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Field evaluation of mulberry accessions for susceptibility to foliar diseases in Uasin-Gishu district, Kenya

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The incidence and severity of foliar disease pathogens in five mulberry accessions (Embu, Thika, Thailand, Kanva-2 and S41) grown in Kenya were evaluated under field conditions in Eldoret. The plants were rated for disease incidence and severity in two mulberry growth cycles over a period of 10 months (May 2009 to February 2010). A high incidence of 58.3% bacterial leaf blight (*Xanthomonas campestris* pv. *mori*) disease prevalence was recorded on Embu accession, 36.6% incidence of fungal leaf blight (*Alternaria alternata*) on Thika and 26.0% for S41 accessions. Black leaf spot (*Pseudocercospora mori*) disease was high (48.93%) on the Thailand accession, while brown leaf spot (*Myrothecium roridum*) was recorded with a low (3.61%) incidence on the Kanva-2. Susceptibility of accessions to diseases was highest in Embu, followed by Thailand, Thika and S41 that were moderately susceptible, while Kanva-2 accession was resistant. Our results suggest that Kanva-2 accession can be utilized in future disease resistance breeding programmes to confer disease resistant trait in mulberry varieties.

Key words: Mulberry, *Xanthomonas campestris* pv. *mori*, *Alternaria alternata*, *Pseudocercospora mori*, *Myrothecium roridum*.

INTRODUCTION

Mulberry (*Morus* sp.) is a hardy deciduous perennial tree or shrub used as a food source for the domesticated silkworm, *Bombyx mori* (Aggarwal et al., 2004). In Kenya, the sericulture industry is an upcoming enterprise that is increasingly being perceived as a promising alternative source of income generation for rural small-scale farmers (Adolkar et al., 2007). However, diseases are some of the limiting factors for successful mulberry cultivation. Like other plants, mulberry is affected by a number of diseases caused by fungi, bacteria, viruses and nematodes (Sengupta et al., 1990; Yashihiko, 1995).

Foliar pathogens alone cause 15 to 20% loss in leaf production, besides severe destruction of leaf lamina to about 20 to 25%. They also cause reduced nutritive value which leads to leaf loss both by quantity and quality (Sengupta, 1990; Philip, 1991; Kumar, 1991; Reddy et

al., 2009). The nutritional quality of mulberry has a predominating influence on the growth and development of *B. mori*'s larva and subsequent cocoon production (Sikdar and Krishnaswami, 1980; Kumar et al., 1993). Foliar diseases are managed by the use of chemical, botanical and biological methods (Gangwar et al., 2000; Chattopadhyay et al., 2002; Maji et al., 2000, 2003b; Pratheesh et al., 2004). However, despite their effectiveness and ease of use, chemical control has been found to cause environmental pollution, residual toxicity to silkworms and non-target organisms (Reddy et al., 2009). Moreover, indiscriminate use of such chemicals has resulted in evolution of resistant strains (Adityachaudhury, 1991).

One of the most promising and economic ways of evaluating disease loss in mulberry is to evaluate germplasm accessions for disease resistance (Philip et al., 1996; Yadav et al., 1993). Screening of mulberry varieties against diseases has been done previously (Govindaiah et al., 1989; Thangavelu et al., 1997; Maji et al., 2005; Maji et al., 2009). Nevertheless, mulberry

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Table 1. Overall rating of five mulberry accessions against foliar diseases

Accession	Mulberry diseases studied	Means of PDI	Rating
Thika	Fungal leaf blight (<i>Alternaria alternata</i>)	36.6	MS
Thailand	Black leaf spot (<i>Pseudocercospora mori</i>)	48.9	MS
Kanva-2	Brown leaf spot (<i>Myrothecium roridum</i>)	3.6	R
S-41	Fungal leaf blight (<i>Alternaria alternata</i>)	26.0	MS
Embu	Bacterial leaf blight (<i>Xanthomonas campestris</i> pv. <i>mori</i>)	58.3	S

R, Resistant; MR, moderately resistant; MS, moderately susceptible; S, susceptible; PDI, percentage of disease incidence.

accessions grown in Kenya have not been screened against diseases. It is therefore imperative to screen Kenyan accessions of Mulberry against diseases for future improvement and use in breeding programmes, as well as for recommendations to farmers and extension staff.

