5z	1.7y	

\*Means within a column (a,b,c) or row (x,y,z) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

effect of PPC.

Bluegrass plants grown under the lower salinity irrigation had significantly less P in leaf tissues than plants grown in potable water or the higher salinity water (Table 4.12). There was no significant difference between the high salinity and potable water regimes. Propiconazole plus Fe was the only treatment that increased P content of bluegrass under potable water. Fortified seaweed extract plus Fe caused a decrease in P content of plant leaves with either low or high salinity irrigation. All the other plant treatments had K contents equivalent to the control plants.

Plants grown with potable water had significantly less Fe content in their tissues than to those that received either low or high salinity irrigation (Table 4.13). There was no significant differences between low and high salinity in relation to Fe content. Iron alone was the only chemical treatment that significantly increased Fe content of bluegrass plants under potable water.

Addition of the Fe to either PPC or FSE significantly decreased Fe content of bluegrass compared to PGR alone and to control plants irrigated with the lower salinity water. All the plant treatments, except chelated Fe, caused a decreased in Fe content of bluegrass leaves grown under high salinity irrigation.

Low, but not high, salinity reduced Mg content of

**TABLE 4.12.** Phosphorus concentration of leaf tissue of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe.

Treatment	Amount		Irrigation Salinity (S m <sup>-1</sup> )		
	PGR	Fe	0	0.15	0.3
	L/ha	kg/ha	-----&-----		
PPC	0.93	0.0	0.51b*	0.41ab	0.53a
FSE	9.3	0.0	0.49b	0.41ab	0.51ab
Fe	0.0	0.11	0.49b	0.44a	0.54a
PPC+Fe	0.93	0.11	0.58a	0.42ab	0.52a
FSE+Fe	9.3	0.11	0.49b	0.40b	0.47b
Control	0.0	0.0	0.49b	0.44a	0.54a
Mean			0.51x*	0.42y	0.52x

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at p=0.1 according to Duncan multiple range test.

**TABLE 4.13.** Iron concentration of leaf tissue of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and water salinity interacted at the  $p=0.1$  level.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )		
	PGR	Fe	0	0.15	0.3
	L/ha	kg/ha	-----§-----		
PPC	0.93	0.0	0.094b*	0.186a	0.160bc
FSE	9.3	0.0	0.095b	0.190a	0.184b
Fe	0.0	0.11	0.230a	0.200a	0.192ab
PPC+Fe	0.93	0.11	0.094b	0.140b	0.135c
FSE+Fe	9.3	0.11	0.084b	0.110c	0.135c
Control	0.0	0.0	0.170b	0.195a	0.220a
Mean			0.130y*	0.170x	0.171x

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

**TABLE 4.14.** Magnesium concentration of leaf tissue of Kentucky bluegrass 10 weeks after chemical treatment, 8 weeks after transplanting, and after growing 7 weeks under various levels of salinity as affected by propiconazole (PPC), fortified seaweed extract (FSE), and Fe. Chemical treatment and water salinity interacted at the  $p=0.1$  level.

Treatment	Amount		Irrigation Salinity ( $S\ m^{-1}$ )		
	PGR	Fe	0	0.15	0.3
	L/ha	kg/ha	-----%-----		
PPC	0.93	0.0	0.52c*	0.47b	0.63a
FSE	9.3	0.0	0.53c	0.42b	0.53b
Fe	0.0	0.11	0.60a	0.29c	0.56b
PPC+Fe	0.93	0.11	0.59a	0.51a	0.55b
FSE+Fe	9.3	0.11	0.50c	0.45b	0.52b
Control	0.0	0.0	0.56b	0.44b	0.55b
Mean			0.55x*	0.43y	0.56x

\*Means within a column (a,b,c) or row (x,y) followed by the same letter are not significantly different at  $p=0.1$  according to Duncan multiple range test.

bluegrass plants when compared to plants irrigated with potable water (Table 4.14). Propiconazole in combination with Fe and chelated Fe alone enhanced Mg content of bluegrass plants grown under potable water, while the other treatments resulted in less Mg content than the control plants. Under low salinity irrigation, plants treated with PPC plus chelated Fe had higher Mg in their leaf tissues than other chemical treatments and the control plants, but chelated Fe alone decreased leaf Mg. Application of PPC to bluegrass grown under high salinity increased leaf Mg content when compared to other chemical treatments and control plants.

