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Monitoring the Dispersal Potential of Bark Beetle, *Hypocryphalus mangiferae* Stebbing (Scolytidae: Coleoptera) in Mango Orchards

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Abstract.- The bark beetle, *Hypocryphalus mangiferae* has been consistently found in Pakistan on mango trees suffering sudden death disease. In this study we estimated the efficiency of different monitoring techniques, dispersal activity and dispersal distance of *H. mangiferae*. The response of the beetle towards coloured sticky traps and ethanol baited traps was almost negligible whereas, the tree-log traps proved to be the most efficient monitoring methods in the orchards. The peak dispersal activity in terms of number of holes was observed in the month of May by using tree-log traps. The activity of *H. mangiferae* was positively related with temperature and negatively related with relative humidity. The dispersal distance of bark beetle was measured with the help of tree log traps on 6 different distances. The maximum attraction was recorded at 5 meters from the source tree which gradually decreased up to 60 meters. Based on these findings, we conclude that monitoring dispersal activity with tree log traps can be an appropriate part of integrated management of mango bark beetles.

Key words: Mango, bark beetle species, monitoring techniques, dispersal and dissemination.

INTRODUCTION

Mango (*Mangifera indica* L) is an important fruit tree of tropical and subtropical regions of the world. Pakistan is ranked third in the world for production of mango (12.25 million tonnes), cultivated on an area of 215000 hectares (Anonymous, 2007). Pakistan earns 9012 million rupees in foreign exchange per year by exporting mangoes to many countries *i.e.* Dubai, Saudi Arabia, UK, Germany, France, Holland, Switzerland, Italy, Singapore and Malaysia (Anonymous, 2008).

In many parts of the world *i.e.* Brazil, USA, Oman *etc*., mango orchards have been damaged by a destructive disease known as the 'quick decline' or 'sudden death'(Batista, 1960; Ploetz *et al*., 1996; Al Adawi *et al*., 2006). Mango quick decline is the most recent rigorous threat to the Pakistan mango industry. The trees die suddenly in large numbers and there is no end in sight (Masood *et al*., 2008b; Masood *et al*., 2009). Initial symptoms of the disease include gummosis from the bark which leads to branch death on affected trees. Later symptoms include vascular discoloration beneath the gummosis which turns into inconspicuous trunk cankers *i.e.* a point where the bark appears darker than normal due to decolourisation (Masood *et al*., 2008b). The diseased tree usually dies within 6 months after the appearance of first symptoms.

The majority of diseased mango trees show the presence of tiny holes made by the bark beetle (*Hypocryphalus mangiferae*). Al Adawi *et al*. (2006) isolated two disease causing fungi *i.e.* *Lasiodiplodia theobromae* and *Ceratocystis fimbriata*, from affected mango trees and *H. mangiferae*. To date, the bark beetle, *H. mangiferae* has not been proved as a vector of the disease causing fungi.

H. mangiferae is phloeophagous, since it mostly feeds on phloem. These are typically soft bodied and yellowish when freshly emerged but soon harden and turn reddish to dark brown. *H. mangiferae* make characteristic irregular galleries with a black appearance (Masood *et al*., 2008a). Populations of the most of bark beetle species increase rapidly when there is abundance of decadent trees (Wood, 1982) *i.e.* wind fallen and trees weakened due to water, disease, nutrient or salt stresses. Masood *et al*. (2009) observed the peak activity of *H. mangiferae* in the month of May and remained active from February to November to variable degrees under subtropical climatic conditions of Punjab, Pakistan. It usually preferred diseased and stressed mango trees and bored tiny holes (1.9-2.0mm in diameter) in the main tree trunk

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from which frass was emitted out. The proper monitoring of *H. mangiferae* is inevitable in the mango orchard to enable management practices to be carried out in time to keep this possible disease vector away form healthy trees (Masood *et al*., 2009).

