

## Alterations in the Phosphorus Metabolism During Seed Germination of *Simarouba glauca*

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Phosphorus (P) is the second most important macronutrient for plant growth and is responsible for plant metabolism. Phosphorus is an important component of nucleic acid and also a component of phospholipids as a basic requirement for cellular organization and function as a membrane building block. Plants respond to Plant Growth Regulators (PGRs) in phosphorus content in a variety of ways. A study was conducted to assess the activities of acid and alkaline phosphatases on seed germination and seedling growth in response to various PGRs of *S. glauca* at different time intervals. Seeds were subjected to 100 ppm solutions of various PGRs, which include GA, 6-BA, CCC, SA, Cysteine, and Methionine. The activity of the enzymes ATPase, acid phosphatase, and alkaline phosphatase increases in response to the most of PGRs.

*Key words:* Phosphorus metabolism, ATPase enzyme, alkaline phosphatase, *Simarouba glauca*

Phosphorus metabolism is significant in plant growth and development across the plant's life cycle. Phosphorus uptake and distribution are completely reliant on a class of enzymes known as phosphatases, Phosphatases or phosphomonoesterases are the hydrolytic enzymes that cleave the ester bond between the phosphate group and the organic residue of the organic phosphates (Dotaniya *et al.*, 2019). ATP is almost exclusively found in the plasma membrane, cytoplasm, as well as cell wall. It has also been identified in vacuolar and other endomembranes such as chloroplast thylakoid membranes and inner mitochondrial membranes, including in the plasma membranes of bacteria and blue green algae (Logan, 2006). The energy stored in ATP's phosphoanhydride bond is used to power a wide range of processes including muscle contraction, cell motility, nerve impulse propagation, and DNA synthesis, among many others (Nirody, 2020). Based on the optimum pH for the activity, phosphatases are of two kinds: acid and alkaline. Acid phosphatases show maximum activity at acidic pH around 6 whereas alkaline phosphatases show maximum activity at alkaline pH around 11 (Dotaniya *et al.*, 2019). Acid phosphatases catalyze the hydrolysis of Pi from a broad range of phosphomonoesters with an acidic pH optimum (Tran *et al.*, 2010). Alkaline phosphatase (ALP; E.C.3.1.3.1.) is a ubiquitous membrane-bound glycoprotein that catalyzes the hydrolysis of phosphate monoesters at basic pH values (Sharma *et al.*, 2013).

## MATERIALS AND METHODS

Freshly harvested seeds of *S. glauca* were purchased from Sri Sri Institute of Agriculture, Bangalore. Surface disinfection with 0.1 percent mercuric chloride has been performed. Six treatments with four replicates have been used. The seeds were soaked in 100 ppm solutions of 6BA, GA, CCC, cysteine, SA and Methionone for 48 hours at room temperature. The twenty five seeds were sown in plastic

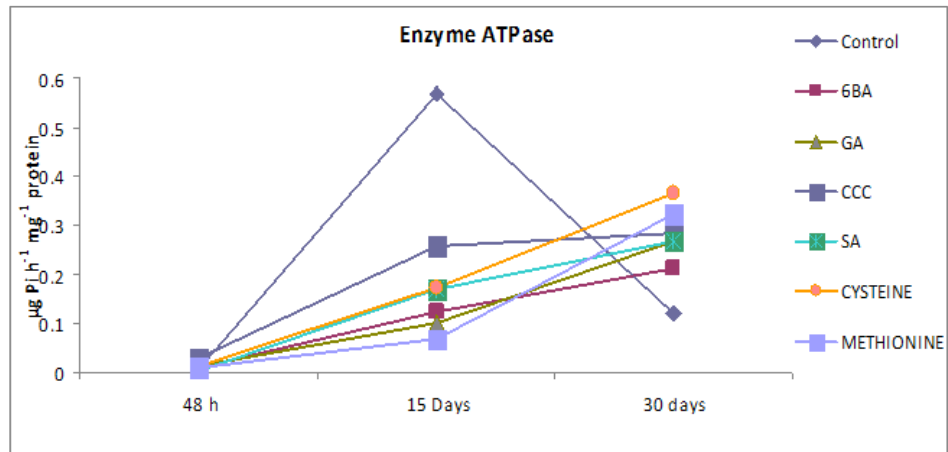
trays that have been densely filled by FYM and soil (1:3).The germinated seeds and seedlings were analyzed to find out the activity of enzymes. The activity of the enzyme ATPase, Acid phosphatase and alkaline phosphatase were determined using the standard published methods.

