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Enzymes Involved in Phenol Metabolism of Gall and Normal Tissues of Insect Induced Leaf Galls on some Economically Important Plants in Rajasthan India

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Abstract: This paper reports biochemical stuldies of some metabolites and enzymes in insect induced leaf galls of *Ficus mysorensis Heyne* induced by unknown Psyllid, *Ficus racemosa Roxb.* induced by *Dyodiplosis fici Rao*, *Ficus religiosa Linn.* induced by *Pipadiplosis pipadiplosis Mani* and *Mangifera indica Linn.* induced by *Amradiplosis bruneigallicola Rao*, are trees of great economic importance. The parameters assayed are total phenols, orthodihydroxy phenols, peroxidase and polyphenol oxidase. Quantitative estimation of these metabolites and enzymes shows distinct variation of leaf galls of different plant studied.The amount of phenolic (total phenols and orthodihydroxy phenols) contents were higher in gall tissues as compared to their normal counterparts. Hyperphenolicity in gall tissues is correlated to high activities of peroxidase and polyphenol oxidase. Higher peroxidase activity was recorded in gall tissues of all the plants tested. *Ficus racemosa* and *Ficus mysorensis* normal tissue showed higher polyphenol oxidase activity while *Ficus religiosa* and *Mangifera indica* gall tissue showed higher peroxidase.

Keywords:Phenolics, phenols, orthodihydroxy phenols, peroxidase, polyphenol oxidase, phenolase, metabolites, leaf galls and neoplast.

INTRODUCTION

Phenolics play an important role both in healthy and diseased plants in their metabolism. Phenolic contents in parts of many plant species raises in response to microorganism. Many experimental data on pathophysiology support the contention that phenolic level is higher in diseased plants than in healthy ones as a result of parasitically enhanced phenol biosynthesis in case of the former and that phenol accumulation takes place in all infected plant tissues (Sequeira and Kelman, 1962, Farkas and Kerlay, 1962).

Specific interactions between animals and plants are very common in nature. Among these interactions, those of gall inducing insects and their host plants are believed to be the most intimate (Mani,2000). Gall causing parasites release growth-regulating chemicals as they feed, causing adjacent plant tissues to form a gall. Entomogenous galls are pathological structures which have originated from neoformed tissues as a mechanical and/or chemical insect stimuli (Kraus et al, 1954).

Gall inducers have the ability to control and manipulate growth of a plant. The degree of such manipulation varies widely from simple cell proliferation to the production of complete structure not normally produced in plants(Ananthakrishnan 1998, Rohfritsch and Antony 1992). Gall inducer may utilize a wide range of mechanism including behavioural, mechinical, chemical or genetic manipulation of the host plant to produce galls (Hori 1992). It has been established that along with the saliva insect inject chemicals into the leaf such as amino acids and plant growth substances (Schllar 1968, and Hori 1976) and the plant produces secondary metabolites as a defense mechanism

Leaf galls of *Ficus mysorensis Heyne* induced by unknown Psyllid, *Ficus recemosa Roxb.* induced by *Dyodiplosis fici Rao*, *Ficus religiosa Linn.* induced by *Pipadiplosis pipadiplosis Mani* and *Mangifera indica Linn.* induced by *Amradiplosis bruneigallicola Rao* have been studied here. Ramani and Kant (1989) have studied the phenolics and enzymes in *Prosopis cineraria* Linn. Druce rachis galls in vitro and in vivo conditions. Gupta (1997) have analysed the biochemical investigation of leaf galls of *Tectona grandis*. Purohit et al. (1979) also studied phenolics, peroxidase and polyphenolase in insect induced plants galls of some arid zone plants. The present observation is to understand the changes in phenolic levels and activities of enzymes involved in phenol metabolism which lead to the development of these neoplastic growths in some economically important plants of Rajasthan.

