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Influence of Ferrous Sulfate and Sodium Nitroprusside on Physiology of Groundnut Genotypes

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ABSTRACT

Lime-induced iron chlorosis is a potential problem on most calcareous soils particularly in arid and semi-arid climates affecting most of the plants grown on them. Iron inactivation in plant tissue and organic anions held are responsible as the mechanism leading to the disorder which is still not fully understood, and there is a lack of agreement as to the primary factor responsible for lime-induced chlorosis. The combined application of sodium nitroprusside which is a source of nitric oxide and iron sulphate reduced the incidence of lime induced iron chlorosis compared to control by promoting higher Fe uptake, translocation and activation;and increased physiological parameters such as photosynthetic rate, transpiration rate and dry matter. Among genotypes GPBD-6 had higher photosynthetic rate and dry matter. These results clearly indicate that both the application of sodium nitroprusside and iron sulphate was effective in alleviating iron chlorosis in groundnut genotypes grown on calcareous soil.

Key words: *Chlorosis, Iron deficiency, Photosynthesis, Dry matter*

INTRODUCTION

Iron is an essential micronutrient for all higher plants. Although abundant in most well-aerated soils, most of this iron is not available for plant uptake[17] primarily because it is very insoluble. The presence of calcium carbonate, bicarbonate, calcium and imbalance of nutrient cations in the growth medium, injudicious addition of phosphates, quality of irrigation water, and other soil and plant factors have been held responsible for the disorder. A peculiarity of iron deficiency is a greater decline in protein synthesis in the chloroplasts of leaf cells than in cytoplasm[18] which leads to drastic decrease in net photosynthesis (Pn). The inhibition of Chl formation under iron deficiency is, in at least part is the result of an impaired protein synthesis. The requirement of protein synthesis is reflected in the leaves by a drastic decline in the number of ribosomes, the sites of protein synthesis[8]. As the severity of iron deficiency increases the protein content per leaf area, the leaf cell volume and the number of chloroplasts remains unaffected, whereas the chloroplast volume and the amount of protein per chloroplast decline.

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To make iron more available, plants have evolved various *adaptation mechanisms* that mobilize iron at the root-soil interface (rhizosphere). The absence of iron deficiency symptoms (chlorosis) in many plant species growing on well-aerated neutral and alkaline soils indicates that these adaptation mechanisms are operating. On calcareous soils (soils rich in calcium carbonate), symptoms of chlorosis frequently occur because certain plant species (genotypes) have only a limited capacity to exhibit adaptive processes when growing in these soils. Amelioration of lime-induced chlorosis by (i) acidification of calcareous soils, (ii) use of iron salts, (iii) use of synthetic iron chelates, and (iv) by management practices including the selection and development of varieties resistant to lime-induced iron chlorosis, is discussed. In Present study used sodium nitroprusside chemical which is a source of nitric oxide along with iron salt (ferrous sulphate). Nitric oxide (NO) is a bioactive free radical which plays important roles in many physiological processes in plants, such as growth, development, senescence and adaptive responses to multiple stresses[1,4,10] oxide is reported to ameliorate stress responses in plants[12]. Nitric oxide can readily form complexes with transition metal ions in aqueous solutions or those present in diverse nucleophylic compounds such as metalloproteins. The Fe^{III} nitric oxide complex appears to undergo a charge transfer reaction to form $Fe^{II}NO^{+}$ and can increase the availability of Fe in plants[3]. The objectve of this study is to know the role of SNP and interaction of both ferrous sulphate and SNP in alleviating iron chlorosis and their effect of physiological parameters of all three groundnut genotypes.

