



## SHORT COMMUNICATION

# IRON AND SULPHUR APPLICATION MAY IMPROVE GROWTH AND YIELD OF SOYBEAN

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**This study was conducted to investigate effect of iron and sulphur application on growth and yield of soybean. Two levels of iron (15 and 20 mg kg<sup>-1</sup> soil) and sulphur (40 and 80 mg kg<sup>-1</sup> soil) were applied as split dose application on soybean var PK-327 individually and in combination. Positive effect of iron and sulphur application was observed on different investigated parameters viz. shoot height, root length, number of leaves plant<sup>-1</sup>, chlorophyll content, leaf nitrogen content, number of pods plant<sup>-1</sup>, length of pods, growth analyses parameters, 100 seed weight and seed protein content. The findings of the study suggest that judicious supplementation of iron and sulphur may improve growth and yield potentiality of soybean.**

**Key words:** Chlorophyll, growth, iron, soybean, sulphur, yield.

India is world's major producer of grain legumes even than it is in adequate to ensure minimum requirements recommended by World Health Organization (W.H.O.) and Food and Agricultural Organization (F.A.O.). Soybean (*Glycine max* L. Merr) occupies important position amongst all pulse crops due to its ability to produce high quality proteins and oil content. Soybean proteins are rich in the sulphur containing amino acids and contains good amount of calcium, iron, other mineral salts and vitamins (thiamin and riboflavin).

High yielding crops need large and regular supply of macro and micronutrient elements to develop high photosynthetic capacity and maintain the proper elements concentration in the leaves (Lawlor 1995). Low availability of plant nutrients limits yield in many crops (Heitholt *et al.* 2003). The effect of micronutrient elements on yield and crop performance has been

reported by many researchers. Fe is one of the most limiting nutrient to agricultural production in the world (Kochian 2000). Fe deficiency in the form of chlorosis is a widespread problem for soybean grown on alkaline, calcareous soils (Caliskan *et al.* 2008). Synthesis of chlorophyll, thylakoid, and some ferrous proteins is dependent on this element (Imsande 1998). Iron deficiency in plants is caused by factors that either restrict its absorption and translocation or impair its utilization in metabolic processes (Fontes and Cox 1998). Wide occurrence of iron chlorosis in plants even in the iron sufficient soils is a general problem observed in different parts of the country. Mostafa *et al.* (2011) reported increased yield and nutrient uptake in sesame (*Sesamum indicum* L.) due to application of sulphur and iron fertilizers.

Similarly sulphur is considered as important nutrient for its role in the production of amino acids cystein, and

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methionine as well as antioxidant glutathione (Jander and Joshi 2010). In wheat, sulphur deficiency resulted in reduced yield and inferior grain quality (Goodrood *et al.* 1989). Mustard (*Brassica juncea*) seed yield increase by application of sulphur, gypsum and ferrous sulphate (Rathore and Maliwal 1990). In chickpea, sulphur application (80 kg ha<sup>-1</sup>) seed yield (Singh and Ram 1990). Application of S containing fertilizer can result in soil acidification which may influence nutrient uptake (Havlin *et al.* 2007). In view of above, Fe and S are considered important nutrients for growth and yield improvement, however, there is need to standardize the optimum dose for different crops. Hence, this study was to investigate the effect of iron and sulfur application on growth, yield and yield components of soybean.

