Research Article

Genotypic variation of forage maize (*Zea mays* L.) hybrids to sources of nitrogenous fertilizer under non-saline and saline conditions

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Abstract

Water shortage and good quality irrigation water are the main alarming issues for forage production of maize crop in the area of arid and semiarid which enforcing the farmers to utilize municipal irrigation and recycled underground brackish water. These constraints reducing the forage yield and resulting into low forage production due to imbalance fertilizer, salinity and solicitation. Nitrogen (N) application is beneficial for the crops in the accordance of its role in crop productivity and nutrient use efficiency. The current study was conducted to study nitrogenous fertilizers role on maize growth under saline conditions. Nitrogenous fertilizers application increased the plant growth parameters in the term of physical para meters like fresh and dry biomass, plant height(cm), leaf area per plant(cm), number of leaves per plant and crude protein (%) under saline conditions. Nitrogen sources application increased the concentration of nitrate (NO₃), ammonium (NH₄), potassium(K), K: Na ratio and decreased sodium (Na), under saline condition. A significant difference was observed in all three maize hybrids while comparing the genotypic performance, the maize hybrid FH-949 showed the considerable increase in all observed parameters as compared to FH-9 22 and Pak-Afgoe which showed a significant reduction in term of all growth parameter under saline conditions.

Keywords: Ammonium; Hybrids; Imbalance fertilizers; K: Na ratio; Nitrate

Introduction

Salinity is becoming one of the alarming issue due to sudden inverse in climate change which reducing the plant yield as well as development [1, 2]. In world the

extent of salt affected soil is 20% and in which 50% of irrigated land is deteriorated just only due to salinity [3]. The main contributing factors is shortage of irrigation water in dry as well as irrigated areas which

enforcing the formers to utilize municipal or brackish water for irrigation purpose [4]. Maize crop is mostly sensitive against salinity at early heading stage which ultimately reduces the forage production as well as quality. For fodder and good forage purpose it is important to harvest crop at two to three weeks before grain maturity. It is also considered as an important raw material for multi manufactured goods and food grain for animals [5].

In agriculture, maize crop contributes 0.4 percent to Gross Domestic Product (GDP) but during the previous year's area maize cultivation decreased 3.30% due to reduction in sown area [6]. It is edible cereal forage which consist of 29.9-31.38% crude fiber, 8.62-10.32% crude protein, 8.45-9.24% total ash and 1.27-1.35% ether extractable fats [7]. In Pakistan, different biotic and abiotic factors are responsible for low production which create problem during lifecycle and genetic material secondly the miss management in balance fertilizer and in irrigation application. So the main constraints for maize yield reduction in Pakistan are imbalance fertilizer application, lack of high yielding genotypes of forage maize, and salinity which influencing the forage productivity of maize crop [8].

Maize crop have a significant forage importance, in earlier duration many of genotypes were introduced and to maximize the production use of these high yielding are best to fulfill the increasing demand of livestock food [7, 9]. The production as well as quality of these genotypes mostly affected due to the ecological factors. It is important to select such type of cultivars by adopting short gun approaches like the suitable nutrient source application under salt stress conditions [10].

Among the essential nutrients, Nitrogen (N) is one of the important nutrient which increase the crop quality and yield, protein contents and seed quality. Nitrogen application also increase the photosynthetic rate and improve the leaf area and enhance

the vegetative growth [11, 12]. The higher dose of N results into ecological contamination so its proper source, its form and doses are needed to consider to prevent the risks from human and soil health [13].

Plants absorb N in the form of anion (NO_3^-) and cation (NH_4^+) which also affected by pH of the root zone (Cheema et al. 2010). The ammonium (NH_4^+) uptake in plants reduced pH of root zone through releasing proton (H^+) which enhance the concentration of (NH_4^+) in vacuole of cell [14].

Nitrate form is less harmful can be accumulated into shoot and other parts. The absorption of N sources effects the N type given to plants also depends on the chemical composition of plants to control the relative ions uptake [15, 16]. The plants which uptake more NO_3 that will reduce the uptake of metals as compared to those provided with NH_4^+ [17-19].

