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Effect of Plant Products on Morphological Parameters of Tukra Affected Mulberry Leaves

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Abstract- Tukra is one of the major problem in mulberry growing areas and it is caused by pink mealy bug *Maconellicoccus hirsutus* which affect both the quality and yield of mulberry leaf. In general the tukra- diseased leaves showed high moisture content compared to normal healthy leaves. A preliminary attempt has been made to know the effect of plant extracts of natural pesticide origin on moisture content, loss and retention capacity of tukra affected mulberry leaves. The seed kernel and leaf extracts of *Azadirachta indica, Pongamia pinnata, Madhuca longifolia* and only leaf extracts of *Lantana camara, Adathoda vasica* were directly used as a foliar spray on M-5 mulberry variety under field conditions. The results revealed that the tukra affected leaves showed high moisture content (75.92%) compared to normal healthy leaves (71.32%). The moisture content was more in NSKE @ 4% (73.83%) PSKE @ 4% (73.80%), MSKE @ 4% (73.30%), and lowest was recorded from MSKE @ 2% (71.76%) followed by control (70.66%). The maximum moisture retention capacity in infested leaves of tukra was observed in NSKE @ 4% (80.60%) sprayed leaves and lowest was recorded from PLE @ 8% (78.46%) compare to control (77.80%). The minimum and maximum moisture loss of 26.06 and 28.20 % was observed in NSKE @ 4% and MSKE @ 2% respectively over the control (29.23%). There was no significant difference among the different treatments with respect to moisture content, loss and retention capacity of tukra affected mulberry leaves.

Index Terms- Pink Mealy Bug, Mulberry, Tukra, Moisture content, Moisture loss, Moisture retention capacity

I. INTRODUCTION

Mulberry is the indispensable food for mulberry silkworm and is known for its luxuriant growth. About 300 insect and non-insect species of pests are known to inflict the damage to mulberry in different parts of the world. Among the pests, sucking pests are considered as major pests causing considerable damage to mulberry in all the growing stages of crop particularly in the apical portion (Reddy and Narayanaswamy, 2003). However, the pink mealy bug *Maconellicoccus hirsutus* Green (Pseudococcidae: Homoptera) is considered as an important cosmopolitan sucking pest and regular in occurrence. During infestation they prefer tender portion of the plant because of succulence. It sucks the sap simultaneously releasing toxins which results in short internodes, curling, wrinkling and crumpling of apical leaves virtually stopping the growth of the plant by suppression of stem elongation affecting the yield of leaves. Further the affected region swells and turns into deep green color. Therefore, the symptoms of mealy bug infestation in mulberry collectively called as tukra (Misra, 1919). Besides, reduction in leaf area, yellowing of leaves, premature leaf fall occurs due to impaired function of the petiole due to mealy bug infestation. The tukra affected mulberry plantations recorded three to six tonnes of leaf yield/ha/ year (Kumar *et al*., 1992). Palanidurai (1996) reported that substantial reduction in number of leaves / plant by 13.6 per cent. Further Satyaprasad *et al.* (2000) reported that, mealy bug incidence caused an estimated loss in leaf yield of 4500 Kg/ha/yr amounting to 34.24 per cent (Manjunath *et al*., 2003), thus depriving the farmer from brushing about 450 dfls/ha/yr, leading to decline in cocoon production by 150 Kg/ha/yr (10-15 per cent). Some studies revealed that the tukra affected mulberry leaves shows the highest moisture content when compared to healthy leaves. Generally insecticides are not advisable for mulberry ecosystem, because of the residual toxicity and also it directly influences the silkworm rearing. Recently non-chemical avenues like botanicals acted as an efficient alternative for the pesticides in mulberry garden (Sathyaseelan and Bhaskaran, 2010**).** The use of plant extracts and plant products is gaining attention due to proven specificity, biodegradability, low toxicity to non target organisms and low residual toxicity in the ecosystem (Fowcett and Spenser, 1970). These are found effective when they are applied in known concentrations and optimum time of application. A number of commonly available plants that are known to have insecticidal and antifungal properties have been tried in mulberry also to develop ecofriendly management strategies for pests and diseases (Tomy Philip *et al.,* 2009). A preliminary attempt has been made to know the effect of plant extracts of natural pesticide origin on moisture content, loss and retention capacity of tukra affected mulberry leaves.

