Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2012, 4(9):4192-4197

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Quantitative Estimation of Some Metabolites and Enzymes in Insect Induced Leaf Galls of *Pongamia pinnata* (L.)

Richa Choudhary* and Sanjay Kumar

Department of Botany, M. S. J. Govt. P.G. College, Bharatpur, Rajasthan, India

---

ABSTRACT

This paper reports the quantitative estimation of some metabolites and enzymes in insect induced leaf galls of Pongamia pinnata. The parameters assayed were total soluble sugar, reducing sugar, starch, alpha-amylase activity, total phenol, o-hydroxy phenol, polyphenol oxidase, peroxidase, ascorbic acid and ascorbic acid oxidase as compared to normal tissues. Quantitative estimation of some metabolites and enzymes showed distinct variations in leaf gall at different ages with their normal counterparts. Present study was thus undertaken on biochemical changes in leaf galls of Pongamia due to infection of Aceria pongamiae.

Key words: *Pongamia pinnata*, Soluble sugar, *Reducing sugar*, Starch, alpha-amylase, total phenol, o-hydroxy phenol, *polyphenol oxidase*, peroxidase, ascorbic acid, *ascorbic acid oxidase*, *Enzymes*.

INTRODUCION

Pongamia pinnata is a fast growing nitrogen fixing tree that produces pleasantly fragrant hanging clusters of pink, purple or white flowers in spring leading to fruit in summer. It is widely distributed in coastal area of India. In India it is present abundantly in Rajasthan, Madhya Pradesh, Utter Pradesh, Gujarat, Bihar and Maharashtra. Its roots, bark, leaves, sap, flowers have medicinal properties. Its seeds and oil is anthelmintic, styptic and depurative which are useful to rheumatism arthritis, whooping cough, skin alinments and scabies. Seed oil is used in cosmetics, in soap making, as a lubricant and also used as insecticidal, nematicidal and bactericidal. Its non edible oil commonly used to fuel lamp and stoves in different parts of India. *Pongamia pinnata* is a biodiesel plant because of its oil has high contents of triglycerides.

A gall is a plant structure formed by abnormal growth within tissues. The growth is a reaction to a parasite attack in the plant's cells. Pongamia suffers galls on its leaf attacking by some pathogens which decrease it seed production to reduce its economic value. Specific interactions between insects and plants are very common in nature. Among in these interactions those of galling insects and their host plants are believed to be the most intimate (Fernandes, 1990, Singh and Sharma, 2008). Most gall forming insects are highly host and organ specific that they induce galls on only one species or a closely related group of host species (Dreger-Jauffref and Shorthouse, 1992). Carbohydrate (starch and cellulose) amounts are higher in gall tissues than normal tissues (Choudhary and Kumar, 2009).

EXPERIMENTAL SECTION

Normal and galled *Pongamia pinnata* leaves of equal size were collected from Keola Deo National park, Bharatpur, Rajasthan and their biochemical study was done. The biochemical parameters were studied in normal and gall leaf at different ages (10days, 20days and old).

The amount of total soluble sugars was estimated by Phenol sulphuric acid reagent method (Dubois *et al.*, 1951), starch by McCready *et al*. (1950), reducing sugar by Miller (1972). Alpha-amylase activity was determined by using

3,5-dinitrosalicylic acid (DNSA) colorimetric procedure of Bernfeld (1955), total phenol and o-hydroxy phenol by Bray and Thorpe (1954), polyphenol oxidase by Sexton and Hall (1978) method, peroxidase by ISSAAC and Winch (1947) method, ascorbic acid and ascorbic acid oxidase by Mahadevan (1982).Quantitative estimation of some metabolites and enzymes in insect induced leaf galls of *Pongamia pinnata.*

RESULTS

The results are presented in Fig. - 1 to 10.

Total soluble sugar contents:
Low amount of total soluble sugars were present in the gall tissues (10days, 20days and old) as compared to normal counterparts. Old gall showed higher amount of total soluble sugar as compared to 10days and 20 days leaf gall. Normal leaf showed increasing total soluble sugar contents with the aging of leaf. This result shows the sequential increase in metabolic activity with the age of normal leaf but overall decrease in gall tissue.

Starch:
High starch content was found in the normal tissue as compared to gall tissues (10days, 20days and old). Old gall showed highest amount of starch as compared to 10days and 20days gall tissues.

Reducing sugar:
Higher amount of reducing sugars was present in normal tissue as compared to old gall tissue. 20days gall showed highest amount of reducing sugar as compared to 10days and old gall tissues.

Alpha-amylase activity:
Alpha-amylase activity was recorded to be more in 10 days gall tissue as compared to normal tissue. Lower amount was recorded in old gall tissue as compared to normal tissue. Highest amount of alpha-amylase activity was observed in 10days gall as compared to 20days and old gall tissues.

