

## OPTIMIZATION OF SULPHUR REQUIREMENT TO SESAME (*SESAMUM INDICUM* L.) GENOTYPES USING TRACER TECHNIQUES

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### Abstract

Four sesame genotypes *viz.*, T6, SM4, SM7 and SM8 were verified with three levels of sulphur 10, 20 and 40 kg/ha including control. Factorial experiment was laid out in a completely randomized design with four replications. Biochemical parameters such as chlorophyll, nitrate reductase, amino acid and total sugar in leaves, total dry mass production, total nitrogen accumulation in seeds and seed yield increased with sulphur application up to 20 kg S/ha. Application of 40 kg S/ha had no significant benefit on biochemical parameters, dry matter and seed yield of sesame though <sup>35</sup>S uptake in seeds increased with increasing sulphur levels till 40 kg/ha. The biochemical traits and nutrients uptake were superior in SM4 than other genotypes which resulted in the highest seed yield of SM4. Among the sulphur levels, 20 kg S/ha showed the maximum seed yield in all the genotypes. Therefore, sulphur @ 20 kg/ha can be recommended for getting maximum productivity of sesame genotypes.

### Introduction

Among the oil crops, sesame (*Sesamum indicum* L.) has the highest oil content of 46 - 64% (Raja *et al.* 2007). In oilseeds, sulphur plays a significant role in the quality and development of seeds. Therefore, crops of oilseeds require a higher quantity of sulphur for proper growth and development for higher yields (Salwa *et al.* 2010). It is reported that sulphur plays an important role in the primary and secondary plant metabolism as a component of proteins, glucosinolates and other compounds that related to several parameters determining the nutritive quality of crops (Ceccotti 1996, Jamal *et al.* 2010). The response of oilseeds to sulphur is increasing due to increasing of cropping intensity (Ghosh *et al.* 2002). Recently, the soils of Indian sub-continent have been reported to be S-deficient and crop response also been reported to S-application (Ghosh *et al.* 2002, Raja *et al.* 2007). However, nitrogen and sulphur metabolism are linked to each other. S-application significantly increased the uptake of N in straw and grain (Badruddin 1999, Fazli *et al.* 2008) thereby increased grain yield. Further, S-deficiency cause decrease in nitrate reductase activity and in the accumulation of chlorophyll, soluble protein, amino acid and sugar (Tandon 1986, Badruddin 1999, Jamal *et al.* 2006, Jamal *et al.* 2009). Thus the synergistic relationship of N and S in plant metabolism and the maximum yield response to these elements is achieved when the supply of them are balanced in oilseed crops (Jaggi *et al.* 1977, Badruddin 1999, Fazli *et al.* 2010). Optimum nutrition, among other agro-techniques is very important for realizing full yield potential and the role of sulfur is next only to nitrogen in the nutrition of this crop. Little effort is made to reclaim optimum S requirement for the growth and yield of sesame using tracer techniques. Hence this study was attempted to assess the importance of sulphur in biochemical activities, nitrogen and sulphur uptake and yield of sesame genotypes.

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## Materials and Methods

Pot experiment was conducted in the glasshouse at Bangladesh Institute of Nuclear Agriculture, Mymensingh, Bangladesh in 2007 to 2008. Earthen pots were lined with polyethylene bags. Each pot was filled with 10 kg sand and the pots were arranged following two factors completely randomized design with four replications. Four variety/mutant lines of sesame *viz.*, T-6 (mother), SM4, SM7 and SM8 were used in this experiment. Ten seeds of each genotype were sown on 05 and 10 March in the year of 2007 and 2008, respectively. After emergence, two seedlings were allowed to grow and the remaining seedlings were removed after seven days of germination. Modified Letcombe nutrient solution (Hewitt 1966) containing @ 1 mM  $\text{KH}_2\text{PO}_4$ , 0.092 mM  $\text{H}_3\text{BO}_3$ , 0.0967 mM Fe-EDTA, 0.0025 mM  $\text{CuCl}_2$ , 0.00016 mM  $\text{NH}_4\text{MO}_7\text{O}_2$ , 0.0928 mM  $\text{MnCl}_2$  and 0.0077 mM  $\text{ZnCl}_2$  were applied for P, B, Fe, Cu, Mo, Mn and Zn, respectively. To maintain 30 kg N/ha  $\text{KNO}_3$ , Ca  $(\text{NO}_3)_2$  and  $\text{NaNO}_3$  @ 2.5, 1 and 2 mM, respectively (jointly equal to 30 kg N/ha) were applied each pot as N-source in every week up to maturity.

