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Quantitative Estimation of Some Metabolites and Enzymes in *Trioza jambolanae* induced leaf galls of *Syzygium cumini* Linn.

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ABSTRACT

In order to test the hypothesis that cecidozoa induced neoplastic formations on trees affect biochemical characteristics of both, the new formed galls and host plant tissues. Biochemical characteristics with a possible adaptive role were determined during Trioza jambolanae (a psyllid) induced cecidogenesis on the host tree Syzygium cumini Linn. (Family- Myrtaceae). Photosynthetic pigments chl.a and b, extractable protein, total sugars, amino acids, phenols, prolines and oxidative enzyme activities were determined and compared in galled and ungalled leaf tissues. Results of biochemical estimation at different stages of cecidogenesis showed variations in the amount of these biochemicals. Gradual decrease in photosynthetic pigments and protein content were observed whereas low content of total sugars, extractable amino acids and prolines were evident during the initial stage of cecidogenesis which showed an increase in 30 days old gall, whereas decrease in amount of these were observed at gall maturity. Besides these quantification of oxidative enzymes activity of IAA and Polyphenol oxidase was also done. An increase in oxidative activities of both the enzymes was evident in 30 days old galls. The present data reflect long term systemic effects of gall formation on host tree physiology, suggesting that gall inducer affects adaptive responses of host plants.

Key Words: *Cecidogenesis, Neoplastic, Ungalled, Polyphenol oxidase, Biochemicals.*

INTRODUCTION

A variety of organisms can programme plant development, producing novel and abnormal structures such as galls. The most fascinating and elaborate galls are those produced by insects. Gall insects modifying minute areas of host plants by soliciting gene expression from adjacent cells in such a way that new developmental events occur [1]. New morphological potentialities of the host plant are brought into play enabling differentiation of new cell types that are unknown in the normal plant organ. New differentiation centres organize which results in the expression of a gall system with well defined nutritional as well as defensive systems. Gall inducers have the ability to control and manipulate the growth of a plant; the degree of such manipulation varies widely from simple cell proliferation to the production of complete structures not normally produced by the plant [2]. Gall inducers may utilize a wide range of mechanisms including behavioural, mechanical, chemical or genetic manipulation of the host plant to produce galls [3]. It would appear that the host plant and its parasites are engaged in a struggle.

It has been established that along with saliva, insects inject chemicals into the leaf such as amino acids and plant growth substance [4,5] and the plant produces secondary metabolites as a defense mechanism. The including molecules are thought to be released by plant cells when insects attack their cell walls [6].

Syzygium cumini is a plant cultivated all over India and propagated by seeds. The gall forming psyllid *Trioza jambolanae* an ectoparasite belonging to order Homoptera, attacks the leaves of most *Syzygium*

trees in late June in Kota district, Rajasthan, indicating large tunnel galls initiation from the base of leaf blade to the margins of leaf lamina.

The objectives of present investigation was to evaluate insect-pest interaction when *Syzygium* attacked by insect, the nature, quantification and comparison of biochemical signals in galled and non galled leaf tissues of the host plant.

MATERIALS AND METHODS

Normal (healthy) and heavily galled *Syzygium cumini* leaves of equal size were randomly collected in replicates from surveyed localities of Kota dist., Rajasthan. Leaf samples were brought to the laboratory, cleaned and surface dried with blotter and kept in refrigerator at 4^{o}C temperature for further processing. Following methodology was followed for estimation of different metabolites:-

Chlorophyll (a&b) were determined by the method of Arnon [7].

The extraction of total sugars was made in 80% ethanol and the assay for sugars was carried out by the Anthrone reagent method [8].

Total Proteins were estimated by the procedure of Lowry [9] using Bovin Serum albumin as the standard.

The amino acids were extracted and estimated by the procedure given by S.K.Sawhney.

Total Phenols were estimated by employing Folin Ciocatteu reagent as described by Bhatia [10] and were calculated from a standard curve. Proline content was extracted with sulphosalycilic acid and estimated spectrophotometrically in fresh shoot systems according to Sadasivam and Manickam [11]. IAA-oxidase and polyphenol oxidase activity in fresh samples was determined by the procedure of Gordon [12].