MATERIALS AND METHODS

The incidence of foliar diseases on mulberry accessions Thailand, Embu, Kanva-2, S41 and Thika was evaluated in two mulberry growth cycles before and after pruning for a period of nine months (June 2009 to February 2010). This was done under field conditions in Uasin Gishu District, Eldoret, Kenya. Five mulberry accessions were planted at a spacing of 3 × 3 m in a randomized complete block design. Farm yard manure was applied at the rate of 7 tons ha⁻¹ prior to planting. Fertilization consisted of a banded application of 16.8 kg nitrogen (N), 7.4 kg phosphorus monoxide (P₂O) and 13.8 kg potassium oxide (K₂O) ha⁻¹ during 60 days post plantation. A booster dose of urea was applied at 45 kg N ha⁻¹ on 35 days after pruning. Weed control was performed by hand and the plots were maintained weed-free to the end of the experiment.

Foliar disease incidence and severity was recorded by randomly selecting 12 plants of each accession. The total numbers of diseased and healthy leaves were recorded in three long branches on the six top fully mature leaves of each plant. Disease severity was recorded using a visual rating scale (KARI, 1996). In this scale, one indicated no spot; two, 1 to 10% few spots on <50% of leaves; three, 1 to 10% spots on >50% of leaves or 10 to 30% spots on < 50% of leaves; and four, >30% spots on >50% of leaves. Percentage of disease incidence (PDI) was scored using one to six scales modified from Mc Kinney (1923) and Teotia et al. (1997). Mulberry accessions were categorized as 1-immune, 2- resistant, 3-moderately resistant, 4- moderately susceptible, 5- susceptible, 6-highly susceptible.

$$\text{PDI} = \frac{\text{Sum of numerical values}}{\text{Total no. of leaves} \times \text{maximum grading}} \times 100$$

Statistical analysis

Statistical analysis of data given in percentage was carried out from angular transformed values. Data was also subjected to analysis of variance using Genstat where the statistical model $Y_{ijk} = \mu + \beta_i + \beta_j + \beta_k + \epsilon_{ijk}$ was used, Least significance difference (LSD) was used to indicate the level of difference between means that were significantly different at $p > 0.05$.

RESULTS

The results indicate that all the mulberry accessions were affected by various diseases as presented in Table 1. However, disease severity varied across the mulberry accessions screened. There was a high incidence of bacterial leaf blight on the Embu accession. The disease was characterized by wilting of the leaf margin and angular yellow areas bounded by the veins. Under severe conditions, the infection led to girdling of the petioles, stem and shoot. Thika and S41 accessions were attacked by fungal leaf blight. The Thika accession showed blackening at the tip of the leaf, while S41 browning at the leaf's tip extended inwards. Under severe conditions, defoliation occurred.

Results presented in (Figure 1) show blight incidence across five mulberry accessions in cycle one, which was low at the initial stages. However, the Embu accession had the highest infection throughout the 28 days. Blight incidence between 21 to 28 days did not differ significantly in Embu and S41 accessions, except for Thika accession. The Embu accession had the highest blight incidence followed by accession S41 and Thika accessions, respectively. Moreover, during the second cycle of the study (Figure 2), observations show maintenance of high incidence of bacterial leaf blight disease on the Embu accession. Thika accession showed a higher incidence and increase in inoculum than in the previous cycle. In cycle two, on all the mulberry accessions blight, incidence gradually increased over the 28 days and throughout this period, Embu accessions was significantly different from Thika and S41 accessions.

Occurrence of leaf spot disease on Thailand accession showed black irregular spots on the surface of the leaves, which eventually enlarged and left large holes with a yellow surrounding. Kanva-2 accession was attacked by brown leaf spot, which formed irregular brown spots. Leaf spot incidence in cycle one (Figure 3) increased gradually throughout the 28 days. In addition, Thailand and Kanva-2 accessions were significantly different in their spot incidence. Observations of leaf spot in cycle two showed a higher incidence on Thailand accession than in the first cycle, with attacked leaves turning yellow, petioles blackening and eventual death of the shoot. Leaf spot