Sulphur @ 0.15, 0.3 and 0.6 mM for first 6 weeks and 0.225, 0.45 and 0.9 mM for next 6 weeks from  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  were applied to maintain 10, 20, and 40 kg S/ha, respectively. In control (-S) pots, magnesium @ 2 mM from  $\text{MgCl}_2$  were applied throughout the growing period. Biochemical parameters such as chlorophyll, nitrate reductase, amino acid and total sugar in leaves were determined at reproductive stage. Chlorophyll, nitrate reductase activity (NR), amino acid and total sugar were determined following the method of Yoshida *et al.* (1976), Stewart and Orebanjo (1979), Badruddin (2005) and Dubois *et al.* (1956), respectively. Grain nitrogen was estimated by micro-Kjeldahl method (AOAC 1980) and converted to total nitrogen per plant. For S-uptake and transport study, isotope of S was applied in the pot at the above mentioned rates during silique filling stage. No  $^{35}\text{S}$  applied in the control pots. For analyses,  $^{35}\text{S}$  extracted by boiling the samples in distilled water in a water bath for 15 minutes and the extract was decanted. The procedure was repeated twice and the radioactivity was counted for 1 minute in a scintillation counter (model LSC 2, NE Technology UK). Biochemical parameters and  $^{35}\text{S}$  isotope uptake were studied only in 2007. The collected data were subjected to statistical analyses as per the design used.

## Results and Discussion

The effect of different levels of sulphur (S) on metabolic activity, growth and yield of sesame was significant (Table 1). Results revealed that chlorophyll, nitrate reductase (NR), amino acid and total sugar were increased with increasing sulphur levels till 20 kg/ha followed by a decline. Similar results were also observed in total dry mass (TDM) and seed yield in both the years (Table 1). The highest TDM and seed yield/plant was recorded in 20 kg S/ha might be due to maximum metabolic activity at this level resulting increased TDM thereby seed yield. The increase in NR activity and chlorophyll content due to S application might be resulted to the availability of S in the plant medium that help in producing S-containing amino acid. As a result, synthesis of NR enzyme increased and it enhanced the reduction of  $\text{NO}_3^-$  to reduce N ultimately resulting higher accumulation of sugar thereby increased TDM and seed yield of sesame. Application of S increased TDM and seed yield in oil seed crops as reported by many workers (Nagwari *et al.* 2001, Amudha *et al.* 2005, Raja *et al.* 2007). Furthermore, it is reported that optimum S increased nutrient uptake which help maximum metabolic activity in plants (Badruddin 1999). In the present study, the maximum metabolic activity was observed in 20 kg S/ha and further increment of S level decreased metabolic activity indicating 20 kg S/ha was the optimum dose for maximum growth and development of sesame plants as well as yield. It indicates that increase in biochemical substances is somehow related to sulphur application and that might lead increase in TDM as well

as seed yield of sulphur treated plants over control. It was expected that plants will not thrive in the control at entire growth period but surprisingly plants survived and thrived well up to maturity. This might be due to contamination in the nutrient solution or may be  $\text{SO}_2$  uptake from air as a scavenger of sulphur. It was reported that plants may also take sulphur as  $\text{SO}_2$  from the air through stomata (Rennenberg *et al.* 1979).

**Table 1. Effect of sulphur on metabolic activities and yields of sesame genotypes.**

| Sulphur levels (kg/ha) | Chlorophyll (mg/g fw) | Nitrate reductase ( $\mu\text{mol NO}_2^-/\text{g fw}$ ) | Amino acid (mg/g fw) | Total sugar (mg/g fw) | Total dry mass/plant (g) |        | Seed yield/plant (g) |        |
|------------------------|-----------------------|--|----------------------|-----------------------|--------------------------|--------|----------------------|--------|
|                        |                       |  |                      |                       | 2009                     | 2010   | 2009                 | 2010   |
| 0                      | 0.71 c                | 1.11 c   | 2.89 c               | 16.51 c               | 3.08 d                   | 3.28 d | 0.26 d               | 0.14 d |
| 10                     | 2.71 b                | 2.92 b   | 4.39 b               | 40.23 b               | 6.50 b                   | 9.22 b | 1.16 c               | 1.83 c |
| 20                     | 3.03 a                | 3.31 a   | 5.27 a               | 50.47 a               | 8.24 a                   | 11.3 a | 2.00 a               | 2.78 a |
| 40                     | 2.45 b                | 3.17 ab  | 4.64 b               | 42.15 b               | 6.06 c                   | 8.71 c | 1.27 b               | 1.94 b |
| F-test                 | **                    | **   | **                   | **                    | **                       | **     | **                   | **     |
| CV (%)                 | 9.51                  | 5.11   | 8.02                 | 5.21                  | 4.87                     | 5.52   | 6.42                 | 8.30   |

Same letter(s) in a column did not differ significantly at  $p \leq 0.05$  by DMRT; \*\* = Significant at 1% level of probability.