## RESULTS

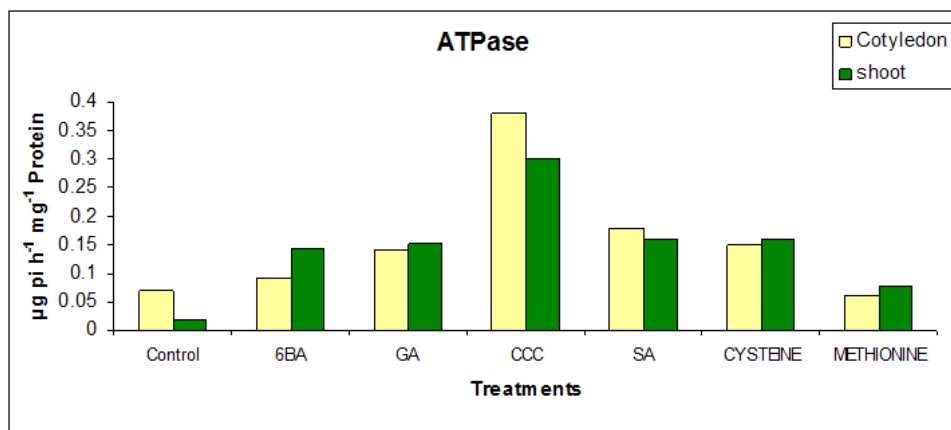
It is evident from fig 1 -2 In 48 h mechanically broken soaking seeds, the activity of enzyme ATPase increases in regard to 6-BA, GA, CCC, cysteine, and methionine treatments, but decreases in response to SA treatment. After 15 days, the activity of the enzyme ATPase in germinating seedlings declines in reaction to all PGRs, whereas after 30 days and 60 days, the activity of the enzyme ATPase decreases in response to all PGRs. In response to presowing soaking treatments, the overall activity of the enzyme ATPase is enhanced in seedlings, cotyledons, and shoots tissue.

From fig 3 and 4 it is observed that After 48 hour of soaking mechanically cracked seeds in PGRs, the enzyme acid phosphatase is activated. It raises in 15-day germination seedlings and then increases again in one-month-old seedlings. After 60 days, the activity of the enzyme acid phosphatase shows a diminishing tendency in cotyledons and an increasing trend in shoot tissues. It's also worth noting that acid phosphatase activity is higher in 15-day-old developing seedlings.

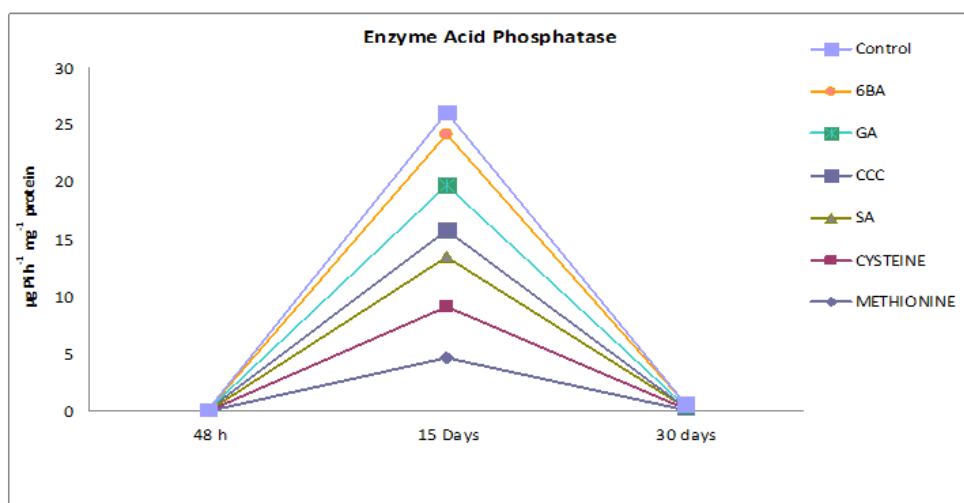
Fig no.5 and 6 indicates that after 48 hours of soaking mechanically split seeds, the enzyme alkaline phosphatase is reduced considerably to GA, CCC, SA, cysteine, and methionine, and induced in response to 6-BA. It is also clear that the activity of alkaline phosphatase is boosted in 15-day-old seedlings in response to all treatments, while the enzyme activity diminishes in 30-day-old seedlings. In 60-day-old seedlings, enzyme activity is decreasing in the cotyledons and increasing in the shoot tissues. It's also worth noting that the overall enzyme activity in 30 day old germinating seedlings is higher.