MATERIAL AND METHODS

A field experiment was conducted during *kharif* 2014 in the Main Agricultural Research Station (MARS), University of Agricultural Sciences, Dharwad. The experiment was laid out in a split plot design consisted of 15 treatment combinations (Three genotypes and five treatments) *viz*., Soil application of SNP @ 5.63 mg+ $FeSO_4$ @ 18.9 mg /g of urea at the time of sowing (0.31kg SNP ha^{-1} + 1.03kg $FeSO_4$ ha^{-1}) (F1), Foliar application of SNP @ 6mM (1.78g $litre^{-1}$) (F2), Foliar spray of $FeSO_4$ @ 0.5% (5g $litre^{-1}$) (F3), Foliar spray SNP @ 6mM + $FeSO_4$ @ 0.5% (F4) Foliar water spray (Control) (F5) and three groundnut genotypes GPBD-4 (G1), TMV-2 (G2), GPBD-6 (G3). Foliar application of these chemicals had taken at 15 days interval for 4 times.The data was statistically analyzed as per the DMRT for various parameters for interpretation of the results.

Physiological parameters

Chlorophyll content (SPAD meter): SPAD (Soil Plant Analytical Development) is a simple diagnostic tool that measures the greenness or relative chlorophyll content in leaves in terms of SPAD values[14]. However, it requires standardization/calibration for each crop because this relationship varies with crop growth stage and variety. The meter readings are given in Minolta Company defined a SPAD value that indicates relative chlorophyll contents.

Photosynthetic rate and transpiration rate: The young leaves were selected to measure net photosynthetic rate (Pn) and transpiration rate (Tr) by using IRGA (Infra red gas analyser) between 9:00–10:00 AM at different stages of growth.

Phenological stages: by counting the days we can note the number of days required to initiation of flowering days to 50% flowering, days to physiological maturity

RESULTS

Photosynthetic rate (µmoles co_2 $m^{-2}sec^{-1}$) & Transpiration rate (m mole of H_2o m^{-2} sec^{-1})

Data on photosynthesis and transpiration shows that the photosynthetic rate & transpiration rate is get increased at pod initiation stage and were decreased slightly during physiological maturity. But the interaction effect of treatments and genotypes for photosynthetic rate was significant at all the stages (Table 1).

the genotype GPBD-6 recorded significantly higher photosynthetic rate & transpiration rate (19.8,7.7) Significantly lower photosynthetic rate was noticed in TMV-2 (12.8) but transpiration rate lower in GPBD-4(6.9) Among all different treatments of iron supply the significantly higher photosynthetic rate is observed in F_4 (19.4) but treatments effect were did not differ for transpiration significantly The interaction effect of genotype GPBD-6 in the treatment F_4 (23.2) recorded significantly higher photosynthetic rate. The photosynthetic rate decreased at maturity as compare to pod filling stage.

SPAD value: Data on SPAD values (chlorophyll content) indicated increase in chlorophyll content from flowering to pod initiation stage. The genotype GPBD-6 (40.1) was recorded significantly highest SPAD values compared to other genotypes and lowest was recorded in TMV-2(33.1). Among the treatments F_4 (41.6) shows a higher chlorophyll content and rest of other treatments and lowest chlorophyll content observed in control (31.8). The interaction effect was significant at this stage in which the genotype GPBD-6 in treatment F_4 (46.1) recorded significantly higher SPAD value.

Phenological stages:

All the phenological stages of crop growth results indicated that the treatments were not significantly differing (Table 3). Days to initiation of flowering varied only among genotypes. The genotype GPBD-4 initiated flowering at 21.53 DAS while in TMV-2 it was initiated at 25.6 DAS. In GPBD-6 flowering was initiated at 30.33 DAS. The interaction effects of genotype GPBD-6 in treatment F_4 initiated flowers late compared to other GPBD-4 and GPBD-6.

Days to 50% of flowering: 50% flowering was observed at 29.8 DAS, 35.20 DAS, and 39.87 DAS in GPBD-4, TMV-2, and GPBD-6 respectively. The interaction effect was significant in which the genotype GPBD-6 in all the treatments took maximum days to 50 percent flowering and genotype GPBD-4 took minimum days to 50 percent flowering in all the treatments.

Days to physiological maturity: The physiologically maturity at 95.1 DAS, 99.7 DAS, 109.7 DAS in TMV-2, GPBD-4, GPBD-6 attained respectively. The interaction effect was significant for days to physiological maturity. The genotype GPBD-6 took maximum days to physiological maturity in all the treatments compared to TMV-2 and GPBD-4. The genotype TMV-2 in all the treatments took minimum days to physiological maturity.