Various methods of monitoring the population dynamic of the bark beetles have been devised. Many scientists have reported that bark beetle species (Scolytidae) are attracted to ethanol and host monoterpenes (Lindelow *et al*., 1993; Kelsey, 1994; Kelsey and Joseph, 1999). Ethanol excreted naturally by plants under stressful conditions plays a significant role in the attraction and the selection of host by bark beetles (Kelsey and Joseph, 2001). Abbasi *et al*. (2007) reported the attraction of bark beetle towards coloured sticky traps *i.e*. yellow, white, green, black and red. The most efficient sampling technique in forests is perhaps the log trapping *i.e*. placement of wood logs in the proximity of infested trees (Leather *et al*., 2005) for a specific period of time, during which naturally occurring bark beetles colonize the logs and eventually produce new generations that can then be collected and counted (Tavakilian *et al.,* 1997). In this paper, we used log traps for monitoring bark beetle in mango orchards for the first time.

Our objectives were to evaluate various monitoring techniques for the dispersal of *H. mangiferae* in mango orchards and to determine if the dispersal of the beetle is associated with temperature and relative humidity. Finally we were also interested to establish the dispersal distance covered by *H. mangiferae* as it could help to formulate its management strategies.

Since the risk of a tree being attacked is not only related to proximity of infestation but also depends on the density of the beetles and the susceptibility of the trees (Wichmann and Ravn, 2001), the dispersal activity is only practicable method to assess their population in time for proper management which can otherwise cause tremendous economic losses (Faccoli and Stergulc, 2006).

MATERIALS AND METHODS

Study site

The study was performed at Faraz farm (30.270º North and 71.250º East) and Nawaz farm (30.271º North and 71.252º East) near Khanewal road, Multan, during the years 2007-09. The orchards were comprised of 2000 and 3000 trees with a total area of 25 and 12.5 acres, respectively. The most frequently grown commercial varieties were Chounsa, late Chounsa, White Chounsa, Ratol, Dosehari, Sindhari and Langra. These were 15-20 years old poorly managed orchards. Seven trees were uprooted during year 2007 due to the attack of mango sudden death syndrome.

Monitoring techniques of H. mangiferae

The monitoring techniques of *H. mangiferae* were included coloured stick traps (yellow, white, green and red), alcohol baited traps and tree-log traps. The experiment was performed under Randomized Complete Block Design (RCBD). The coloured sticky traps were made up of plastic sheets (30.48x 30.48 cm) and hung in front of bark beetle infested tree at a height of 3 feet from ground; grease being used as the sticky substance. Ethanol baited trap was specially designed by making a window-hole in the sidewall of a plastic water bottle containing water at the bottom; 95% ethanol was used as an attractant in a small vial (Fig. 1). Mango tree-log traps (50-60 cm in length and 30- 40 cm in width) were obtained from healthy tree surrogates (Tavakilian *et al.,* 1997).

Monitoring the dispersal activity of H. mangiferae

From 1st April to 30th July, 2007, the beetles caught using sticky and ethanol baited traps were recorded at fortnight intervals whereas; the entrance holes made by *H. mangiferae* in tree log traps were recorded at one month interval. The older logs were replaced with fresh ones after each recording event. The installation of fresh logs was done around the same or some other infected trees depending on tree conditions (completely dried and dead trees were avoided). Since the tree-log traps exhibited highest efficiency in year 2007, we repeated this method exclusively during 2008 (January to December,). In each month, ten fresh tree-logs were placed near the trunk of the infected living tree with clear symptoms of mango sudden death (oozing, wilting *etc*.) and bark beetle attack *i.e.* circular holes.

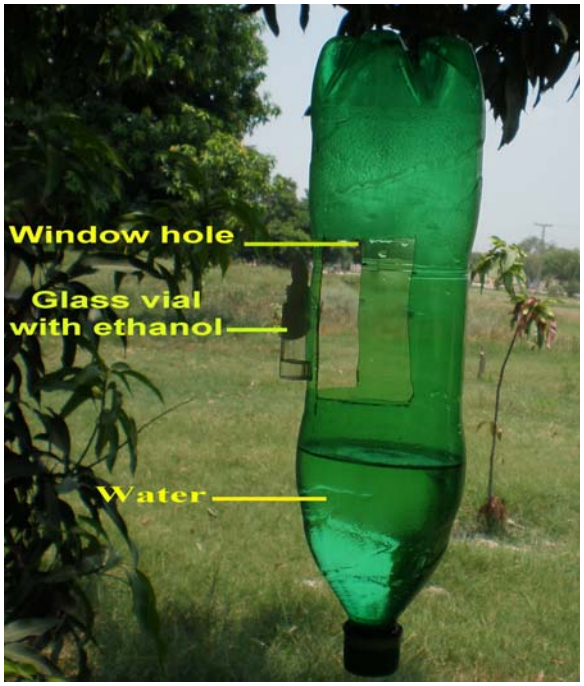

Fig 1. Ethanol baited trap for mango bark beetle.