II. MATERIALS AND METHODS

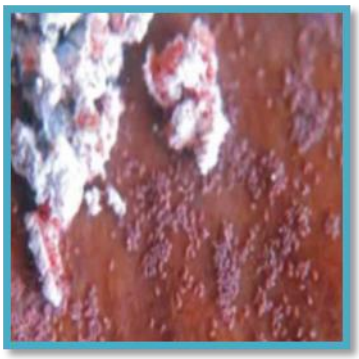
This experiment was laid out during post rainy season of 2011-2012 in Randomized Block Design with three replications. Well established M5 variety mulberry garden were selected at UAS, GKVK, Bangalore. All the agronomic practices were followed as per the package of practices for higher yields except the plant protection schedule.

Culturing of pink mealy bug

The pure culture was released on well matured pumpkin which was cleaned using water and treated with 0.1% Bavistin 50 WP. The wounds present on pumpkins were plugged using wax. The culture was maintained through out the research period without contamination. Chacko *et al.* (1978) and Singh (1978).

Inoculation studies were carried out on unaffected plants by transferring 25 crawlers to the growing tips with an interval of 30 days. The symptom developed due to mealy bug was assessed by colonisation and infestation by the coccids after 15 days of release in the infested (Sriharan *et al.*, 1979). Total of four sprays were given with an interval of 7 days.

Crawlers



Infested pumpkin

Preparation of plant extracts and spraying

Plant extract was prepared by homogenizing 10 g of plant material (leaf) in 100 ml of distilled water using pestle and mortar. The homogenate was filtered through three layered muslin cloth. The resulting clear solution is used as foliar spray (8% and 10 %) on M_5 mulberry plants with an interval of 7 days. Further the seed kernel extracts @ 2 and 4% (4 g of seed kernel powder in 100 ml of distilled water) along with one per cent soap solution also sprayed on each plant. The leaf and seed kernel extracts were sprayed (using hand sprayer) for 28 days on mulberry sapling as drenching spray of extracts both on ventral and dorsal surface of leaves. The control batch (sprayed with water) was also maintained within the net. All the mulberry plants were treated (5 ml) with botanical at a time. Care was taken to wash the hand sprayer with water thoroughly well before using another botanical to avoid contamination.

Table:1 Treatments Details

Tr. No	Treatments
T1: NSKE @ 4%	Neem (*Azadirachta indica*) seed kernel extract @4% + Soap powder @1%
T2: NSKE @ 2%	Neem (*Azadirachta indica*) seed kernel extract @2% + Soap powder @1%
T3: NLE @ 10%	Neem (*Azadirachta indica*) leaf extract @ 10%
T4: NLE @ 8%	Neem (*Azadirachta indica*) leaf extract @8%
T5: PSKE @ 4%	Honge *(Pongamia pinnata)* seed kernel extract @ 4% + Soap powder @1%
T6: PSKE @ 2%	Honge *(Pongamia pinnata)* seed kernel extract @2%+ Soap powder @1%
T7: PLE @ 10%	Honge (*Pongamia pinnata*) leaf extract @ 10%
T8: PLE @ 8%	Honge (*Pongamia pinnata*) leaf extract@ 8%
T9: MSKE @ 4%	Mahua (*Madhuca longifolia*) seed kernal extract @4% + Soap powder @1%
T10: MSKE @ 2%	Mahua (*Madhuca longifolia*) seed kernal extract@2% + Soap powder @1%
T11: MLE @ 10%	Mahua (*Madhuca longifolia*) leaf extract @10%
T12: MLE @ 8%	Mahua (*Madhuca longifolia*) leaf extract@8%
T13: LLE @ 10%	*Lantana (Lantana camara)* leaf extract@ 10%
T14: LLE @ 8%	*Lantana (Lantana camara)* leaf extract @8%
T15: ALE @ 10%	Adusoge (*Adathoda vasica)* leaf extract @10%
T16: ALE @ 8%	Adusoge (*Adathoda vasica)* leaf extract @ 8%
T17	Control