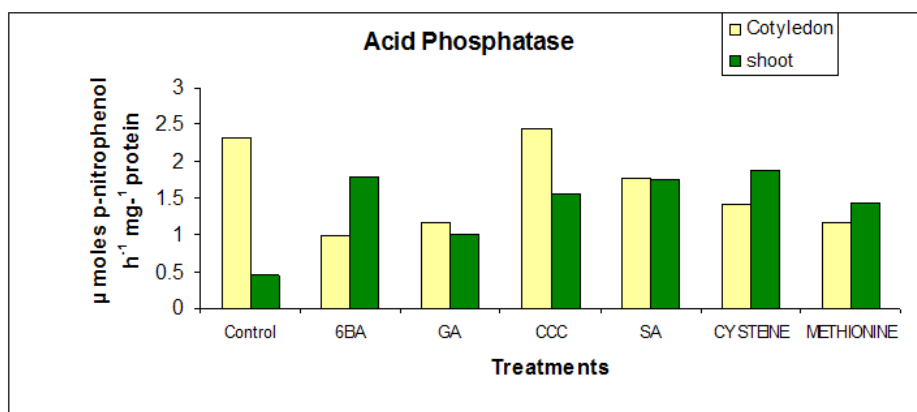
**Figure 1** Effect of presowing soaking treatments of PGRs on the activity of enzyme ATPase in seedlings of *S. glauca*



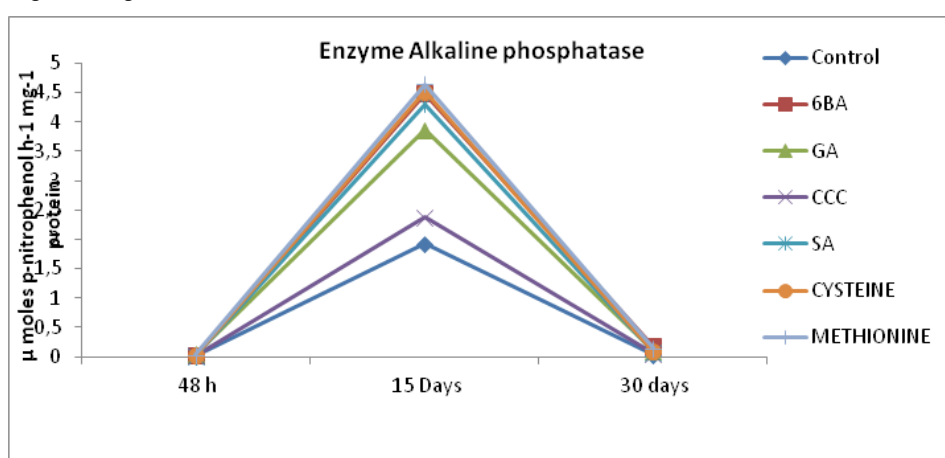
**Figure 2** Effect of presowing soaking treatments of PGRs on the activity of enzyme ATPase in 60 days old seedlings of *S. glauca*



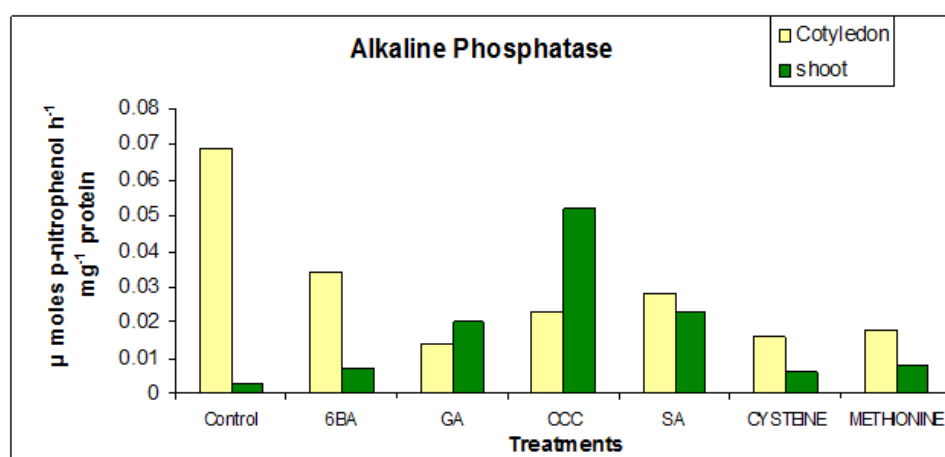
**Figure 3** Effect of presowing soaking treatments of PGRs on the activity of enzyme acid phosphatase in seedlings of *S. glauca*



