Journal of Mosquito Research 2014, Vol.4, No.16, 1-5
http://jmr.biopublisher.ca

Research Report Open Access

Distribution of Dengue Vectors during Pre- and Post-Monsoon Seasons in three districts of Punjab, Pakistan

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Journal of Mosquito Research, 2014, Vol.4, No.16 doi: 10.5376/jmr.2014.04.0016
Received: 29 Jun., 2014
Accepted: 30 Jul., 2014
Published: 11 Sep., 2014

Preferred citation for this article:
Muhammad et al., 2014, Distribution of Dengue Vectors during Pre- and Post-Monsoon Seasons in three districts of Punjab, Pakistan, Journal of Mosquito Research, Vol.4, No.16 1−5 (doi: 10.5376/jmr.2014.04.0016)

Abstract Entomological surveys were carried out on the dengue vectors (*Aedes aegypti* and *Aedes albopictus*) during the pre- and post-monsoon seasons in 2013 from different breeding places in and around the residential areas of three Punjab districts, namely Lahore (217 m), Sheikhupura (214 m) and Faisalabad (184 m) located at different altitudes. Four vector indices (house index, container index, breteau index and premises index) were used to assess the breeding potential of vectors in each district. The house index ranged from 12 to 18 % during pre-monsoon while it was from 14 and 29% due to the post-monsoon season. The container index ranged from 11.40 to 13.17% for the pre-monsoon and from 11.22 to 30.39% for the post-monsoon. Breteau index varied from 11 and 69% during both seasons and the premises index ranged from 24.40 to 44.32%. Entomological survey also revealed that in both seasons, the highest breeding potentiality was recorded in Lahore followed by Sheikhupura and Faisalabad. The high vector indices of *Aedes* in these areas warrant intensification of vector surveillance activities along with habitat reduction and health education.
Keywords Mosquito; *Aedes aegypti*; *Aedes albopictus*; pre-monsoon; post-monsoon

Introduction

Aedes mosquitoes are generally prevalent in tropical and subtropical zones of Asia. Globally, *Aedes* (*Stegomyia*) *aegypti* (Linnaeus) *and Aedes* (*Stegomyia*) *albopictus* (Skuse) are the principal mosquito vectors of Dengue and Dengue Hemorrhagic Fever (DHF) viruses. It is considered highly invasive in nature and can carry a variety of pathogens that can be transmitted to humans. *Ae. aegypti* is the main vector that transmits the viruses. It is also known to transmit filarial infection of Wuchereria bancrofti, Dirofilaria immitis and avian parasite *Plasmodium gallinaceum* (Russell et al., 2005). This species commonly transmit Dengue virus (DENV) around the globe, with an estimated 50-100 million new dengue fever infection occur each year, causing ≥ ~500,000 cases DHF and >20,000 deaths (Gubler., 2001; Bhatt et al., 2013). It also transmits Yellow Fever, Chikungunya and Ross River viruses. The three districts selected for studies are highly diverse in terms of culture and ecology. The climatic conditions as humidity, temperature; rainfall, the forest cover, and availability of the breeding habitats render the area suitable for the existence of mosquitoes (Balakrishnan et al., 1995).

Aslam Khan (1971) reported 134 mosquito species within the boundaries of Pakistan. There was reported the presence of 22 mosquitoes species in Lahore (WHO, 2003). While Suleman et al. (1993) reported 29 mosquitoes species in Punjab. Qasim et al. (2014) reported the presence of *Ae. aegypti* and *Ae. albopictus* in Lahore and adjoining areas along with their relative abundance with special reference to the dengue fever survey. Nusrat and Naila (2011) also reported the presence of *Ae. aegypti* in Lahore. In view of uncertainty of the current remedial measures, a thorough study of human environmental interrelation, climate wise distribution, and changing pattern of occupancy have to be studied with the utmost priority before deciding control programs against mosquitoes in this ecologically fragile environment of the three districts of Punjab. There is no available information on the status of dengue vectors in Pakistan. The present study was conducted to examine the distribution and seasonal abundance