Isotopic sulfur uptake in seeds by the sesame genotypes was increased with increasing sulphur levels (Fig. 1). However, there was no significant difference in  $^{35}\text{S}$  uptake by seed between 20 and 40 kg S/ha. Similarly, application of sulphur @ 20 kg/ha had the highest accumulation of N in seeds (280.2 mg/plant) followed by 40 kg S/ha (189.0 mg/plant) (Fig. 1). This result indicated that application of 20 kg S/ha was optimum dose for sesame cultivation. The lowest accumulation of

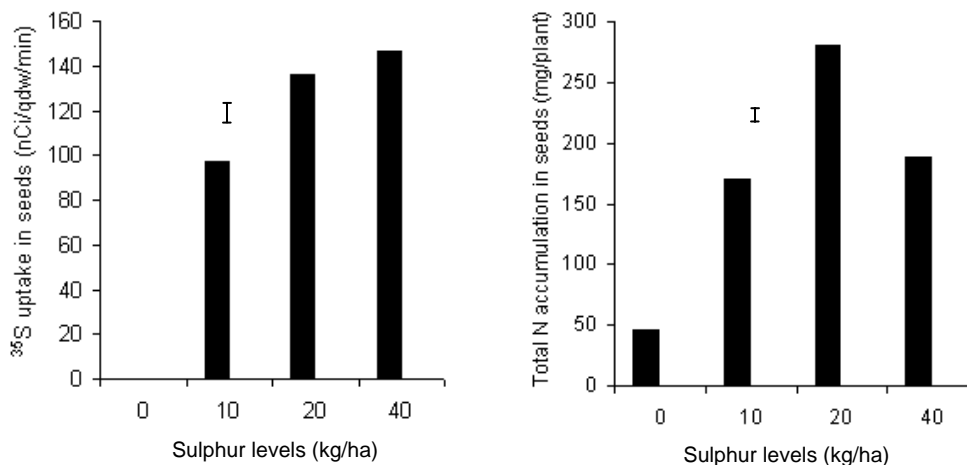


Fig. 1. Effects of sulphur levels on sulphur and nitrogen uptake in grains of sesame genotypes. Vertical bar represents LSD (0.05).

total N in seeds (46.5 mg/plant) was found in the control. Sulphur and nitrogen are linked to each other in metabolic activity. Control showed the lowest N-metabolism in the seed because no S was supplied there. Judicious application of S significantly increased the uptake of N in straw and grain as reported by many workers (Badrudin 1999, Fazli *et al.* 2008). Furthermore, it was reported that optimum S increased nutrient uptake which helped maximum metabolic activity in

plants (Badruddin 1999). In the present study, the maximum metabolic activity was observed in 20 kg S/ha and further increment of S level decreased metabolic activity indicating 20 kg S/ha was optimum dose for maximum growth and development of sesame.

Biochemical parameters,  $^{35}\text{S}$  uptake and total N accumulation in seeds, TDM production and seed yield in sesame except total sugar in leaves were also significantly influenced by different genotypes (Table 2, Fig. 2). The genotype SM4 produced the highest TDM as well as seed yield/plant due to superiority in biochemical activity in leaves,  $^{35}\text{S}$  uptake and total N-accumulation in seeds. In contrast, the genotype T6 showed the lowest seed yield though it performed lower to medium biochemical activities. Badruddin *et al.* (2002) reported that uptake of sulphur was higher in high yielding genotypes than those of low yielding ones in sunflower which is in agreement with the present findings. The performance of genotypes was in the following order: SM4>SM7>SM8>T6.