Observations:

Moisture content

Fresh, healthy and infested leaves (10 from each replication) before and after spray of plant extracts were collected and weighed immediately for fresh weight determination (mf) and then the leaves were dried at 80^0c in an oven until a constant dry weight was obtained. The moisture content (mc) was calculated using the formula (Turner, 1981).

$$\text{Moisture content (\%)} = \frac{\text{Fresh wt.} - \text{Dry wt.}}{\text{Fresh wt.}} \times 100$$

Moisture Loss

Moisture loss (%) = 100 - Moisture content

Moisture retention capacity

To determine moisture retention capacity, fresh leaves were weighed (WI) and incubated at 27^0c ± 1^0c with a relative humidity of 45-50% for 12 hours in a BOD incubator and weighed (WF) again. Further, leaves were dried in an oven and weighed.

Moisture Retention Capacity (MRC)

$$MRC\ (\%) = \frac{\text{Moisture content after 12 hr of incubation}}{\text{Moisture content}} \times 100$$

III. Results and Discussion

A small study was conducted on moisture content of tukra and healthy leaves of mulberry. The results revealed that the tukra affected leaves showed high moisture content (75.92%) compared to normal healthy leaves (71.32%). Further, an attempt were made to know the influence of botanicals on moisture content of tukra affected mulberry leaves.

Moisture content:

The moisture content was more in NSKE @ 4% (73.83%) PSKE @ 4% (73.80%), MSKE @ 4% (73.30%), and lowest was recorded from MSKE @ 2% (71.76%) followed by control (70.66%). The order of treatments which contain more moisture content was NSKE @ 4% > PSKE @ 4% > MSKE @ 4% > LLE @ 8% >PLE @ 8% > NLE @ 10% > NLE @ 8% > NSKE @ 2% > LLE @ 10% > PSKE @ 2% > PLE @ 10% > MLE @ 10% > ALE @ 10% > ALE @ 8% > MLE @ 8% > MSKE @ 2% > control. There was no significant difference among the different treatments with respect to moisture content of tukra affected mulberry leaves.

Moisture loss and moisture retention (%) in mulberry leaves

The morpho physiological parameters of mulberry leaves showed variation in the infested leaves of mealy bug *Maconellicoccus hirsutus* after different days of observations. The observation made on 7, 14, 21 and 28 days showed variation in the moisture loss as well as moisture retention capacity in the infested mulberry leaves. However on the 14 days of observation significantly more moisture loss (29.73 and 29.23%) was noticed in the leaves sprayed with MLE @ 8% the same observation noticed on 21st day in PLE @ 10% sprayed leaves. The other seed kernel extracts *viz.*, NSKE @ 4% (26.97%), NSKE @ 2% (27.72%), PSKE @ 4% (26.56%), PSKE @ 2% (27.82%), MSKE @ 4% (27.51%) and MSKE @ 2% (28.96%) also registered significant difference the same trend seen even in 21st day of observation NSKE @ 4% (26.36%), NSKE @ 2% (27.38%), PSKE @ 4% (27.00%), PSKE @ 2% (27.90%), MSKE @ 4% (26.71%) and MSKE @ 2% (28.41%). The observations noticed on 7th day and 28th day did not yield any significant results but the same effect was noticed in moisture loss of mulberry leaves sprayed with seed kernel and leaf extracts.