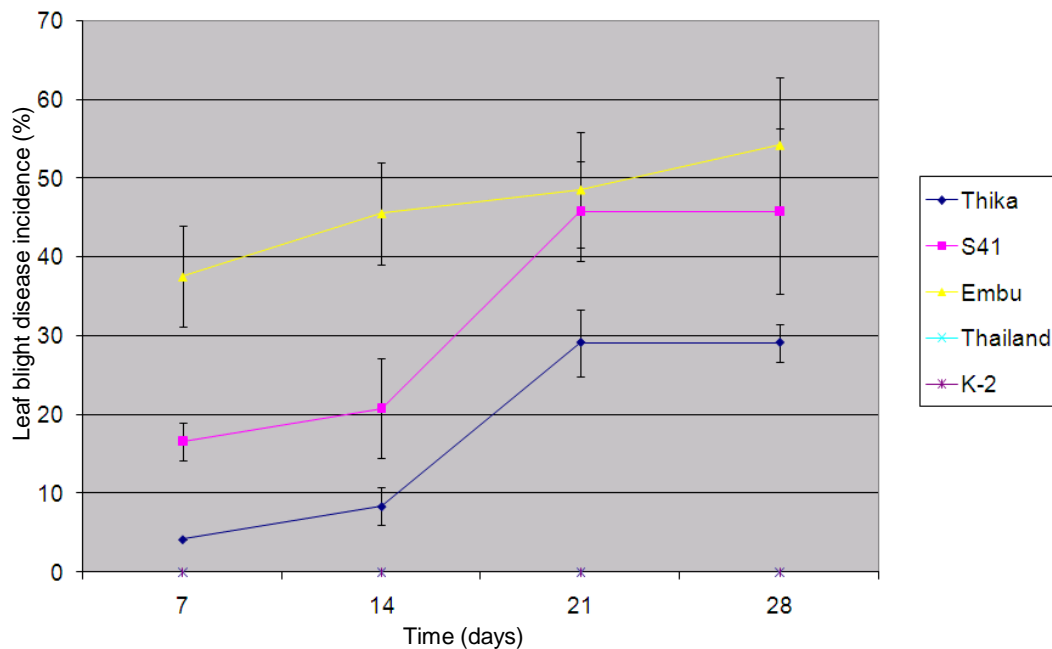


Figure 1. Leaf blight incidence (%), across three mulberry accessions over time in cycle 1.

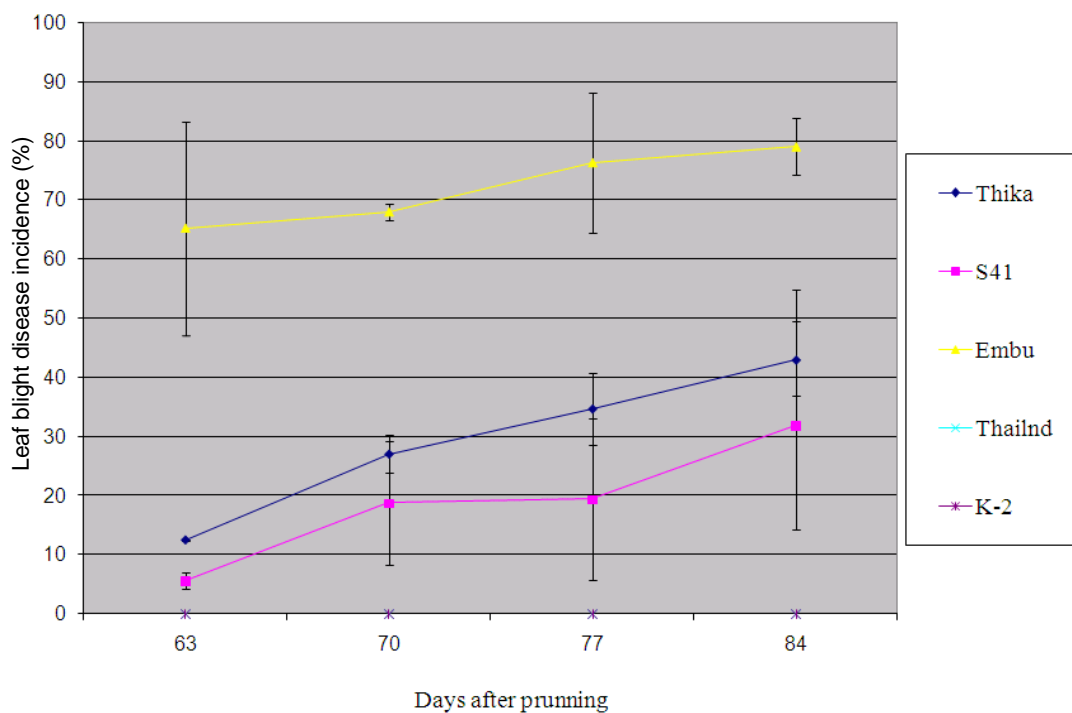


Figure 2. Leaf blight incidence (%), across three mulberry accessions over time in cycle 2.