(pre- and post-monsoons) of dengue vectors and identify their active breeding sites in these districts

1 Material and Methods

1.1 The Study Area

An entomological survey was undertaken in and out of domestic premises in three different districts of Punjab, i.e. Lahore (217 m), Sheikhupura (214 m), and Faisalabad (184 m) (Figure 1) during May-June (pre-monsoon) and October- November 2013 (post-monsoon). During last five years large number of dengue cases were recorded in the selected three districts. Different places /houses were surveyed for *Aedes* breeding sites in water storage containers following the method of Sanchez et al. (2006). A total of 100 places /houses were surveyed from each study district and the larval density was expressed as house index (HI), container index (CI), breteau index (BI) and premises index (PI).

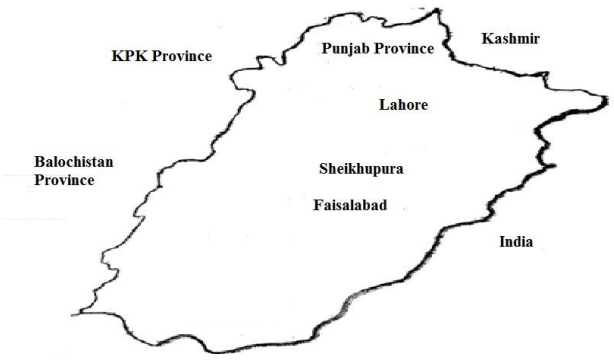

Figure 1 Location where *Aedes albopictus* and *Aedes aegypti* were collected from Punjab, Pakistan

Following three indices were calculated to determine the larval density levels of the particular locality:

1.2 House/Premise Index (HI)

Percentage of houses or premises infested with larvae/ or pupae

$$HI = \frac{\text{Infested houses}}{\text{Houses inspected}} \times 100$$

1.3 Container Index (CI)

Percentage of water holding containers infested with Aedes larvae / or pupae

$$CI = \frac{\text{Containers positive}}{\text{Containers inspected}} \times 100$$

1.4 Breteau Index (BI)

Number of positive containers per 100 houses inspected

$$BI = \frac{\text{No. of positive containers}}{\text{Houses examined}} \times 100$$

1.5 Collection of mosquito larvae and pupae

Larvae and pupae were collected from different breeding sources as cement tanks, earthen-pots, grinding stones, discarded tin, plastic container, tree holes, coconut shells, tyres, axils of leaves, drainages canals, and so forth, by the standard dipping method (Sulesco et al., 2013). Immature stages were placed in plastic containers (500 ml capacity) and were transported to the laboratory.

1.6 Preservation and Identification

In the laboratory, immature samples were allowed to emerge into adults. The adults were immobilized with chloroform and identified following the keys of Barraud (1931and 1934).

1.7 Statistical Analysis

Kolmogorov-Smirnov (with Dallal-Wilkinson-Lilliefor P value), D'Agostino & Pearson Omnibus and Shapiro-Wilk normality test has been performed to see whether the data follow a Gaussian distribution and implemented appropriate statistical tests and calculated the corresponding P values. Larval survey data of different breeding sites based on the name /type of the containers (viz. tyres, cooler, fire extinguisher buckets, etc.), construction materials (viz. tin, cement tanks, clay pots, and plastic container) were analyzed. Plain vanilla probabilistic and chi-square frequency tests were used to judge the significance of chance of getting positive containers (i.e. containers containing single *Aedes* larvae) in different areas were applied. For all areas different entomological indices, (viz. HI, CI, BI, and/or premises index) were calculated.

