eISSN: 09748369, www.biolmedonline.com

Nutritional significance of sulphur in pulse cropping system

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Abstract

Sulphur is a part of every living cell and is a constituent of two of the 21 amino acids which form proteins. Of all the macronutrients, sulphur is perhaps the nutrient which has attracted the most attention in soil science and plant nutrition due to its potential defensive characteristics to pests, good nutritive potentiality to crops and its relative immobility in the soil-plant system. The benefits from sulphur fertilisation of crops can be traced to its role in protein development, to improvement of nitrogen use, etc. However, the availability of sulphur needed for profitable crop production continues to decline. This review highlights the prominent role of microbes in sulphur availability to crop plants as well as includes the mechanism of its uptake, translocation and assimilation. Moreover, it provides new insights leading us to revisit the hypothesis of sulphur significance in pulse cropping and regulatory mechanisms in sulphur assimilation.

Keywords: Pyrite; ATP; sulphurylase; 5-adenyl-sulphate; phytochelatine; glutathione; *Rhizobium*; *Thiobacillus*; pulse; chemical oxidation.

Abbreviations: H_2SO_4, Sulphuric acid; BGA, Blue green algae; APS, Adenosine-5-phosphosulfate; OAS; O-acetyl-serine; Ser, Serine; Cys, Cysteine; SAM, S-adenosyl-methionine; PAPS, 3-Phospho-adenosine-5-phosphate.

Introduction

Sulphur represents the ninth and least abundant essential macronutrient in plants, preceded by C, O, H, N, K, Ca, Mg and P. The dry matter of sulphur in plants is only about one-fifteen of that of nitrogen. Both sulphur and nitrogen absorption from soil through roots is necessary to be assimilated into organic metabolites. Sulphur plays various critical roles in catalytic or electrochemical function of biomolecules in the cells (Saito, 2004). Among the macronutrient N, K, Ca are non toxic to an extent at low to moderate concentrations while P and S seems to be more or less toxic to plants at higher concentration (Table 1 and 2) (Goldbold and Huttermann, 1985; Niess, 1999).

Recently observed lower sulphur emission to the atmosphere decreased the amount of sulphur in soil and caused worse sulphur nutrition of crop plant. It has long been known that in regions where sulphur deficient soil occur, legumes specially pulses are particularly responsive to sulphur containing fertilizers and that elementary sulphur or sulphates increase the percentage nitrogen as well as yield on such deficient soils. However, until 15 years ago there was little concern for sulphur deficiency, even though the ability of soil to retain and to release sulphur to crops is small and the input of high analysis through sulphur containing fertilizers increased and the SO_2 emission from industrial sources were reduced. Now areas of sulphur deficiency are becoming widespread throughout the world (Irwin *et al.*, 2002; Scherer, 2001). Sulphur is one of the limiting plant nutrients threatening the sustainability of crop production in semi arid tropical regions of India cover 73 million ha of vertisols and associated soils (Kanwar, 1988; Rao and Ganeshamurthy, 1994). Sulphur as a fertilizer or as a constituent of other fertilizers is generally not applied by farmers. As a result, large areas of S deficiency are reported from this agro-ecological region (Ganeshmurthy and Saha, 1999).

Understanding the role of sulphur in pulses growth is important from the point of view that the deficiency of the sulphur containing amino acids cysteine, cystine and methionine may limit the nutritional value of food and feed (Sexton *et al.*, 1998). Studies with medicagoativa indicate that with suboptimum sulphur supply, the mole percent of both amino acids significantly decreased (DeBoer and Duka, 1982), resulting in lower protein concentration (Rendig *et al.,* 1976), while non-protein N is accumulated. Also in *Pisum sativum*, sulphur deficiency resulted in a decreased synthesis of sulphur- containing storage protein albumin and legumin. However, according to Sexton *et al.* (1998) the protein quality of *glycine max* can be enhanced by increasing the concentration of sulphur-containing amino acids.

Sulphur is found in amino acids (Cys and Met), oligopeptides (glutathione and phytochelatins), vitamins and cofactors (biotin, thiamine, CoA and S-adenosyl-Met), and a

variety of secondary products (Leustek, 2000). Sulphur containing amino acids and stress response-related compounds, such as GSH, are derived from reduction of root-absorbed SO_4^{2-}. SO_4^{2-} distribution in cell compartments necessitates specific transport systems. The low-affinity SO_4^{2-} transporters SULTR4;1 and SULTR4; 2 have been localized to the vacuolar membrane, where they may facilitate SO_4^{2-} efflux from the vacuole. The study of Zuber *et al.* (2010) revealed a role for SULTR4; 1 in determining SO_4^{2-} content of mature *Arabidopsis* seeds. Moreover, the adaptive response of sultr4; 1 mutant seeds as revealed by proteomics suggests a function of SULTR4; 1 in redox homeostasis, a mechanism that has to be tightly controlled during development of orthodox seeds. The thiol (sulfhydryl) group of Cys in proteins takes the job of maintaining protein structure by forming disulfide bonds between two Cys residues via oxidation. The thiol of Cys and GSH in often involved in redox cycle by two thiol ↔ di-sulphide conversions. This interchange is versatile for redox control and mitigation against oxidative stress in nearly all plants including pulses (Leustek and Saito, 1999).

GSH contents increased in response to NaCl stress in leaves but not in roots, the primary site of salt exposure. The increasing leaf GSH concentrations correlated with stress-induced decreases in transpiration and net CO_2 assimilation rates at light saturation. Enhanced rates of photorespiration could also be involved in preventing ROS formation in chloroplasts and, thus, in protecting PS II from damage. Accumulation of Gly and Ser in leaves indeed indicates increasing rates of photorespiration. Since Ser and Gly are both immediate precursors of GSH that can limit GSH synthesis (Herschbach *et al.*, 2010). The nucleophilicity of the thiol group and in particular GSH plays a role in detoxification of xenobiotics by direct conjugation with –SH and mediated by GSH-transferase. Phytochelatins (a polymerised version of GSH) are involved in detoxification of heavy metals by serving as chelating secondary ligands through thiol groups. The role of sulphur along with other macronutrients like nitrogen, phosphorus, sulphur and calcium and micronutrients like zinc, iron, manganese and silicon has play a role in decreasing Cd uptake and accumulation in crop plants (Sarwar *et al.*, 2010). Excessive S supply may result in loss of rice yield, but it could effectively reduce Cd accumulation in brown rice exposed to Cd contaminated soils (Fan *et al.*, 2010). Results of Zhang *et al.* (2010) suggest that H_2S could increase antioxidant capability in wheat seeds leading to the alleviation of Al^{3+} stress. Sulphur containing secondary products often have a characteristic smell and are regarded only as defensive compound against herbivores and pathogenic micro-organisms but also as signalling molecules for fundamental cellular functions (Matsubsyashi *et al.*, 2002).

Sulphur transformation in soil mediated by microbes

The dominant form of sulphur taken up by plants and soil microbes is sulphate (SO_4^{2-}). Biological oxidation of H_2S to SO_4^{2-} is one of the major reactions of global sulphur-cycle. Reduced inorganic sulphur are exclusively oxidised by prokaryotes and SO_4^{2-} is the major oxidation product. The sulphur may originate from the weathering of soil minerals, from the atmosphere and from originally bound sulphur. Transformation of elemental sulphur to SO_4^{2-} form was necessary for sulphur to be available for crop uptake (Singh, 1988). The oxidation was faster in coarse textured soils and was completed in 3-4 weeks. Sulphur transformations in soil are considered to result primarily from microbial activity which involves process of mineralization, immobilization, oxidation and reduction (Kapoor and Mishra, 1989). Reduced sulphur formed by assimilatory SO_4^{2-} reduction is cell-constituents and are therefore protected within the cells against reaction with oxygen. The reduction of SO_4^{2-} to H_2S is mediated by anaerobic sulphate reducing bacteria. SO_4^{2-} reduction is a major component of the sulphur cycle in soils exposed to waterlogging condition or provides flooding especially where readily decomposable plant residues are present.

The biogeochemical transformation of sulphur has been formulated by Vernadski (1927). In the magnetic crust of our planet, sulphur is always present in its reduced form as sulphide of metals. Oxidation of the magnetic metal sulphides reaching the crust settling in ore deposits preceded with the participation of *Thiobacillus ferrooxidans* into the oceans. In arid regions, lagoons or inland seas, they may form gypsum deposits. In waterlogged soils, SO_4^{2-} are reduced to H_2S by *Disulphovibrio de*sulphur*icans* (Shooner and Tyagi, 1995).

The transfer of sulphur between the inorganic and organic pool is entirely caused by the activity of the soil microbial biomass, which has two greatest potential for both mineralization and for subsequent transformations of the oxidation state of sulphur. Thiobacilli bacteria play an important role in sulphur oxidation in soil. Sulphur

oxidation is the most important step of sulphur cycle which improves soil fertility. It results in the formation of sulphate which can be used while acidity produced by oxidation helps to solubilise plant nutrients and improves alkali soils (Wainwright, 1984; Sokolova *et al.*, 1968; Chapman, 1990).

Microbial production of sulphate

Starkey (1934) reported that sulphate production from elemental sulphur in sulphur media ranged from 31.7 mg per 100 cc. to 118.9 mg per 100 cc. SO_4^{2-} sulphur over the range of 100-200 µg per ml with a precision of 10 µg was obtained on the oxidation of elemental sulphur by *Thiobacillus thiooxidans* (Volger, 1951).

In an experiment involving the biological oxidation of pyrite conducted in mine spoil, Pitchel and Dick (1991) noticed concentration of SO_4^{2-} was found to be 81 m. Mol kg^{-1} (at controlled atmospheric conditions) spoil after 28 days of incubation.

Symptoms of pulse crops under sulphur stress

Adequate supplies of all essential plant nutrients are essential for high crop yield and profits. Since sulphur is immobile or less mobile than N in the plants and does not readily move from old to new growth, leads clorosis of younger leaves and at later stages, leaves show necrotic symptoms and die (Singh *et al.*, 1994). Sulphur concentration in most plants should range from about 0.2 to 0.5%. When Sulphur is deficient, nitrate-nitrogen is accumulating. Plants suffering from severe S deficiency show characteristic symptoms, leads to reduction in growth rate of the plant and generally the growth of shoots were more affected than that of roots and plants showing yellowing, small and spindle with short slender stalks (Nateson *et al.*, 1985). The classical symptom was the yellowing of younger leaves, while old leaves remained green (Tandon, 1991). Nodule formation is affected and N_2 fixation was reduced in leaves (Joseph *et al.*, 1994).

Reliable early diagnosis of Sulphur deficiency is required in order to minimise reductions in yield and quality. Diagnostic plant indicators such as total S concentration, SO_4^{2-} concentration, N/S ratio, SO_4^{2-} to total S and glutathionine concentration have been used in many crops to predict sulphur deficiency (Zho *et al.*, 1996; Spencer *et al.*, 1980). Sulphur-induced resistance is also known as sulphur-enhanced defense (SIR/SED). Sufficient SO_4^{2-} supply resulted in a suppressed and delayed symptom development and diminished virus accumulation over a period of time (14 days) after inoculation as compared with -S conditions. SO_4^{2-} withdrawal from the soil was accelerated at the beginning of the infection, whereas it declined in the long term, leading to an accumulation of sulphur in the soil of plants grown with SO_4^{2-}. Results of Holler *et al.* (2010) demonstrates a link between the activation of cysteine and glutathione metabolism and the induction of SIR/SED during a compatible plant-virus interaction in tobacco plants, indicating a general mechanism behind SIR/SED.