incidence in cycle two (Figure 4) increased gradually over the 28 days. Accessions Thailand and Kanva-2 were significantly different, with Kanva-2 accession having a lower percentage spot incidence than in the previous cycle.

Rating of the different mulberry accessions against foliar diseases

Rating of mulberry accessions against foliar diseases was taken using the PDI means. Different mulberry

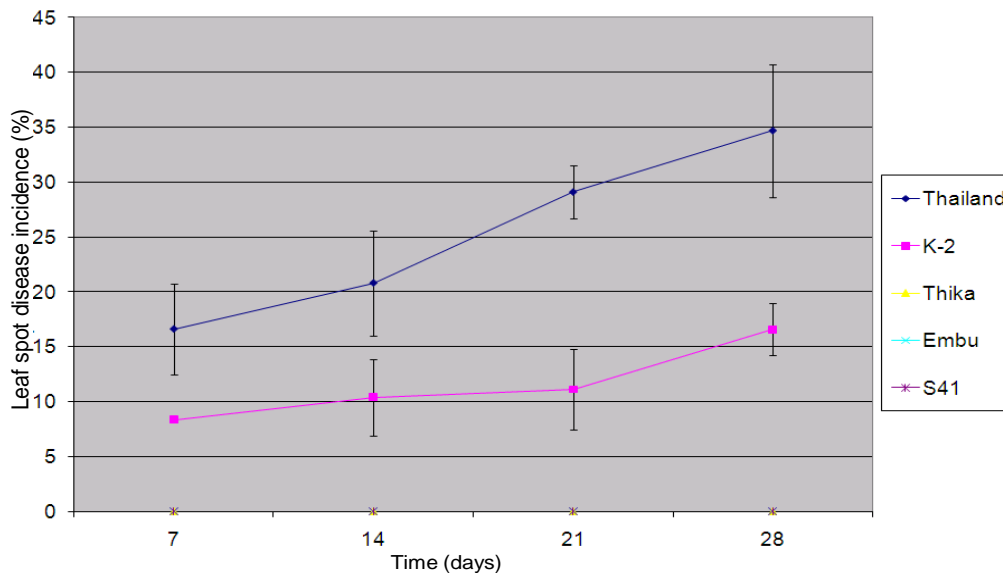


Figure 3. Leaf spot incidence (%), across three mulberry accessions over time in cycle 1.