Dispersal distance of H. mangiferae

To find the potential dispersal distance among *H. mangiferae* population, tree log traps were installed at six distances from the infested bark beetle trees *i.e*. 5, 15, 25, 35, 45 and 60 m. At each dispersal distance, five mango logs were installed during the peak activity period (15^{th} April to 15^{th} May) for two consecutive years (in 2008 at Fraz farm and in 2009 at Nawaz farm). The attraction of beetles was determined by monthly counts of the number of holes on each log at the six distances from the source bark beetle infested tree.

Statistical analysis

The number of adult beetles caught by coloured sticky, alcohol baited traps and holes on log traps were subjected to statistical analysis using analysis of variance (ANOVA). The means were compared using a Least Significant Difference (LSD) test with confidence interval of 95%. We used linear regression analysis to find the relationship between bark beetle activity in terms of number of holes and temperature and relative humidity during the year. The data was analyzed using XLSTAT computer software (XLSTAT, 2008).

RESULTS AND DISCUSSION

Monitoring techniques of H. mangiferae

There was a significant difference in bark beetle attraction to the six tested monitoring treatments (green, yellow, white and red sticky traps, alcohol baited traps and tree-log traps) during four observation months *i.e*. April to July, 2007 (Table I). The attraction of bark beetle towards all the four coloured stick traps and alcohol baited trap was statistically non-significant during all the observation months, whereas the maximum attraction of beetles was observed in tree-log traps in terms of number of holes (207.55). As a whole, the attraction of beetles towards coloured and alcohol baited traps showed their lack of preference towards these treatments. The response of *H. mangiferae* towards different coloured traps has been reported for the first time. Saint-Germain *et al.* (2006) reported varying degrees of non-preference among wood feeding beetles (Scolytidae and Cerambycidae: Coleoptera) towards coloured sticky traps in Canadian jack pine (*Pinus banksiana* Lambert). The reduced response of bark beetles towards alcohol baited traps was due to the difference of ethanol concentration, since we used absolute ethanol (99%). Saint-Germain *et al*. (2007) also reported wood inhabiting beetles (Scolytidae: Coleoptera) as more attracted to a high-release commercial blend of ethanol and α-pinene (Phero Tech Inc., Canada) which produced a stronger volatile concentration. Insects usually use various cues for the selection of their host, either visual or olfactory (Bernays and Chapman, 1994). The effective attraction radius of a baited trap also depends on the release rate of the attractants and species of bark beetles involved (Byers *et al.,* 1989). Primary and random attraction has also been reported as a principal mechanism of host finding behaviour in different bark beetle species (Person, 1931; Moeck *et al*., 1981). Primary attraction is the relative response of beetles to volatiles produced by the host, which is a very important step in host selection and colonization (Person, 1931). Random attraction relies on insects flying and landing on trees by chance and then assessing the condition of potential hosts for further colonization, which is based on olfactory and gustatory cues (Byers, 1995).

In contrast to coloured and alcohol baited traps, tree-log traps attracted more *H. mangiferae* (Table I). Within Scolytidae, host selection behaviour is mostly related to the physical and physiological state of the preferred host (Wood, 1982). Log trapping is considered an efficient method of monitoring bark beetle populations, because wood inhabiting insects are attracted to stressed, moribund or dead timber; healthy trees are normally resistant to borer attack by chemical or physical defences (Leather *et al*., 2005). Therefore, bark beetle species are attracted to recently cut logs, due to their weakened defences and the emission of volatiles (alcohol or monoterpenes) under stressed condition (Wood, 1982; Brattli *et al*., 1998; Hanks, 1999; Pureswaran and Borden, 2003). The emission of volatiles from cut tree logs has also been demonstrated in different field (Tavakilian *et al.,* 1997) and laboratory studies (Mustaparta *et al*., 1979; Huber *et al*., 2000; Pureswaran *et al*., 2004).