## **DISCUSSION**

Salinity regimes influenced growth and plant nutrient content when compared to plants irrigated with potable water. Leaf discoloration and other salinity damage symptoms on leaves were visible under both salinity levels as salinization was prolonged. Foliage elongation and shoot dry weight tended to decline under salinity regimes, in particular under high salinity. Plant growth regulator applications either with or without Fe and Fe alone tended to improve LWS under either low or high salinity regimes compared to controls. Foliar application of PGR and/or chelated Fe to bluegrass plants stimulated root growth under both low and high salinity

levels.

Joseph and Khan (1976) and Braun et al. (1976) also reported that applications of kinetin and fusicocin improved seedling development and root growth in lettuce grown under saline soil conditions. Application of proline, an amino acid, to several plant species and synthetic gibberellic acid to Suaeda usuriensis grown under high salinities increased growth and development (Helkal and Shaddad, 1982; Munns and Termaat, 1986). Turner and Kramer (1980) noted improvement of osmotic pressure, leaf permeability, and extensive root growth as major factors for plant survival under saline conditions. The present investigation, demonstrated that PGR with or without chelated Fe have a tendency to improve growth under saline water. Plant canopy growth, leaf color, LWS, and biomass production were 10% to 15% higher in the lower than in the higher salinity irrigation. There was no indication of significant differences in root-pull strength between low and high salinity regimes.

Nieman and Bernstein (1959) and Shaddad (1990) reported that proline generally alleviated the effects of salinization treatments on radish growth. Hasson-Porath et al. (1972) and Weimberg et al. (1984) documented that low saline conditions stimulated growth and development, and such growth promotion was achieved by hyperadaptation and increased turgor pressure for cell elongation. Nieman and Bernstein (1959) reported that

exogenous application of gibberellin to kidney beans grown under low saline soil increased stem length and yield production. The present study, also showed that PGR with or without Fe can potentially improve bluegrass growth and development under saline conditions.

The adverse effects of saline irrigation on bluegrass growth could be mainly due to Na toxicity in leaf tissues. Zekri and Pearson (1990) and Kent and Lauchli (1985) demonstrated that the addition of NaCl to the nutrient solution without addition of Ca can potentially increase leaf Na and Cl but decrease K, Ca, and Mg and cause a decline in crop productivity. High Na concentrations in the plant produced a Ca deficiency and may also inhibit enzymatic oxidation of polyphenol. Brown pigmentation and reduced carbon in leaf tissues then result (Kent and Lauchli, 1985; Aloni et al., 1976). Results of investigation, in this experiment indicated that Na, Ca, and Fe concentrations increased under high salinity regime; but K concentrations decreased, and no significant differences occurred in P and Mg concentrations with saline irrigation. Propiconazole-treated plants at the higher salinity and PPC plus chelated Fe at the lower salinity increased the Mg concentrations in the leaves. The reason for these results remains unclear.

Even with increasing Na concentration associated with increase salinity in the leaves of plants, we found no decline

in foliage dry weight and root-pull strength. Turner and Kramer (1980) proposed that plants with excess Na content in their leaf tissues exhibit positive growth responses, possibly related to cell membrane permeability.