MATERIALS AND METHODS

Normal and gall tissues of *Ficus mysorensis Heyne* induced by unknown Psyllid, *Ficus racemosa Roxb.* induced by *Dyodiplosis fici Rao*, *Ficus religiosa Linn.* induced by *Pipadiplosis pipadiplosis Mani* and *Mangifera indica Linn.* induced by *Amradiplosis*

bruneigallicola Rao were collected form jaipur and adjoining areas. Quantative analysis of leaf gall and normal tissues were made adopting following methods. Estimation of total phenols by Bray and Thorpe 1954, orthodihydroxy phenols by Johnson and Schall 1952, peroxidase by Worthington Enzymes Manual 1972 and polyphenol oxidase by Shinshi and Moguch 1975.

RESULTS AND DISCUSSION

Total phenolic contents:

The results, presented in Fig. i showed that diseased part of *Ficus racemosa, Ficus mysorensis, Ficus religiosa* and *Mangifera indica* contain higher amount of total phenolic contents as compared to that of normal tissues. *Ficus racemosa* gall tissue showed maximum total phenols followed by, *Ficus mysorensis, Mangifera indica* and *Ficus religiosa*. Maximum difference in total phenolic contents between gall and normal tissues was recorded in *Ficus racemosa* and minimum in *Ficus religiosa*.

Oztho-dihydroxyphenolic contents:

Results obtained are presented in Fig. ii. Gall tissues of all the plants tested showed increased ortho-dihydroxyphenolic contents as compared to their normal counterparts. Maximum ortho-dihydroxyphenolic contents were recorded in gall tissue of *Ficus mysorensis* followed by *Ficus racemosa, Ficus religiosa* and *Mangifera indica* gall tissues. Maximum difference in ortho-dihydroxyphenolic contents between gall and normal tissue was recorded in *Ficus mysorensis.*

Peroxidase activity:

The results obtained are presented in Fig.iii. gall tissues of all the plants examined showed increased peroxidase activity as compared to their normal counter parts. Maximum activity of this enzyme was recorded in *Ficus religiosa* gall tissue followed by *Ficus mysorensis*, *Ficus racemosa* and *Mangifera indica*. Normal tissue showed maximum proxidase activity in *Ficus religiosa* followed by *Ficus racemosa*, *Ficus mysorensis* and *Mangifera indica*. Maximum difference in the activity of peroxidase enzyme between gall and normal tissue was recorded in *Ficus mysorensis* and minimum in *Mangifera indica*.

Polyphenol oxidase activity:

The results obtained are presented in Fig iv. Higher polyphenol oxidase activity was recorded in gall tissues of *Mangifera indica* and *Ficus religiosa* as compared to that of normal tissues. While normal tissues of *Ficus racemosa* and *Ficus mysorensis* showed higher polyphenol oxidase activity as compared to gall tissues. Maximum activity of this enzyme among the normal tissues was recorded in *Ficus mysorensis* followed by and *Ficus racemosa*, *Ficus religiosa* and *Mangifera indica*. *Ficus religiosa* showed maximum polyphenol oxidase activity among the gall samples tested followed by *Mangifera indica, Ficus mysorensis*, and *Ficus racemosa*.

The results obtained during present investigation indicate interesting aspects of phenol metabolism of gall and normal tissues. Gall tissues of all the plants tested showed higher total phenolic and ortho-dihydroxyphenolic contents as compared to their normal counterparts. Maximum total phenolic contents were recorded in *Ficus racemosa* and ortho-dihydroxy phenolic in *Ficus mysorensis* gall tissue as compared to all other plant samples tested. The higher peroxidase activity was recorded in gall tissues of all the plants tested as compared to the normal tissues, *Ficus religiosa* gall tissue showed highest peroxidase activity among all the plant samples tested. A definite correlation was found between phenolic contents and oxidative enzymes.