The applied form of N a key role on plant vegetative and reproductive growth which also dependent on growing site, fertilizer dose and crop specie. Keeping in view the N role in plant growth under saline condition, the current study conducted with the objectives to evaluate the comparative effects of $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ and to study the comparative performance of maize hybrids under saline condition.

Material and Methods

Experimental conditions and plant material

The experiment was conducted in the research area of Agronomy department. Soil samples were analyzed in the laboratory for physico chemical analysis. Soils samples ECe and pH were measured by using EC meter (DDS -307A) and pH meter and cation exchange capacity (CE C) was measured by using ammonium acetate method wet oxidation method used for the measurement of organic matter contents. The soil analysis before the experiment conducting are given inn (Table 1). Salinity was artificially developed into pots by using the method of U.S. Salinity Lab Staff [18]. The levels of nitrogenous fertilizers

were applied by using ammonium sulphate (as a source of NH_4) and calcium nitrate (as a source of NO_3).

There were six treatment i.e. T₁: control T₂: EC=10.00 dS m⁻¹, T₃: ammonium sulphate (75 mg kg⁻¹ soil), T₄: calcium nitrate (75.00 mg per kg soil), T₅: ammonium sulphate + EC = 10.00 dS m⁻¹, T₆: calcium nitrate + EC= 10.00 dS m⁻¹ with triplicates. All the treatments arranged factorial with three replications. Three maize hybrids i.e. FH-949, FH-922, and Pak Afgoe sown for this experiment. Seed of all maize hybrids imported from AARI, Faisalabad. After sieving of soil 20 kg pots were filled with sieved soil. The recommended dose of NP K was thoroughly mixed into the soil. The first dose of N, all P and K were applied at the pot filling. Maize seeds were soaked in distilled water, five seeds were sown in each pot. Plants were thinned to maintain two plants in each pot. For maintain the field capacity, pots were irrigated with distilled water for more than fifty days.

Plant parameters

After the duration of 45 days, plants were harvested, and left for sun drying. Samples were oven dried to constant weight and weighed with a spring balance and yield per plant was recorded. After harvesting, shoot samples were oven dried at 70°C to obtain a constant weight. A homogenous

portion of finely ground shoot samples were passed through 40-mesh sieve and samples were digested in a di-acid mixture which was prepared by mixing the ratio of HNO_3 : HClO_4 (2:1) respectively [20].

The sample of 0.5 g dry plant material added into a 100-mL digestion tube. The plant material was digested in the mixture of sulfuric acid and hydrogen peroxide. Leaf NO_3 -N concentration was determined by the method as suggested by [21]. For the determination of NH_4 -N, plant samples were digested in the mixture of K_2SO_4 and HgO as a catalyst and it was determined by the colorimetric method using auto-analyser system. Ammonium in the digested plant samples was determined by spectrophotometer (Shimadzu, UV - 1201, Kyoto, Japan). The absorbance was read in spectrophotometer at 650 nm. The concentration of K and Na were determined by using flame photometer (Model 410, Thermo Electron Limited, Cambridge, UK) respectively.

Statistical analysis

The mean data regarding shoot dry matter and mineral content were analyzed statistically by using *Statistix 9*®. Least significance difference (LSD) test used for mean separation. The variations of the data were expressed as standard deviation and significance of the data was calculated at the $p < 0.05$ [22].

Table 1. Soil analysis used for experiment

Soil Characteristics	Values	Units
Nitrogen	10	mg kg ⁻¹
Organic matter	0.90	%
SAR	3.45	(mmol L ⁻¹) ^{1/2}
Soil Texture	Clayey soil	
Saturation percentage	60.00	%
pH _s	7.50	
EC _e	1.90	dS m ⁻¹

Results and Discussion

Plant height (cm²) and number of leaves per plant

According to forage importance point of plant height and number of leaves per plant

are considered one of the important parameters. Maize hybrid FH-949 produced maximum numbers of leaves per plant (9.49) and plant height (144.00 cm) while minimum leaves (7.30) and plant height

(92.33 cm) was observed in Pak-Afgo under non-saline conditions (Fig. 1). Plant height and number of leaves per plant reduced adversely under saline conditions. Maximum plant height (67.33cm) was observed in FH- 949in the treatment where ammonical form of N while minimum plant height (55.60 cm) was observed in Pak-Afgoe where no N was applied. So the genotype FH-949 performed best under both of the conditions as compared to FH-922 and Pak-Afgoe.

