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INFLUENCE OF THRIPS (*PSEUDODENDROTHRIPS MORI*) INFESTATION ON THE BIOCHEMICAL CONSTITUENTS AND PHOTOSYNTHETIC PIGMENTS OF MULBERRY (*MORUS* SPP.) LEAVES

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ABSTRACT: The occurrence of fringed wings insect, *Pseudodendrothrips mori* is a major pest of mulberry, which causes a considerable leaf yield loss and inturn damages its quality. Hence, an attempt was made to know the changes in biochemical constituents (Free amino acids, total soluble proteins, total reducing sugars, total soluble sugars, starch and total phenols) and photosynthetic pigments (total chlorophyll, chlorophyll – a, chlorophyll – b, chlorophyll – a/b ratio and carotenoids) in thrips infested mulberry foliage of six popular varieties (M_5, MR_2, Mysore local, S_{36}, S_{54} and V_1). It was found that there was a significant variation in biochemical constituents and photosynthetic pigments in the thrips infested mulberry leaves compare to healthy. These post-infestational changes in these components leads to imbalance in nutritional status of mulberry leaves which adversely influence the growth and development of silkworm as well as quality of silk produced.
Key words: biochemical constituents, infestation, mulberry, photosynthetic pigments, pest, thrips

INTRODUCTION

Mulberry (*Morus* spp.) is a perennial, deep rooted, fast growing and high biomass producing foliage plant. It forms the basic food material for the silkworm, *Bombyx mori* L. Increased production of raw silk, to large extent, depends on timely supply of quality mulberry leaves to silkworms. It is therefore clear that mulberry leaf plays a dominant role in cocoon production as a source of nutrition to the silkworm. The quality of mulberry leaf is influenced by several factors such as variety, agronomic practices, biotic and abiotic components (Krishnaswami *et al*. 1970). In-spite of adopting all these, sometimes, the nutritive values are degraded due to diseases and pest damage. Insect pests take a heavy toll of mulberry. Thrips (*Pseudodendrothrips mori* Niwa) are one of the important sap sucking insect pests of mulberry, belonging to the order Thysanoptera : Tripiidae. The estimated leaf loss due to this pest is about 40 – 50 % of the total leaf produced. Thrips causes a damage on a single leaf blade by using their mouth parts, rasp the epidermis on the ventral side. During laceration, they secret saliva, which coagulates the sap resulting in the formation of white streaks in the early stage followed by salivary blotches (Hadimani *et al*. 2006). The usual transpiration process of leaves is quickened through these wounds especially during high temperature seasons. Finally the leaves are dwindled and dried just like a piece of paper, being unfit to be used as feed to silkworms (Ye and Gu, 1990). An attempt was made to know the influence of thrips infestation on the biochemical components and photosynthetic pigments of mulberry leaves.

MATERIALS AND METHODS

Six popularly cultivated indigenous mulberry varieties (M_5, MR_2, Mysore local, S_{36}, S_{54} and V_1) were chosen. The healthy and thrips-infested (*Pseudodendrothrips mori* Niwa) leaves were collected from mulberry gardens situated in and around Bangalore rural (Kanakapura and Ramanagaram taluk) and Tumkur district (Karnataka State), India. The leaves were oven - dried and processed to analyze the free amino acids by Ninhydrin method (Moore and Stein, 1948) and total soluble proteins (Lowry *et al*. 1951) The reducing sugars were estimated by using DNS (Dinitro-salysilic acid) method (Miller, 1972).

While the soluble sugars and starch contents were determined by anthrone method (Yemm and Willies, 1954). Folin-ciocalteau was used to determine the total phenols (Bray and Thorpe, 1954). The photosynthetic pigments viz., chlorophyll (total chlorophyll, chlorophyll – a, chlorophyll – b and chlorophyll – a/b ratio) and carotenoids were estimated in fresh leaves by following Arnon's (1949), Mahadevan and Sridhar's (1984) procedures respectively. The results were analysed statistically by applying Student's t-test.

RESULTS AND DISCUSSION

BIOCHEMICAL COMPONENTS (Table – 1)

Table – 1: Biochemical changes (dry weight) in the thrips infested mulberry leaves.