Dry matter partitioning

Stem dry weight (g): The stem dry weight did not significantly differ among the genotypes. Among the treatments highest stem dry weight recorded in F_4 (7.5). Significantly lowest stem dry weight recorded in control (5.6). The interaction effect of genotype GPBD-4, GPBD-6, TMV-2 in the treatment F_4 (7.6, 7.5, 7.4 respectively) recorded significantly higher stem dry weight as compared to other interactions (Table 2).

Leaf dry weight (g): The data on leaf dry weight significant differences among the genotypes and treatments at all the growth stages. The genotype GPBD-6 recorded significantly highest leaf dry weight (6.5) Among all the treatments, leaf dry weight was significantly higher in F_4 (6.8) Where as significantly lower stem dry weight was recorded in control (5.7) compared to other treatments. The interaction effect of genotype GPBD-6 in the treatment F_4 (7.0) recorded significantly higher leaf dry weight as compared to other genotypes.

Pod dry weight (g): The data on pod dry weight was minimum at 60 DAS and it was maximum at harvest. The pod dry weight was not significantly differ among the genotypes. Among the treatments highest pod dry weight recorded in F_4 (4.2). The interaction effect of genotype GPBD-6 in the treatment F_4 (4.4) recorded significantly higher pod dry weight as compared to other genotypes.

DISCUSSION

Biophysical parameters

Nutrient uptake came to a climax at podding stage, the pegs interposed into the soil to have

pod, and the photosynthesis was most vigorous. Fe deficiency resulted in a decrease of photosynthetic rate and transpiration rate. The supply of $FeSO_4$ and NO maintained higher photosynthetic rate as compared to control. Transpiration pull is the prime impetus of Fe transport in xylem. The present experiment showed that NO supplementation promoted Transpiration in leaves. Therefore, the increase in transpiration promoted both Fe transport in xylem as well as Fe absorption from the soil[19].

The photosynthetic rate in the present investigation during peak flowering (40 to 60 DAS). Photosynthetic rate was higher in FeSO4 and SNP foliar spray alone. Though Bertamini, *et al.*[2] reported that photosynthetic rate was low in chlorotic leaves of grapevine. Here in our present study it increased (fig. 1) due to supply of FeSO4 and SNP. Peanut plants subjected to Fe deficiency exhibited a dramatic decrease in net photosynthetic (Pn) and transpiration rates (Tr)[7].

SPAD values: SPAD (soil plant analysis development) values instantly measures chlorophyll content or "greenness" of plants. Chlorophyll has been rightly designated as "pigment of life" because of their central role in living systems responsible for harvesting sunlight and transforming its energy into biochemical energy essential for life on earth. SPAD values quantify the relative chlorophyll content in plants and are mainly used to indicate the current leaf N status. In the present study, SPAD values differed significantly among the different treatments. The higher SPAD value was recorded under foliar spray of SNP and $FeSO_4$ and SNP alone foliar spray. FeSO4 and SNP donated Fe and increased active Fe(ferrous iron) and leaf chlorophyll content caused re-greening of leaves in Fe-deficient conditions[10,16].

The genotype GPBD-4 and GPBD-6 had higher SPAD values when compared to TMV-2 which is chlorosis susceptible. The results obtained were in conformation with the findings of Prasad *et al.*[15] he revealed that TMV-2 and ICGS-11 were susceptible to Fe chlorosis and produced significantly lower haulm and pod yield.

Per cent chlorosis was assessed by adopting the formula given by[5]. Per cent chlorosis also closely followed the trend of dry matter production. The least number of chlorotic plants were observed in FeSO4 and SNP foliar spray followed by SNP foliar spray. This is due to; Fe deficiency resulted in decrease of chlorophyll content and cause leaf interveinal chlorosis. The chlorosis can be ascribed to the role of Fe in the formation of δ-aminolevulinic acid and protochlorophyllide, the precursors of the chlorophyll biosynthesis.