The study was carried out in sandy loam soil at Agricultural Research Farm of Banaras Hindu University, Varanasi located in the North Indian belt of semi arid to sub humid climate. Before conducting experiments soil was analyzed for the level of sulphate sulphur and available iron by the method of A.O.A.C. (1970) 8.2 and 2.9 ppm and found. Seeds of soybean (*Glycine max* L. Merr) var PK – 327 were sown in randomized block design which consisted of nine treatments and four replications. Treatments were applied in experimental plots considering 2.0 million kg soil ha<sup>-1</sup>. Ferrous sulphate and sodium sulphate were taken as source of iron and sulphur respectively. Observations for different morpho-physiological parameters were recorded at regular interval of 30 days up to 90 days after germination (DAG) i.e., at pre-flowering (30 DAG), flowering (60 DAG) and post-flowering (90 DAG) stage and at harvest. Leaf area was calculated as described by Puttaswamy *et al.* (1976). Absolute growth rate (AGR), relative growth rate (RGR), leaf relative growth rate (LRGR), leaf area duration (LAD) and net assimilation rate (NAR) was analysed according to formulae given by Radford (1967). Chlorophyll pigment were determined as per method of Vernon and Seely (1966). Total nitrogen in leaves was estimated by Nesslerers reagent method (Jackson, 1958). Seed protein contents were estimated by the method of Lowry *et al.* (1951). Data recorded were subjected to statistical analyses and recourse was taken to Fischer's analysis of variance (ANOVA) as suggested by Rangaswamy (2000).

Morpho-physiological attributes of plants are affected by a battery of variables, amongst them balanced nutrition is an important variable. Soybean is an annual crop species, and during the growth stages, remobilization of micronutrients occurs from root to shoot (leaves and stems), and finally to seeds. The rate of mobilization is affected by source-sink relationships in plant, environmental conditions, availability of nutrients and genetic characteristics of plant (Kobraee *et al.* 2011). Availability of nutrients might be improved by exogenous supplementation. Iron and sulphur play crucial role in plant metabolism. The requirement of iron in the development of normal photosynthetic activity in higher plants has long been recognized. Chloroplast have been shown to be accumulation site of the largest fraction of leaf iron and during period of iron stress the fine plastid structure has been observed to undergo deformations resulting in decrease in photochemical capacity of the plastid decreases (Shick *et al.* 2011). The ability to accumulate iron from rhizosphere is also dependent on the redox state of iron. Assimilated iron is re-oxidized at the xylem and transported to foliar tissues of dicotyledonous plants as Fe citrate (Conte and Walker 2010). The translocated Fe citrate appears to be reduced in the foliage to become biologically active.

The maximum length of roots was recorded in plants treated with combined application of iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil which was 12.97 cm as compared to 9.70 cm in control. Similarly, shoot height increased from 23.12 cm in control to 30.50 cm in plants treated with combined application of iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil. Number of leaves plant<sup>-1</sup>, chlorophyll content and leaf nitrogen content increased from 27.25 to 47.25, 0.19 to 1.42 mg g<sup>-1</sup> fresh wt. and 1.48 to 2.04% respectively in control and plants treated with combined application of iron @ 20 mg kg<sup>-1</sup> soil and sulphur @ 80 mg kg<sup>-1</sup> soil. An increase from 2.87 pods plant<sup>-1</sup> in control to 4.65 pods plant<sup>-1</sup> in plants treated with combined application of iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil was also recorded. Length of pods increased from 2.87 cm in control to 4.65 cm in plants treated with combined application of iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil. Moreover, 100 seed wt. increased from 9.40 g to 10.30 g and seed protein 36.60% to 38.55% in control and in plants treated with combined application of iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil respectively. As

compared to iron, sulphur was found to be more conducive for better root or shoot growth might be due to its role in increasing cambial activity (Mc Naught and During 1970).

The experimental soil was slightly alkaline and had high total but low available Fe. Present findings reveal that application of iron and sulphur individually as well as in combination improves height of shoot, length of root, number of leaves, chlorophyll content, leaf nitrogen content, number of pods plant<sup>-1</sup>, length of pods, 100 seed weight and seed protein content (Table 1). In conformity with the present findings, Christin *et al.* (2009) found that application of iron along with other nutrients increases growth and yield of sorghum and sunflower. There are two groups of well defined iron containing proteins; hemeprotein and iron sulphur protein (Sandman and Boger 1983). The most well known hemeproteins are the cytochromes (a<sub>1</sub>+a<sub>3</sub>, b<sub>559</sub>, b<sub>6</sub>, b and c) which contain a heme iron porphyrin complex as a prosthetic group. Cytochromes are constituents of the redox system in chloroplast and mitochondria and in the form of cytochrome oxidase, participate in the terminal step of the respiratory chain. The rest of the heme iron is found in nitrate reductase, nitrite reductase, sulphite reductase, catalase and peroxidase. Non heme iron proteins contain