**Figure 4** Effect of presowing soaking treatments of PGRs on the activity of enzyme acid phosphatase in 60 days old seedlings of *S. glauca*



**Figure 5** Effect of presowing soaking treatments of PGRs on the activity of enzyme alkaline phosphatase in seedlings of *S. glauca*



**Figure 6** Effect of presowing soaking treatments of PGRs on the activity of enzyme alkaline phosphatase in 60 days old seedlings of *S. glauca*

## DISCUSSION

Sen and Mukharji (2004) investigated ATPase and ATP content in Okra at two developmental stages (28 and 58 days after planting). To deal with the low

phosphorus level, the activities of both acid and alkaline phosphatases raised dramatically in the winter. The effect of hormones on sucrose absorption and ATPase activity in *Citrus sinensis* L. Osbeck leaves was

examined by Martinez-cortina and Sanz (1994). They discovered that IAA suppressed ATPase activity in isolated plasma membrane vesicles, but GA and ABA had no effect. SA had no influence on the level of ATPase in sugar beet, according to Bourbouloux *et al.* (1998). Salicylic acid, rather than transcription, may have an impact on plasma membrane H<sup>+</sup> ATPase activity. Salicylic acid concentration of 100M had no effect on the plasma membrane's cation permeability. SA treated *Vitis vinifera* L. cv. Jingxiu plants have stronger plasmalemma H<sup>+</sup> and Ca<sup>2+</sup>-ATPase activity than control plants (Liu *et al.*, 2008). When sucrose, indole butyric acid, or 6-BA were applied exogenously to the seeds of *Acacia mangium* following 3 days of colchicine treatment, the activities of H<sup>+</sup> -PPase, H<sup>+</sup> -ATPase, PPI, and ATP-dependent H<sup>+</sup> transport were stimulated in 6 day old seedlings, as per Wang *et al.* (2001). They furthermore concluded that IAA, 6-BA, and sucrose could overcome colchicine's inhibitory effect. Jain *et al.* (2004) noticed an increase in acid phosphatase activity in pearl millet seeds treated with GA, ABA, and NaCl. In embryos of *Sorghum bicolor* (L) Moench seeds pretreated with GA and NaCl, acid phosphatase activity increased (Sharma *et al.*, 2004). El-Shora and Metwally (2009) investigated the effect of phytohormones and group selective reagents on *Cladosporium cladosporioides* acid phosphatase. According to the researchers, adding GA to the growth medium stimulated acid phosphatase activity. In addition, they discovered that cysteine, L-ascorbic acid, DTT, and GSH act as reducing agents, stimulating enzyme activity in *Cladosporium cladosporioides*. It has also been reported that alkaline phosphatase is involved in the breakdown and mobilisation of starch and sucrose for the biosynthesis of essential oil in lemongrass (*Cymbopogon flexuosus* Steud) Wats (Ganjewala *et al.*, 2010). Jain *et al.* (2004) explored the role of plant growth hormones and salt stress on acid and alkaline phosphatase activities in pearl millet seeds. They realized an increase in alkaline phosphatase activity in the endosperm after treatments with ABA, GA, and NaCl.

## CONCLUSION

The higher amount of ATPase activity during germination indicates that ATP is available to carry out metabolic operations during seed germination, this increased activity of ATPase might play a contribution in plant nutrition and growth by sustaining the trans membrane, electrochemical gradient for nutrient absorption and mobilization. The stimulation of acid phosphatase activity during all stages of seed germination of *S. glauca* might be playing some role in breaking dormancy as indicated. While an increase in activity of acid phosphatase during early stages of germination may act on phytate in cotyledons which will help to mobilize in the form of organic phosphate during the growth of embryonic axis. The low levels of acid phosphatase activity in germinated seeds also indicated that the activity of acid phosphatase is good indicator of germination. It is noticed that the alkaline phosphatases are found to occur in lower levels during initial period of germination and during shoot formation, while, during vigorous seed germination stage the activity of alkaline phosphatase is elevated. As the alkaline phosphatase play a role in formation of inorganic phosphate for metabolic hydrolysis of metaphosphate to orthophosphate. An increased level of alkaline phosphatase might be beneficial in breakdown of these phosphates or carbohydrate compounds essentially required during germination.

## CONFLICTS OF INTEREST

The authors declare that they have no potential conflicts of interest.

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