The observation on chlorosis clearly indicated TMV-2 was "Fe susceptible" and inefficient in extracting the native Fe from the calcareous soils. This result is agreement with the findings of Nagartnamma *et al.*[13]. They reported that genotype TMV-2 was susceptible to iron chlorosis and recorded higher percent chlorosis and lower SPAD values.

Dry matter production and its partitioning

The amount of dry matter produced is an indication of the overall efficiency of utilization of resources and better light interception. The partitioning of dry matter in leaf, stem and pod varied and was influenced by chlorosis. The chlorotic leaves had less light interception rate. Increased DM has been attributed to higher photosynthetic rate as well as increased nutrient acquision and utilization efficiency[9].

Pod yield was positively related with DM, which emphasized the need of maintenance of higher DM for increased pod yield under nutrient stress like lime induced iron chlorosis. The genotypes differed significantly for DM accumulation. Under iron deficiency, the leaf dry weight, stem dry weight and DM decreased as was observed in the susceptible genotype TMV-2. The genotype GPBD-6 had significantly higher stem and leaf dry weight at all the stages with moderately higher iron and chlorophyll contents but the pod dry weight was low. This may be due to the disproportionate partitioning

of photosynthates between vegetative and reproductive parts as influenced by iron chlorosis[6]. The other two genotypes *viz.*, TMV-2 and GPBD-4 had disproportionate partitioning of photosynthates.

The interaction effect of treatment with genotypes was significant for pod yield (fig 2) which might be due to iron supplementation under iron deficiency conditions.

Table 1: Effect of soil and foliar application of $FeSO_4$ and SNP on physiologuical parameters of groundnut in calcareous soil

Treatments	Physiological parameters		
	Photosynthetic rate(µmole of co_2 m^{-2} sec^{-1})	**Transpiaration rate(mmoles of H_2O m^{-2} sec^{-1})**	**SPAD value**
Fe supply(F)			
Soil application of SNP @ 5.63 mg+ $FeSO_4$ @ 18.9 mg /g of urea(F_1)	15.40^{b}	7.21	37.75^{b}
Foliar application of SNP @ 6mM(F_2)	18.15^{a}	7.13	38.27^{b}
Foliar spray of $FeSO_4$ @ 0.5%(F_3)	15.75^{b}	7.82	36.44^{b}
Foliar spray SNP @ 6mM + $FeSO_4$ 0.5%(F_4)	19.40^{a}	7.01	41.66^{a}
Foliar water spray (F_5)	12.95^{c}	7.88	31.76^{c}
S.Em±	**0.53**	**0.278**	**0.57**
LSD @ 5%	**1.74**	**NS**	**1.85**
Genotypes(G)			
GPBD-4(G_1)	16.38^{b}	6.98^{b}	38.30^{b}
TMV-2(G_2)	12.79^{c}	7.35^{ab}	33.11^{c}
GPBD-6(G_3)	19.82^{a}	7.90^{a}	40.12^{a}
S.Em±	**0.41**	**0.31**	**0.44**
LSD @ 5%	**1.35**	**0.99**	**1.43**
Interactions(FXG)			
F_1G_1	15.33^{de}	6.97^{bc}	38.97^{cd}
F_1G_2	11.98^{g}	7.07^{bc}	34.24^{e}
F_1G_3	18.90^{c}	7.60^{ab}	40.05^{bc}
F_2G_1	18.96^{c}	6.50^{cd}	40.08^{bc}
F_2G_2	14.25^{ef}	7.03^{bc}	33.20^{e}
F_2G_3	21.25^{b}	7.85^{ab}	41.53^{b}
F_3G_1	15.38^{de}	7.67^{ab}	37.23^{d}
F_3G_2	12.42^{fg}	7.97^{ab}	33.57^{e}
F_3G_3	19.45^{c}	7.83^{ab}	38.51^{cd}
F_4G_1	19.38^{c}	5.97^{d}	41.77^{b}
F_4G_2	15.59^{de}	7.13^{bc}	37.10^{d}
F_4G_3	23.22^{a}	7.93^{ab}	46.12^{a}
F_5G_1	12.84^{fg}	7.80^{ab}	33.45^{e}
F_5G_2	9.717^{h}	7.53^{ab}	27.43^{f}
F_5G_3	16.30^{d}	8.30^{a}	34.40^{e}
S.Em±	**0.59**	**0.30**	**0.74**
LSD @ 5%	**1.76**	**0.88**	**2.20**