about 19% of the total iron and are coordinated to the third group of cystine and / or inorganic sulphur the most prominent one is ferridoxin which act as an electron transmitter in a number of basic metabolic processes (Marschner 1986). The improved growth and yield of plants treated with iron individually or in combination with sulphur might be due to role of iron in these metabolic processes. Sulphur application with iron was found to be conducive in elimination of symptoms of chlorosis to the great extent, and used to correct iron chlorosis (Kalbasi *et al.* 1988). Chlorophyll is formed in higher plants from the precursor amino levulinic acid (ALA). In iron deficient plants, ferredoxin would be limiting and might affect chlorophyll biosynthesis (Miller *et al.* 1984). Sulphur fertilization helps to maintain active iron level in plants, which further takes part in metabolic process and improve growth and yield. Therefore, simultaneous availability of both the elements is essential for optimum growth, development and yield.

Increased yield may be correlated with increase in growth analyses parameters viz., AGR, RGR, LRGR, and LAD of treated plants as compared to control (Table 2). A significant increase in these growth attributes may be responsible for better yield. At pre-flowering stage (30-60 DAG) AGR increased from 0.2606 g day<sup>-1</sup> in

**Table 1.** Influence of iron and sulphur application on growth and yield of soybean.

Treatments	Root Length (cm)	Shoot Height (cm)	No. of leaves plant <sup>-1</sup>	Chlorophyll (mg.g <sup>-1</sup> fresh wt.)	Leaf nitrogen content (%)	No. of pods plant <sup>-1</sup>	Length of pods (cm)	100 seed wt. (g)	Seed Protein content (%)
Control	9.70	23.12	27.25	0.19	1.48	2.87	2.87	9.40	36.60
Fe 15	11.00	24.15	30.00	0.27	1.53	3.17	3.17	9.68	37.17
Fe 20	11.47	24.75	29.50	0.31	1.55	3.45	3.45	9.80	37.32
S 40	10.25	23.30	28.50	0.21	1.60	2.95	2.95	9.98	37.47
S 80	11.25	23.50	29.75	0.23	1.65	3.12	3.12	9.93	37.55
S 40 + Fe 15	11.38	26.90	33.25	0.40	1.75	3.67	3.67	10.05	38.20
S 80 + Fe 15	11.50	26.40	33.00	0.46	1.93	3.87	3.87	10.07	38.40
S 40 + Fe 20	12.15	28.90	39.25	0.66	1.88	4.07	4.07	10.20	38.35
S 80 + Fe 20	12.97	30.50	42.25	1.42	2.04	4.65	4.65	10.30	38.55
LSD 5%	0.52	3.59	5.42	NS	NS	0.07	0.08	0.61	0.08

\*NS = Non Significant

**Table 2.** Influence of iron and sulphur application on growth analyses of soybean.

Treatments	AGR (g day <sup>-1</sup> )			RGR (g g <sup>-1</sup> day <sup>-1</sup> )		NAR (g dm <sup>-2</sup> day <sup>-1</sup> )		LRGR (dm <sup>2</sup> dm <sup>-2</sup> day)		LAD (dm <sup>2</sup> day plant <sup>-1</sup> )	
	30-60 Days	60-90 Days	90 Days- Harvest	30-60 Days	60-90 Days	30-60 Days	60-90 Days	30-60 Days	60-90 Days	30-60 Days	60-90 Days
Control	0.2606	0.0358	0.0168	0.0697	0.0009	0.0164	0.0014	0.0211	0.005	0.515	0.7974
Fe 15	0.3855	0.0376	0.0183	0.0851	0.0021	0.0199	0.0036	0.0232	0.0078	0.993	1.5848
Fe 20	0.4152	0.0384	0.0189	0.0877	0.0025	0.0211	0.0042	0.0238	0.0085	0.998	1.6651
S 40	0.3650	0.0362	0.0172	0.0847	0.0014	0.0187	0.0022	0.0222	0.0061	0.608	0.9919
S 80	0.3793	0.0369	0.0178	0.0865	0.0019	0.0195	0.0025	0.0225	0.0065	0.688	1.1317
S 40 + Fe 15	0.4557	0.0416	0.0345	0.0926	0.0035	0.0221	0.0047	0.0259	0.0092	1.134	1.6882
S 80 + Fe 15	0.4864	0.0433	0.0349	0.0947	0.0039	0.0232	0.0051	0.0268	0.0096	1.471	1.8821
S 40 + Fe 20	0.5283	0.0512	0.0365	0.0967	0.0042	0.0254	0.0056	0.0279	0.0099	1.622	2.2800
S 80 + Fe 20	0.5446	0.0638	0.0388	0.0987	0.0049	0.0262	0.0065	0.0315	0.0102	2.116	2.4823
LSD 5%	0.018	0.012	0.011	0.005	0.0004	0.0007	0.0002	0.006	0.0003	0.005	0.013