Importance of sulphur in pulse cropping

Sulphur is a controversial chemical element (Ostowska, 2008). It causes the acidification of atmosphere on one hand while a necessary component of amino acids e.g. methionine and cysteine is a requisite for protein synthesis necessary for biomass and growth on other hand. It classified as secondary nutrient, necessary for all plants and is indispensable for the growth and metabolism (Vidyalakshmi *et al.*, 2009). The concentration was found to be highest in oil seed (1.1-1.7%), intermediate in pulses (0.24-0.32%) and the lowest (0.12-0.20%) in cereals (Singh, 2001). It has a number of oxidising functions in plant nutrition and a constituent of Fe-S proteins called Ferridoxin, responsible for transfer of electrons during the first phase of photosynthesis (light dependent) reactions (Randall, 1988; Goswami, 1988; Petrovic and Kastori, 1994; Marschner, 1995). Sulphur has a profound effect on creating assimilation area absorbing PAR and as a consequence on yield of crops.

Total number of nodules and active nodules significantly increased with application of S up to 20 kg S/ha (Ganeshamurthy and Readly, 2000). Formation of nodule in black gram was increased due to sulphur application in black gram (Khandkar *et al.*, 1985) and is involved in the formation of nitrogenase enzyme known to promote nitrogen fixation in legumes (Saraf, 1988; Scherer *et al.*, 2006). It is involved in the formation of chlorophyll (Mehta *et al.*, 1979; Jamal *et al.*, 2006), and associated with production of crops of superior nutritional and market quality (Hanesklaus and Schnug, 1992; Sexton *et al.*, 1998). Sulphur application increased the total chlorophyll content of green gram and pea (Poorani, 1992; Spencer *et al.*, 1990). Sulphur plays dominant role in improving the quality of pulses (Paricha *et al.*, 1993). Varin *et al.* (2010) examined whether the effect of sulphate addition on N fixation resulted from a stimulation of host plant growth, a specific effect of Sulphur on nodulation, or a specific effect of Sulphur on

nodule metabolism. The application of SO_4^{2-} increased whole plant dry mass, root length, and nodule biomass, expressed on a root-length basis. N uptake proved less sensitive than N_2 fixation to the effects of S-deficiency, and decreased as a consequence of the lower root length observed in S-deficient plants. N_2 fixation was drastically reduced in S-deficient plants as a consequence of a low nodule development, but also due to low nitrogenase and leghaemoglobin production. This effect is likely to be due to down-regulation by a N-feedback mechanism, as, under severe S-deficiency, the high concentration of whole plant N and the accumulation of N-rich amino acids (such as asparagine) indicated that the assimilation of N exceeded the amount required for plant growth.

Application of sulphur at the rate of 40kg/ha enhanced plant height, branches, pod per plant and 1000 gram weight in green gram (Sharma and Singh, 1979). Application of gypsum at the rate of 60Kg/ha produced significantly higher pod length, seed/pod to 1000 seed weight in black gram (Singh and Aggarwal, 1998), they also reported that application of 30 kg sulphur/ha significantly increased the yield contributing characters and yield of black gram. Application of 20 S kg/ha also significantly increased the dry matters yield of soya bean (Ganeshamurthy and Reddy, 2000). Although the dry weight content of the nodules at higher levels of applied S showed a tendency to increase but this was not significantly beyond 20 kg S/ha (Ganeshhamurthy and Takkar, 1997, 2000) . Total effect of S fertilization can be estimated by using relative growth rate index, which presents relative biomass increment rate (Lamber *et al.*, 1990; Black-kalff *et al.*, 1998).

Interaction of sulphur with other nutrients

The interaction of sulphur with other nutrients was studied (Tiwari, 1997) and reported that S had synergistic relationship with N, P, Mg, and Zn. Acidity produced on oxidation of reduced inorganic S compounds in soil was known to increase the solubility of micronutrients, like Fe, Zn, and Mn. Phosphorous along with S were found to improve nodulative activity. Sulphur fertilization significantly increased the cys, GSH, and alliin contents of leaves and bulbs, while nitrogen fertilization had no significant influence. The alliin concentration in bulbs increased with Sulphur fertilization significantly at all harvesting dates and at maturity. High sulphur application in combination with low N fertilization increased the alliin concentration in garlic significantly during main growth until the beginning of ripening (Bloem *et al.*, 2010). In addition, Sulphur fertilisation increased the content of the dominant glucosinolate but it did not significantly change levels of the saponins 3-0-beta-cellobiosylhederagenin and 3-0-beta-cellobiosyloleanolic acid - a best-known feeding detergent for *Plutella xylosteella* larvae (Badenes-Perez *et al.*, 2010). General responses to sulphur limitation along with C and N are reduced growth and photosynthesis (Davies and Grossman, 1990). Since these compounds are integrated into major constituents of the cell, a converging sequence of events can be expected. The regulation of C and N metabolism has been extensively investigated and found to be intimately related on physiological and molecular level with S (Huppe and Turpin, 1994; Kaiser, 1997). For example, the chemical form of N and its concentration modulate photosynthesis due to direct link existing between C skeleton availability and N fixation into amino acids (Huppe and Turpin, 1994; Giordano and Bowes, 1997). In turn, the availability of inorganic carbon strongly affects the ability to assimilate nitrate(NO_3^{2-}) and ammonium (NH^{4+}) (Larson *et al.*, 1985). Sulphur deficiency caused a decrease in accumulating in N^{15} in the root and shoot of non-modulating and modulating chickpea seedlings (Badruddin and Karmokee, 2001) and decreased accumulating of K, Fe^{2+} and Zn^{2+} while increased that of Mg^{2+} and NO_3^{2-} in both the genotypes (Badruddin and Karmokee, 2001). The co-regulation of NO_3^{2-} and SO_4^{2-} uptake, the down regulation of nitrate reductase (NR) and changes in the level of O-acetyl-ser are the main elements of N-S metabolism interaction (Clakson *et al.*, 1989; Brunold, 1993; Davies and Grossman, 1998).

Sulphur fertilisation and *Rhizobium* interaction

Studies in Pigeon Pea indicated increased uptake of N, P and K due to Sulphur nutrition (Umarani, 1994). Sulphur application (40 kg/ha) with *Rhizobium* inoculation in green gram, increased the bacterial population significantly (Naidu *et al.*, 1995, 1998). The mean total number of nodules and active nodules significantly increased with application of Sulphur only up to 20 Kg S/ha but beyond the 20 kg/ha, the mean nodule production reached a plateau and did not increase further (Ganeshhamurthy and Reddy, 2000).

Effect of sulphur application on phosphorous availability in soil

Since legumes usually require almost equal amounts of P and Sulphur. When P and

Sulphur are present below the critical level in soil, plant growth and quality of production are affected adversely (Dube *et al.*, 1970). S was found to improve nodulation activity (Andrew and Robins, 1969; De Moy *et al.*, 1973; Lluch *et al.*, 1982). Sulphur oxidation through soil microbes lead to production of H_2SO_4 which solubilises P. The acidity generated on oxidation of pyrite can be coupled to solubilisation of rock phosphate or to reclaim alkali soil (Kapoor *et al.*, 1989). The solubilisation of mussoorie rocks phosphate on addition of elemental sulphur and pyrite and on inoculation with Sulphur and Fe oxidising bacteria in soil was studied (Kapoor *et al.*, 1991; Costa *et al.*, 1992).

Also, the grain yield of green gram was highest when 50 Kg P_2O_5 + 40 kg S/ha was applied (Paricha, 1993). Application of 40 kg Sulphur along with a 40 kg P_2O_5 /ha resulted in higher grain yield than Sulphur alone in chickpea (Shivakumar, 2001). Thus, P and Sulphur are reported to have synergistic effect on the productivity of pulse crops.

Sulphur absorption and translocation

Sulphur is an essential nutrient, taken up as SO_4^{2-} from soil, reduced and incorporated into bioorganic compounds in plant cells. The pathway of SO_4^{2-} assimilation is highly regulated in a demand-driven manner in seed plants (Hermsel *et al.*, 2010). Plants as autotrophic organisms have a set of transporters and enzymes that mediate uptake and assimilation of inorganic SO_4^{2-} and subsequent metabolic conversion to organic sulphur compounds. Studies in higher plants indicate the individual components of SO_4^{2-} transport systems and enzymes for SO_4^{2-} assimilation are consisted of multiple isoforms. Among these isoforms, several essential components are shown to have specific biochemical properties and localize in specific cellular and subcellular compartments. Recent findings of Takahashi (2010) provided evidence that the regulatory pathways are highly organized to balance the uptake, storage, and assimilation of SO_4^{2-} in plants. In addition to the physiological and biochemical functions diversified among the isoforms of SO_4^{2-} transporters, regulatory elements in transcriptional and posttranscriptional mechanisms were suggested to play significant roles in coordinating the assimilatory functions to adapt with varying sulphur nutritional status that fluctuates in the environment.

Sulphur availability plays an important role in growth and development of higher plants (Hell and Hillebrand, 2001). Plant mainly absorbs sulphur in the form of SO_4^{2-}, comes predominantly from weathering of parents rocks material. Industrialization, however, adds an addition source of SO_4^{2-}, atmospheric pollution. The burning of fossil fuels releases several gaseous forms of sulphur including SO_2 and H_2S, which find their way to soil in rain. When dissolved in water, SO_2 is hydrolysed to become H_2SO_4, a strong acid which is major source of acid rains. Plant can also metabolize SO_2 taken up in the gaseous form through their stomata.

General responses to Sulphur limitation are reduced growth and photosynthesis (Davies and Grossman, 1998). Sulphur is often referring to as the essential component of important metabolic and structural compounds. Sulphur is a constituent of protein, lipids, carbohydrates, electron transport system and many other cellular constituents and intermediate metabolites (Leustek, 2000). Most sulphur is imported into cells as SO_4^{2-} and then translocated into chloroplasts. Before SO_4^{2-} is metabolized, it is adenylated by ATP sulphurylase to form 5-adenyl-sulphate, APS. APS is a branch point intermediate that can be phosphorylated by APS kinase and used in sulfation reactions or used to synthesize cys, to form cysteine, APS is reduced by sequential reactions catalysed by APS reductase and sulphite reductase to form sulphide, is subsequently combined with OAS to form Cys by OAS (thiol) lyase. Cys is incorporated into protein and is precursor of methionine (Met) and S-adenosyl-l-met in one set of reactions and glutathione (GSH) and phytochelatins (PCs) in another. Little is known about the interaction of N metabolism with Sulphur metabolism. Elements of this interaction may be the co-regulation of NO_3^{2-} and SO_4^{2-} uptake, the down regulation of NR and changes in the level of OAS, the intermediate in cys metabolism under Sulphur limitation (Clarkson *et al.*, 1989b; Giordano *et al.*, 2000). Even less information is available on the interaction of Sulphur and C metabolism. So far, mostly isolated steps of the corresponding pathways have been investigated during limitation of SO_4^{2-} as a nutrient, although Sulphur is a key component of essential cell constituents (Hell, 1997).

Sulphur availability plays an important role in the growth and development of higher plants (Hell and Hillerbrand, 2001). In the pH range to which roots are normally exposed, uptake is not very pH sensitive. Multiple transport steps through different membrane are involved. Plasma membrane transporters in root present in the outermost cell layers for initial uptake. Plasma membrane transporters of vascular tissues for long distance

translocation and of leaf mesophyll cells for assimilation coupled with photosynthesis and inside the cells. Transporters associated with organelle in particular plastids and vacuoles (Hawkesford, 2003). However, Selenate which is closely chemically related to SO_4^{2-} , depresses SO_4^{2-} uptake substantially. Plasma membrane SO_4^{2-} transporters are classified as H^+/ SO_4^{2-} co-transporters that mediate active SO_4^{2-} driven by the transmembrane H^+ gradient. Thus, the uptake mediated by these transporters is pH dependent, and the H^+ gradient is generated by the plasma membrane H^+ATPase. The SO_4^{2-} transporters possess 12 membrane-spanning domains and belong to a large family of cation co-transporters (Saito, 2000; Hawkesford, 2003). According to investigators, with sunflower SO_4^{2-} is absorbed and translocated against an electrochemical gradient which suggest that SO_4^{2-} uptake is an active process. Lin (1983) studied that the uptake was promoted by a decrease in the pH of the outer solution. The author suggests that SO_4^{2-} may be taken via H^+/SO_4^{2-} co transport or an OH^-/SO_4^{2-} anti-transport. A number of genes encoding SO_4^{2-} transporters of *Arabidopsis* (Yashimato *et al.*, 2002), inducible by SO_4^{2-} depletion localized to root epidermal cells and thus responsible for initial uptake of SO_4^{2-} from the rhizosphere (Takashi *et al.*, 2000; Yoshimato *et al.*, 2002). A high affinity transporter named SULTRI 3, is localised to the phloem and mediates long distance translocation from source organs (roots) to sinks (leaves and shoots) (Yoshionoto *et al.*, 2003).