Table 2: Effect of soil and foliar application of $FeSO_4$ and SNP on dry matter partitioning of groundnut in calcareous soil

Treatments	Dry matter partitioning		
	Stem dry weight(g)	**Leaf dry weight(g)**	**Pod dry weight(g)**
Fe supply(F)			
Soil application of SNP @ 5.63 mg+ $FeSO_4$ @ 18.9 mg /g of urea(F_1)	5.92^{bc}	6.19^{bc}	3.56^{bc}
Foliar application of SNP @ 6mM(F_2)	6.47^{b}	6.34^{ab}	3.79^{b}
Foliar spray of $FeSO_4$ @ 0.5%(F_3)	6.12^{bc}	6.03^{bc}	3.22^{d}
Foliar spray SNP @ 6mM + $FeSO_4$ 0.5%(F_4)	7.49^{a}	6.78^{a}	4.19^{a}
Foliar water spray (F_5)	5.61^{c}	5.66^{c}	3.24^{cd}
S.Em±	**0.23**	**0.17**	**0.09**
LSD @ 5%	**0.75**	**0.55**	**0.33**
Genotypes(G)			
GPBD-4(G_1)	6.45	6.26^{ab}	3.64
TMV-2(G_2)	6.21	5.86^{b}	3.49
GPBD-6(G_3)	6.31	6.47^{a}	3.67
S.Em±	**0.18**	**0.13**	**0.08**
LSD @ 5%	**NS**	**0.43**	**NS**
Interactions(FXG)			
F_1G_1	6.06^{b}	6.20^{a-c}	3.44^{c-f}
F_1G_2	5.87^{b}	5.96^{a-c}	3.70^{b-e}
F_1G_3	5.81^{b}	6.43^{ab}	3.53^{b-f}
F_2G_1	6.53^{ab}	6.25^{a-c}	3.82^{b-d}
F_2G_2	6.19^{ab}	6.19^{a-c}	3.47^{c-f}
F_2G_3	6.69^{ab}	6.58^{ab}	4.09^{ab}
F_3G_1	6.44^{ab}	6.12^{a-c}	3.58^{b-e}
F_3G_2	5.98^{b}	5.70^{bc}	3.13^{ef}
F_3G_3	5.92^{b}	6.26^{a-c}	2.94^{f}
F_4G_1	7.55^{a}	6.90^{ab}	4.11^{ab}
F_4G_2	7.44^{a}	6.43^{ab}	3.98^{a-c}
F_4G_3	7.47^{a}	7.00^{a}	4.49^{a}
F_5G_1	5.63^{b}	5.83^{a-c}	3.25^{d-f}
F_5G_2	5.53^{b}	5.07^{c}	3.18^{ef}
F_5G_3	5.65^{b}	6.06^{a-c}	3.30^{d-f}
S.Em±	**0.40**	**0.35**	**0.18**
LSD @ 5%	**1.20**	**1.05**	**0.54**

Table 3: Effect of soil and foliar application of $FeSO_4$ and SNP Phenological stages of groundnut in calcareous soil