control to 0.5446 g day<sup>-1</sup> in plants treated with a combined application of iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil. NAR represents the net production of dry matter in terms of unit leaf area. At 30-60 DAG stage NAR increased from 0.0082 g dm<sup>-2</sup>.day in control to 0.0164 g dm<sup>-2</sup> day in plants treated with iron @ 20 and sulphur @ 80 mg kg<sup>-1</sup> soil. Significant increase in LAD might be due to a longer retention of chlorophyll in treated plants. Improvement in the above parameters in treated plants indicates a greater efficiency for photosynthetic assimilation and partitioning towards sink development. This might be the reason of higher yield of treated plants. Therefore, application of iron along with sulphur (@ Fe 20 + S 80 mg kg<sup>-1</sup> soil) may be helpful for increasing quality and yield potential of soybean which is otherwise very low.

## REFERENCES

- A.O.A.C. (1970). Official methods of analysis of the association of official agricultural chemists.
- Caliskan, S., Ozkaya, I., Caliskan, M. E. and Arslan, M. (2008). The effects of nitrogen and iron fertilization on growth, yield and fertilizer use efficiency of soybean in a Mediterranean-type soil. *Field Crops Res.* **108**: 126-132.
- Christin, H., Petty, P., Khaled, Q., Burgado, S., Lawrence, C., and Kassem, M. A. (2009). Influence of iron, potassium, magnesium and nitrogen deficiencies on the growth and development of sorghum (*Sorghum bicolor* L.) and sunflower (*Helianthus annuus* L.) seedlings. *J. Biotech. Res.* **1**: 64-71.
- Conte, S.S. and Walker, E.L. (2011). Transporters contributing to iron trafficking in plants. *Mol. Plant.* **4**: 464-476.
- Dhillon, N.S. and Dev, G. (1982). Studies on sulphur nutrition of soyabean from three sulphate sources. *Soils Fert.* **45**: 2450.
- Fontes, R.L.F. and Cox, F.R. (1998). Iron deficiency and zinc toxicity in soybean grown in nutrient solution with different levels of sulfur. *J. Plant Nutr.* **2**: 1715-1722.
- Goodrood, L.K., Okhi, K. and Wilson, D.O. (1989). Sulphur (s) deficiencies in soft red winter wheat (*Triticum aestivum* L.). *J. Plant Nutr.* **12**: 1029-1039.
- Havlin, J.L., Beaton, J.D., Tisdale, S.L. and Nelson, W.L. (2007). Soil Fertility and Fertilizers, An Introduction to Nutrient Management, 7th edition, Pearson Education Inc. Singapore, p. 221.
- Heitholt, J.J., Sloan, J.J., MacKown, C.T. and Cabrera, R.I. (2003). Soybean growth on calcareous soil as affected by three iron sources. *J. Plant Nutr.* **26**: 935-948.

