Research Paper Botany

Change The Activity of Enzyme Catalase (Ec 1.11.1.6) During Leaf Senescence in Sericultural Crop Morus Alba Linn

S. K. Khade

Department of Botany, Dattajirao Kadam Arts Science and Commerce College, Ichalkaranji, Dist: Kolhapur. (Maharashtra) – 416 115.

ABSTRACT

The feeding of the silkworms on the leaves of mulberry is one of the significant components of sericulture. Study the changes in the activity of enzyme catalase (EC 1.11.1.6) during leaf senescence, in the three mulberry cultivars namely – M5 (K2), V1 and S36. Among the three categories of leaf the activity of enzyme catalase is highest in young leaves in case of all the three cultivars of mulberry. While, a decline in catalase activity in senescent leaves has been noticed in all the three cultivars and the extent of this decline is significant in case of S36 cultivars.

KEYWORDS Enzyme Catalase (EC 1.11.1.6), Morus alba. Linn

Introduction-

The important agro industry sericulture involves rearing of silkworms for commercial production of the silk. Mulberry leaves are used as food for rearing monophagous silkworm (*Bombyx mori* L) (Ullal and Narasimhanna, 1981). Mulberry leaves used as food for rearing of silkworms, larvae growth and development of silkworm and subsequent cacoon production depends mainly on the nutrient composition of mulberry leaves (Krishnaswami *et al*., 1971; Bhuyian, 1981). Many aspects like health and growth of the larvae, cacoon 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), stated that, due to low protein level, declining (i.e. over mature, yellowing) leaves should be discarded. But at the same time there are several reports which indicate that leaf senescence in plants is promoted by several environmental constituents.

Material and method-

Enzyme Catalase activity was assayed by following the method of Luck (1974) as described by Sadasivam and Manickam (1992). Fresh leaves of three mulberry cultivars namely – M5 (K2), V1 and S36 (Different categories such as young, mature and senescent leaves) were collected, washed and blotted to dry and cut into small segments. 500 milligrams of leaf material was homogenized in 10 ml (1/15 M) phosphate buffer (pH 6.8) and filtered through four layered muslin cloth. The filtrate was centrifuged at 10,000 rpm for 20 minutes at 4^0C and supernatant was used as an enzyme source. The reaction mixture contained 3ml of 10% H_2O_2 [0.16 ml of H_2O_2 (6% w/v) were diluted to 100 ml with phosphate buffer (pH^7)] and 0.1 ml enzyme extract. It was mixed well and ΔOD was recorded at 240 nm. The enzyme activity was expressed as unit ΔOD $min^{-1}mg^{-1}$ protein as described by Bergmeyer (1974).

Result and discussion

The changes in the activity of enzyme catalase (EC 1.11.1.6) during leaf senescence in the three mulberry cultivars namely – M5 (K2), V1 and S36 are shown in the Fig. 1. Among the three leaf categories the activity of enzyme catalase is highest in young leaves in case of all the three cultivars of mulberry. While, a decline in catalase activity in senescent leaves has been noticed in all the three cultivars and the extent of this decline is significant in case of S36 cultivars.

Catalase (E.C. 1.11.1.6) hydrogen peroxide oxidoreductase represents one of the most important oxidoreductase enzymes involved in protective processes in the cell as it brings about degradation of harmful metabolic hydrogen peroxide (H_2O_2). It is indicated by Willekens *et al.,* (1997), that catalase can serve as a sink for H_2O_2. Lopez-Nicolas *et al.,* (2002) have demonstrated that in etiolated lupin hypocotyls, the H_2O_2 consumption by catalase was 20 to 30 times higher than guaiacol peroxidase and 200-300 times higher than ascorbate peroxidase indicating very clearly that catalase is a major H_2O_2 detoxifying enzyme. At the catalytic centre each monomer has heme as prosthetic group (Boon *et al.,* 2001). Thus typical catalase enzyme is a three dimensional structure having homotetrameric heme proteins. Catalase shows reactions.