Treatments	Phenological stages (days)		
	Days to flower initiation	Days to 50% flowering	Days to physiological maturity
Fe supply(F)			
Soil application of SNP @ 5.63 mg+ $FeSO_4$ @ 18.9 mg /g of urea(F_1)	25.78	35.00	101.4
Foliar application of SNP @ 6mM(F_2)	25.56	34.67	101.6
Foliar spray of $FeSO_4$ @ 0.5%(F_3)	26.00	35.00	101.4
Foliar spray SNP @ 6mM + $FeSO_4$ 0.5%(F_4)	26.00	35.00	101.6
Foliar water spray (F_5)	25.78	35.11	101.4
S.Em±	**0.24**	**0.14**	**0.16**
LSD @ 5%	**NS**	**NS**	**NS**
Genotypes(G)			
GPBD-4(G_1)	21.53^{c}	29.80^{c}	99.7^{b}
TMV-2(G_2)	25.60^{b}	35.20^{b}	95.1^{c}
GPBD-6(G	30.33^{a}	39.87^{a}	109.7^{a}
S.Em±	**0.18**	**0.11**	**0.12**
LSD @ 5%	**0.60**	**0.37**	**0.39**
Interactions(FXG)			
F_1G_1	21.67^{de}	29.67^{cd}	99.7^{b}
F_1G_2	25.67^{c}	35.33^{b}	95.3^{c}
F_1G_3	30.00^{b}	40.00^{a}	109.3^{a}
F_2G_1	21.00^{e}	29.00^{d}	99.7^{b}
F_2G_2	25.67^{c}	35.00^{b}	95.3^{c}
F_2G_3	30.00^{b}	40.00^{a}	109.7^{a}
F_3G_1	22.33^{d}	30.00^{c}	99.7^{b}
F_3G_2	25.33^{c}	35.33^{b}	95.0^{c}
F_3G_3	30.33^{ab}	39.67^{a}	109.7^{a}
F_4G_1	21.33^{e}	30.00^{c}	100.0^{b}
F_4G_2	25.67^{c}	35.33b	95.0^{c}
F_4G_3	31.00^{a}	39.67^{a}	109.7^{a}
F_5G_1	21.33^{e}	30.33^{c}	99.7^{b}
F_5G_2	25.67^{c}	35.00^{b}	94.7^{c}
F_5G_3	30.33^{ab}	40.00^{a}	110.0^{a}
S.Em±	**0.26**	**0.23**	**0.3**
LSD @ 5%	**0.76**	**0.68**	**0.9**