Total phenol:
Lower amount of phenol was found in the gall tissue (20days and old) as compared to normal tissue. 10days gall tissue showed highest amount of phenol as compared to normal and 20days and old gall tissues.

o-hydroxy phenol:
o-hydroxy phenol was recorded to be more in normal tissue as compared to gall tissues (10days, 20days and old). Highest o-hydroxy phenol was observed in old gall as compared to 10days and 20days.

Poly phenol oxidase:
Higher amount of Poly phenol oxidase was recorded in gall tissues (10days, 20days and old) as compared to normal tissue. Highest Poly phenol oxidase was observed in 20 days gall as compared to 10 days and old gall tissues.

Peroxidase:
Peroxidase was recorded to be more in gall tissues (10 days, 20 days and old) as compared to normal tissue. Lower amount was recorded in 20days and old gall tissues as compared to 10 days gall tissue.

Ascorbic acid:
Ascorbic acid was recorded to be more in 20 days gall tissue as compared to normal tissues. Lower amount of ascorbic acid was recorded in gall tissues (10 days and old) as compared to normal tissues. Highest amount of ascorbic acid was observed in 20days gall as compared to 10days and old.

Ascorbic acid oxidase:
Lower amount of Ascorbic acid oxidase was recorded in normal tissue as compared to gall tissues (10days, 20 days and old).

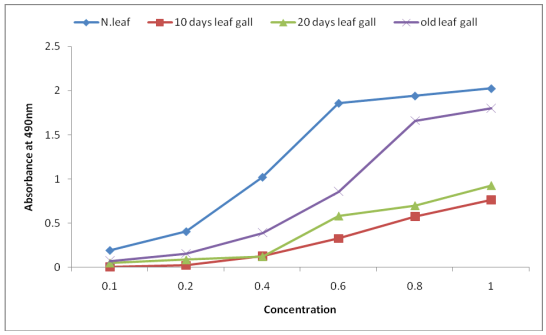

Fig: 1 showing amount of total soluble sugar contents in normal and gall (10days, 20days and Old) leaves.

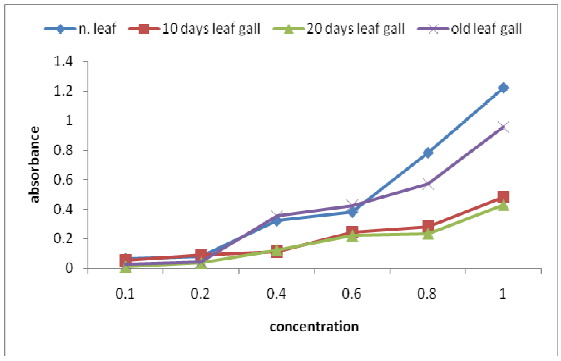

Fig: 2 showing amount of starch in normal and gall (10days, 20days and Old) leaves.

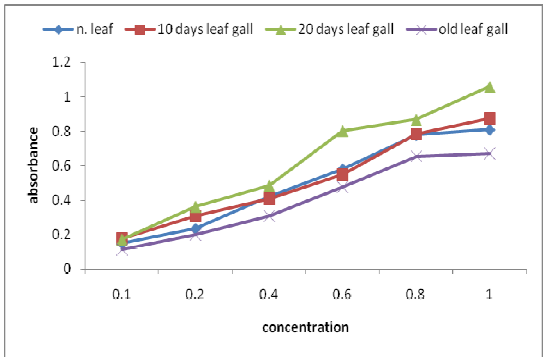

Fig: 3 showing amount of reducing sugar in normal and gall (10days, 20days and Old) leaves.

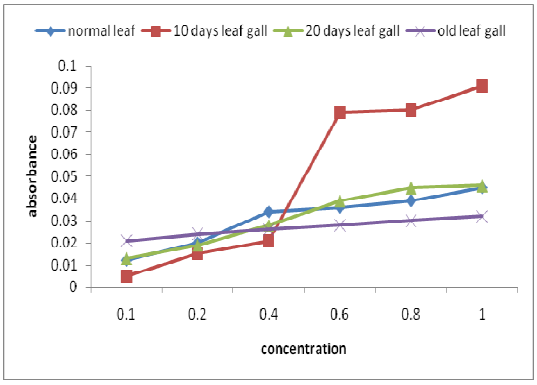

Fig: 4 showing amount of á-amylase activity in normal and gall (10days, 20days and Old) leaves.