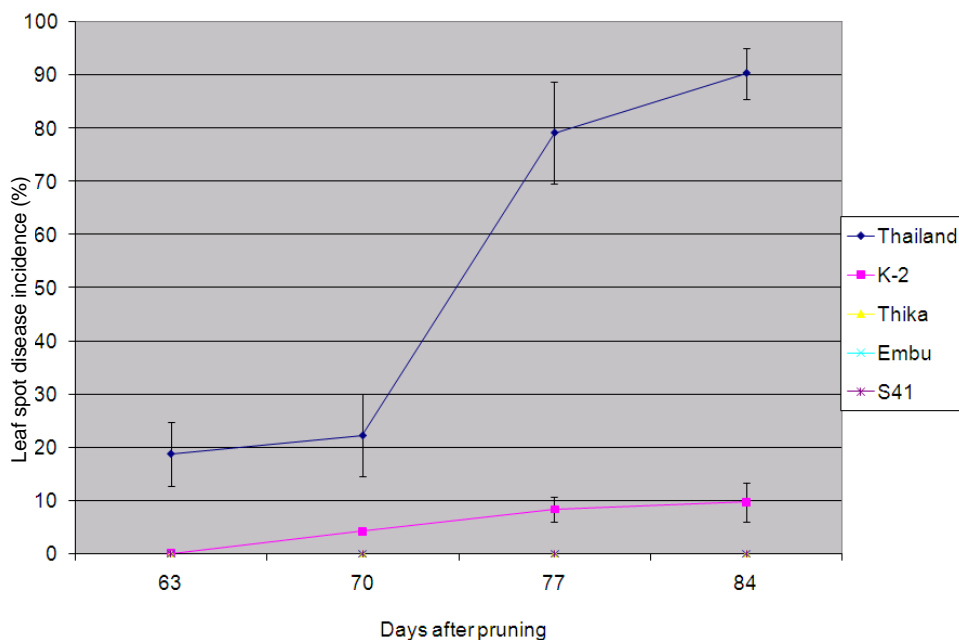


Figure 4. Leaf spot incidence (%), across three mulberry accessions over time in cycle 2.

accessions were affected by different foliar diseases (Table 1).

DISCUSSION

Mulberry leaf yield and quality affect silkworm rearing and ultimate cocoon yield and silk production. In this study,

leaf spot (*Myrothecium roridum*; *Pseudocercospora mori*) and leaf blight (*Alternaria alternata*; *Xanthomonas campestris* pv. *mori*) are some of the diseases studied. These diseases are similar to those reported by Ghosh et al. (2003), Baiyewu et al. (2005) and Maji et al. (2009) in Bangladesh, Nigeria and India on mulberry, respectively.

Observations of bacterial leaf blight in this study are similar to those reported by Pataky (2010) on bacterial

leaf blight of geranium and Scot (2004) in *Morinda citrifolia*. Symptoms of fungal leaf blight observed in this study are also similar to those reported by Reddy et al. (2007) on fungal leaf blight of mulberry. A gradual increase in leaf blight and leaf spot disease incidence was observed in this study during cycle one. Meteorological factors have a predominant effect in the development of disease (Wolf and Verret, 2005). Furthermore, the intensity and duration of atmospheric precipitation plays a fundamental role in disease progress (Awasthi and Kolte, 1994). At this stage of the cycle, temperatures varied from 11.1 to 24.7°C, while humidity ranged from 73 to 87%. Embu, S41 and Thika accessions were affected by bacterial and fungal leaf blight diseases. Sener and Fatih (2005) reported survival of septoria blight of parsley at 15 to 32°C temperature, while disease progressed at 20 to 23°C. Thailand and Kanva-2 accessions were affected by fungal leaf spot diseases. An average of 24°C and a relative humidity of not less than 60% are favorable for the development of leaf spot diseases in mulberry (Sikder and Bose, 1975; Gunasekhar et al., 1994; Srikantaswamy et al., 2000).

Moreover in cycle two, percentage leaf blight incidence increased in Embu and Thika accessions, while S41 declined in its incidence to the disease. On the other hand, percent leaf spot incidence in Thailand accession increased as Kanva-2 decreased. These may be attributed to the spread and colonization of the pathogen with increasing crop age. Srikantaswamy et al. (2006) reported a similar trend of increase in leaf rust development with increasing crop age in mulberry. However, with increased utilization of available food material by the pathogen, this may lead to a decline in infection at the later stages. Hedge and Anahosur (1994) reported a similar trend of decreased white rust infection in mustard. Nevertheless, temperatures in cycle two varied from 12.8 to 24.3°C, while relative humidity ranged from 92 to 97% thereby providing a favorable environment for disease development.

The difference in infection rate of different mulberry accessions can be attributed to varietal characteristics ascribed to genetic inheritance (Jindal and Bhavani, 2002). Environmental factors, crop (leaf) age and host susceptibility influence disease progress (Biswas et al., 1995). In this study, the overall rating of the mulberry accessions show that the Embu accession was susceptible. Thika, S41 and Thailand accessions were moderately susceptible, while Kanva-2 was resistant. Kanva-2 accession has been noted to be resistant to leaf spot (Maji et al., 2009).

Conclusion

Bacterial leaf blight was most severe on Embu accession, thereby showing susceptibility. Thika and S41 accessions had incidences of fungal leaf blight hence their reaction

was moderately susceptible. Thailand accession had the highest incidence of black leaf spot, hence being moderately susceptible, while Kanva-2 accession had low incidences of brown leaf spot, thus showing resistance. Kanva-2 accession therefore proved to be a suitable accession that can be utilized in mulberry disease resistance breeding programmes.

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