Since, the yield and quality of legume seeds are limited by the amount of sulphur partitioned to the seeds. The amino acid S-methylmethionine (SMM), a methionine derivative and a long-distance transport form of reduced Sulphur and whether SMM phloem loading and source-sink translocation are important for the metabolism and growth of pea (*Pisum sativum*) plants. The changes in SMM phloem loading affected plant growth and seed number, leading to an overall increase in seed Sulphur, N, and protein content. The phloem loading and source-sink partitioning of SMM are important for plant Sulphur and N metabolism and transport as well as seed set (Tan *et al.*, 2010). Low affinity transporters belonging to the SULTR2 and SULTR3 subfamilies are localised to vascular tissues and are presumably involved in the uptake of SO_4^{2-} from plant apoplasts into vascular cells. The transporters of SULTR4 subfamily were initially thought to be involved in plastid uptake. However, these are responsible for efflux of SO_4^{2-} from the vacuoles to the cytoplasm (Kataoka *et al.*, 2004). Besides H^+/ SO_4^{2-} co transporters, anion channels, ABC proteins and oxaloacetate/SO_4^{2-} transporters may mediate sulphate transport in plant cells (Leutek, 2002). Today increasing sulphate utilization efficiency (SUE) is an important issue for crop improvement. Little is known about the genetic determinants of sulphate utilization efficiency. The function of SUE3 and SUE4 in low sulphur tolerance was confirmed either by multiple mutant alleles or by recapitulation analysis. Results of Wu *et al.* (2010) demonstrate that this recapitulation analysis is a genetic screen or a reasonable approach to isolate *Arabidopsis* mutants with improved low sulphur tolerance and potentially with enhanced SO_4^{2-} utilization efficiency.

Molecular identification of these additional transport systems remains an open question. SO_4^{2-} is mainly translocated in an upward (acropetal) direction and the capability of higher plants to move Sulphur in a downward (basipetal) direction is relatively poor (Schobert *et al.,* 1995). Sulphur in the root and petiole was translocated toward the younger leaves (Bouma, 1967). This shows that translocation against the transpiration stream did not occur. Sulphur (SO_4^{2-}) along with K, Mg, P and Cl are almost completely included from the phloem (Zimmermann and Brown, 1967; Fonday, 1975; Vanobel and Gamalei, 1992). There are now a considerable body of evidence to show that plants can utilise atmospheric SO_2 as part of their S supply. Growth of various plant species in growth chambers with defined SO_2 concentration as the sole S source, found that growth was reduced in the treatment where SO_2 was absent (Faller *et al.*, 1970). The beneficial effect of SO_2 in alleviating S deficiency has also been reported by Cowling and Lockyer (1976). SO_2 is absorbed through the entire plant and detected in various sulphur fractions such as protein Sulphur, amino acid Sulphur and sulphate Sulphur (De Cormis, 1968).

Activation of Sulphur transport systems is critical for plant growth under a low sulphur environment, when the soil environment is adequately fertilized with SO_4^{2-} plant will sustain their growth by increasing the capacities of sulphate uptake systems in roots (Clarkson *et al.*, 1983, Deane-Drummond, 1987; Smith *et al.*, 1995, 1997). In *Arabidopsis thaliana*, high affinity SO_4^{2-} transporters that facilitate the initial uptake of SO_4^{2-} serve this purpose (Takahashi *et al.*, 2000; Shibagaki *et al.*, 2002; Yoshimoto *et al.*, 2002). Transporters mediating vascular transport of

sulphate and release of vascular SO_4^{2-} can also contribute for efficient use of SO_4^{2-} pools under Sulphur conditions (Kataoka *et al.*, 2004b). In addition, catabolic recycling of secondary sulphur metabolites and storage compounds may become necessary for adaptation to a Sulphur environment (Hirai *et al.*, 1995, 2005; Kutz *et al.,* 2002).

The high affinity SO_4^{2-} transport system predominates under the Sulphur environment for efficient acquisition of SO_4^{2-} from the soil. This phenomenon has been well characterized by physiological experiment (Clarkson *et al.*, 1983; Deane-Drummond, 1987). More recent molecular biological studies suggested that this Sulphur inducible transport system can be attributed to the function of two sulphates transporter genes (SULTRI 1 and SULTRI 2) in arasidopsin (Takahashi *et al.*, 2000; Shibaki *et al.*, 2002; Yoshimoto *et al.,* 2002). Such similarly inducible SO_4^{2-} transporters exist in other plant species as well (Smith *et al.*, 1995, 1997; Vidmer *et al.*, 1999; Howarth *et al.*, 2003; Buchner *et al.,* 2004a, 2004b; Hopkins *et al.*, 2005).

Sulphur assimilation

Sulphur is among the most versatile elements in living organism (Hell, 1997) and essential macronutrient required for plant growth (Nakashita *et al.,* 2004b). The incorporation of mineral nutrients into organic substances such as pigments, enzyme, cofactors, lipids, nucleic acid or amino acids is termed nutrient assimilation. The most important sources of S for higher plants is sulphate (SO_4^{2-}) the oxidized form of S existing in the soil, as a sulphur source (Crowford *et al.*, 2000; Leustek *et al.*, 2000; Saito, 2004). In several respects, S assimilation resembles with that of NO_3^{2-}, through the detailed mechanism is not so well understood but in all postulated pathways SO_4^{2-} assimilation consumes about 14 ATP per SO_4^{2-} converted to the amino acids cysteine (Hell, 1997). The first step of S assimilation is the activation of Sulphur by the enzyme ATP-sulphurylase, and in cascade of enzymatic steps inorganic S is converted to the important non-protein tripeptide, glutathione (Tausz *et al.*, 2004). Significance of plant SO_4^{2-} assimilatory pathway is manifested by its ability to fill this metabolic gap in the global S cycle in nature (Crawford *et al.*, 2000). In addition to its basic nutritional importance, S is present in number of plant metabolites representing important biological activities as redox controllers, vitamins, coenzymes, flavours and defence chemicals (Crawford *et al.*, 2000, Leustek *et al.*, 2000; Saito, 2004; Grabb and Abel, 2006; Halkier and Gershenzon, 2006).

(i) *Sulphur assimilation, cascade of various steps*

SO_4^{2-} is the most oxidative and thus stable form of S present in the soil. Uptake of S into roots from the soil is almost exclusively via SO_4^{2-} uptake. The form of sulphur found in xylem and phloem sap is also primarily SO_4^{2-}, thus translocation of S throughout the plant is mostly via unmetabolised SO_4^{2-}.

SO_4^{2-} assimilation occurs mostly in leaves, the SO_4^{2-} reduction is carried out by a number of organisms including higher plants, algae, fungi, BGA and cyanobacteria. The reduction of SO_4^{2-} to cysteine changes the oxidation number of S from +6 to –4, thus entailing the transfer of 10 electrons. Glutathione, thioredoxin, ferridoxin, NADPH or OAS may serve as electron donors at various steps of the pathway. Leaves are generally much more active than roots in Sulphur assimilation, presumably because photosynthesis provides reduced thioredoxin, ferredoxin, and photorespiration generator serine that may stimulate the production of OAS. Sulphur assimilated in leaves is exported via the phloem to sites of proteins synthesis mainly as glutathione (Rennenberg, 1993). Glutathione also acts as a signal that co-ordinates the absorption of SO_4^{-2} by the roots and the assimilation of SO_4^{2-} by the shoot, mostly via unmetabolized SO_4^{2-}. Subsequently, SO_4^{2-} is subjected to adenosine 5–phosphosulfate [5– adenylyl sulfate (APS)] for further conversion. The major assimilatory pathway is APS to SO_3^{2-} (Sulfite) and then sulfide (S^{2-}). The overall reduction from SO_2^{2-} to S^{2-} requires one ATP and 8 electrons (Schiff and Hodson, 1973). S^{2-} is then coupled with OAS that is formed from Ser, yielding cys. A relatively minor branch point in this pathway is from APS to PAPS, which is restricted for sulfation.

Cys is the central compound for production of a variety of metabolites containing reduced S, such as Met, GSH, PCs, and glucosinolates. Although the turnover rate of Cys is high, the cellular concentration of free Cys is maintained at a low level (approx. 20µM), compared with high levels of GSH (approx. up to 10mM) (Leustek *et al.*, 2000; Saito, 2000). The antioxidant function of 2-Cys peroxiredoxin (Prx) involves the oxidation of its conserved peroxidative cysteine to sulphenic acid that is recycled by a reducing agent. In conditions of oxidative stress, the peroxidative cysteine can be overoxidized to sulphinic acid inactivating the Prx. An enzyme recently

discovered, named sulfiredoxin (Srx), reduces the sulphinic 2-Cys Prx (Prx-SO(2)H). The activity of sulfiredoxin dependent on the concentration of the sulphinic form of Prx and the conserved Srx is capable of regenerating the functionality of both pea and *Arabidopsis* Prx-SO(2)H (Iglesias-Baena *et al.*, 2010).

(ii) *Activation is a pre-requisite for SO_4^{2-} assimilation*

SO_4^{2-} is very stable and thus needs to be activated before any subsequent reactions may proceed. Therefore, for assimilation, SO_4^{2-} must be activated to APS, in which SO_4^{2-} is linked by an anhydride bond to a phosphate residue by consumption of ATP and concomitant release of Pyrophosphate (PPi). This reaction is catalyzed by ATP sulphurylase, has been assured as the rate limiting step enabling and initiating sulphur metabolism (Hofgen *et al.*, 2001). The sole entry step for the metabolism of SO_4^{2-} APS formed in the cytosol may not be directly involved in assimilation but may participate in sulfation after being converted to PAPS (Rotte and Leustek, 2000).

$$SO_4^{2-} + ATP \longrightarrow APS + PPi,\ \Delta G^{01} = 45\ KJ/mole$$

ATP Sulphurylase, Las Cytosolic and plastid forms in spinach and potato, but it seems restricted to plastids in *Arabidopsis thaliana* (Leustek, 1996). However, all four ATP Sulphurylase genes in *Arabidopsis* likely encode plastidic forms. The cytosolic isoforms is presumably produced from one of those four genes by using different translational start codons (Hatzfeld *et al.*, 2000a).

$$APS + ATP \longrightarrow PAPS + ADP\ \Delta G^{01} = 25\ KJ/mole$$

PAPS are then reduced to SO_3^{2-} and then to S^{2-}. This is the major pathway for SO_4^{2-} assimilation in bacteria and fungi. In addition, PAPS serves as a source of activated SO_4^{2-} for sulfotransferases that catalyzes sulfation of a variety of compounds such as flavonoids, glucosinolates and fasmonates. Since the reaction equilibrium of ATP sulphurylase favours the reverse direction, i.e. the formation of ATP and SO_4^{2-}, the products of the forward reaction, i.e. APS and pyrophosphate, must be further metabolized immediately by the enzymes APS reductase, APS kinase and pyrophosphatase. Alternatively, the sulphur in APS may be converted to an enzyme bound thiosulfonate ($R\text{-}SO_3^-$), which in turn is reduced to a thiosulphide ($R-\overline{S}$).