Moisture retention (%)

The maximum moisture retention capacity in infested leaves of tukra was observed in NSKE @ 4% (80.60%) sprayed leaves and lowest was recorded from PLE @ 8% (78.46%) compare to control (77.80%). The order of treatments which contain more moisture retention capacity was NSKE @ 4% > NSKE @ 2% > NLE @ 10% > PSKE @ 2% > PSKE @ 4% > ALE @ 8% > ALE @ 10% > MSKE @ 4% > PLE @ 10% > MLE @ 8% > NLE @ 8% > MSKE @ 2% > LLE @ 8% > MLE @ 10% > LLE @ 10% > PLE @ 8% > control. There was no significant difference among the different treatments with respect to moisture retention capacity of tukra affected mulberry leaves.

These results were agreement with Babu *et al*. (1994) showed that tukra affected leaves have more moisture content (73.50%) when compared to healthy leaves(68.90%) and stated that in general the tukra affected leaves showed high moisture content compared to normal healthy leaves. Further Shree *et al.* (1989) also reported that tukra affected leaves showed higher moisture content (74.50%) compared to healthy mulberry leaves (70.30%) in Kanva-2 mulberry variety. The same trend was also followed in Kajili variety where tukra affected has 38.00 per cent moisture content and 37.60 per cent was recorded from healthy leaves. However, the moisture content was almost identical in the Kajli variety whereas in Kanva-2 there was an increase in the tukra affected leaves compared to healthy ones.

Bose *et al.* (1992) reported that the tukra affected mulberry leaves showed higher moisture content when compare to healthy. After conducting tests on six mulberry varieties *viz.*, Local (74.15 and 70.10%), K2 (73.40 and 72.14%), S30 (72.53 and 72.43%), S36 (73.92 and 72.28%), S41 (73.08 and 71.37%) and S54 (75.49 and 73.71 %). The similar observations were recorded on M5 mulberry variety in the present investigation. Babu *et al.* (1994) observed significant variation among biochemical constituents of both tukra diseased and healthy leaves. Significant variation in moisture content was found in tukra affected leaves of all test varieties *viz.*, M5 (60 %), I-Chinose (70 %), Gosoerami (75%), Kosen (55%), BC2-59 (68%), TR4 (69%) except S13 (68%).

Table:2 Influence of plant extracts on moisture content (%) in mulberry leaves with mealy bug infestation

Treatments	Days After Spray (DAS)			
	7 DAS	14 DAS	21 DAS	28 DAS
NSKE @4%	72.86	73.02	73.63	73.87
NSKE @2%	72.33	72.28	72.62	72.89
NLE @10%	72.35	72.98	72.54	73.15
NLE @ 8%	72.52	72.85	72.2	72.96
PSKE @4%	72.04	73.44	73.00	73.81
PSKE @2%	70.81	72.18	72.10	72.64
PLE @ 10%	69.88	71.08	70.77	72.63
PLE @ 8%	71.26	72.63	72.76	73.22
MSKE @4%	70.83	72.49	73.28	73.34
MSKE @2%	70.14	71.03	71.59	71.77
MLE @ 10%	70.27	71.27	71.22	72.36
MLE @ 8%	70.16	70.26	71.10	71.95
LLE @ 10%	70.67	70.82	70.99	72.71
LLE @ 8%	72.20	71.47	72.25	73.29
ALE @ 10%	71.99	71.76	71.92	72.40
ALE @ 8%	69.96	71.77	72.04	72.22
Control	70.33	70.51	70.86	70.69
F- test	*	*	NS	NS
S.Em±	0.52	0.48	0.45	0.50
CD at 5%	1.51	1.40	-	-

* Significant at 5%, NS- Non significant

Table:3 Influence of plant extracts on moisture loss (%) from mulberry leaves with mealy bug infestation