2 Results

2.1 Dengue Vector Abundance before the Monsoon

A total of 206 immature specimens (larvae and pupae) were collected from the three districts of Punjab before monsoon (Table 1). The results revealed that in Lahore highest collection of *Ae. aegypti* (n=66) and *Ae. albopictus* (n=41) was recorded. In Sheikhupura, *Ae. aegypti* (n=45) was higher than *Ae. albopictus* (n=32). In Faisalabad, *Ae. aegypti* (n=14) was higher than *Ae.*

Table 1 Dengue vectors collected during pre- and post-monsoon seasons from three districts of the Punjab

Locality (Punjab)	Pre-monsoon		Post-monsoon	
	Aedes aegypti	*Aedes albopictus*	*Aedes aegypti*	*Aedes albopictus*
Lahore 217(m)	66	41	96	54
Sheikhupura 214(m)	45	32	62	48
Faisalabad 184(m)	14	08	39	28
Total	125	81	197	130

albopictus (n=8) as well. Overall, *Ae. aegypti* (60.68%) was found to be higher than *Ae. albopictus* (39.32%).

2.2 Dengue Vector Abundance after the Monsoon

A total of 327 immature specimens (larvae and pupae) were collected from the three districts during post-monsoon seasons (Table 1), which was higher than the pre-monsoon collection. The highest number of *Ae. aegypti* (*n*=96) was recorded in Lahore followed by Sheikhupura (*n*=62), and Faisalabad (*n*=39). Likewise, *Ae. albopictus* was found to be abundant in Lahore (*n*=54), followed by Sheikhupura (*n*=48) and Faisalabad (*n*=28). A total number of *Ae. aegypti* larvae (60.24%) in containers was higher than *Ae. albopictus* larvae (39.76%) after the monsoon. The main breeding sources were the plastic containers, grinding stones, discarded tins, earthen pots, tyres, coconut shells, drainage canals and cement tanks, for both *Ae. aegypti* and *Ae. albopictus* after the monsoon (Table 2, Figure 2).

Figure 2 Surveyed breeding sites of dengue vectors. Positive (a), (b), (d), (e) and (f) and negative (c). (Note: (a) plastic container, (b) coconut shell (c) discarded tin (d) Grinding stone, (e) tyre (e) cement tank

2.3 Pre- and Post-Monsoon Indices of Aedes

The house index (HI) of Lahore was 18.00%; CI, BI and PI were 13.17, 27.00 and 40.27 % respectively, during the pre-monsoon season. The post-monsoon HI, CI, BI and PI indices of Lahore were 29, 30.39, 69 and 44.32%, respectively. The house index of Sheikhupura was 16%; while CI, BI and PI were 12.25,19 and 31.25%, respectively, during the pre-monsoon season. The post-monsoon HI, CI, BI and PI indices of Sheikhupura were 17, 14.09, 21 and 34.21%, respectively. The pre-monsoon house index of Faisalabad was 12%; while CI,BI and PI were 11.40,13, and 24.40% respectively while the post-monsoon HI,CI,BI and PI indices of Faisalabad were 14,11.22,11 and 27.92% respectively (Tables 3 and 4).

3 Discussion

A total of 533 immature specimens was collected from different habitats of three districts of Punjab and reared the adult stages in the laboratory. The diversity of dengue vector fauna differed among these metropolitan areas, indicating the existence of variation in the spatial distribution pattern. The variation in the diversity and distribution are mainly associated with specific ecological condition of the selected sites. Altogether two species were recorded from total emerged adults (*n* = 533) that belong to the genera *Aedes*. Among the recorded species, i.e. *Ae. aegypti* and *Aedes albopictus*, we detect the positive sample with DENV, but this was perhaps due to the high viral load in the post-monsoon season (peak season). There was no mortality recorded during the transport of larvae /pupae to the laboratory. Plastic containers and discarded tins have contributed a maximum number of dengue vector specimens. The number of recorded *Ae. aegypti* immature was higher (*n*=197) during the post-monsoon season than pre-monsoon season (*n*=125). Likewise, *Ae. albopictus* collection was also higher during post-monsoon (*n*=130) than pre-monsoon collection (*n*=81). In total, *Aedes aegypti* (*n*=322) was dominant,