Mulberry varieties	Free amino acids (μg/g)		Total proteins (mg/g)		Total reducing sugars (mg/g)		Total soluble sugars (mg/g)		Starch (mg/g)		Total Phenols (mg/g)	
	Healthy	Infested	Healthy	Infested	Healthy	Infested	Healthy	Infested	Healthy	Infested	Healthy	Infested
M_5	15.48	18.04**	123.50	104.00**	40.40	28.00**	1.58	1.30*	1.50	1.58**	2.42	2.02**
	(+16.54)		(-15.79)		(-30.69)		(-18.00)		(+5.36)		(-16.53)	
MR_2	4.08	6.48	136.50	134.00	78.00	71.80	1.52	1.35*	2.39	2.20	3.44	3.12**
	(+58.83)		(-1.90)		(-9.23)		(-11.46)		(-7.87)		(-9.30)	
Mysore local	17.44	14.28**	122.20	83.90**	66.00	48.40**	1.76	1.49**	1.66	1.74	3.12	3.48
	(-37.39)		(-31.39)		(-26.67)		(-15.32)		(+4.840)		(+11.54)	
S_{36}	9.00	9.36**	96.85	96.90	42.40	32.40**	2.25	1.95**	1.31	1.34	2.90	2.90
	(+4.00)		(+0.89)		(-23.58)		(-13.38)		(+2.04)		(---)	
S_{54}	14.40	13.20**	81.90	74.10**	64.40	44.00**	2.44	2.44	1.02	1.02	1.14	1.02
	(-8.33)		(-9.52)		(-31.68)		(---)		(---)		(-10.53)	
V_1	9.28	6.96**	104.00	127.00**	31.60	72.80	2.68	2.36**	0.99	0.05	2.68	2.76
	(-25.00)		(+22.50)		(+130.38)		(-11.83)		(+5.41)		(+2.99)	

** Significant at 1% level; * Significant at 5% level ; Values in parenthesis indicate difference over healthy (+ = more than; - less than; --- No changes/ alteration).

It is an established fact that the biochemical constituents and photosynthetic pigments of mulberry leaves play a major role in the improved production of silk. They were changed drastically due to thrips – infestation in all the mulberry varieties considered.

Free amino acids

The free amino acids were increased significantly in thrips infested leaves of M_5 and S_{36} (4.00 %) and non-significantly in the leaves of MR_2 (58.33 %) variety. The thrips infested leaves having a significant decreased free amino acid contents in the leaves of Mysore local, S_{54} and V_1. The decrease was minimum in S_{54} (8.33 %) and maximum (37.39%) in the leaves of Mysore local variety.

The amino acid content was increased (Kurangi, Thysong-3, Vietnam and Thailand) as well as decreased (M_5, Kajali, Hakkikalu and *M. nigra*) in the leaves of mulberry varieties due to thrips attack (Latha, 1999). The silkworm, *B. mori* requires 10 essential amino acids which can be synthesised by silkworm only when their precursors are present in its diet i.e., mulberry leaves (Bajpeyi *et al.* 1991). Variation in the free amino acid level in mulberry leaves due to thrips infestation may alter its requirement for the silkworm growth and development. This leads poor biosynthesis of silk.

Total soluble proteins

The *P. mori* infested leaves showing significantly decreased total soluble proteins in the leaves of M_5, Mysore local and $S_{54.}$ But, the decrease was non-significant in leaves of MR_2 (1.90%) variety. The reduction was maximum (31.39%) in the leaves of Mysore local. The increased total soluble proteins were observed in the leaves of S_{36} (0.89 %) and it was significant in V_1 (22.50%) variety.

The protein content was increased in the leaves of M_5, Kurangi, Kajali, Vietnam, Thailand and *M. nigra* and decreased in Hakkikalu and Thysong-3 due to thrips infestation (Latha, 1999). The decrease in the protein content may be attributed to the damage caused by the insect through sucking of the leaf sap; thus altering the metabolic functions leading to either decline in protein synthesize or mobilization of the proteins for repair of the damaged tissues in order to develop resistance to insect bite (Sathya Prasad *et al*. 2002). Reduction in the protein content of infested leaves may be partly due to utilization by the insect pest at a faster rate for its multiplication. Hydrolysis of proteins by proteolytic enzymes secreted by the pest itself, may be the other cause for lowering of proteins (Sengupta *et al*. 1990). Increase in the protein content may be a result of possible change in its synthesis pattern to overcome the injury and develop resistance (Satya Prasad *et al*. 2002).

Total reducing sugars

The total reducing sugars were declined significantly in the leaves of M_5, Mysore local, S_{36} and S_{54} and non-significantly in the leaves of MR_2 variety due to thrips infestation. The thrips infested leaves of MR_2 and S_{54} respectively showed a lower (9.23 %) and higher (31.68 %) percentage of decrease in the total reducing sugars. The increased total reducing sugars were noticed in the leaves of V_1 (130.38 %).

Latha (1999) noticed the increased reducing sugar content in the thrips infested leaves of four indigenous (M_5, Kurangi, Kajali and Hakkikalu) and four exotic (Thysong-3, Vietnam, Thailand and *M. nigra*) mulberry varieties. The reducing sugars are required for the biosynthesis of fibroin and sericin by the silkworm along with amino acids (Dorcus and Vivekanandan, 1997). Alteration in the reducing sugars may be due to reduction in leaf lamina and malformation of leaves in pest affected plants resulting in less productivity (Shree and Umesh Kumar, 1989).