Yet a third possibility is that APS is directly reduced to SO_3^{2-} and then to S^{2-} (Hell, 1997). In all three pathways, the resultant thiosulfide or sulfide reacts with OAS to form cys and acetate. The enzymes involved in cys synthesis have been found in the cytosol, plastids and mitochondria of various plants, probably reflecting the inability of organelles to transport cysteine across their membrances (fig. 1) (Hell, 1997).

(iii) *Reduction of activated SO_4^{2-}*

The SO_4^{2-} residue of APS is reduced to SO_3^{2-} by APS reductase that has been previously referred to as APS sulfotransferase (Suter *et al.*, 2000). The *in vivo* APS reductase is present as a homodimer most probably linked by a disulfide bond of the conserved Cys residue (Kopriva and Koprivova, 2004). The mature APS reductase consists of two distinct domains. The N-terminal domain exhibits homology to thioredoxin and acts as a gluta-redoxin using reduced GSH as the electron donor (Bick *et al.*, 1998). APS reductase catalyzes a thiol- dependent two-electron reduction of APS to SO_3^{2-}. The enzyme bound S-Sulfo intermediate is presumed to be involved in the reduction of APS (Weber *et al.*, 2000). In some lower plants such as the moss *Physcomitrella pattern*, in addition to an APS reductase-dependent pathway, PAPS can be reduced to SO_3^{2-} by PAPS reductase as in bacteria (Kopriva and Koprivova, 2004).

(iv) *Ferredoxin involved in reduction of sulfite (SO_3^{2-}) to sulphide (S^{2-}).*

Sulfite reductase catalyzes the transfer of six electrons from ferredoxin to SO_3^{2-} to produce S^{2-} found in plant cells and consists of a homo-oligomer containing a siroheme and an Fe-S cluster per subunit. Electrons are supplied to ferredoxin from PS-I in photosynthetic cells and from NADPH to non-photosynthetic cells. The proper combination of different isoforms of ferredoxin, ferredoxin $NADP^+$ reductase, and sulphate reductase, is

critical for efficient SO_3^{2-} reduction (Yone Kura-SakaKibara *et al.*, 2000).

S^{2-} is directly utilized as the sulphur donor for the formation of UDP-Sulfo-quinovose from UDP-G/c (Sanda *et al.*, 2001); a precursor of the sulfolipid, Sulfo quinovosyl diacyl glycerol, present in photosynthetic membranes representing one of the few naturally occurring sulfonic acid ($R–CH_2 – SO_3^{2-}$) derivatives. Sulfite oxidase (a Mo enzyme), catalyses oxidation of S^{2-} to SO_4^{2-} in peroxisomes (Eilers *et al.*, 2001) and widely distributed in higher plants and is likely to responsible for detoxification of S^{2-} rather than for chloroplast based Sulphur assimilation (fig. 1). Sulfite oxidase catalyses the physiologically vital oxidation of sulfite to SO_4^{2-}, the terminal reaction in degradation of sulphur containing amino acids, cys and methionine. Sulfite oxidase from vertebrate sources is among the best studied MO enzymes. Existence of this enzyme in plants has been established recently by identification of a cDNA from *Arabidopsis thaliana* encoding a functional SO (Ahmad and Sarfraz, 2010).

(v) *Role of serine to incorporate sulfide into cysteine*

Incorporation of $S^{2–}$ into the β-positions of amino acids is the terminal step of sulphur assimilation, leading to the formation of cys. Two enzymes, ser acetyl-transferable and cys synthase are committed for this step and found in three major compartments of the plant cells (Saito, 2000). Cytosol, chloroplast and mitochondria in contrast to specific localization of the enzymes of SO_4^{2-} reduction in plastids. Five genes encoding Ser acetyl-transferases are found in the *Arabidopsis* genome (Hell *et al.*, 2002; Kawashima *et al.*, 2004). Three of these isoforms seems to play major roles in OAS formation, judging from their kinetic properties under normal growth conditions (Noji *et al.*, 1998).

Two minor forms of Ser acetyl transferase found in *Arabidopsis* and induced under stress conditions, i.e. sulphur deficiency and heavy metal stress, and thus may play a role in adoptive responses to stresses (Kawashima *et al.*, 2004).

(vi) *Cysteine in* sulphur *assimilatory pathway*

Cys is the pivotal sulphur containing compound regarded as the terminal metabolite of sulphur assimilation and the starting point for production of Met, GSH, and a variety of other sulphur metabolites. OAS-lyase catalyzing the formation of cys from OAS and H_2S. The catalytic activity of OAS-lyase requires pyridoxal phosphate as a cofactor and it belong to a large family of enzymes catalyzing the reaction of β-substitution of amino acids, including β-cyano-Ala Synthase responsible for β-Cyano-Ala formation (Hatzfeld *et al.*, 2000b). The mechanism and genes for degradation/recycling of cys are not yet well understood. However, cysteine lyase catalysing the first step of cystine breakdown has been recently characterized in *Arabidopsis* (Jones *et al.*, 2003). Cysteine-rich PR proteins, such as non-specific lipid transfer proteins (nsLTPs) and metallocarboxypeptidase inhibitors are candidates for the sequestration of metals (Harada *et al.*, 2010). Cysteine is a component in organic compounds including GSH that have been implicated in the adaptation of plants to stresses. O-acetylserine (thiol) lyase (OAS-TL) catalyses the final step of cysteine biosynthesis. OAS-TL enzyme isoforms are localised in the cytoplasm, the plastids and mitochondria but the contribution of individual OAS-TL isoforms to plant sulphur metabolism has not yet been fully clarified. OAS-A1 (gene encoding the cytosolic OAS-TL) is involved in maintaining sulphur and thiol levels and is required for resistance against cadmium stress. The cytosolic OAS-TL is involved in maintaining organic sulphur levels (Shirzadian-Khorramabad *et al.*, 2010).

(vii) *Role of multiple regulatory circuits for Cys assimilation*

Cys formation is controlled through a multiple regulatory circuit involving Ser acetyl transferase and OAS (Thiol) Lyase. OAS is not only a rate-limiting metabolite of the Cys biosynthetic pathway (Saito *et al.*, 1994) but also a positive regulatory factor for gene expression (Leusteak *et al.*, 2000; Saito, 2000; Leutek, 2002).

A unique regulatory mechanism operates through the formation of an enzyme complex involving Ser acetyl-transferase and OAS (thiol)-lyase. The OAS lyase concentration is far in excess of Ser acetyl-transferase concentration (approx. 300 fold), indicating that only a fraction of the OAS lyase forms a complex with Ser-acetyl-transferase (Droux *et al.*, 1998). The large amount of free OAS–lyase is responsible for the actual catalytic function of Cys formation. OAS accumulation stimulated by sulphur deficiency promotes dissociation of the complex to attenuate the activity of Ser acetyl transferase, resulting in reduced OAS formation. Furthermore, OAS formation in controlled by isoform specific of Ser acetyl transferase activity by L-cys, the product of this pathway. The importance of this feedback regulation is

supported by the following observations: (i) overexpression of a feedback insensitive Ser acetyl transferase gene results in elevated levels of Cys in bacteria and plants (Noji and Saito, 2002) and (ii) higher levels of cys were found in Chinese chive (*Allium tuberosum*), in which a cytosolic Ser acetyl transferase was inhibited by a higher concentration of cys than other plants (fig. 2) (Urano *et al.*, 2000).

Regulatory mechanism in sulphur assimilation

(i) *Sulphur starvation*

It is well known that SO_4^{2-} uptake and assimilation activity is induced (derepressed) under conditions of sulphur starvation or high demand for sulphur metabolites. This induction correlates with the inducible accumulation of steady state mRNA, of a particular set of genes that encode SO_4^{2-} transporter and APS reductase, but not all genes in the sulfate assimilation pathway (Saito, 2000; Leustek, 2002) Sulphur starvation causes an increase in OAS levels, which in turn induces the expression of genes encoding SO_4^{2-} transporters and APS reductase, thereby overriding the repressive effect of Sulphur. *Arabidopsis* plants were exposed to sulphur depletion alone, changes of photosynthetic parameters and metabolite abundances were quantified. Photosynthetic electron transport rates (ETRs) of plants exposed to sulphur depletion and high light decreased strongly at day 2 of the acclimation period. However, at metabolic level, the stress combination had a profound effect on central metabolic pathways such as the tricarboxylic acid (TCA) cycle, glycolysis, pentose phosphate cycle and large parts of amino acid metabolism. Under these conditions, central metabolites, such as sugars and their phosphates, increased, while sulphur-containing compounds were decreased (Wulff-Zottele *et al.*, 2010).

Sulphur starvation causes an increase in OAS levels, which in term induces the expression of genes encoding SO_4^{2-} transporter and APS reductase, thereby overriding the repressive effect of sulphur sufficient nutritional conditions (Smith *et al.*, 1997; Koprivova *et al.*, 2000). Addition of OAS mimics sulphur deficiency stress for sulphur-related gene expression and global cellular changes in terms of the transcriptome and metabolome. Similar sets of genes and metabolites are modulated by sulphur deficiency and OAS supplementation (Hirai *et al.*, 2003, 2004). In contrast to OAS's positive effect, GSH acts as a negative regulator (repressor) of sulphur metabolism. GSH is thought to be a phloem-translocated signal molecule that represses the genes of sulphur assimilation (Lappartient *et al.*, 1999). Confocal laser scanning microscopy (CLSM) showed that GSH levels in tip cells of both long and short trichomes were higher than those in other types of leaf cells, indicating the presence of an active sulphur-dependent protective system in trichomes (Harada *et al.*, 2010). More generally, thiols such as Cys and GSH act negatively on gene expression (Vauclare *et al.*, 2002). All these observations indicate that OAS and thiol are positive and negative regulators, respectively, on gene expression responding to sulphur starvation. However, it does not necessarily follow that these metabolites directly act on gene expression as shown for OAS in bacteria, but could involve multiple and complex regulatory pathways (fig. 2) (Hartman *et al.*, 2004).

(ii) *Plant hormones*

Some plant hormones control gene expression related to sulphur metabolism (Kutz *et al.*, 2002). Several studies on transcriptome analysis suggest that methyl-jasmonate and auxin are involved as signals of sulphur-deficiency stress (Hirai *et al.*, 2003; Maruyama-Nakashita *et al.*, 2003, Niki Forova *et al.*, 2003). Indeed, methyl-jasmonate induces a cluster of sulphur assimilation genes but not sulfate transporters, as well as well-known jasmonate–inducible genes. Cytokinin signalling also appears to be involved in gene expression related to sulphur metabolism. Cytokinins have been shown to down regulate the expression of high affinity transporter genes in *Arabidopsis* roots (Maruyama-Nakahita *et al.*, 2004). By contrast, a sulphur deficiency responsive promoter of soyabean (Glycene max) β-conglycinin responds positively to cytokinin (Ohkama *et al.*, 2002). Since cellular cytokinin levels do not significantly change in response to suffer status, further investigations are needed on the signal perceptions and downstream gene expression events.

(iii) *Abiotic stresses and biological clock*

Sulphur assimilation is highly active in growing tissues where high levels of Cys and Met are required for protein synthesis. Indeed, gene expression of plastidic ATP sulphurylase and APS reductase is high in young leaves of *Arabidopsis* (Rotte and Leustek, 2000). Developing seeds seem to assimilate sulphur directly from SO_4^{2-} inside the seed, rather than translocating Cys or GSH from other tissues (Tabe and Droux, 2001). Some suffer assimilating genes, but not all, are regulated

by circadian rhythm (Kopriva *et al.*, 1999; Harmes *et al.*, 2000). The expression of SO_4^{2-} transporters, APS reductase, Ser acetyl transferase, and 3–Phosphoglycerate dehydrogenase, are at a peak just before the onset of the light period.

