

Changes in The Activity of Enzyme ATPase (E.c. 3.6.1.3) During Leaf Senescence in Sericultural Crop *Morus Alba* Linn.



Botany

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ABSTRACT

An attempt has been made to study changes in the enzyme ATPase during leaf senescence in mulberry (Morus alba Linn.). The leaf senescence was accompanied with the changes in the activity of enzyme ATPase (E.C. 3.6.1.3) during leaf senescence in the three mulberry cultivars namely M5 (K2), V1 and S36 is shown in Fig. It is evident from the figure that, the young leaves of three cultivars have the highest enzyme activity as compared to mature and senescent leaves. All the three categories of leaves of cultivar V1 show the highest enzyme activity as compared to the corresponding leaf categories in cultivars M5 (K2) and S36. In the senescent leaves of all the three cultivars a general decline in the enzyme activity is noticeable.

INTRODUCTION-

The important agro industry sericulture involves rearing of silkworms for the commercial production of the silk. Mulberry (*Morus alba* Linn.) leaves are used as food while rearing monophagous silkworm, *Bombyx mori* Linn. (Ullal and Narasimhana, 1981). Cocoon production depends mainly on nutrient composition of mulberry leaves. (Krishnaswami *et al.*, 1971; Bhuyan, 1981). Many aspects like health and growth of the larvae, cocoon quality and raw silk quality are also influenced by quality of leaf. In addition to involving verities, different practices have been worked out to raise leaf production including irrigation, pruning and training types, application of fertilizers, etc. (Koul and Bhagat, 1991; Singh and Koul, 1997; Pandit *et al.*, 1999). Ganga (2003) suggested that, over mature and yellow leaves with low protein content should be discarded to other nutritious feed to the worms. Mulberry leaves are extensively utilized as silkworm feed, these appears an excellent forage for feeding and supplementing ruminants, in fact, there are several places where mulberry leaves are used traditionally as feed in mixed forage diets for ruminants and there have also been several studies on the use of mulberry for cows and other domestic animals (Sanchez, 2000). A team of nutritional scientists in India has even suggested that the powdered leaves of white mulberry (*Morus alba* L.) might make good, nutritious, non toxic and low cost food ingredients for *Paratha*, the traditional food item of breakfast and dinner of the Indian diet (Srivastava *et al.*, 2003). The leaves also are reported to have some medicinal value and mulberry leaf tea is taken in some parts of India (Zepada, 1997 and Bellini *et al.*, 2000). Due to all the above reasons, understanding the study of senescence process of this economically important leaf material was thought worthwhile. For this purpose three promising cultivars of mulberry (M5 (K2), V1 and S36) have been selected. During present study nutritional constituents of young, mature and senescent leaves from three cultivars of mulberry (viz- M5, V1 and S36) studied has been compared. Hence, in order to have further insight in to the above problem, a fate of activity of enzyme ATPase during leaf senescence in the three cultivars of mulberry (viz.M5 (K2), V1 and S36) has been studied in the present investigation.

MATERIAL AND METHOD -

Activity of enzyme ATPase (E.C. 3.6.1.3) was determined following the method of Jha and Sinha (1989). One gram leaf material (of different categories) of each cultivar was homogenized in 10 ml cold tris-HCl buffer (0.1M, pH 7.5). The extract was filtered through a four layered muslin cloth and centrifuged at 5000 rpm for 10 minutes and the supernatant was used as the source of enzyme. Assay mixture was prepared by adding tris-HCl buffer (pH 7.5), ATP (3mM) and enzyme in equal volumes. The reaction was terminated by adding 10 % TCA after appropriate time. The resultant reaction mixture was centrifuged and the supernatant was analyzed for inorganic phosphate following method of Sekine *et al.*, (1965). One ml reaction mixture and 2 ml 2N HNO₃

were mixed with 1 ml of freshly prepared Molybdate Vanadate reagent. Volume was made to 10 ml with distilled water. After 20 minutes absorbance was measured at 420 nm against a blank prepared without phosphorus source. Phosphorus content was calculated by using a standard curve of phosphorus (0.025mg ml⁻¹). Enzyme activity was expressed μ mole phosphorus h⁻¹ mg⁻¹ protein.