Total soluble sugars

Total soluble sugars were decreased in the thrips infested leaves of M_5, MR_2, Mysore local, S_{36} and V_1. The decrease was maximum (18.00%) in the leaves of M_5 and minimum (11.46 %) in MR_2 variety. In S_{54} variety, the total soluble sugars were not varied even after *P. mori* infestation. Latha (1999) observed the increased soluble sugar content in M_5, Kurangi, Hakkikalu, Thysong-3, Vietnam, Thailand and *M. nigra* and decreased in the Kajali mulberry variety due to thrips.

Starch

The starch content was lowered in the leaves of MR_2 (7.87 %) and increased in the leaves of M_5, Mysore local, S_{36} and V_1 in the thrips infested mulberry leaves. The leaves of S_{54} variety showed a no-alteration in the starch content even after thrips attack. The thrips infested leaves of S_{36} variety showed lower (2.04 %) and higher in the leaves of V_1 (5.41 %) variety.

Suma (2001) noticed the decreased enzymes activity (protease and amylase) and their substrate levels (proteins and starch) in thrips infested leaves of eight indigenous (C_6, Hakkikalu, M_5, Mysore local, S_{30}, S_{36}, S_{41} and S_{54}) and four exotic (Kosen, KNG, Tsukasaguwa and Vietnam) mulberry varieties.

Total phenols

The total phenolic contents were decreased in the thrips infested leaves of M_5, MR_2 (16.53%) and S_{54} (10.53 %). The increase was maximum (11.54 %) and minimum (2.99%) in the leaves of Mysore local and V_1 varieties respectively. The total phenols were not altered in the leaves of S_{36} variety due to pest injury.

The phenolic content was increased significantly in thrips infested leaves of M_5, Hakkikalu and *M. nigra* and decreased in Kurangi, Kajali, Thysong-3, Vietnam and Thailand varieties. The accumulation of phenolics in the host may inactivate the enzyme which inhibits the further advance of the pathogenic organism by limiting its source of nutrients (Uritani, 1961). The most important phenolic compounds implicated in defence mechanism of plants against pathogens are coumaric acid, phloretin, umbelliferous, caffeic acid, chlorogenic acid and ferulic acid (Agrios, 1969). Hadimani *et al*. (2006) reported early maturity, depletion of moisture and reduction in foliar constituent (crude protein, total sugars and carbohydrates etc.,) in the thrips infested mulberry leaves. Alterations in the biochemical constituents were observed in other cases also, where mulberry leaves were infested by various pests - such - as jassids (Shree and Mahadeva, 2005 a), leaf roller (Narayanaswamy, 2003), mealy bugs (Shree and Umesh Kumar, 1989: Veeranna, 1997) and thrips (Sengupta *et al*. 1990; Satya Prasad *et al*. 2002). This alteration in the biochemical composition may be because of disturbed host metabolism due to pest damage (Umesh Kumar *et al*. 1990). The altered biochemical constituent leads to imbalance in the nutritional components of mulberry foliage which when fed to silkworms exert a harmful affect on their growth and development.

The chlorophylls are the essential catalysts of photosynthesis and occur universally as green pigments in all the autotrophic plant tissues. In the present work, drastic changes were noticed in the photosynthetic pigments of all the mulberry varieties considered due to thrips - attack.

The total chlorophyll contents were decreased significantly in the thrips infested mulberry leaves of six varieties. The reduction was maximum (57.01 %) in Mysore local and minimum (20.20 %) in the leaves of S_{36} variety.

The thrips infested leaves of all the maturity levels in six mulberry varieties the chlorophyll – a content decreased significantly. The reduction was in the range of 22.62 % to 56.83 % in the leaves of S_{36} and Mysore local varieties respectively.

PHOTOSYNTHETIC PIGMENTS (Table - 2)

Table – 2 : Changes in the photosynthetic pigments (mg/g. fresh weight) of thrips infested mulberry leaves.