RESULT AND DISCUSSION

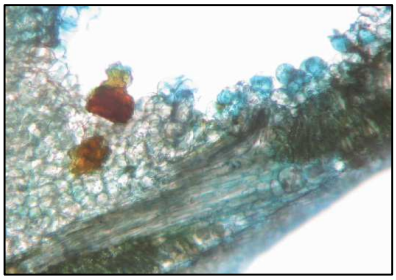
Data presented in Figure-1 depict the comparative quantities of Chlorophylls, proteins, sugars, amino acids, phenols, prolines and oxidative enzymes in healthy and galled leaf tissues at 15, 30 and 45 days of cecidogenesis. It is apparent from the data presented that, there was a marked decline in the quantities of chlorophyll a & b contents at different stages of cecidogenesis. Degradation of chlorophylls compounds in aphid induced galls have earlier been reported by Miles and Purohit [13,14].

Initially there was increase in Protein content as compared to healthy in 15 days old young galls after that there was marked decline in protein content in galled tissues.These results are in accordance with the findings of Anders [15] who found that the, saliva of aphid contains proteolytic enzymes like protease and peptidase which degrade proteins into amino acids.In the present study same may be the cause of decrease in Protein content at maturity of galls for providing nutrition to the insect. As illustrated in figure-1, the amount of amino acids and proline was higher in galled tissues as compared to normal leaf, which showed marked decline in mature galls. The deficiency of amino acids may be due to their utilization by the developing insect. Similar possibilities were given by Anders [15]. Significant increase in proline content just before gall maturity, can be explained on the basis of response given by the host plant against mechanical and physiological stress [16]. The higher content of Carbohydrate was also observed in galled tissues that abruptly decreased in fully mature galls. This increase may be providing absorbable nutrition to the insect. Amount of Phenols and oxidative activity of enzymes gradually increases from healthy to gall maturity.A marked accumulation is shown in mature galls which help in gall formation. Accumulation of phenols at increased level might be the tendency of the host to isolate the pathogen at original site of infection. A higher level of phenol affected adversely the IAA oxidase activity in plant tissue resulting in a higher level of IAA [17,18,19,20,21], thus leading to hyperauxinity and gall formation. Increased polyphenol oxidase converts phenolics into quinones [22].

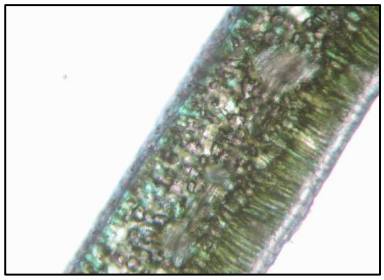
Table. 1. Biochemical Metabolites in healthy and galled leaf tissues during cecidogenesis (Mean value of 3 replicates)

Contents → Tissue ↓	Protein	Carbohydrate	Proline	Amino Acids	Phenol	IAA	PPO	Chl. A
Healthy	3.24	1.14	1.385	0.72	0.64	2.44	2.21	0.76
15 days	3.41	1.21	2.61	0.85	0.645	2.51	2.89	0.79
30 days	2.68	1.8	5.54	1.54	0.671	2.68	3.2	0.56
45 days	1.64	0.28	5.85	0.16	0.656	3.9	3.56	0.42

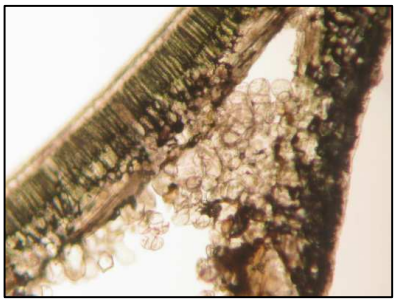
PLATE- Biochemicals Metabolites in Healthy and Galled leaf tissues of *Syzygium cumini.*