(I) Decomposition of hydrogen peroxide to give water and oxygen.

$$2H_2O_2 \xrightarrow{\text{catalase}} 2H_2O + O_2 \uparrow$$

(ii) Oxidation of H donors for example methanol, formic acid, phenol with the consumption of one mole of peroxidase.

$$ROOH + AH_2 \xrightarrow{\text{catalase}} H_2O + ROH + A$$

Recent cytochemical and biochemical findings indicate that catalase in plant cell is located only in microbodies (peroxisomes, glyoxysomes). Catalase brings about H_2O_2 decomposition in the leaf peroxisomes, where H_2O_2 is generated during photorespiration by glycolate oxidase; photorespiration rates are quite significant in C_3 plants (Dat *et al.,* 2000) in contrast of C_4 species. In all organisms with aerobic metabolism catalase plays an important role of protecting living cells against toxic oxygen derivatives derived in the metabolism (Marcel, 1998). Santucci *et al.,* (2002) emphasized that catalase forms a major part of enzymic groups of oxidoreductase as it scavenges superoxide dismutase. H_2O_2 generation is also promoted in plants following exposure to a wide variety of abiotic and biotic stimuli such as, very high and very low of temperatures, UV radiations ozone exposure, excess light stress, plant growth inhibitors like ABA, water deficit, wounding and pathogenesis (Neill *et al.,* 2002). Bolwell *et al.,* (2002) suggested that potential sources of H_2O_2 generation include NADPH oxidase, cell wall peroxidases, amine oxidase, oxalate oxidase and flonin containing oxidase. H_2O_2 is relatively long lived molecule that can diffuse some distances from its production site. Sarkar and Choudhary (1981) have noticed enhancement of senescence of detached leaves due to H_2O_2. H_2O_2 is reported to bring about programmed cell death. (Desikan *et al.,* 1998) Hence, regulation of H_2O_2 level is of great significance for the

cellular survival. A decline in catalase activity during aging of the leaves has been reported by Braber, (1980) in bean leaves. Lin and Kao (2000) investigated the role of catalase in water stress promoted senescence of detached rice leaves. The decreased catalase activity was noticeable only when senescence was observed. Patra *et al.,* (1978) also noticed a decrease in catalase activity during leaf senescence of several species. Almeselmani *et al.*, (2006) made an extensive study of protwctive role of antioxideant enzymes under high temperature stress. The age dependent increase in the production of ROS and free radicals with a concurrent decrease in the levels of antioxidants and activities of free radical scanvenging enzymes cause oxidative damage to vital biomolecules (Wei *et al.,* 1998). The experimental results of Wang and Zhang (2000) indicate that the decreasing rate of the activities of catalase in the flag leaf of CTW (cold type wheat) is lower and the activities are higher than that of warm type wheat (WTW). According to Sangeetha and Ramarethinam (2000), the activity of enzyme which decompose activated oxygen species or hydrogen peroxide such as catalase, on investigation, showed variation during the different stages of leaf development. Such decline would lead to the accumulation of toxic molecular species H_2O_2 in the leaf tissue (Parida *et al.,* 1978).

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

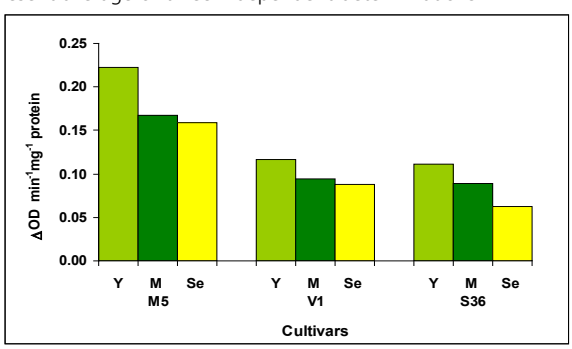

Fig. 1: Change the activity of enzyme catalase during leaf senescence in Sericultural crop *Morus alba* **Linn. (Y = Young, M = Mature and Se = Senescent).**

CONCLUSION-

In the senescent leaves of three mulberry varieties analyzed in the present investigation however a marked decline in catalase is noticeable. The activity catalase did not show any significant variation between that of the mature and senescent leaves of mulberry, even though, slight variations were there in the other stages of the leaves.

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