Abiotic stresses such as heavy metals and oxidative stress affect sulphur assimilation. Once plants are exposed to heavy metals such as cadmium, phytochelatins $(\gamma\text{-Glu-Cys})_n$ – Gly are synthesized from GSH, which consequently consumes cys. In fact, heavy metal stress promotes the expression of sulphur assimilation and transporter genes (Dominguez-Solis *et al.*, 2001; Nacito *et al.*, 2002).

Plant responses to sulphur stress

Nutritional stress of Sulphur most notably modulates SO_4^{2-} Sulphur assimilation. Upon Sulphur deficiency, sulphur is remobilized less efficiently than nitrogen, resulting in chlorosis of young leaves by sulphur deficiency; while the first appearance of nitrogen deficiency is in old leaves (Hawkesford, 2000). Analysis of transcriptome (Hirai *et al.*, 2003; 2004; Maruyama-Nakashita *et al.*, 2003; Nikiforova *et al.*, 2003; Wang *et al.*, 2003) and metabolome (Hirai *et al.*, 2004) in *Arabidopsis* indicate the presence of global multiple networks responding to nutritional deficient stress. Long-term sulphur deficiency results in similar global changes in the transcriptome and metabolome, as Doe's nitrogen deficiency (Hirai *et al.*, 2004). These are general responses common to sulphur deficiencies. These general nutritional responses differ in leaves and roots. Signalling pathways involving methyl jasmonate and auxin seem to be involved in the sulphur stress response. Primary and secondary metabolic pathways, involving amino acids, carbohydrates, and glucosinolates, are modulated in response to sulphur deficiency stress (Hirai *et al.*, 2004). Glucosinolates are regarded as storage and possibly mobilizing forms of assimilated sulphur in response to acute sulphur deficiency. Since integrated proteomics and genomics studies have just started, further investigation will provide more detailed information on holistic networks of sulphur metabolism.

Acknowledgement

The authors are highly thankful for the facilities provided at AMU, Aligarh. Financial support from the Department of Science and Technology, New Delhi in the form of project (SR/FT/LS-087/2007) is gratefully acknowledged.

References

Ahmad A, Sarfraz A, 2010. Screening and partial immunochemical characterization of sulfite oxidase from plant source. Indian Journal of Experimental Biology, 48(1): 83-86.

Anderson AJ, Spencer D, 1950. Sulphur in nitrogen metabolism of legumes and non-legumes. Australian Journal of Science Research B, 3: 431–449.

Jamal A, Fazli IS, Ahmad S, Kim KT, Oh DG, Abdin MZ, 2006. Effect of sulphur on nitrate reductase and ATP sulurylase activities in groundnut (*Arachis hypogea* L.). Journal of Plant Biology, 49(6): 513-517.

Badenes-Perez FR, Reichelt M, Heckel DG, 2010. Can sulphur fertilisation improve the effectiveness of trap crops for diamondback moth, Plutella xylostella (L.) (Lepidoptera: Plutellidae)? Pest Management Science, 66(8): 832-838.

Badruddin M, Karmoker JL, 2001. The effect of sulphur deficiency on ion accumulation with special references to ^{15}N and ^{35}S transport in chickpea. Developments in Plant and Soil Sciences, 92: 116-117.

Bergmann L, Rennenberg H, 1993. Glutathione metabolism in plants. In sulphur nutrition and assimilation in higher plants. Regulatory, agriculture and environmental aspects. L.J. De Kok, I. Stulen, H. Rennenberg, C. Brunold and W.E. Rauser, eds., SPB Academic Publishers, The Hague, Netherlands, pp. 102-123.

Bick JA, Slund F, Chen Y, Leustek T, 1998. Glutaredoxin function for the carboxyl terminal domain of the plant-type 5-adenylylsulfate (APS) reductase. Proceedings of the National Academy of Sciences of the USA, 95: 8404–8409.

Blake-Kalff MMA, Harrison KR, Hawkesford MJ, Zhao FJ, McGrath SP, 1998. Distribution of sulphur with in oilseed rape leaves in response to sulphur deficiency during vegetative growth. Plant Physiology, 118(4): 1337-1344.

Bloem E, Haneklaus S, Schnug E, 2010. Influence of fertilizer practices on S-containing metabolites in garlic (Allium sativum L.) under field conditions. Journal of Agriculture and Food Chemistry, 58(19): 10690-10696.

Bouma D, 1967. Nutrient uptake and distribution in subterranean clover during recovery from nutritional stresses. II. Experiment with sulphur. Australian Journal of Biological Sciences, 20: 613-621.

Brunold C, 1993. Regulatory interactions between sulphate and nitrate assimilation. *In* LJ De Kok, I Stulen, H Rennenberg,C Brunold, WE Rauser, eds, Sulphur Nutrition and Assimilation in Higher Plants. SPB Academic Publishing, The Hague, The Netherlands, pp 61–75.

Buchner P, Stuiver CE, Westerman S, Wirtz M, Hell R, Hawkesford MJ, De Kok LJ, 2004a. Regulation of sulphate uptake and expression of sulfate transporter genes in Brassica oleracea as affected by atmospheric H2S and pedospheric sulphate nutrition. Plant Physiology, 136: 3396–3408.

Buchner P, Takahashi H, Hawkesford MJ, 2004b. Plant sulphate transporters: Co-ordination of uptake, intracellular and longdistance transport. Journal of Experimental Botany, 55: 1765–1773.

Chapman SJ, 1990. *Thiobacillus* population in some agricultural soils. Soil Biology and Biochemistry, 22: 479-482.

Clarkson DT, Saker LR, Purves JV, Lee RB, 1989b. Depression of nitrate and ammonium transport in barley plants with diminished sulphate status: Evidence of co-regulation of nitrogen and sulphate intake. Journal of Experimental Botany, 40: 953-963.

Clarkson DT, Smith FW, Vanden Berg PJ, 1983. Regulation of sulfate transport in a tropical legume, Macroptilium atropurpureum cv. Siratro. Journal of Experimental Botany, 34: 1463–1483.

Costa ACA, Medronhe RA, Pecanha RP, 1992. Phosphate rock bioleaching. Biotechnology Letters, 5: 53-58.

Cowling DW, Locker DR, 1976. Growth of perennial ryegrass (Lolium perchne L.) exposed to a low concentration sulphur dioxide. Journal of Experimental Botany, 27: 411-417.

Crawford NM, Kahn ML, Leustek T, Long SR, 2000. Nitrogen and sulphur. In Biochemistry and Molecular Biology of Plants, B.B. Buchanan, W. Gruissem, and R.L. Jones, eds (Rockville, MD: American Society of Plant Biologists), pp. 824–849.

Davies JP, Grossman AR, 1998. Responses to deficiencies in macronutrients. *In* M Goldschmidt-Clermont, S Merchant, eds, The Molecular Biology of Chloroplast and Mitochondria in Chlamydomonas. Kluwer Academic Publishers, Amsterdam, the Netherlands, pp 613–633.

Deane-Drummond CE, 1987. The regulation of sulfate uptake followinggrowth of Pisum sativum L. seedlings in S nutrient limiting conditions. Interaction between nitrate and sulphate transport. Plant Science, 50: 27–35.

Deboer DL, Duke SH, 1982. Effects of sulphur nutrition on nitrogen and carbon metabolism in lucerne (Medicago sativa L.). Plant Physiology, 54: 343-350.

Decormis L, 1968. Contribution to the study of sulphur absorption by plants exposed to an atmosphere of SO_2. Annual Physiologie Vegetables, 10(2): 99-112.

Dominguez-Solis JR, Gutierrez-Alcala G, Romero LC, Gotor C, 2001. The cytosolic O-acetylserine (thiol) lyase gene is regulated by heavy-metals and can function in cadmium tolerance. Journal of Biological Chemistry, 276: 9297–9302.

Droux M, Ruffet ML, Douce R, Job D, 1998. Interactions between serine acetyltransferase and O-acetylserine (thiol) lyase in higher plants: structural and kinetic properties of the free and bound enzymes. European Journal of Biochemistry, 225: 235–245.

Dube SD, Mishra PH, 1970. Effect of S deficiency on growth, yield and quality of some important leguminous crops. Journal of the Indian Society of Soil Science, 18: 375-378.

Eilers T, Schwarz G, Brinkmann H, Witt C, Richter T, Nieder J, Koch B, Hillem R, Hansch R, Mendel RR, 2001. Identification and biochemical characterization of *Arabidopsis* thaliana sulfite oxidase: a new member in plant sulphur metabolism. Journal of Biological Chemistry, 276: 46989–46994.

Epstein E, 1972. Mineral nutrition of plants: Principles and Perceptives. Wiley, New York.

Epstein E, 1994. The anomaly of silicon inplant biology. Proceeding of the National Academy of Sciences of the USA, 91: 11-17.

Faller N, Hervig K, Kuhn H, 1970. The uptake of $^{35}SO_2$ from the air. Effect on crop yield. Plant and Soil, 33: 177-191.

Fonday BR, 1975. Sugar selectivity of phloem loading in beta vulgaris, L and Fraxinus Americanus, L. Thesis, University of Dayton, Dayton, OH.

Foyer CH, Theodoulou FL, Delrot S, 2001. The functions of inter- and intracellular glutathione transport systems in plants. Trends in Plant Science, 6: 486–492.

Ganeshamurthy AN, Sammi Reddy K, 2000. Effect of integrated used of farmyard Manure and Sulphur in a soyabean and wheat cropping systems on nodulation dry matter production and chlorophyll content of soyabean on swell-shrink soil in central india. Journal of Agronomy and Crop Science, 185: 91-97.

Ganeshamurthy AN, Saha JK, 1999. Status in the soils of agroecological regions of India. Fertilizer News, 44: 57-66.

Ganeshamurthy AN, Subba Rao A, Sammi Reddy K, 1994. Phosphorus and sulphur constraints for

Soybean- wheat and gram production on some swell- shrink soils. Agropedology, 4: 99-106.

Ganeshamurthy AN, Takkar PN, 1997. Residual management of sulphur applied to soybean or wheat in soybean wheat cropping system on Vertisols. Australian Journal of Soil Research, 35: 199-208.

Ganeshamurthy AN, Takkar PN, 2000. Integrated use of FYM and S in soybean-wheat cropping system in relation to substitution of S in Vertisols of Central India. Australian Journal of Agriculture Research, in press.

Giordano M, Bowes G, 1997. Gas exchange and allocation in *Dunaliella salina* cells in response to the N source and CO2 concentration used for growth. Plant Physiology, 115: 1049–1056.

Giordano M, Pezzoni V, Hell R, 2000. Strategies for the allocation of resources under sulphur limitation in the green alga *Dunaliella salina.* Plant Physiology, 1124: 857-864.

Goldbol DL, Hutterman A, 1985. Effect of Zinc cadmium and mercury on root, elongation on picea abies (karst) seedling and the significance of these metals to forest die-back. Environmental Pollution, 38: 375-381.

Goswami NN, 1988. Sulphur in Indian agriculture. Proc. T'SI-FAI sym. On sulphur in Indian agriculture, New Delhi, pp: 1-90.

Grubb C, Abel S, 2006. Glucosinolate metabolism and its control. Trends in Plant Science, 11: 89–100.

Halkier BA, Gershenzon J, 2006. Biology and biochemistry of glucosinolates. Annual Review of Plant Biology, 57: 303–333.

Hanesklaus S, Schnug E, 1992. Baking quality and sulphur content of wheat. Sulphur in Agriculture, 16: 35-36.

Harada E, Kim JA, Meyer AJ, Hell R, Clemens S, Choi YE, 2010. Expression profiling of tobacco leaf trichomes identifies genes for biotic and abiotic stresses. Plant and Cell Physiology, 51(10): 1627-1637.