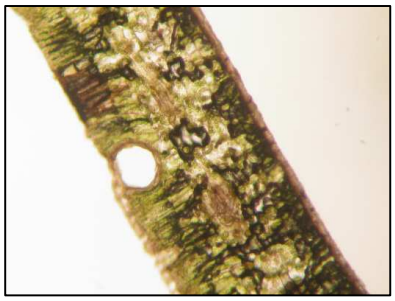
Lipid in galled tissue



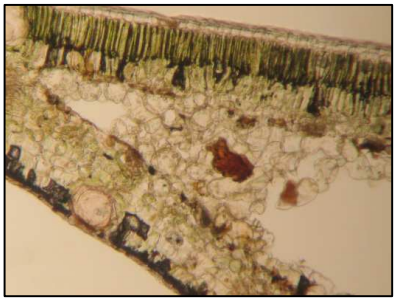
Lipid in healthy tissue



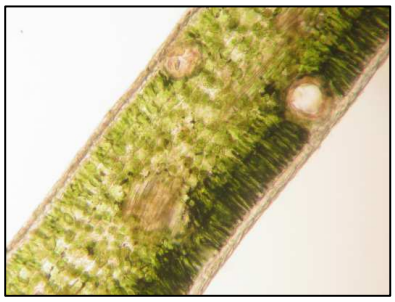
Phenol in galled tissue



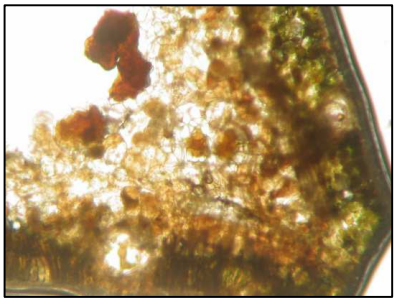
Phenol in healthy tissue



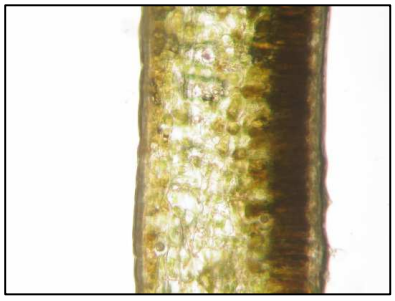
Polysaccharide in galled tissue



Polysaccharide in healthy tissue

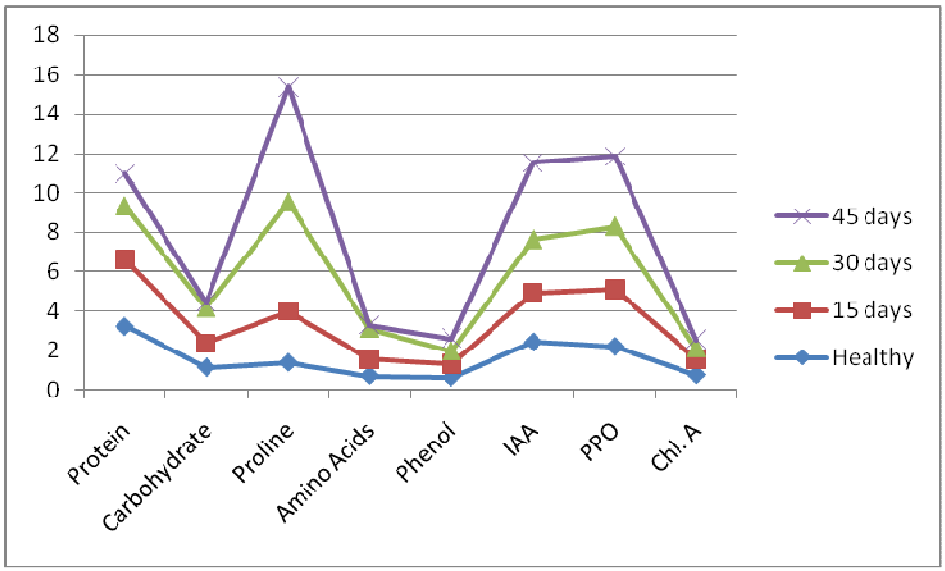


Starch in galled tissue



Starch in healthy tissue

Fig. 1: Graph representing biochemical metabolites



CONCLUSION

In the present study decrease in Protein content at maturity of galls is due to providing nutrition to the insect. The deficiency of amino acids may be due to their utilization by the developing insect. On the other hand increase in proline content just before gall maturity, indicates that the host plant gives response against mechanical and physiological stress. The increase in carbohydrate may be providing absorbable nutrition to the insect. Accumulation of phenol and IAA oxidase helps in gall formation.

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