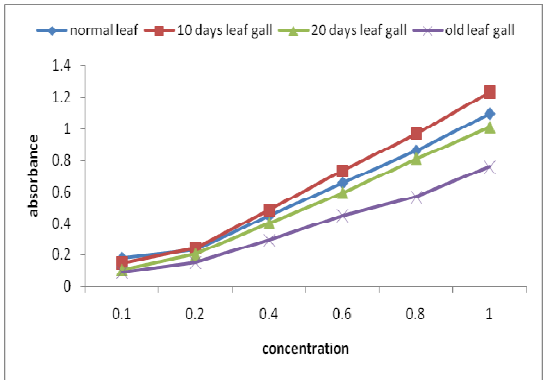

Fig: 5 showing amount of total phenol in normal and gall (10days, 20days and Old) leaves.

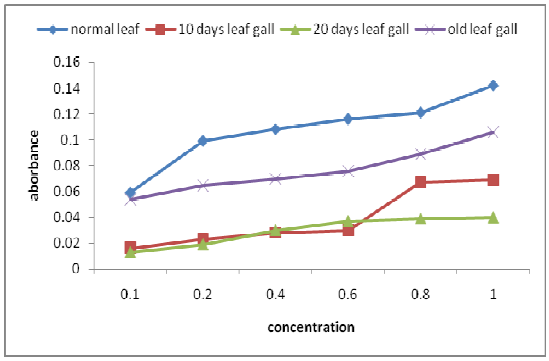

Fig: 6 showing amount of o-hydroxy phenol in normal and gall (10days, 20days and Old) leaves.

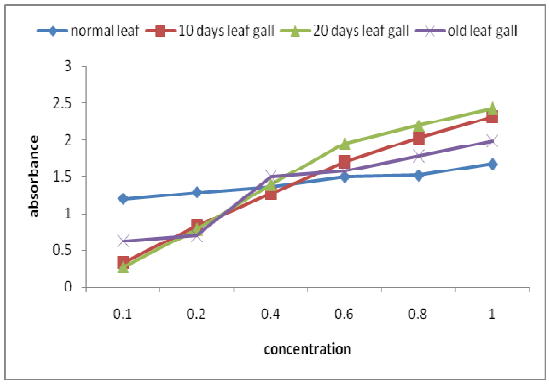

Fig: 7 showing amount of polyphenol oxidase in normal and gall (10days, 20days and Old) leaves.

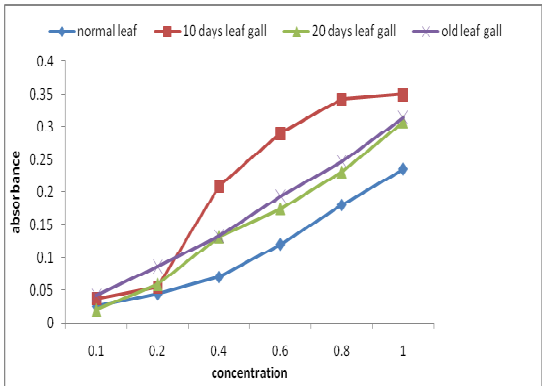

Fig: 8 showing amount of peroxidase in normal and gall (10days, 20days and Old) leaves.

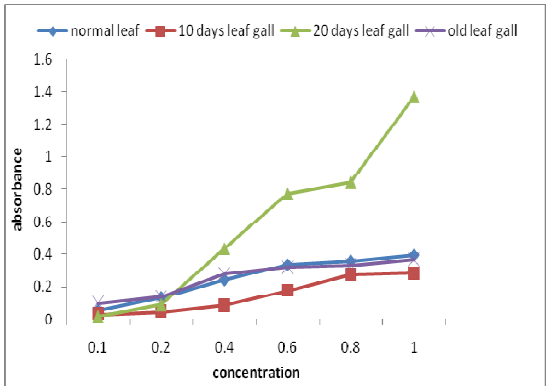

Fig: 9 showing amount of ascorbic acid in normal and gall (10days, 20days and Old) leaves.

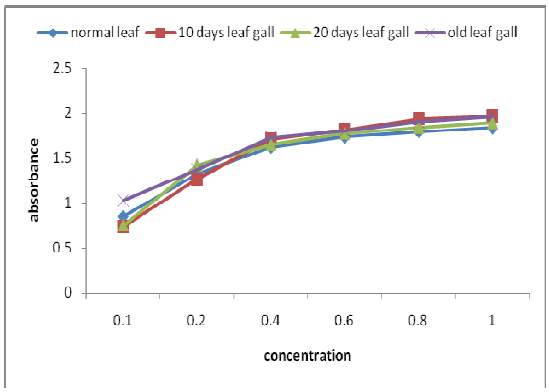

Fig: 10 showing amount of ascorbic acid oxidase in normal and gall (10days, 20days and Old) leaves.

DISCUSSION

The quantity of total soluble sugar was considerably high in normal tissue as compared to gall tissue. According to Mehrotra and Agarwal (2003), sugar has large numbers of stereo-isomer, because they contain several Asymmetric carbon atoms (Lindhrost and Thisbe, 2003).Galls have often been described as physiological sinks. Similarly carbohydrates may also accumulate by depletion of starch due to the activated alpha-amylase activity and other enzymes (Garg and Mandhar, 1975; Shekhawat, 1980 and Rao, 1989).