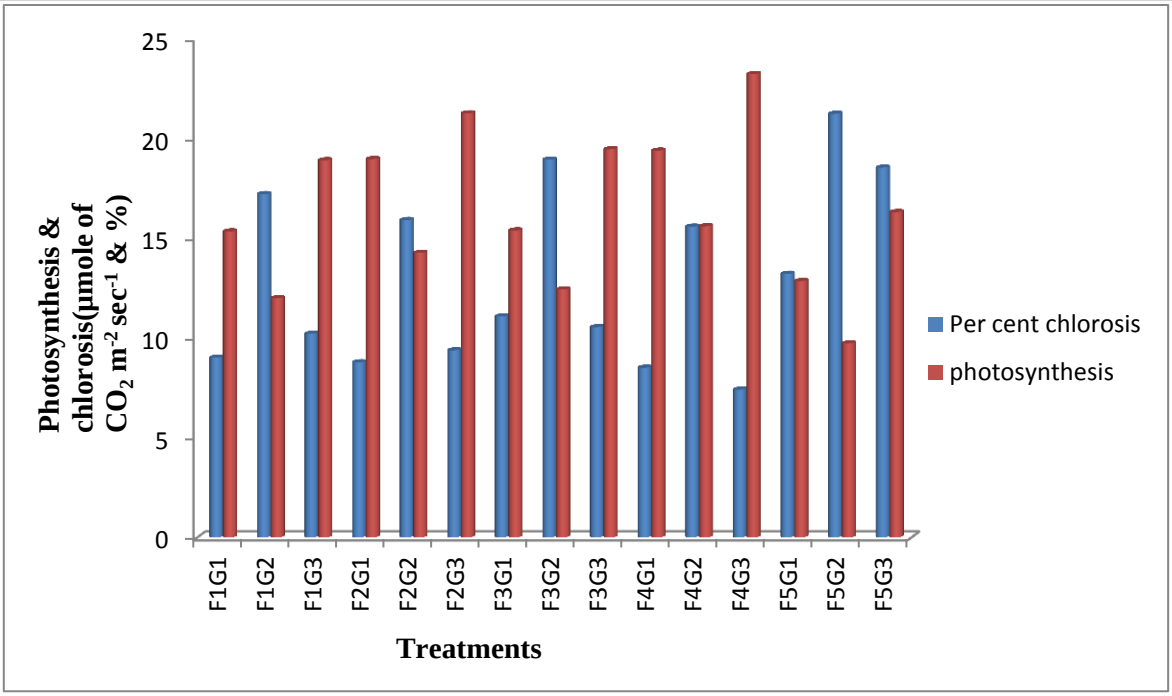

Fig. 1:

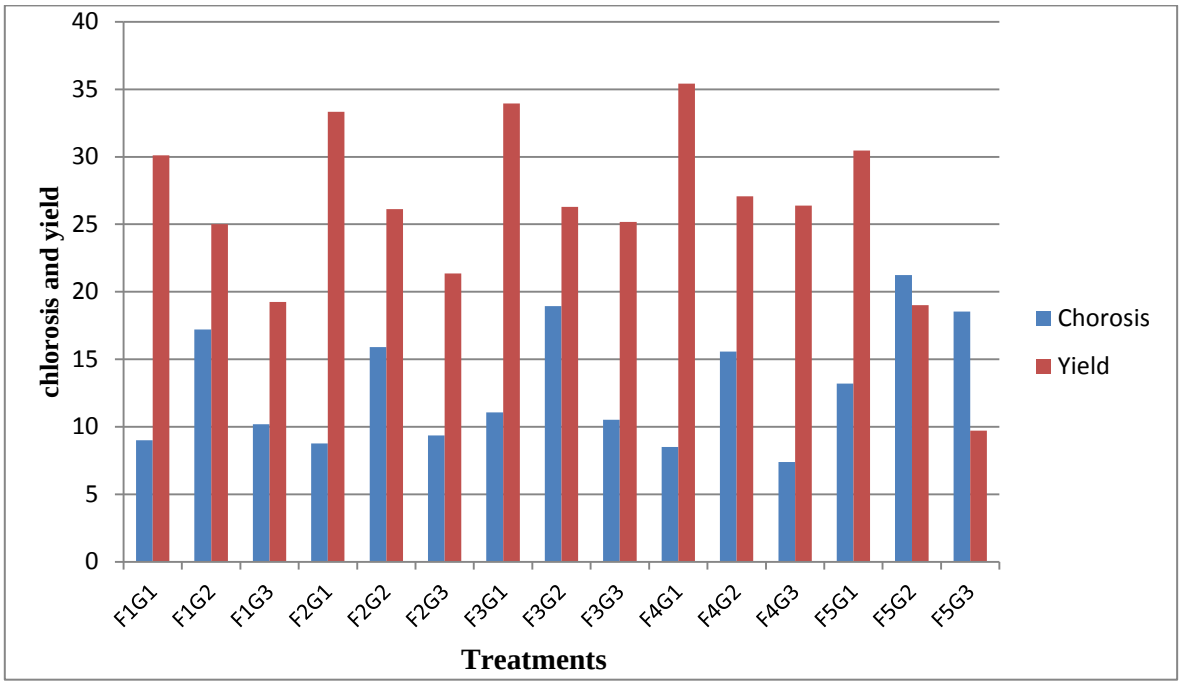

Fig. 2:

CONCLUSION

In present investigation showed that, rather than alone foliar spray of SNP both the combination of $FeSO_4$ and SNP foliar spray showed better results and confirmed that it acts as alleviating agent against iron chlorosis and also shows in both combination SNP had played a main role than $FeSO_4$ in alleviating a lime induced iron chlorosis. In future line of work suggested confirming the rate at low concentration in alleviating the iron deficiency in lime soil because SNP is a donar of nitric oxide is known to be a toxic chemical contains a ferrocynide molecule in it so it has to be used at low concentration.

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