Application of PGR with or without Fe was accompanied by a significant increase in cationic content, especially Ca that may have alleviated the inhibitory effects of Na toxicity and restored growth when compared to control plants. These results support previous work of Zekri and Pearson (1990) and Shaddad (1990) who believed that excess of some cations, in particular Ca, offset the inhibitory effects of Na. Previous work has shown that the addition of Ca to nutrient solutions can alleviate the effects of salinity, improve essential mineral nutrients in leaf tissues, and promote growth (Epstein, 1972; Deo and Kanwar, 1969; Lahaye and Epstein, 1971; Hansen and Munns, 1988; Kent and Lauchli, 1985; Muhammed et al., 1987). Harding et al. (1958) and Pearson and Huberty (1959) also demonstrated that gypsum application to saline soil significantly reduced soluble Na in the soil and in citrus tissues. The present investigation also indicated that plants receiving potable water had generally lower Ca concentrations than with either low or high salinity regimes. Explanation of these results remain unclear.

Plant growth regulators, especially those containing cytokinins may activate root cells and at the same time

stimulate biosynthesis of endogenous cytokinins from roots. Higher cytokinins in the root systems accelerate the transportation of essential elements and amino acids from roots to the sites where these substances are utilized (Nelson and Van Staden, 1984). The alleviation of Kentucky bluegrass salinity stress was possibly associated with the accumulation of Ca in leaves that was stimulated by the application of PGR plus chelated Fe. Bernstein (1975) found the accumulation of Ca counteracted Na toxicity in plant tissues. When the Na to Ca ratio increased in plant tissues under non-saline soil without additive Ca (Bernstein, 1975), severe necrotic burn resulted in part due to higher Na contents and Ca displacement in the leaf (Zid and Grognon, 1985).

Plants grew well and appeared to have a better color, especially at the lower salinity. The higher salinity induced a low rate of discoloration on plants without visible burning or dark grayish color symptoms on leaves. Gaugh and Wadleeigh (1944) reported that high salinity caused leaf discoloration and often leaf burning, because of excess amounts of Na and Cl in leaves.

Plant growth regulators either alone or in combination with chelated Fe not only improved LWS but also protected green pigmentation in leaf tissues during salinity stresses. Plant growth regulators with or without chelated Fe were found to increase bluegrass salt tolerance as evidenced by enhanced

LWS, some plant nutrients, shoot growth, and root-pull strength. Improvement of these agronomic characteristics by PGR and chelated Fe application are highly important for grass species to survive in saline environments.

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## CHAPTER 5

### SUMMARY

A series of studies were conducted in field, greenhouse, and laboratory to evaluate responses of Kentucky bluegrass and creeping bentgrass to N fertility, plant growth regulators (PGR), and chelated Fe when the plants were grown under various soil moisture regimes or with saline irrigations. The two PGR used were Roots, a cold-water extract of seaweed and peat humus fortified with "intermediate metabolites" and thiamine marketed by Roots Inc. It was applied at 9.3 L/ha. The other PGR was the systemic fungicide Propiconazole (Banner) [1-((2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl)methyl-1 H-1,2,4-triazole)], produced by Ciba Geigy, was also tested for its PGR-like properties. It was applied at 0.93 L a.i/ha. Each PGR was applied alone or in conjunction with Na-diethylene-triamine-pentaactate chelated Fe (Ciba Geigy 330Fe) at 0.11 kg a.i/ha. Plant growth regulators and/or chelated Fe enhanced growth and development of these grasses especially when they were under environmental stresses.

Salinity and drought may act in similar and dissimilar ways to inhibit cell differentiation and elongation. Leaves exposed to salinity or drought may grow more slowly because of a lower turgor pressure in their tissues. Lower turgor pressures translate to reduced stomatal apertures, and that

can limit CO<sub>2</sub> uptake and lower photosynthetic rates and sugar contents in plant tissues. If saline irrigation or drought continue for long, K, Ca, and other integral components of in the middle lamella may be reduced, and the result may be declining leaf elongation and leaf permeability. That also can reduce growth of the plant. During drought, N can escape from the soil in the form of gases; and chloride can interfere with N in leaf tissues during saline irrigation.