**Table 2. Genotypic effect on metabolic activities and yields of sesame.**

| Geno-<br>types | Chloro-<br>phyll<br>(mg/g fw) | Nitrate<br>reductase ( $\mu\text{mol}$<br>$\text{NO}_2^-/\text{g fw}$ ) | Amino acid<br>(mg/g fw) | Total sugar<br>(mg/g fw) | Total dry mass/<br>plant (g) |        | Seed yield/<br>plant (g) |        |
|----------------|-------------------------------|---|-------------------------|--------------------------|------------------------------|--------|--------------------------|--------|
|                |                               |   |                         |                          | 2009                         | 2010   | 2009                     | 2010   |
| T6             | 2.38 b                        | 2.78 ab   | 3.87 c                  | 36.21                    | 4.99 c                       | 7.00 c | 0.77 d                   | 1.04 d |
| SM4            | 3.02 a                        | 2.95 a  | 5.04 a                  | 38.23                    | 8.05 a                       | 10.3 a | 1.72 a                   | 2.47 a |
| SM7            | 1.97 c                        | 2.53 b  | 4.00 b                  | 36.80                    | 5.96 b                       | 7.12 c | 1.16 b                   | 1.73 b |
| SM8            | 1.38 d                        | 1.95 c  | 4.16 b                  | 37.12                    | 4.88 c                       | 8.01 b | 1.05 c                   | 1.45 c |
| F-test         | **                            | **  | **                      | NS                       | **                           | **     | **                       | **     |
| CV (%)         | 9.51                          | 5.11  | 8.02                    | 5.21                     | 5.55                         | 5.22   | 6.42                     | 8.30   |

Same letter(s) in a column did not differ significantly at  $p \leq 0.05$  by DMRT; \*\* = Significant at 1% level of probability; NS = Not significant.

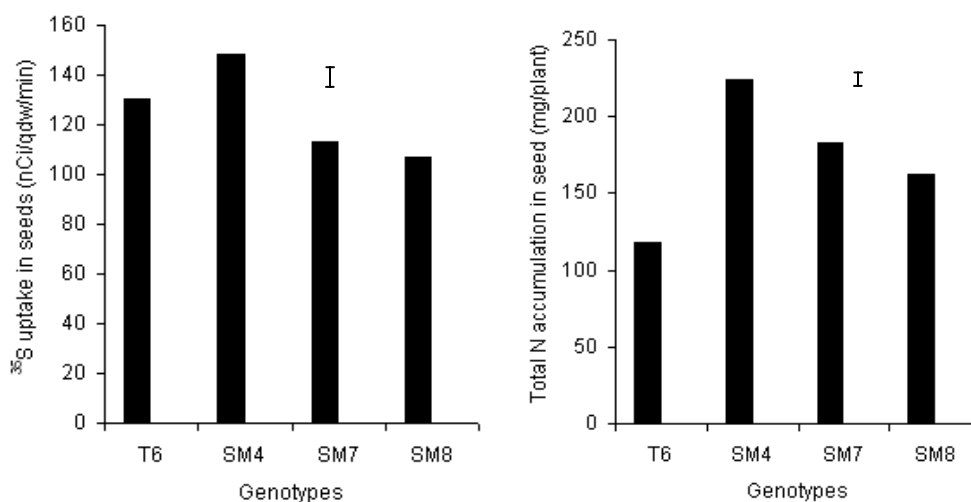


Fig. 2. Genotypic performance on  $^{35}\text{S}$  uptake and total nitrogen accumulation in seeds of sesame. Vertical bar represents LSD (0.05).

Interaction effect between genotypes and sulphur levels showed statistically significant differences in  $^{35}\text{S}$  uptake, total N accumulation in seeds, TDM production and seed yield of sesame (Table 3). Sulphur utilization by the genotypes was increased with the increase of S levels

except the genotype T6. In T6, the highest sulphur uptake was observed in 20 kg S/ha. The highest total nitrogen accumulation by the seeds was found in 20 kg S/ha in all the genotypes except the genotype SM8. In SM8, the maximum total nitrogen accumulation by the seeds was observed at 40 kg S/ha. The lowest nitrogen uptake by the seeds was found in control where no sulphur was applied for all the genotypes. The highest total nitrogen accumulation (385 mg/plant), total dry mass production (11.6 and 14.3 g/plant for 2007 and 2008, respectively) and seed yield (3.09