Harmer SL, Hogenesch JB, Straume M, Chang HS, Han B, Zhu T, Wang X, Kreps JA, Kay SA, 2000. Orchestrated transcription of key pathways in *Arabidopsis* by the circadian clock. Science, 290: 2110–2113.

Hartmann T, Honicke P, Wirtz M, Hell R, Rennenberg H, Kopriva S, 2004. Regulation of sulphate assimilation by glutathione in poplars (Populus tremula X P. alba) of wild type and overexpressing g-glutamylcysteine synthetase in the cytosol. Journal of Experimental Botany, 55: 837–845.

Hatzfeld Y, Lee S, Lee M, Leustek T, Saito K, 2000a. Functional characterization of a gene encoding a fourth ATP sulphurylase isoform from *Arabidopsis* thaliana. Gene, 248: 51–58.

Hatzfeld Y, Maruyama A, Schmidt A, Noji M, Saito K, 2000b. B-Cyanoalanine synthase is a mitochondrial cysteine synthase-like protein in spinach and *Arabidopsis* thaliana. Plant Physiology, 123: 1163–1171.

Hawkesford MJ, 2003. Transporter gene families in plants: the sulphate transporter gene family: redundancy or specialization? Physiologia Plantarum, 117: 155–163.

Hell R, 1997. Molecular physiology of plant sulphur metabolism. Planta, 202: 138-148.

Hell R, Hillebrand H, 2001. Plant concepts for mineral acquisitionand allocation. Current Opinion in Biotechnology, 12: 161–168.

Hell R, Jost R, Berkowitz O, Wirtz M, 2002. Molecular and biochemical analysis of the enzymes of cysteine biosynthesis in the plant *Arabidopsis* thaliana. Amino Acids, 22: 245–257.

Hermsen C, Koprivova A, Matthewman C, Wesenberg D, Krauss GJ, Kopriva S, 2010. Regulation of sulfate assimilation in Physcomitrella patens: mosses are different! Planta. 232(2): 461-470.

Herschbach C, Teuber M, Eiblmeier M, Ehlting B, Ache P, Polle A, Schnitzler JP, Rennenberg H, 2010. Changes in sulphur metabolism of grey poplar (Populus x canescens) leaves during salt stress: a metabolic link to photorespiration. Tree Physiology, 30(9): 1161-1173.

Hirai MY, Klein M, Fujikawa Y, Yano M, Goodenowe DB, Yamazaki Y, Kanaya S, Nakamura Y, Kitayama M, Suzuki H, Sakurai N, Shibata D, Tokuhisa J, Reichelt M, Gershenzon J, Papenbrock J, Saito K, 2005. Elucidation of gene-to-gene and metaboliteto- gene networks in *Arabidopsis* by integration of metabolomics and transcriptomics. Journal of Biological Chemistry, 280: 25590–25595.

Hirai MY, Fujiwara T, Awazuhara M, Kimura T, Noji M, Saito K, 2003. Global expression profiling of sulphur-starved *Arabidopsis* by DNA macroarray reveals the role of O-acetyl-L-serine as a general regulator of gene expression in response to sulphur nutrition. Plant Journal, 33: 651–663.

Hirai MY, Fujiwara T, Chino M, Naito S, 1995. Effects of sulfate concentrations on the expression of a soybean seed storage protein gene and its reversibility in transgenic *Arabidopsis* thaliana. Plant and Cell Physiology, 36: 1331–1339.

Hirai MY, Yano M, Goodenowe DB, Kanaya S, Kimura T, Awazuhara M, Arita M, Fujiwara T, Saito K, 2004. Integration of transcriptomics and metabolomics for understanding of global

responses to nut ritional stresses in *Arabidopsis* thaliana. Proceedings of the National Academy of Sciences of the USA, 101: 10205–10210.

Hofgen R, Krept O, Willmitzer L, Hesse H, 2001. Manipulation of thiol content in plants. Amino acids, 20: 291-299.

Holler K, Kiraly L, Kunstler A, Muller M, Gullner G, Fattinger M, Zechmann B, 2010. Enhanced glutathione metabolism is correlated with sulphur-induced resistance in Tobacco mosaic virus-infected genetically susceptible Nicotiana tabacum plants. Molecular Plant Microbe Interaction, 23(11): 1448-1459.

Hopkins L, Parmar S, Blaszczyk A, Hesse H, Hoefgen R, Hawkesford MJ, 2005. O-acetylserine and the regulation of expression of genes encoding components for sulfate uptake and assimilation in potato. Plant Physiology, 138: 433–440.

Howarth JR, Fourcroy P, Davidian JC, Smith FW, Hawkesford MJ 2003. Cloning of two contrasting high-affinity sulphate transporters from tomato induced by low sulphate and infection by the vascular pathogen Verticillium dahlia. Planta, 218: 58–64.

Huppe HC, Turpin DH, 1994. Integration of carbon and nitrogen metabolism in plants and algal cells. Annual Review of Plant Physiology and Plant Molecular Biology, 45: 577–607.

Iglesias-Baena I, Barranco-Medina S, Lazaro-Payo A, Lopez-Jaramillo FJ, Sevilla F, Lázaro JJ, 2010. Characterization of plant sulfiredoxin and role of sulphinic form of 2-Cys peroxiredoxin. Journal of Experimental Botany, 61(5): 1509-1521.

Irwin JG, Campbell G, Vincent K, 2002. Trends in sulphate and nitrate wet deposition over the united kingdom. Atmospheric Envirnment, 36: 2867-2879.

Jones P, Manabe T, Awazuhara M, Saito K, 2003. A new member of plant CS-lyases: cystine lyase from *Arabidopsis* thaliana. Journal of Biological Chemistry, 278: 10291–10296.

Joseph B, Verma SC, 1994. Response of rainfed chickpea to jalsakthi incorporation and P and S fertilization. Indian Journal of Agronomy, 39: 312-314.

Kaiser WM**,** 1997. Regulatory interaction of carbon- and nitrogen metabolism. Program in Botany, 58: 150–163.

Kanwar JS, 1976. Calcium, magnesium and sulphur in soil fertility-theory and Practice. ICAR, New Delhi, 202228.

Kapoor KK, Mishra MM, 1989. Microbial transformation of sulphur and plant nutrition. In: Soil microorganisms and crop growth. L.L. Somani and S.L. Bhandari. (Eds.). Diyajyoti Prakasam, India, pp: 1-30.

Kapoor KK, Mishra MM, Malik RS, Banger KC, 1991. Solubilization of Mussoorie rock phosphate by use of pyrite and *Thiobacilli*. Environment and Ecology, 9: 635-637.

Kataoka T, Watanabe-Takahashi A, Hayashi N, Ohnishi M, Mimura T, Buchner P, Hawkesford MJ, Yamaya T, Takahashi H, 2004b. Vacuolar sulfate transporters are essential determinants controlling internal distribution of sulfate in *Arabidopsis*. Plant Cell, 16: 2693-2704.

Kawashima CG, Berkowitz O, Hell R, Noji M, Saito K, 2004. Characterization and expression analysis of a serine acetyltransferase gene familyinvolved in a key step of the sulphur assimilation pathway in *Arabidopsis* thaliana. Plant Physiology (in press).

Khandkar MR, Shinde DA, Kanekar VS, Jambley NR, Jain NK, 1985. Growth, nodulation and yield of rainfed black gram as influenced by phosphate and sulphate fertilization in vertisol. Indian Journal of Plant Physiology, 28: 318-322.

Kocsis MG, Ranocha P, Gage DA, Simon ES, Rhodes D, Peel GJ,Mellema S, Saito K, Awazuhara M, Li C, Meeley RB, Tarczynski MC, Wagner C, Hanson AD, 2003. Insertional inactivation of the methionine S-methyltransferase gene eliminates the S-methylmethionine cycle and increases the methylation ratio. Plant Physiology, 131: 1808–1815.

Kopriva S, Koprivova A, 2004. Plant adenosine 5-phosphosulfate reductase: the past, the present, and the future. Journal of Experimental Botany, 55: 1775–1783.

Kutz A, Muller A, Hennig P, Kaiser WM, Piotrowski M, Weiler EW, 2002. A role for nitrilase 3 in the regulation of root morphology in sulphur-starving *Arabidopsis* thaliana. Plant Journal, 30: 95–106.

Lambers HM, Cambridge L, Konings H, Pons TL, 1990. Causes and consequences of variation in growth rate and productivity of higher plants. SPB Acad. Publ. The Hague, pp:364.

Lappartient AG, Vidmar JJ, Leustek T, Glass ADM, Touraine B, 1999. Inter organ signaling in plants: regulation of ATP sulphurylase and sulphate transporter genes expression in roots mediated by phloem-translocated compounds. Plant Journal, 18: 89–95.

Larsson M, Olsson T, Larsson CM, 1985. Distribution of reducing power between photosynthetic carbon and nitrogen assimilation in *Scenedesmus*. Planta, 164: 246–253.

Leustek T, 1996. Molecular genetics of sulphate assimilation in plants. Physiologia Plantarum, 97: 411-419.

Leustek T, 2002. Sulfate metabolism. In CR Somerville, EM Meyerowitz, eds, The *Arabidopsis*

Book. American Society of Plant Biologists, Rockville, MD, doi/10.1199/tab.0009.

Leustek T, Martin MN, Bick JA, Davies JP, 2000. Pathways and regulation of sulphur metabolism revealed through molecular and genetic studies. Annual Review of Plant Physiology and Plant Molecular Biology, 51: 141–165.

Leustek T, Saito K, 1999. Sulfate transport and assimilation in plants. Plant Physiology, 120: 637–643.

Lin M, Turpin DH, Plaxton W, 1989. Pyruvate kinase isozymes from the green alga *Selenastrum minutum*: II. Kinetic and regulatory properties. Achieves of Biochemistry and Biophysics, 269: 228–238.

Mahendra S, 1988. Sulphur management in coarse textured alluvial soils. Proc. of TSI-FAI symp. Sulphur in agriculture, Mar. 9-11. New Delhi. pp: S III/2(1-9).

Marchner H, 1995. Mineral nutrition of higher plants. 2 nd Ed. Academic press, San Diego.

Maruyama-Nakashita A, Inoue E, Watanabe-Takahashi A, Yamaya T, Takahashi H, 2003. Transcriptome profiling of sulphur-responsive genes in *Arabidopsis* reveals global effects of sulphur nutrition on multiple metabolic pathways. Plant Physiology, 132: 597–605.

Maruyama-Nakashita A, Nakamura Y, Yamaya T, Takahashi H, 2004b. A novel regulatory pathway of sulfate uptake in *Arabidopsis* roots: Implication of CRE1/WOL/AHK4-mediated cytokinindependent regulation. Plant Journal, 38: 779–789.

Matsubayashi Y, Ogawa M, Morita A, Sakagami Y, 2002. An LRR receptor kinase involved in perception of a peptide plant hormone, phytosulfokine. Science, 296: 1470–1472.

Mehta UR Singh HG, 1979. Response of greengram to sulphur in calcareous soils. Indian Journal of Agriculture Sciences, 49: 703-706.

Naidu MV, Ram H, 1995. Effect of sulphur and *Rhizobium* inoculation on rhizosphere and non rhizosphere microflora. Agropedology, 5: 53-58.

Naidu MVS, Ram H, Prasad J, 1998. Effect of sulphur application and *Rhizobium* inoculation to green gram on nitrifying capacity of rhizosphere and non rhizosphere soils. Journal of the Indian Society of Soil Sciences, 46: 313-314.

Natesan S, Palani N, Ranganathan V, 1985. Sulphur for tea. Proc. of TNAU-FACT National seminar on Sulphur in agriculture, Oct. 18-19, 1985, TNAU, Coimbatore. pp: 129.