Kentucky bluegrass and creeping bentgrass are C3 plants, which lack the "defense" mechanisms of C4 plants when subjected to limited water. The strategies for altering these C3 grass species' responses to water deficit involve improvement of plant characters by breeding techniques or by exogenous chemical application.

Nitrogen is an essential element for growth and development. When plants received N, above-ground vegetative parts, but not necessarily roots, grow more vigorously.

The use of FSE, PPC, and/or chelated Fe on plants grown under nonstressful conditions has been shown in other studies to increase photosynthetic rate, stomatal aperture, leaf elongation, leaf number, root initiation and elongation, and dry weight. The present investigations indicate that foliar application of these compounds can also be beneficial to plants growing under adverse environments. This study shows that these chemicals may be an important tool for increasing

plant tolerance to salinity and limited water. Chelated Fe was equally or more beneficial than PPC or FSE, but the Fe and the PGR did not cause synergistic or additive effects.

### **Foliage and root growth**

Chemically treated bluegrass grew than untreated plants under lowered soil moisture or saline irrigations. The increased foliage growth was likely due to stimulated root development following chemical treatments. Larger, deeper root systems are able to exploit a larger volume of soil and provide more moisture to plants when needed. The treatments that most consistently stimulated leaf growth across all three soil moisture were PPC and chelated Fe. Fortified seaweed extract, although not significantly different from PPC, was the most active plant treatment in enhancing shoot growth with salinity treatments.

Bentgrass plants grown under low and high N fertility levels behaved differently following drought. The higher rate of N caused a greater growth of above-ground parts than roots. This resulted in enhanced transpiration and wilting when plants were exposed to drought. This had a negative effect on plants and caused a slower growth recovery when irrigation was reinitiated. Root strength/development did differ between N fertility levels. Fortified seaweed extract was the most

active chemical treatment in lowering transpiration, while PPC plus Fe increased transpiration at the higher N fertility level. Propiconazole was the least active chemical in reducing wilting in bentgrass.

#### **Leaf water status, color, and shoot dry weight**

Bluegrass plants that had good growth and development under salinity and limited water had better leaf water status (LWS). Improvement of LWS presumably means increased turgor pressures and stomatal apertures. This along with greater chlorophyll content should lead to enhanced photosynthetic rates. The increased photosynthesis should consequently stimulate tillering, leaf elongation, leaf number, and higher yields.

Chemical treatments, except PPC plus Fe, resulted in a better LWS and enhanced bluegrass growth under three soil moisture tensions. Improvement of LWS in plants grown under saline irrigation also was correlated with increased shoot dry weight under each salinity level.

Bluegrass plants grown under drought were able to maintain more moisture in their leaf tissues at the lower than the higher rate of N fertility. Propiconazole and FSE (at the low N) and PPC plus Fe (at high N) consistently enhanced LWS and resulted in higher shoot dry weights during and after

drought stress.

### **Mineral composition of leaf tissues**

Improvement in root growth, leaf elongation, and leaf water status following chemical treatments promoted nutrition of bluegrass under three different soil moisture levels and saline irrigations. Leaf P and K were enhanced by PGR and/or chelated Fe. Potassium and P are essential elements for the activity of guard cells and leaf conductivity. Leaf Ca, Mg, and Fe concentrations of treated bluegrass were also enhanced under limited soil moisture. These cations are important in enzyme and carbohydrate biosynthesis that conserve energy in plant tissues and protect plant seedlings during stress.

Mineral nutrition bluegrass grown under saline irrigations varied with chemical treatment and salinity level. Potassium content of leaves decreased, while Mg concentration remained unchanged with either salinity treatment. On the other hand, leaf Ca, Fe, P, and Na contents of treated plants increased under salinity in response to application of PGR and/or chelated Fe.

Under salinity, Ca became more important than K for growth response of treated bluegrass. A reason for that may be due to accumulation of Na content in leaf tissues, which caused the plants to become more selective in mineral

nutrients, in particular Ca to offset ion toxicity in plant tissues.

## VITA

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