Treatments	Days After Spray (DAS)			
	7 DAS	14 DAS	21 DAS	28 DAS
NSKE @4%	27.14	26.97	26.36	26.12
NSKE @2%	27.67	27.72	27.38	27.10
NLE @10%	27.64	27.02	27.46	26.85
NLE @ 8%	27.48	27.15	27.80	27.04
PSKE @4%	27.95	26.56	27.00	26.18
PSKE @2%	29.19	27.82	27.90	27.35
PLE @ 10%	30.11	28.92	29.23	27.37
PLE @ 8%	28.73	27.37	27.24	26.77
MSKE@4%	29.16	27.51	26.71	26.66
MSKE @2%	29.86	28.96	28.41	28.22
MLE @ 10%	29.73	28.73	28.77	27.64
MLE @ 8%	29.83	29.73	28.90	28.05
LLE @ 10%	29.33	29.17	29.01	27.29
LLE @ 8%	27.79	28.52	27.75	26.70
ALE @ 10%	28.00	28.23	28.08	27.59
ALE @ 8%	30.03	28.22	27.95	27.78
Control	29.67	29.49	29.13	29.30
F- test	NS	*	*	NS
S.Em±	0.55	0.49	0.45	0.50
CD at 5%	-	1.41	1.31	-

* Significant at 5%, NS- Non significant

Table:4 Influence of plant extracts on moisture retention capacity (%) in mulberry leaves with mealy bug infestation

Treatments	Days After Spray (DAS)			
	7 DAS	14 DAS	21 DAS	28 DAS

NSKE @4%	79.28	79.40	80.46	80.64
NSKE @2%	79.12	79.91	79.74	79.96
NLE @10%	78.02	78.69	79.32	79.66
NLE @ 8%	77.25	77.24	78.50	78.98
PSKE @4%	77.25	77.37	78.54	79.44
PSKE @2%	77.94	78.98	79.00	79.54
PLE @ 10%	76.80	77.13	78.66	79.14
PLE @ 8%	77.26	77.61	78.25	78.50
MSKE@4%	78.62	78.81	79.09	79.23
MSKE @2%	77.09	78.01	78.72	78.95
MLE @ 10%	77.15	77.49	78.22	78.60
MLE @ 8%	77.31	78.35	78.20	79.16
LLE @ 10%	77.26	77.01	78.39	78.52
LLE @ 8%	77.33	77.11	78.86	78.86
ALE @ 10%	78.58	79.80	78.97	79.37
ALE @ 8%	77.43	77.93	78.78	79.42
Control	77.17	77.71	78.31	77.85
F- test	NS	NS	NS	NS
S.Em±	0.68	0.52	0.43	0.40
CD at 5%	-	-	-	-

NS- Non significant

Based on the study the reasons for increased moisture content in tukra affected mulberry leaves may be due to following reasons:

1. The tender part of affected mulberry plant exhibit curling, wrinkling and crumpling of the leaves virtually stopping the growth of plant by the suppression of elongation of stem.
2. The growing region became thick and dark coloured due to the presence of abundant chloroplasts in the entire mesophyll of the tukra infested leaves imparted their characteristic dark green colour.
3. Due to curling, wrinkling and crumpling of affected leaves the area exposed to the sunlight was less when compared to the healthy leaves so there is a minimum water loss due to the transpiration. It is the one of the reason that moisture content is more in tukra affected leaves compare to healthy leaves.
4. A heavy, black, sooty mould may develop on infested leaves and stems as a result of which heavy honeydew secretions of the mealy bug**.** The sooty mould may cover the leaf surface which donot allow the transpiration and minimises the water loss.
5. In tukra affected leaves the mesophyll cells were compactly packed with very little intercellular spaces, which can hinder the normal photosynthetic function of the leaves so it permit little gas exchange when compare to healthy leaves where intercellular spaces in mesophyll, facilitate rapid gas exchange and not affect the normal photosynthetic activity.
6. The formation of abundant grandular and non grandular trichomes on the newly formed leaves of infected plants seemed to be a natural defense reaction of the plant which covers the entire leaf surface and minimise the exposure to sunlight which leads to minimum water loss from tukra affected leaves. The trichome density is more in tukra infected leaves when compare to normal healthy leaves.

The tukra affected mulberry leaves recorded high moisture content compared to healthy leaves. After botanical spray, the affected leaves did not show any alteration with respect to moisture content, moisture loss and moisture retention capacity in M5 mulberry leaves. There is no significant difference among the botanicals with respect to above morphological parameters in the affected mulberry leaves.

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