Niiess DH, 1999. Microbial metal and non-metal resistance. Applied Microbiology and Biotechnology, 51: 730-750.

Nikiforova V, Freitag J, Kempa S, Adamik M, Hesse H, Hoefgen R, 2003. Transcriptome analysis of sulphur depletion in *Arabidopsis* thaliana: interlacing of biosynthetic pathways provides response specificity. Plant Journal, 33: 633–650.

Nocito FF, Pirovano L, Cocucci M, Sacchi GA, 2002. Cadmium-induced sulfate uptake in maize roots. Plant Physiology, 129: 1872–1879.

Noctor G, Gomez L, Vanacker H, Foyer CH, 2002. Interactions between biosynthesis, compartmentation and transport in the control of glutathione homeostasis and signaling. Journal of Experimental Botany, 53: 1283–1304.

Noji M, Inoue K, Kimura N, Gouda A, Saito K, 1998. Isoform-dependent differences in feedback regulation and subcellular localization of serineacetyltransferase involved in cysteine biosynthesis from *Arabidopsis thaliana*. Journal of Biological Chemistry, 273: 32739–32745.

Noji M, Saito K, 2002. Molecular and biochemical analysis of serine acetyltransferase and cysteine synthase towards sulphur metabolic engineering in plants. Amino Acids, 22: 231–243.

Ohkama N, Takei K, Sakakibara H, Hayashi H, Yoneyama T, Fujiwara T, 2002. Regulation of sulphur-responsive gene expression by exogenously applied cytokinins in *Arabidopsis* thaliana. Plant and Cell Physiology, 43: 1493–1501.

Ostowska D, Pietkiewicz S, Ciesinski M, Kucinska K, Gozdowaski D, 2008. Biomass accumulation and absorption of Photosynthetic active radiation by rapeseed Plantd depending on sulphur fertilization. World Journal of Agriculture Sciences, 4 (2): 133-136.

Pasricha NS, Fox RL, 1993. Plant nutrient, sulphur in the tropics and subtropics. Advances in Agronomy, 50: 209-255.

Petrovic N, kastori R, 1994. Effect of sulphur on nitrogen assimilation in young sugar beet plants (beta vulgaris L.). Biologia Plantarum 36(suppl.): 201.

Pitchel JR, Dick WA, 1991. Sulphur, iron and solid phase transformations during the biological oxidation of pyretic mine spoil. Soil Biology and Biochemistry, 23: 101-107.

Poorani B, 1992. Radiotracer studies on the interaction effects of application of P and S on available nutrients, yield and quality of greengram. M.Sc. (Ag.) Thesis Tamil Nadu Agricultural University, Coimbatore.

Randall PJ, 1988. Evaluation of the sulphur status of soils and plants. Techniques and interpretation. Proc. of TSI-FAI symp. Sulphur in Indian agriculture. New Delhi, pp: Sl/3 (1-15).

Rendig VV, Oputa C, McComb EA, 1976. Effect of Sulphur deficiency on non protein nitrogen, soluble sugrs and N/S ratio in young corn (*Zea mays*) plants. Plant Soil, 44: 423-437.

Rotte C, Leustek T, 2000. Differential subcellular localization and expression of ATP sulphurylase and APS reductase during ontogenesis of *Arabidopsis* thaliana leaves indicates that cytosolic and plastid forms of ATP sulphurylase may have specialized functions. Plant Physiology, 124: 715–724.

Saito K, 2000. Regulation of sulfate transport and synthesis of sulphurcontaining amino acids. Current Opinions in Plant Biology, 3: 188–195.

Saito K, 2004. Sulphur assimilatory metabolism. The long and smelling road. Plant Physiology, 136: 2443–2450.

Saito K, Kurosawa M, Tatsuguchi K, Takagi Y, Murakoshi I, 1994. Modulation of cysteine biosynthesis in chloroplasts of transgenic tobacco over expressing cysteine synthase. O-acetylserine (thiol) lyase. Plant Physiology, 106: 887–895.

Sanda S, Leustek T, Theisen MJ, Garavito RM, Benning C, 2001. Recombinant *Arabidopsis* SQD1 converts UDP-glucose and sulfite to the sulfolipid head group precursor UDP-sulfoquinovose in vitro. Journal of Biological Chemistry, 276: 3941–3946.

Saraf CS, 1988. Sulphur fertilization to pulses in yield and quality. Proceedings of TSI-FAI symposium. Sulphur in Indian agriculture, New Delhi. pp: S II/2-1.

Sarwar N, Saifullah, Malhi SS, Zia MH, Naeem A, Bibi S, Farid G, 2010. Role of mineral nutrition in minimizing cadmium accumulation by plants. Journal of Science, Food and Agriculture, 90(6): 925-937.

Scherer HW, 2001. Sulphur in crop production – invited paper. European Journal of Agronomy, 14: 81-111.

Scherer HW, Pacyna S, Manthey N, Schulz M, 2006. Slphur supply to peas (*Pisum Sativum L*.) influences symbiotic N2 fixation. Plant Soil Environment, 52 (2): 72-77.

Schobert C, Gromann P, Gottschalk M, Komor E, Pacsvaradi A, Zur Nieden U, 1995. Sieve tube exudates from Ricinus communis L. Seedling contain ubiquitin and chaperones. Planta, 196: 205-210.

Sexton J, 1996. The effects of sulphur deficiency on nitrogen metabolism in sugar beet. *PhD Thesis*, University of Wolverhampton.

Sexton PJ, Paek NC, Shibles R, 1998. Soybean sulphur and nitrogen balance under varying levels of available sulphur. Crop Science, 37: 1801-1806.

Sharma MP, Singh R, 1997. Effect of phosphorus and sulphur on greengram *(Phaseolus radiatus).* Indian Journal of Agronomy, 42: 650-652.

Shibagaki N, Rose A, Mcdermott JP, Fujiwara T, Hayashi H, Yoneyama T, Davies JP, 2002. Selenate-resistant mutants of *Arabidopsis* thaliana identify SULTR1;2, a sulfate transporter required or efficient transport of sulfate into roots. Plant Journal, 29: 475–486.

Shirzadian-Khorramabad R, Jing HC, Everts GE, Schippers JH, Hille J, Dijkwel PP, 2010. A mutation in the cytosolic O-acetylserine (thiol) lyase induces a genome-dependent early leaf death phenotype in *Arabidopsis*. BMC Plant Biology, 10: 80-88.

Shivakumar BG, 2001. Performance of chickpea (*Cicer arietinum*) varieties as influenced by sulphur with and without phosphorus. Indian Journal of Agronomy, 46: 273-376.

Shooner R, Tyagi RD, 1995. Microbial ecology of simultaneous thermophilic microbial leaching and digestion of sewage sludge. Canadian Journal of Microbiology, 41: 1071-1080.

Singh MV, 2001. Importance of sulphur balanced fertilizer use in India. Fertilizer News, 46: 13-18.

Singh SK, Singh R, Singh HP, 1994. Effect of sulphur on growth and yield of summer moong. Legumes Research, 17: 53-56.

Singh, Y.P. Aggarwal RL, 1998. Effect of sulphur and levels on yield, nutrient uptake and quality of black gram *(Phaseolus mungo).* Indian Journal of Agronomy, 43: 448-452.

Smith FW, Ealing PM, Hawkesford MJ, Clarkson DT, 1995. Plant members of a family of sulfate transporters reveal functional subtypes. Proceedings of the National Academy of Sciences of the USA, 92: 9373–9377.

Smith FW, Hawkesford MJ, Ealing PM, Clarkson DT, Vanden Berg PJ, Belcher AR, Warrilow AGS, 1997. Regulation of expression of a cDNA from barley roots encoding a high affinity sulfate transporter. Plant Journal, 12: 875–884.

Sokolova GA, Karavaiko GI, 1968. Physiology and geochemical activity of black gram *Thiobacilli.* In: Sulphur bacteria J.R. Postgate and D.P. Kelley (Eds). University Press. Cambridge. pp: 133-152.

Spencer D, Rerie WG, Randall PJ, Higgins TJV, 1990. The regulation of pea seed storage protein genes by sulphur stress. Australian Journal of Plant Physiology, 17: 355–363.

Spencer K and Freney, JR, 1980. Assessing the sulphur status of field grown wheat by plant analysis. Agronomy, 72: 469-472.

Starkey RL, 1966. Oxidation and reduction of sulphur compounds in soils. Soil Science, 101: 297-306.

Suter M, Von Ballmoos P, Kopriva S, Op den Camp R, Schaller J, Kuhlemeier C, Schu¨rmann P, Brunold C, 2000. Adenosine 5-phosphosulfate sulfotransferase and adenosine 5-phosphosulfate reductase are identical enzymes. Journal of Biological Chemistry, 275: 930–936.

Tabe LM, Droux M, 2001. Sulphur assimilation in developing lupin cotyledonscould contribute significantly to the accumulation of organic sulphur reserves in the seed. Plant Physiology, 126: 176–187.

Takahashi H, 2010. Regulation of sulfate transport and assimilation in plants. International Review of Cell and Molecular Biology, 281: 129-159.

Takahashi H, Watanabe-Takahashi A, Smith FW, Blake-Kalff M, Hawkesford MJ, Saito K, 2000. The role of three functional sulfate transporters involved in uptake and translocation of sulfate in *Arabidopsis* thaliana. Plant Journal, 23: 171–182.

Tandon HLS, 1991. In: Sulphur research and agricultural production in India. 3rd edition, TSI, Washington. pp: 140.

Tausz M, Sircelj H, Drill, 2004. The glutathione system as a strees marker in plant ecophsiology : is a stess response concept valid ? Journal of Experimental Botany, 55: 1955-1962.

Tiwari KM, 1997. Sulphur in balanced fertilisation in Northern India. Proc. TSI/PM/IFA symposium on sulphur in balanced fertilisation. New Delhi. pp: SI-I/I-SI-1/15.

Umarani N, 1994. Response of gram genotype to varying plant densities, sulphur application and rationing .MSc (Ag.) Thesis .Tamil Nadu Agricultural University, Combatore.

Urano Y, Manabe T, Noji M, Saito K, 2000. Molecular cloning and functional characterization of cDNAs encoding cysteine synthase and serine acetyltransferase thatmay be responsive for high cellular cysteine content in Allium tuberosum. Gene, 257: 269–277.

Varin S, Cliquet JB, Personeni E, Avice JC, Lemauviel-Lavenant S, 2010. How does sulphur availability modify N acquisition of white clover (Trifolium repens L.)? Journal of Experimental Botany, 61(1): 225-234.

Vauclare P, Kopriva S, Fell D, Suter M, Sticher L, Von Ballmoos P, Krahenbuhl U, Op den Camp R, Brunold C, 2002. Flux control of sulphate assimilation in *Arabidopsis* thaliana: adenosine 5-phosphosulphate reductase is more susceptible than ATP sulphurylase to negative control by thiols. Plant Journal, 31: 729–740.

Vernadskii VI, 1927. Istoriya mineralov zemnoi kory (Hisotry of the minerals of the earth crust), Vol.1, No: 2 (see Izbrannye Trudy, Vol. 3, Izd. An SSSR. 1955).

Vidmar JJ, Schjoerring JK, Touraine B, Glass ADM, 1999. Regulation of the hvst1 gene encoding a high-affinity sulphate transporter from Hordeum vulgare. Plant Molecular Biology, 40: 883–892.

Vidyalakshmi R, Paranthaman R, Bhakyaraj R, 2009. Sulphur Oxidizing Bacteria and Pulse Nutrition - A Review. World Journal of Agricultural Sciences, 5(3): 270-278.

Vogler KG, Umbreit WW, 1941. The necessity for direct contact in sulphur oxidation by *Thiobacillus thiooxidans.* Soil Science, 51: 331-337.