RESULT AND DISCUSSION-

The changes in the activity of enzyme ATPase during leaf senescence in the three mulberry cultivars namely M5 (K2), V1 and S36 is shown in Fig.1. It is evident from the figure that, the young leaves of three cultivars have the highest enzyme activity as compared to mature and senescent leaves. All the three categories of leaves of cultivar V1 show the highest enzyme activity as compared to the corresponding leaf categories in cultivars M5 (K2) and S36. In the senescent leaves of all the three cultivars a general decline in the enzyme activity is noticeable. In the biological systems existing on this planet, Adenosine triphosphate (ATP) is one of the most important compounds. This is due to the fact that the cellular energetic depends on the state of this compound. Hence, the most of turnover of this compound is of paramount significance in deciding the proper functioning of any cell. It is obvious that ATP synthase, the enzyme responsible for ATP synthesis and ATPase, the enzyme responsible for ATP breakdown are very important in the bioenergetics of the cell. The plasma membrane H⁺ATPase (P-type H⁺ ATPase) is the principle primary active transport of plant cells. Vacuolar membrane also contain a vacuolar type (V-type H⁺ ATPase) which catalyses proton pumping. Such enzymes are also present in membranes from ER, Golgi bodies and coated vesicles of plant cells. ATPases from all sources have a complex multisubunit structure (Sanders and Bethke, 2000). The plasma membrane ATPase is specific to Mg²⁺ as well as Mn²⁺ ions. Spanwick, (1981) reported that in plants, plasma membrane ATPase play a role in acidification of cell walls, regulation of cytoplasmic pH and maintenance of the proton motive force. Palmgren, (1991) noted that when plant tissues are exposed to plant growth factors such as plant hormones, light and pathogens the activity of plasma membrane H⁺ATPase is altered rapidly. Abscisic acid has been reported to accelerate the onset and enhance the magnitude of the increase in ATPase activity which accompanies leaf senescence (De Leo and Sacher, 1970). Decompartmentation of the vacuolar ATPase has been proposed to provide a mechanism for Pi mobilization from endogenous phosphorylated compounds of senescent plant tissue (De Leo and Sacher, 1970). But one cannot ignore the fact that there are many other phosphatases present in the plant cells who can contribute to the mobilization of phosphorus from senescent leaves. The energy charge of the senescent tissue is also an important factor since, it regulates many metabolic processes in the cells. The available analytical data shows that in senescing fruits and leaves, the level of ATP increases sharply during the respiratory rise; (Solomos and Laties, 1976; Malik

and Thiamann, 1980; Warman and Solomos, 1988 and Bennett *et al.*, 1987). In senescing ivy leaves (*Hedera helix*) the increase in ATP is associated with a net increase in the total adenylate pool (Turner and Turner, 1980). The decrease in the rate of ATP hydrolysis can also contribute to such an increase in the ATP level. A decline in ATPase activity in the senescent leaves may probably contribute to such increase of ATP status. Further decrease in ATPase activity can affect the active uptake of various ions and solutes in the leaf cells from other cells as well as ion distribution within the cells of senescent leaves.

The values presented in the part –‘Results and Discussion’ represent average of three independent determinations.

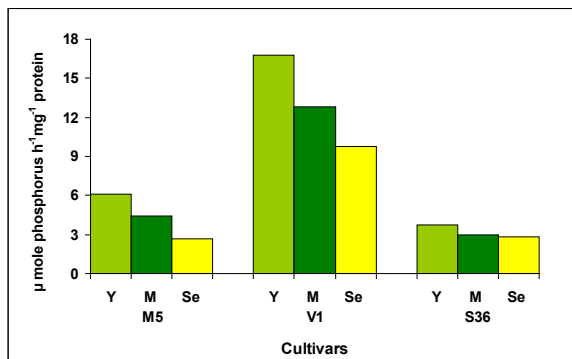


Fig. 1: Changes in the activity of enzyme ATPase during leaf senescence in sericultural crop *Morus alba* Linn. (Y = Young, M = Mature and Se = Senescent)

CONCLUSION-

A decline in ATPase activity in the senescent leaves may probably contribute to such increase of ATP status. Further decrease in ATPase activity can affect the active uptake of various ions and solutes in the leaf cells from other cells as well as ion distribution within the cells of senescent leaves

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