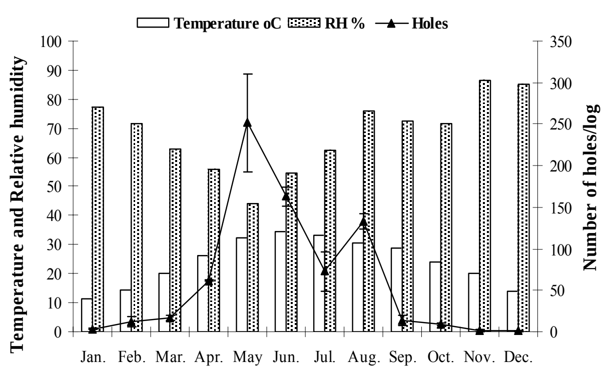

Fig 2. Activity of bark beetle, *H. mangiferae* in relation to Temperature (°C) during the year, 2008.

Dispersal activity of H. mangiferae

At the end of spring (end of March), when temperature increases and humidity decreases, the beetles start dispersal flights. The activity of bark beetles, as measured by counting numbers of holes, started in the months of February to April (Fig. 2) and reached a peak in May *i.e.* 252 holes. A sharp decline in dispersal activity was observed in the month of June *i.e.* 163 holes, which might be due to sudden decrease in relative humidity (Fig. 2). The boring activity almost ceased in September but continued at a low level until December. This suggests that the hibernation period of *H. mangiferae* tends to extend from September to February and is induced at temperatures lower than 20°C and relative humidity higher than 75%. The linear regression analysis also revealed that beetle activity was positively related between temperatures (Fig. 3) and negatively with relative humidity (Fig. 4). Masood *et al.* (2009) studied the life cycle and biology of *H. mangiferae* and reported that it over-winters as a pale yellow immature adult under the phloem portion of the mango tree. Tree-log trapping is a very effective sampling technique used by many forest ecologists under different climatic and geographical conditions. Lawson (1993) estimated the seasonal activity of bark beetle, *Ips gradicollis* on Australian pine by using tree-logs in infested forest, whereas Tavakilian *et al.* (1997) successfully monitored tropical longhorn beetles (Cerambycidae: Coleoptera) in French Guiana using the same technique. In agricultural landscapes, the same method has also been tested by Buhroo and Lakatos (2007) for monitoring the dispersal activity of, *Scolytus nitidus* (Coleoptera: Scolytidae) in apple orchards.

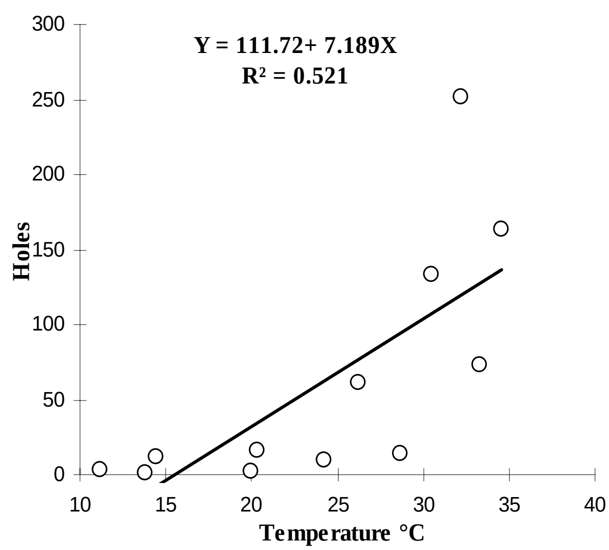

Fig 3. Relationship between number of holes per log and temperature from January to December, 2008 at Multan.

Table I.- Comparison of monitoring techniques efficacy in trapping *H. mangiferae* from April-July, 2008 in orchard. Mean values sharing similar letters show non-significant differences (P<0.05) by using LSD test.