Vonbel AJE, Gamalei YV, 1992. Ecophysiology of Phloem loading in source leaves. Plant Cell and Environment, 15: 265-270.

Wainwright M, 1984. Sulphur oxidation in soils. Advances in Agronomy, 37: 350-392.

Weber M, Suter M, Brunold C, Kopriva S, 2000. Sulfate assimilation in higher plants characterization of a stable intermediate in the adenosine 5-phosphosulfate reductase reaction. European Journal of Biochemistry, 267: 3647–3653.

Wu Y, Zhao Q, Gao L, Yu XM, Fang P, Oliver DJ, Xiang CB, 2010. Isolation and characterization of low-sulphur-tolerant mutants of *Arabidopsis*. Journal of Experimental Botany, 61(12): 3407-3422.

Wulff-Zottele C, Gatzke N, Kopka J, Orellana A, Hoefgen R, Fisahn J, Hesse H, 2010. Photosynthesis and metabolism interact during acclimation of *Arabidopsis* thaliana to high irradiance and sulphur depletion. Plant Cell and Environment, 33(11): 1974-1988.

Yonekura-Sakakibara K, Onda Y, Ashikari T, Tanaka Y, Kusumi T, Hase T, 2000. Analysis of reductant supply systems for ferredoxin-dependent sulfite reductase in photosynthetic and nonphotosynthetic organs of maize. Plant Physiology, 122: 887–894.

Yoshimoto N, Inoue E, Saito S, Yamaya T, Takahashi H, 2003. Phloemlocalizing sulfate transporter, Sultr1;3, mediates re-distribution of sulphur from source to sink organs in *Arabidopsis*. Plant Physiology, 131: 1511–1517.

Yoshimoto N, Takahashi H, Smith FW, Yamaya T, Saito K, 2002. Two distinct high-affinity sulfate transporters with different inducibilities mediate uptake of sulfate in *Arabidopsis* roots. Plant Journal, 29: 465–473.

Zhang H, Tan ZQ, Hu LY, Wang SH, Luo JP, Jones RL, 2010. Hydrogen sulfide alleviates aluminum

toxicity in germinating wheat seedlings. Journal of Integrative Plant Biology, 52(6): 556-567

Zhao FJ, Hawkesford MJ, Warrilow AGS, McGrath SP and Clarkson DT, 1996. Responses of two wheat varieties to sulphur addition and diagnosis of sulphur deficiency. Plant soil, 181: 317-327.

Zimmermann MH, Brown CL, 1971. Trees: Structure and function Springer-Verlag, Berlin.

Zuber H, Davidian JC, Wirtz M, Hell R, Belghazi M, Thompson R, Gallardo K, 2010. Sultr4; 1 mutant seeds of *Arabidopsis* have an enhanced sulphate content and modified proteome suggesting metabolic adaptations to altered sulphate compartmentalization. BMC Plant Biology, 10: 78.

Table 1. Adequate tissue levels of macronutrients that may be required by plants (Epstein, 1972, 1994).

Element	Atomic Wt.	Conc. in dry matter	%	Relative no. of atoms
N	14.01	1,000	1.5	1,000,000
K	39.10	250	1.0	250,000
Ca	40.08	125	0.5	125,000
Mg	24.32	80	0.2	80,000
P	30.58	60	0.2	60,000
S	32.07	30	0.1	30,000
Si	28.09	30	0.1	30,000

Table 2. Macro-elements classified on the basis of their mobility within a plant and their tendency to re-translocate during deficiencies. Elements are listed in the order of their abundance in the plant.

Mobile	Immobile
N	Ca
K	S
Mg	Si
P	

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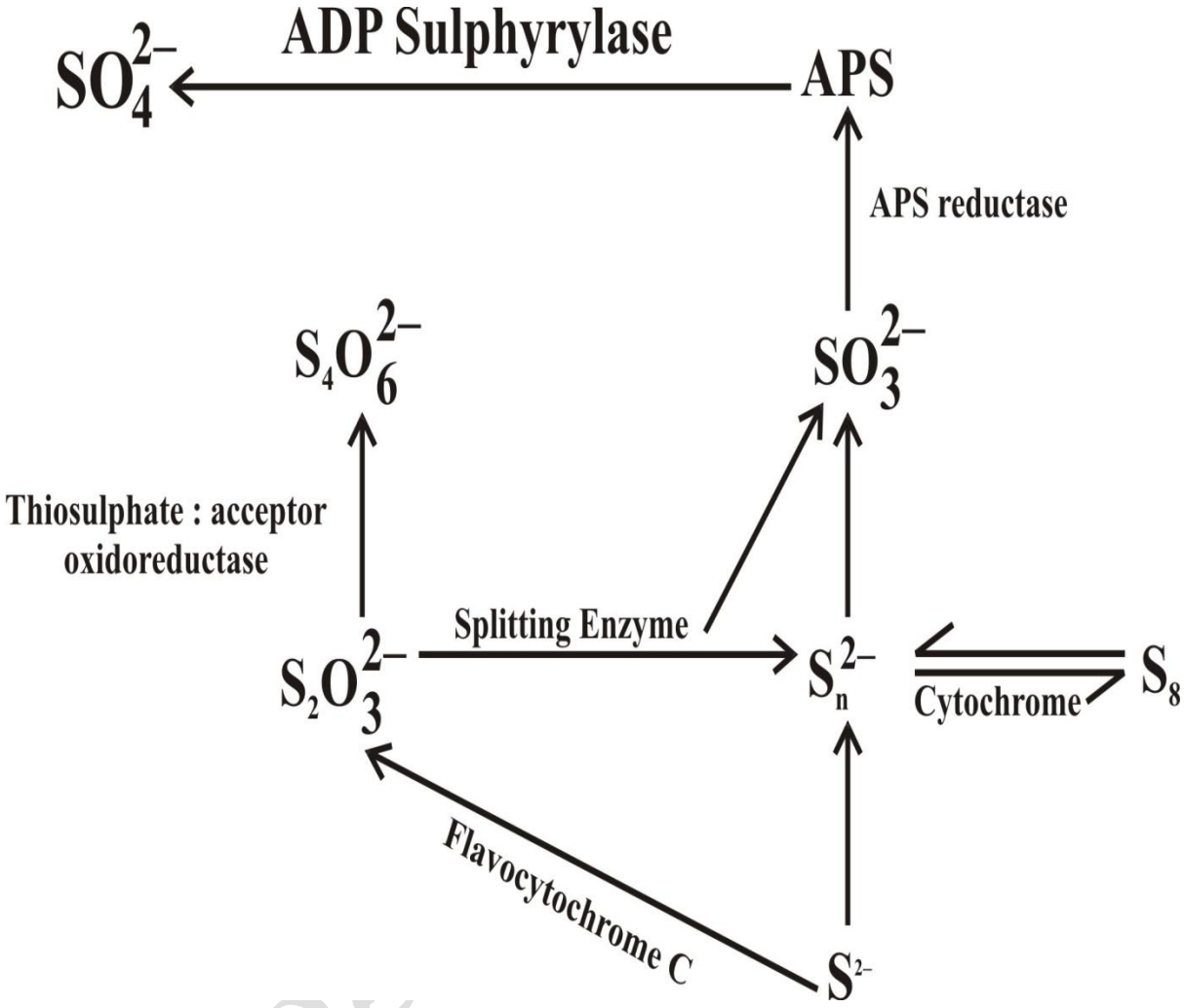

Figure 1. Sulphur transformation by action of enzyme acceptor oxidoreductase could be considered as a bypass of the APS reductase / ADP sulphurylase system. APS reductase occurs in sulphate reducing and in sulphur-oxidising bacteria.

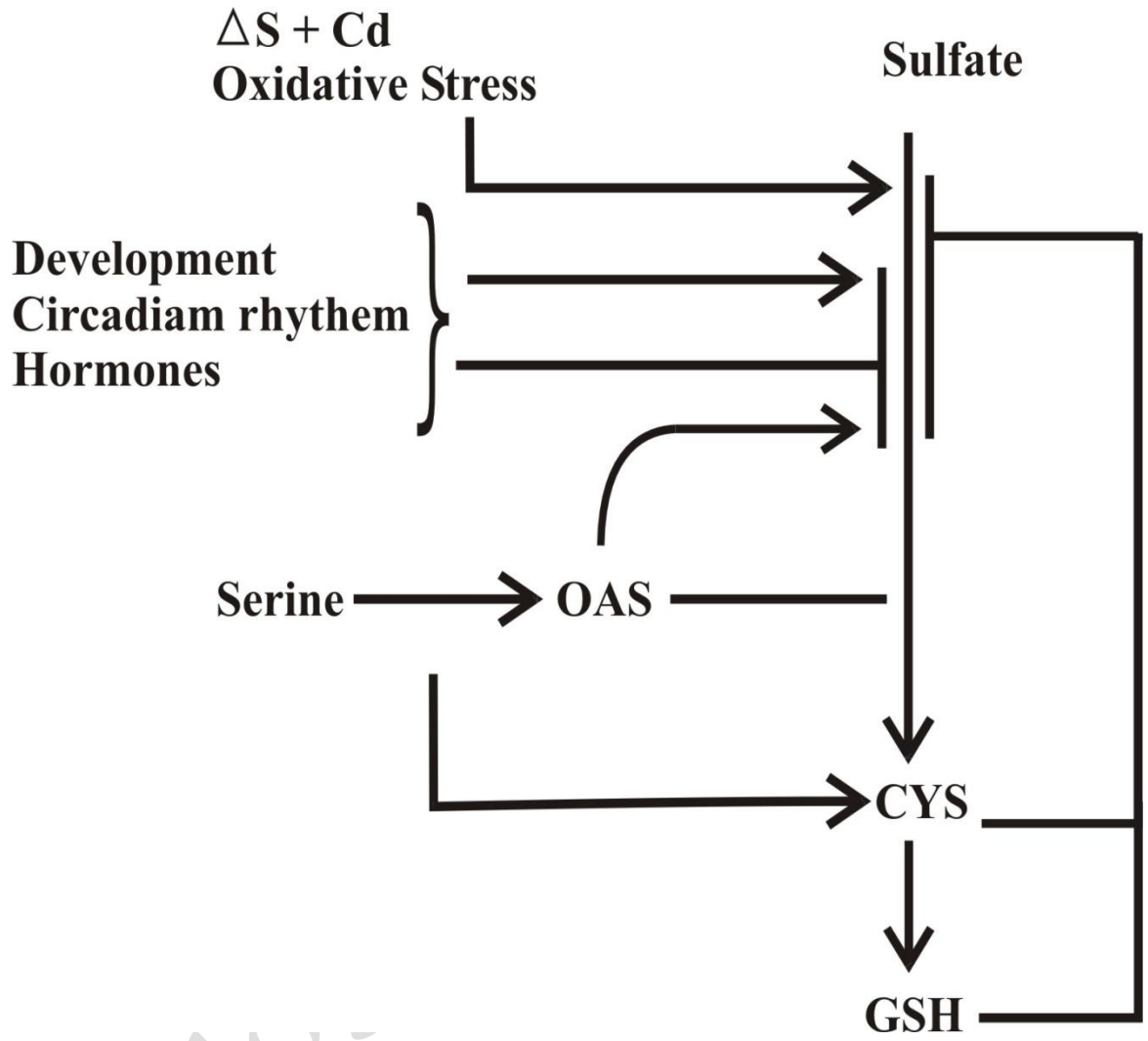

Figure 2. Positive and negative regulation of sulphur assimilatory metabolism, sulphur starvation and increased demand of sulphur metabolites induce assimilatory metabolism. OAS also acts as a positive factor for induction. Plant development, circadian rhythms, and hormones influence sulphur metabolism either positively or negatively. As negative factors, Cys and GSH regulate specific steps of sulphur metabolism. Arrow indicates the positive effect and bar indicates the negative effect.