## INFLUENCE OF IRON AND SULPHUR ON SOYBEAN

- Imsande, J. (1998). Iron, sulfate, and chlorophyll deficiencies: A need for an integrative approach in plant physiology. *Physiol. Plant.* **103**: 139-144.
- Jackson, M.L. (1958). Soil chemical analysis, Prentice Hall of India Private Limited, New Delhi, pp. 134 - 204.
- Jander, G and Joshi, V. (2010). Recent progress in deciphering the biosynthesis of aspartate derived amino acids in plants. *Mol. Plant* **3**: 54 – 65.
- Kalbasi, M., Filsoof, F. and Rezai-Nejad, Y. (1988). Effect of sulphur treatments on yield and uptake of Fe, Zn and Mn by corn, sorghum and soyabeans. *J. Plant Nutr.* **11**: 1353-1360.
- Kobraee, S., NoorMohamadi, G., HeidariSharifabad, H., DarvishKajori, F. and Delkhosh, B. (2011). Influence of micronutrient fertilizer on soybean nutrient composition. *Indian J. Sci. Tech.* **4**: 763 – 769.
- Kochian, L.V. (2000). Molecular physiology of mineral nutrient acquisition, transport, and utilization. In: B.B. Buchanan, W. Gruissem and R.L. Jones (eds.), Biochemistry and molecular biology of plants, Am. Soc. of Plant Biology. Rockville, MD, pp. 1204 – 1249.
- Lawlor, D.W. (1995). The effects of water deficit on photosynthesis In: Smirnoff (ed.) Environment and plant metabolism, Bios. Sci. Publishers, pp. 129-160.
- Lowry, O.H., N.J. Rosebrogh, A.L. Farr and R.J. Randall (1951). Protein measurement with folin reagent. *J. Biol. Chem.* **193**: 265-275.
- Marschner, H. (1986). Mineral nutrition in higher plants. Academic Press, London, pp. 195-287.
- Mc Naught, J.K. and During, C. (1970). Relations between nutrient concentration in plant tissues and response of white clover to fertilizer on a grey podzol near west port. *N.Z.J. Agr. Res.* **13**: 567 - 590.
- Miller, G.W., Denny, A., Pushnik, J. and Yu. M.H. (1984). The transformation of delta-amino-levulinate, a precursor for chlorophyll in barley and the role of iron. *J. Plant Nutr.* **5**: 289-300.
- Mostafa, H., Galavi, M. and Hassani, M. (2011). Effect of sulfur and iron fertilizers on yield, yield components and nutrient uptake in sesame (*Sesamum indicum* L.) under water stress. *African J. Biotech.* **10**: 8816–8822.
- Puttaswamy, T., Gowda and K. Krishnamurthy (1976). Determination of leaf karea in pulses. *Curr. Res.* **5**: 48-49.
- Radford, P.J. (1967). Growth analysis formula- their use and abuse. *Crop Sci.* **7**: 171-175.
- Rangaswamy, R. (2000). A Textbook of Agricultural Statistics. New Age International Pvt. Ltd. New Delhi.
- Rathore, P.S. and Maliwal, P.L. (1990). Response of mustard to soil and foliar application of sulphur. *Haryana J. Agron.* **35**: 96-97.
- Rehm, G. and Albert, S. (2006). Micronutrients and production of hard red spring wheat. *Minnesota Crop News.* pp. 1-3.
- Sandmann, G. and Boger, P. (1983). The enzymomatological function of heavy metals and their role in electron transfer process of plants. In: A . Lauchli and R. L. Bieleski (eds.), Encyclopedia of Plant Physiology, New series, 15A pp. 563-596.
- Shick, J.M., Iglie, K., Wells, M.L., Trick, C.G., Doyle, J. and Dunlap, W.C. (2011). Responses to iron limitation in two colonies of *Stylophora pistillata* exposed to high temperature: Implications for coral bleaching. *Limnol. Oceanogr.* **56**: 813–828.
- Singh, P.N. and Ram, H. (1990). Effect of phosphorus and sulphur application on protein and amino acid contents in chickpea. *Indian J. Pulse Res.* **3**: 36-39.
- Vernon, L. P. and Seely, G.R. (1966). The “chlorophylls”. Academic Press, New York.