Application of N increased the plan height and number of leaves under non saline conditions which results due to higher fertilizer levels which ultimately facilitate the diffusion and increased nutrient availability. This increase in plant height also observed by some other researchers [23-26]. Some other researchers [27-29] also reported that it might be due to genetic variations and easily facilitation of nutrient in root zone which ultimately improve plant growth in term of an increase in number of leaves and plant height.

Figure 1. Effect of NO₃-N and NH₄-Non A) leaves per plant and B) plant height of maize hybrids

Leaf Area (cm) and Crude protein (%)

The application of nitrogenous fertilizers increased the leaf area of all maize hybrids under non-saline conditions. Among the maize genotype maximum leaf area (80.86 cm) and crude protein (9.02%) was observed in FH-949 followed by FH-922 while in Pak-Afgoe minimum leaf area (70.00 cm) and crude protein (6.76%) was observed (Fig. 2). Application of NO₃-N increased the leaf area and crude protein as compared to applied NH₄-N form.

To estimate the quality of forage contents of crude protein are very important. The quality of forage is considered best when crude protein contents existing in higher quantity in forage composition. Salinity stress adversely reduced the plant growth as a result of physiological dehydration which thick the chlorophyll contents and

decreased leaf area. In case of saline environment, maximum leaf area (67.33 cm) was observed in FH-949 through the application of NH₄-N and minimum leaf area (55.60 cm) was observed in Pak-Afgoe in the treatment where no N was applied.

The response of maize hybrids against nitrogenous fertilizers was different as a result of variation in genetic potential. Difference in crude protein of maize hybrids also reported by [29, 30] reported contradict as they found non-significant differences between hybrids of sorghum. Plants which were fertilized with NH₄-N under non-saline condition might be due to the uptake of more nitrogen resulting in more nitrogen in plants. Similar findings as observed in current study as observed by [31] in maize. In case of leaf area similar observation were also observed by [32, 33]

they reported that the leaf area increased in maize and millet as a result of nitrogenous fertilizer application.

Figure 2. Effect of NO₃-N and NH₄-Non A) leaf area(cm) and B) crude protein (%) of maize hybrids

Fresh and dry biomass per plant (g)

Production of green forage is also dependent on the fresh biomass production. The highest (199.05g) fresh biomass and dry biomass (84.59g) was observed in FH - 949 through NH₄ form of N application while “Pak-Afgoe” maize genotype produced minimum fresh (143g) and dry biomass (81.00 g) under non-saline condition (Fig. 3). The highest fresh and dry biomass were (140.79 g) and (60.45 g) respectively while lowest fresh and dry weight were (108.28 g) and (43.40g) which was observed in Pak-Afgoe under saline conditions. So the application of NH₄ enhance the fresh and dry biomass as compared to NO₃-N form.

The application of N increased the chlorophyll contents and facilitate the photosynthesis process also have a key role in protein contents all these improve the plant growth and development and increase the biomass of maize crop. Similar findings in accordance to maize and sorghum cultivars. It has been confirmed by the previous findings of [29, 30, 34, 35] they reported that maize plant fresh weight enhanced with the application of ammonium sulphate as a source of nitrogen fertilizer under normal and salt stressed conditions.

The increase in plant fresh and dry biomass through the NH₄-N application results actually due to net photosynthesis, plant water potential and improvement in plant water use efficiency. Similar findings were also reported by some other researchers [26, 27, 36] as they reported that application of N in the form of ammonium fertilizer increased the plant growth as it has key role in soil medium acidification and other nutrient availability which ultimately results into more forage production.