Table 2 Details of different habitat collection of dengue vectors from three districts of Punjab

Locality (Punjab)	Altitude in meters	Habitat	*Ae. aegypti*	*Ae. albopictus*	Total
Lahore	217	Plastic container	36	25	61
		Grinding stones	23	12	35
		Discarded tin	24	21	45
		Earthen pots	26	18	44
		Tyres	21	9	30
		Coconut shells	12	10	22
		Drains canals	14	12	26
		Cement tanks	18	16	34
Sheikhupura	214	Tyres	12	11	23
		Axils of leaves	11	10	21
		Plastic container	21	18	39
		Tree holes	12	10	22
		Drains canals	10	11	21
Faisalabad	184	Plastic container	12	11	23
		Grinding stones	08	06	14
		Discarded tin	11	6	17
		Earthen pots	09	06	15
		Tyres	12	09	21
		Coconut shells	02	03	05
		Drains canals	04	05	09
		Cement tanks	02	04	06
Total			300	233	533

Table 3 Index profile for dengue vectors during pre-monsoon collection

Study area	No of containers surveyed (positive containers)	Indices (%)			
		HI[a]	CI[b]	BI[c]	PI[d]
Lahore	205(27)	18	13.17	27	40.27
Sheikhupura	155(19)	16	12.25	19	31.25
Faisalabad	114(13)	12	11.40	13	24.40

Note: [a]The houses Index=% of houses positive for *Aedes* larvae. [b]The containers Index= % of positive for *Aedes* larvae. [c]The Breteau Index =number of positive containers per 100 houses. [d] The Premesis Index= % of Premesis positive per 100 houses

Table 4 Index profile for dengue vectors during post-monsoon collection

Study area	No of containers surveyed (positive containers)	Indices (%)			
		HI[a]	CI[b]	BI[c]	PI[d]
Lahore	227(69)	19	30.39	69	44.32
Sheikhupura	149(21)	17	14.09	21	34.31
Faisalabad	98(11)	14	11.22	11	27.92

Note: [a]The houses Index=% of houses positive for *Aedes* larvae. [b]The containers Index= % of positive for *Aedes* larvae. [c]The Breteau Index =number of positive containers per 100 houses. [d] The Premesis Index= % of Premesis positive per 100 houses

followed by *Aedes albopictus* (n = 211). The highest *Aedes* indices were found in Lahore in post-monsoon season, where HI was 29.00, CI was 30.39, BI was 69.00 and PI was 44.32. These results support the previous studies, in which dengue vectors are being reported as major in other parts of the world (Tewari et al., 2004; Amala et al., 2011). The earlier studies conducted regarding the metropolitan area in the Asia also showed high entomological indices and even during the dry months (Gill *et* al., 2000; Sharma et al., 2005). Therefore, it is quite clear that higher authorities need to give more attention to implement control measures to overcome the mosquito-breeding problem. Mosquito breeding surrounding the

metropolitan area is not just a simple local health problem or biting nuisance, it is a serious threat to global health security. Thus, a careful invigilation by the trained scientists /vector control personnel is recommended to prevent the mosquito breeding. Moreover, future research priorities should include regular surveillance of arthropod vectors, in the metropolitan areas. A rapid action team of trained scientist and health workers, equipped with conventional and modern scientific instrumentations and suitable vector control tools, would be able to counter vector breeding during any epidemics and outbreaks situation.

4 Conclusion

In conclusion, the occurrence of dengue vectors *Aedes aegypti* and *Aedes albopictus* in both pre- and post-monsoon seasons may have epidemiological importance.

Acknowledgment

The authors are thankful to Dr. Faisal Hafeez (Mosquito specialist), Department of Entomological Research Institute, AARI, Faisalabad, Punjab, Pakistan, for his assistance during field collection and species identification.

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