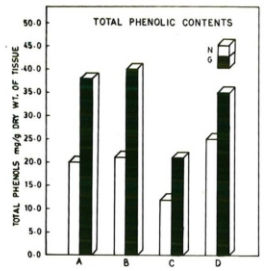

Fig. (i)

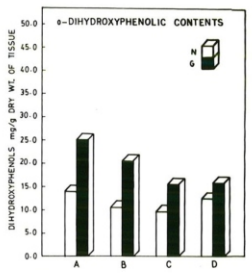

Fig. (ii)

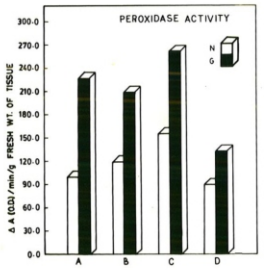

Fig. (iii)

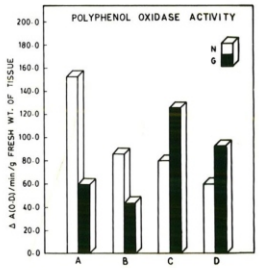

Fig. (iv)

Abbreviations used in figuers i to iv

A = Ficus mysorensis
B = Ficus racemosa
C = Ficus religiosa
D = Mangifera indica

myxa (Ramawat et al. 1979),leaf gall of *Zizyphus mauritiana*, *Achyranthes aspera* and *Ficus mysorensis* (Purohit et al 1980a). A higher level of phenol affected adversely the IAA oxidase activity in

plant tissue resulting in a higher level of IAA (Andreae 1952, Tomaszeniski and Thimann 1966 Ramani and Kant 1989, Sisodia and Patni 2006), thus leading to hyperauxinity and gall formation. Phenolics also act as substrates for some enzymes such as peroxidase. *Gopinathan* and Ananthakrishnan (1985) have reported increased polyphenol oxidase and peroxidase activities in galls of *Mimusops elengi* and *Calycopteris floribundus*. Increase in severity of gall disease with increased peroxidase activity has been recorded (Ramani 1987). Hence in the present study increased activity of peroxidase indicates that the cecidozoan has the capacity of detoxifying the effect of oxidized phenols.

Peroxidases are responsible for oxidation of phenolics (Kosuge 1969). Lower polyphenol oxidase activity in the gall tissue might be another factor for hyperphenolic level. Therefore, the phenolic compounds are regulated by polyphenol oxidase (Stonier *et al*. 1970). The higher level of orthodihydroxy phenols in the diseased tissues is responsible for lower polyphenol oxidase activity (Webb 1966). Increase in peroxidase activity is due to increased phenol concentration, which plays an important role in oxidising enzymes (*Kant et al*. 1992, Arora and Patni 2001). Increased peroxidase and polyphenol oxidase converts phenolics into quinones (Farkas and Kiraly 1962). Increase activity of these oxidative enzymes indicates a state of high catabolism induced during pathogenesis.

The quantity of total phenols was significantly higher in all four leaves gall tissues as compared to normal leaf tissues (Fig. 1). The increase in levels of phenols may be attributed to defense mechanism. According to Rana et al. (2005) the resistance to disease caused by aphids is due to the presence of high amount of phenols. Motta et al. (2000) assayed higher contents of soluble phenols, tannins, lignins, lipids etc. in galls and suggested that these substances represent the main energy source for the insect. This increase may be due to the action of enzymes such as polyphenol oxidase, peroxidase or pre-existing phenolic compounds or through the release of bound phenolic compounds. Activity of oxidative enzymes viz. peroxidase, polyphenol oxidase was found to be higher in galls of the cynipid wasp on chestnut oak had significantly greater peroxidase and polyphenol oxidase activities (Gasper et al.1985).Peroxidases play an important role in plant metabolism and physiology and are involved in the responses of plants to infectious and abiotic stress stimuli. Increased peroxidase activity could be due to increased phenol concentration, where phenols are factors of peroxidases and hence influence resistance in the host.

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