

## SUMMARY AND CONCLUSIONS

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The pernicious exotic weed Parthenium hysterophorus (L.) of the family Asteraceae (Compositae) is a native of tropical South and North America and West Indies. The weed has entered in India in 50s and spread like wild fire in almost all states. The rapid encroachment of the weed caused agricultural damage all over India, and even threatned the very existance of human life causing allergic reactions. Large seed output and its wide range of ecological adaptation to extreme environment are the major factors responsible for its fast spreading.

Since the weed P. hysterophorus competes with useful crop plants and causes a number of allergic diseases, its erradication is alarmingly important and has been attempted by number of workers hitherto. Though the weed appeared to be sturdier than any other weed, and has immunity to fungal and insect attack, it exhibited symptoms of phyllody disease.

Most of the workers have tried to concentrate upon studying the causal organism of phyllody disease of Parthenium and were successful in identifying mycoplasma like organism as a causal organism using electron microscope (Phatak et al. 1975). The careful observation of phyllody of Parthenium revealed that the disease transforms reproductive part into vegetative

part and the entire plant appear like a witch's broom. Varma and his colleagues (1974) have reported mycoplasmal etiology for P. hysterophorus and proposed its utility for the biological control. However, the phyllody of Parthenium has not been looked for the biochemical changes that may take place in host plant due to MLO infection. This has prompted us to select this problem to carry out experimentation on biochemical changes caused due to MLO infection.

Following experiments were carried out during the year 1992-93.

1. Frequency, abundance and density of phyllody disease of Parthenium.
2. Organic constituents such as chlorophylls and polyphenols.
3. Oxidative enzymes like polyphenol oxidase and IAA oxidase.
4. Detection of polyphenols and amino acids using paper chromatography.
5. Isolation and characterization of sesquiterpene lactone. and
6. Allelopathic effects of sesquiterpene lactone.

Method of approach -

A huge fallow land having dense population of P. hysterophorus was selected for the collection of plant material. Healthy and diseased plants were harvested from the same area and brought to the laboratory. Morphological changes induced by the disease were observed with naked eye and also under light microscope. The identity of MLOs was established by staining the trans-sections with Diene's stain. The parameters such as chlorophyll content, starch test and oxidative enzymes were performed using fresh tissue whereas polyphenols, amino acids and sesquiterpene lactone were analysed from dry tissue and the allelopathic effects of sesquiterpene lactone were studied by using bioassay technique. The parameters studied were compared with healthy plant.

The results are discussed under the light of available up-to-date literature.

**Conclusions** -

- 1) The frequency of the phyllody disease observed in P. hysterophorus is in increasing order and the maximum disease intensity occur in the month of June to December when the climate is humid and cool.

2. Microscopic observation of phyllody disease of P. hysterophorus revealed complete transformation of reproductive part into vegetative form, where stamens and stigma converted in to leaf like structure and flowers fail to produce any seeds. This indicate induction of hyperauxinity in host tissue by the causal organism.
3. The positive histochemical test with Diene's stain in the phloem tissue of infected plant confirmed the association of MLOs with phyllody of P. hysterophorus.
4. Reduction in chlorophyll content due to MLO infection in P. hysterophorus possibly be due to disruption of photosynthetic apparatus and loss of capacity to harness solar energy efficiently by the chlrophyll molecules.
5. Increase in chl. a/b ratio in infected plant is attributed to drastic reduction in chl. 'b' concentration caused due to MLO infection.
6. Photo-oxidative reduction of chlorophyll in MLO infected plant is comparatively at faster rate than that of healthy plants.