**Table 3. Interaction effect of genotypes and sulphur levels on  $^{35}\text{S}$  uptake and total nitrogen accumulation in seeds, total dry mass and seed yield in sesame.**

| Interaction |                 | $^{35}\text{S}$ uptake in seeds<br>( $\mu\text{Ci/gdw/min}$ ) | Total nitrogen accumulation in seeds<br>(mg/plant) | Total dry mass/plant (g) |         | Seed yield/plant (g) |         |
|-------------|-----------------|---|--|--------------------------|---------|----------------------|---------|
| Genotype    | S level (kg/ha) |   |  | 2009                     | 2010    | 2009                 | 2010    |
| T6          | 0               | 0   | 55 g   | 2.87 i                   | 3.20 hi | 0.28 j               | 0.10 h  |
|             | 10              | 108.0 f   | 100 f  | 4.92 g                   | 7.18 g  | 0.77 hi              | 1.13 g  |
|             | 20              | 148.5 c   | 200 d  | 6.95 d                   | 10.4 cd | 1.28 f               | 1.91 de |
|             | 40              | 135.2 d   | 115 f  | 5.23 fg                  | 7.17 g  | 0.73 i               | 1.00 g  |
| SM4         | 0               | 0   | 50 g   | 4.01 h                   | 3.81 h  | 0.24 jk              | 0.14 h  |
|             | 10              | 118.5 e   | 275 d  | 9.41 b                   | 12.4 b  | 1.61 d               | 2.66 c  |
|             | 20              | 158.0 b   | 385 a  | 11.6 a                   | 14.3 a  | 3.09 a               | 4.16 a  |
|             | 40              | 168.5 a   | 245 c  | 7.18 d                   | 10.8 c  | 1.93 c               | 2.93 b  |
| SM7         | 0               | 0   | 48 g   | 3.12 i                   | 2.42 i  | 0.34 j               | 0.20 h  |
|             | 10              | 80.0 g  | 200 d  | 6.05 e                   | 8.43 f  | 1.22 f               | 1.83 ef |
|             | 20              | 120.0 e   | 330 b  | 7.02 d                   | 9.93 de | 2.24 b               | 3.00 b  |
|             | 40              | 140.1 cd  | 153 e  | 7.65 c                   | 7.28 g  | 0.85 h               | 1.90 de |
| SM8         | 0               | 0   | 33 g   | 2.32 j                   | 3.29 hi | 0.16 k               | 0.10 h  |
|             | 10              | 83.1 g  | 165 e  | 5.62 ef                  | 8.88 f  | 1.05 g               | 1.68 f  |
|             | 20              | 117.2 e   | 206 d  | 7.41 cd                  | 10.3 cd | 1.40 e               | 2.06 d  |
|             | 40              | 120.0 e   | 243 c  | 4.18 h                   | 9.56 e  | 1.59 d               | 1.96 de |
| F-test      |                 | **  | **   | **                       | **      | **                   | **      |
| CV (%)      |                 | 6.49  | 8.38   | 4.87                     | 5.52    | 6.42                 | 8.30    |

Same letter(s) in a column did not differ significantly at  $p \leq 0.05$  by DMRT; \*\* = Significant at 1% level of probability.

and 4.16 g/plant for 2007 and 2008, respectively) was observed in SM4 with 20 kg S/ha. The lowest value of the above mentioned parameters was observed in SM8 with 0 kg S/ha. The increase in seed yield with 20 kg S/ha was the highest in SM4. Thus, SM4 genotype was more responsive to sulfur than other genotypes. The seed yield was the highest at 20 kg S/ha with any genotypes was due to increase uptake of S and N accumulation that stimulated biochemical activity thereby increased TDM. The lowest seed yield was observed in control plants with any genotype due to lack of sulphur. Furthermore, application of 40 kg S/ha with any genotype decreased total nitrogen accumulation in seeds due to lower biochemical activity and resulting lower seed yield than 20 kg S/ha indicating 40 kg S/ha may be toxic for plant growth and development in sesame. Many authors reported that under field condition, the optimum sulphur dose for sesame cultivation was 30 - 60 kg/ha depending on the soil and cultivation techniques (Tandon 1986, Ghosh *et al.* 2002, Nagwari *et al.* 2001, Amudha *et al.* 2005, Raja *et al.* 2007, Jamal *et al.* 2010). The present experiment was conducted in sand culture and nutrients were

applied frequently at 7 days interval up to maturity. As a result, nutrients use efficiency by the plants may be near 70 - 80% under sand culture. On the other hand, under field condition, sulphur is applied as gypsum and its use efficiency range 20 - 40%, depending on land and cultural condition (Jaggi and Sharma 1999). Singh and Saha (1995) reported that 40 kg S/ha was optimum dose for most of the crops under field condition. Therefore, it can be summarized that sulfur @ 20 kg/ha is the optimum dose for sufficient plant growth and development of sesame.

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