Feeny 1976 suggested that phenolics serve as chemical defense agents through their astringent test and by interfering with the digestive enzymes of the predators.Phenolics compounds plays important role in the defence mechanism of the plant. The results revealed that during the initial infections phenolics synthesized in more amounts but in the later stage polyphenol oxidase increased and phenolic compounds decreased or it may be possible the infecting agent may die naturally or comes out from the gall.

The natural sugars are usually considered to be the ultimate precursors of ascorbic acid, since they produce an increase in this acid when administered through the conductive tissues or the roots. Sucrose is often singled out as the principal precursor, and a quantitative relationship seems to exist between these two substances under a variety of experimental conditions.Ascorbic acid is linked to cell growth, being involved in the cell cycle and other mechanisms of plant cell growth and division, as well as acting as a co-factor for many enzymes (Smirnoff, 1996; Lee and Kader, 2000).

Preliminary insect infections in *Pongamiapinnata* alter the metabolism of the infecting part and almost biochemical increases. As the galls become old the enzyme activity increases the biochemical contents decreases. It may happen due to the aging of infected part or aging of insect.

CONCLUSION

Since the study was conducted in a controlled manner, the amount ofstarch content, reducing sugars, Alpha-amylase activity, Poly phenol oxidase, Peroxidase, Ascorbic acid were found higher in the galls in which insects were present. This study is preliminary and these results may be useful to check the insect infection on this bio-diesel

plant to increase the seed production for bio-diesel in future. The infected part of the plant can be used as a natural source of auxin content for the growth and development o f useful plants.

To conclude the present study, I have found that most of the biochemicals and enzymes were higher in the galls part of the plant as compared to their normal counterpart.

Acknowledgement
Financial assistance granted by UGC as major research project to carry out this research work is great fully acknowledged.

REFERENCES

[1] GW Fernandes, *Environ. Entomol*., **1990**,19, 1173-1182.
[2] MK Singh; SLSharma,*Asian J. Exp. Sci*., **2008**, 22(3), 343-346.
[3] F Dreger-Jauffret; JDShothouse. Diversity of gall-inducing insects and their galls. In: Biology of insect-induced galls JD. **1992.**
[4] R Choudhary; SKumar, *J. Phytol. Res*., **2009**, 22(2), 229-223.
[5] M Dubios; K Gilles; JK Hamilton; PA Rebers; FSmith, *Analyt. Chem*., **1951**, 26, 351-356.
[6] RM McCready; J Guggolz; V Silviera; HSOwens,*Anal. Chem*., **1950**, 22, 1156-1158.
[7] GL Miller,*Anal. Chem*., **1972**, 31, 426-428.
[8] P Bernfeld,*Amylases α and β. Method Enzymol*., **1955,**1, 149-158.
[9] HGBray; WV Thorpe, *Meth. Biochem. Anal.,* **1954,** 1, 27-52.
[10] R Sexton; JL Hall. Enzyme cytochemistry. In: Electron microscopy and cytochemistry of plant cells (ed.) J. L. Hall, (Amsterdam El-sevier North Holland Botanical Press), **1978**, 63-148.
[11] WE ISSAAC; NHWinch, *J. Pomol. Horti. Sci.*, **1947**, 37, 23-27.
[12] Mahadevan. The role of proteolytic enzymes in disease development. In: *Plant Disease Problems.* Proceedings First Int Symp PI Pathol. SP Raychauhuri, ed, **1982**, 179-184, Indian Agricultural Research Institute, New Delhi.
[13] RS Mehrotra; AAgarwal. Plant Pathology. TataMchraw-Hill publishing Co. Ltd. New Delhi, **2003**.
[14] TK Lindhorst; K Thisbe. Essentials of carbohydrate chemistry and biochemistry. New Delhi. Wiley Eastern Ltd., **2003**.
[15] ID Garg; CLMandhar,*Indian Phytopathl.*,**1975**,28, 565-566.
[16] NS Shekhawat. Studies on the nature of abnormal growth in plants during pathogenesis *in vitro* and *in vivo* state with special reference to green ear in pearl millet. Ph.D.Thesis, Jodhpur University, Jodhpur, India. **1980**.
[17] P Feeny. Plant apparency and chemical defense.In: J W Wallace and R L Mansell (eds). *Biochemical inter actions between plants and insects. Recent Advances in Phytochemistry*, **1976,** Vol. 10. Plenum, New York.
[18] N Smirnoff, *Ann. Bot.*, **1996**, 78, 661-669.
[19] SK Lee; AAKader,*Post Harv. Biol. Technol*., **2000**, 20, 207-220.