Monitoring techniques	Number of beetles or holes observed				Cumulative beetles or holes
	April	May	June	July	
Green sticky trap	5.00b	4.40b	2.60b	3.00b	3.75 b
Yellow sticky trap	2.80b	3.00b	1.60b	1.40b	2.20 b
White sticky trap	1.60b	1.60b	1.20b	0.80b	1.30 b
Red sticky trap	0.80b	0.80b	0.20b	0.80b	0.60 b
Alcohol trap	6.00b	4.40b	2.60b	7.00b	5.00 b
Tree-log traps	72.60a	55.60a	314.6a	387.4a	207.55a
ANOVA results *F; df; P*	7.2; 5; 0.000	9.8; 5; 0.000	23.6; 5; 0.000	66.6; 5; 0.000	98.3; 5; 0.000

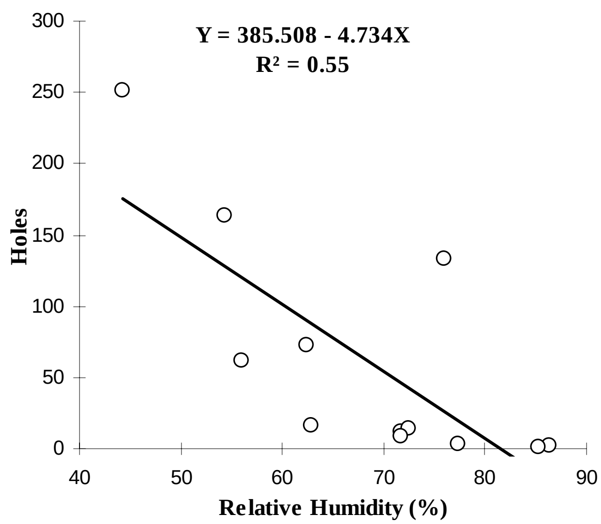

Fig 4. Relationship between number of holes per log and relative humidity from January to December, 2008 at Multan.

Dispersal distance of H. mangiferae

There was a significant difference in dispersal potential of *H. mangiferae* among the six tested distances during two consecutive years *i.e.* 2008 and 2009 (F=4.38 P=0.006 and F=12.36 P <0.0001, respectively). The maximum attraction in terms of number of holes was observed at a distance of 5 meters from the source tree (Fig. 5). The attraction gradually decreased with the increase in distance from the source tree. At Fraz farm, the maximum number of holes was observed at a 5 meter distance followed by 15, 25 and 35 meters *i.e.* 43, 23 and 14, respectively. There was no significant difference in number of holes at 45 and 60 meters. The dispersal pattern at Nawaz farm was similar to that at Fraz farm. The maximum number of holes was observed at 5 meters distance followed by 15 meter distance from the source tree. There was no significant difference in other four distances.

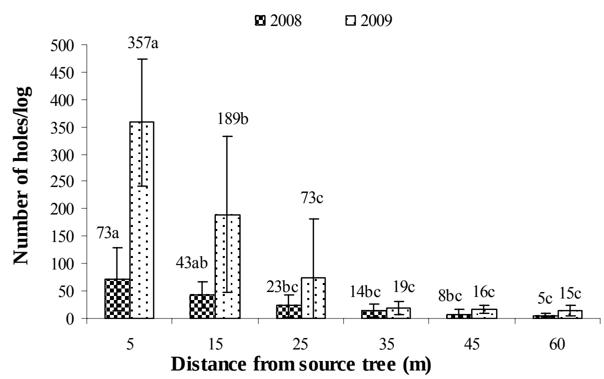

Fig. 5. Dispersal potential of *H. mangiferae* recorded at the peak activity period during April-May, 2008-09. Treatments sharing the common letters are statistically non-significant (P<0.05) by LSD test.

The dispersal distance of bark beetle depends on the release rate of volatile chemicals from cut logs and their flight capacity (Byers *et al.*, 1989). By understanding the host selection behaviour of beetles based on dispersal distances, the monitoring and management of potential damage causing bark beetle species can be implemented (Borden *et al.*, 2003;Wermelinger, 2004; Faccoli and Stergulc, 2006). This basic knowledge of dispersal distance can be helpful in making well timed management strategy decisions *i.e.* pruning of unhealthy or dried branches and chemical treatments (Civantos, 1998; Lozeno *et al*., 2001). Furthermore, the tree-log traps themselves can be used as a management tool by trapping and destroying the colonized beetles.

We conclude that tree-log traps can be utilized efficiently in an integrated control program for monitoring bark beetle and for the application of proper management practices. Furthermore, this study provides the basic information about the dispersal potential of mango bark beetle, which provides the groundwork for future studies.

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