Mulberry varieties	Total chlorophyll		Chlorophyll (- a		Chlorophyll (- b		Chlorophyll – a/b		Carotenoids	
	Healthy	Infested	Healthy	Infested	Healthy	Infested	Healthy	Infested	Healthy	Infested
M_5	2.45	1.66**	2.05	1.45**	0.40	0.21**	5.08	7.08**	1.24	0.87**
	(-32.28)		(-28.99)		(-49.13)		(+39.57)		(-29.99)	
MR_2	2.82	1.95**	2.32	1.59**	0.50	0.36**	4.66	4.41	1.35	0.93**
	(-30.81)		(-31.51)		(-27.41)		(-5.53)		(-30.85)	
Mysore local	1.20	0.86**	1.63	0.70**	0.37	0.16**	4.38	4.50	0.97	0.39**
	(-57.01)		(-56.83)		(-57.95)		(+2.62)		(-59.59)	
S_{36}	2.50	1.99**	2.03	1.57**	0.47	0.42**	4.32	3.71**	1.11	0.92**
	(-20.20)		(-22.62)		(-9.81)		(-14.20)		(-17.27)	
S_{54}	3.51	1.63**	2.82	1.22**	0.69	0.41**	4.10	2.90*	1.53	0.72**
	(-53.67)		(-56.69)		(-41.08)		(-29.29)		(-53.00)	
V_1	2.36	1.70**	2.02	1.42**	0.41	0.27*	5.01	5.15	1.04	0.62**
	(-27.99)		(-29.53)		(-35.06)		(+2.71)		(-40.59)	

* Significant at 1% level; * Significant at 5% level; Values parenthesis indicate difference over healthy (+ = more than; - less than).

The chlorophyll – b contents were decreased significantly in the leaves of all the six varieties due to thrips infestation. The reduction was in the range of 9.81 % to 57.95 % in leaves of S_{36} and Mysore local varieties respectively.

The chlorophyll – a/b ratio reduced in the leaves of MR_2, S_{36} and S_{54} due to thrips infestation. The reduction was low (5.53 %) in MR_2 and higher (29.29%) in the leaves of S_{54} variety. The thrips infested leaves of M_5, Mysore local and V_1 varieties showed an increased chlorophyll – a/b ratio. However, it was significant only in leaves of M_5 variety. The leaves of M_5 and Mysore local varieties showed a maximum (39.57%) and minimum (2.62%) increase of chlorophyll – a/b ratio respectively.

The carotenoids were decreased significantly in all the leaves of six mulberry varieties due to thrips infestation. The reductions of carotenoids were maximum (59.59%) in Mysore local and minimum (17.27%) in the leaves of S_{36} mulberry variety.

Das *et al*., (1994) observed a combined loss of total chlorophyll and carotenoids (57.44 and 52.07 % respectively) in the thrips - infested mulberry leaves. Satya Prasad *et al.* (2002) observed a non-significant change in the total chlorophyll, chlorophyll – a, chlorophyll – b and carotenoids in K_2, S_{13}, S_{34}, S_{36} and V_1 mulberry varieties due to thrips - infestation. Decrease in chlorophyll and carotenoid contents in infested leaves may have been due to loss in chlorophyll synthetase activity in response to thrips infestation (Das *et al.*, 1994).

Etebari and Bizhannia (2006) observed that there was a significant decrease in total cocoon production, cocoon weight, pupal weight, and shell weight due to feeding of thrips infested leaves.

Thus, the content of photosynthetic pigments varied depending upon the pest, intensity of pest-infestation, extent of damage and mulberry varieties. The reduction in the photosynthetic pigment(s) and/or laminar area caused due to feeding nature of insect pests will result in decreased photosynthetic efficiency and thereby the productivity. Sengupta *et al.* (1999) observed a significant difference in the photosynthetic rate between the healthy and thrips - infested leaf as well as in different positions of the diseased leaves. As a result, mulberry leaves suffer from nutritional inferiority. Similar observations were made in other cases when mulberry leaves were infested by various pests such as mealy bugs (Shree and Umesh Kumar, 1989), jassids (Shree and Mahadeva, 2005 a), spiralling whitefly (Narayanaswamy *et al.* 1999; Chandramohan *et al*. 2001) and leaf rollers (Narayanaswamy, 2003; Mahadeva and Shree, 2005).

Most sap sucking insects, such as adult leaf hoppers, aphids, or thrips cause minimal direct tissue destruction. These insects use a specialized mouth part, the stylet, to locate, penetrate and drain sap from the phloem sieve elements of the plants vascular tissue. Heavy infestation caused by them leads to chronic shortages of photosynthates and thus severely reduce the growth potential of the plant (Kim Hammond-Kosack and Jonathan Jones, 2000).

The altered chlorophyll content adversely affected the photosynthetic activity (Heldt, 1997), productivity which ultimately leads to reduced protein synthesis (Burd and Elliot, 1996). Consequently, the nutritional status of mulberry foliage comes down (Shree and Umesh Kumar, 1989). Therefore, the pest - attacked leaves when fed to silkworms will exert an adverse impact on their growth and development, leading to cocoon crop failures (Pradeep Kumar *et al*. 1992; Doureswamy and Chandramohan, 1999; Shree and Mahadeva, 2005 b; Muthuswami *et al*. 2010). The pest - injured mulberry leaves should not be used for silkworm feeding as they are known to affect the commercial characters of cocoons (Mahadeva and Shree, 2004).

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