7. MLO infection lower down the starch forming ability of plant leaves by affecting the leaf chloroplasts, however this conclusion needs study of chloroplast structure and function under electron microscope.
8. MLO infection in P. hysterophorus accelerate synthesis of polyphenols which is evidenced by increase in polyphenol content by 31.22% over control.
9. Increase in the activity of an enzyme polyphenol oxidase in MLO infected P. hysterophorus is the result of increased level of polyphenols induced by MLO infection.
10. Decrease in the activity of an enzyme IAA oxidase in MLO infected plant is mainly due to increased level of polyphenols and the inhibitory action of caffeic acid detected in the MLO infected Parthenium plant. Thus induction of hyperauxinity due to MLO infection can be correlated with inhibition of IAA oxidase activity.
11. MLO infection induces synthesis of more number of phenolic compounds which is evidenced by

chromatographic analysis of phenolic compounds from healthy and MLO infected plant extract.

12. The dominant phenolic compounds in MLO infected plant extract were tannic acid, caffeic acid, ferulic acid, quercetin, myricetin, catechol and quinic acid.
13. MLO infection results in accumulation of amino acids viz. glutamate, asparatate, alanine, tyrosine and tryptophan.
14. The accumulation of tryptophan in MLO infected plant is an interesting observation and can be attributed its visilance as a pre-curser of auxin IAA and possibly the changes taking place in the auxin concentration due to MLO infection may be responsible for induction of phyllody disease in P. hysterophorus.
15. The sesquiterpene lactone (SL) isolated from both healthy and MLO infected plant exhibited yellowish white mass with radiating type of crystals.
16. The solubility test revealed that the SL is sparingly soluble in acetone, methanol,

chloroform, while insoluble in solvent ether, hexane, benzene, toluene, xylene and water.

17. MLO infection reduces the yield of SL by 58.3% as compared with healthy P. hysterophorus.
18. The SL has maximum absorption in the UV region at 215 nm which is the average  $\lambda_{\text{max}}$  of SL isolated from P. hysterophorus.
19. MLO infection has adverse effect on allelopathic effect which is evident from wheat germination study in different concentrations of SL.
20. The poor allelopathic effects exhibited by SL isolated from MLO infected plant may be due to either structural change in SL or due to inhibition of biological compound which is responsible for allelopathic effect.
21. The allergic reactions caused due to Parthenium plant are because of formation of adduct of SL with sulfur containing amino acids in which  $\alpha$ -methylene group reacts with  $\alpha$ -cysteine to form mono adduct with the endocyclic double bond on the cyclopentenone ring and become involved in the allergic reaction.
22. • Characterization of SL on NMR confirmed presence



of major sesquiterpene lactone 'parthenin' with structural formula  $C_{15}H_{18}O_4$ .

23. Although NMR analysis proved that both healthy and MLO infected plant contain the same SL parthenin, has exhibited marked difference in allelopathic effects.
24. HPLC analysis of SL isolated from healthy and MLO infected plant exhibited marked difference, which is evident from variation in the peak numbers. (11 peaks in SL isolated from healthy plant and only 6 peaks in SL isolated from infected plant.)
25. The peaks which are missing in SL isolated from MLO infected plant may be responsible for poor allelopathic effects.

Thus, the overall message of the dissertation is : MLO infection not only induce morphological changes but also hamper the physiological and biochemical mechanism of host plant. The most interesting observations noticed in the present piece of work in MLO infected P. hysterophorus are : accumulation of tryptophan, reduction in IAA oxidase activity, lowering of photosynthetic pigment, enhanced level of polyphenol

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compounds, poor allelopathic effects and variation in peak numbers in HPLC analysis. This study has created number of questions in the mind which are as follows.

1. Does really MLO induces hyperauxinity?
2. Whether MLO cause structural change in photosynthetic apparatus?
3. Is there any relation of micronutrient in causing hyperauxinity in MLO infected plant?
4. Though NMR spectra of SL isolated from healthy and MLO infected plant proved that the major SL in both the plant is parthenin, then why SL isolated from infected plant shows poor allelopathic effects?
5. What are those additional peaks observed in HPLC analysis of SL isolated from healthy P. hysterophorus ?

The study on these above points will throw much more light to understand the induction of physiological and biochemical changes in the MLO infected P. hysterophorus plant, which are under investigation in this laboratory.