Ionic contents (m mol g⁻¹dry wt)

Salt stress causes osmotic effect which limits the nutrients and water uptake because of higher concentration of Na in soil solution. The highest content of Na under saline condition (Fig. 4). Maximum sodium content (396.50 mM) was observed in Pak afgoe genotype and while maximum K content (179.78 mM) was observed in FH-922 maize genotype through the application of NH₄-N. In contrast, lowest sodium contents (325.55mM) and maximum K (198.56 mM) was observed in FH-949. Comparing the N sources impact it was observed that the application of NH₄-N application increased the K concentration and reduced the Na concentration as compared to NO₃-N form.

Figure 3. Effect of NO₃-N and NH₄-N on A) fresh biomass (g) B) dry biomass (g) of maize hybrids

Figure 4. Effect of NO₃-N and NH₄-N on A) sodium concentration B) potassium concentration of maize hybrids

Nitrogenous fertilizer application increases the stomatal conductance and ultimately net photosynthetic process. An increase in K⁺ concentration and suppressing of Na⁺ also argued by other researchers [37, 38]. In case of salt stressed conditions, the excess amount of toxic Na having osmotic effect as well as oxidative damage of maize causing stunted growth and development. Similar findings were also reported by some other researchers [26, 39-43].

A similar trend as observed in case of K:Na ratio as observed for K concentration under non-saline conditions but in the saline conditions. K:Na ratio and K concentration decreased as a result of higher concentration of Na under saline condition. Application of NH₄-N increased K:Na ratio in FH-949 while minimum K:Na ratio was observed in Pak-Afgoe under salt stressed conditions (Fig. 5). The application of NH₄-N increased the K:Na ratio and reduced

the Na concentration as compared to $\text{NO}_3\text{-N}$ form. Similar observation was also observed by researchers [41, 42] they observed that variable response of K: Na ratio of different fertilizers applications in maize crop under saline exist due to antagonistic effect of K and Na. The application of N in the form of $\text{NH}_4\text{-N}$ fertilizers increased the K and K: Na ratio which was also reported by other researchers [43-45] in case of maize crop.

Leaf NH_4 and NO_3 concentration

The NO_3 concentration increased through Nitrate-N application while application of $\text{NH}_4\text{-N}$ increased NH_4 contents in non-saline and saline conditions. The highest NO_3 (455.67%) and NH_4 concentration (56.66%) was observed in FH-949 through the N sources application respectively

while minimum NO_3 (319.33 %) and NH_4 concentration (23.33%) was observed in Pak-Afgoee under salt stressed conditions (Fig. 6). Comparing the genotypic performance, FH-949 uptake maximum NO_3 and NH_4 concentration followed by FH-922 while minimum NO_3 and NH_4 concentration was observed in Pak-Afgoee maize hybrids.

Nitrate form of nutrition is not so much effective as compared to NH_4 and mixing of NO_3 and NH_4 form of fertilizer under saline conditions while comparing the individual application $\text{NH}_4\text{-N}$ form is most beneficial for maize crop [46, 47] also reported similar arguments while working on maize under normal and salt application. Similar findings were also reported by [25, 48, 49].

Figure 5. Effect of $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ on maize hybrids potassium to sodium ratio

Figure 6. Effect of $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ on A) NH_4 concentration B) NO_3 concentration of maize hybrids

Conclusion

In Current study, it was observed that NH_4^+ -N increased the maize hybrids growth in the term of fresh and dry biomass, plant height, leaf area, crude protein, K concentration and K: Na ratio. Furthermore, salt stress either without application of any N source adversely affected the growth of forage maize. The maize plants fertilized with nitrogen application as NH_4 form produced significantly higher forage yields as it has a key role in acidification of root zone which reduced the alkaline conditions and increased the nutrient availability. Comparing the genotypic performance of maize hybrids, FH-949 proved to be the best genotype for fodder production. The adaptation of such approaches in future under saline conditions are helpful for getting good forage yield in saline agriculture especially in calcareous and arid and semiarid regions.

Authors' contributions

Conceived and designed the experiments: M Ibrahim & M Jan, Performed the experiments: MM Maqbool & B Tariq, Analyzed the data: M Nadeem & A Rauf, Contributed reagents/ materials/ analysis tools and wrote the paper: AN Abbas.

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