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Impact of rainfall on larval density of malaria vectors in district Baghpat, Uttar Pradesh.

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ABSTRACT

Mosquito density is influenced by various climatic factors such as temperature, humidity and rainfall. Rainfall affects the mosquito population dynamics either by creation of breeding habitats or by flushing off larvae out of their habitats. Here, we explore the impact of rainfall on larval density of malaria vectors in permanent and temporary breeding habitats. Study was carried out in district Baghpat, Uttar Pradesh from July 2014 to June 2015. Larval sampling was done by dipping method before and after rainfall within a period of 2-3 days. The amount of rainfall (mm) was measured using a rain gauge. The correlation analysis was done for relationship between anopheline larval density and rainfall (mm). Maximum larval density of anopheline mosquito was 17.52 in permanent habitats which reduced to 9.2 after a rainfall of 45.6 mm. In temporary habitats the larval density was reduced from 4.02 to 1.86. The results of correlation analysis showed positive correlation between larval density and rainfall both in permanent ($r = 0.20$) and temporary breeding habitats ($r = 0.31$).The study demonstrated impact of rainfall on larval density of malaria vectors in permanent and temporary breeding habitats. An average rainfall of 45.6mm and above flushed out larvae and pupae from their breeding habitats resulting in the reduction of larval density. Further studies from different geographical areas are required to find out the cutoff value of rainfall for flushing out the mosquito larvae.

Keywords: Malaria vectors, rainfall, larval density and breeding habitat.

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INTRODUCTION

Malaria is a vector-borne disease associated with climatic factors. It is transmitted by female *Anopheles* mosquito but the disease transmission depends upon vector density, longevity, biting rate, parasite load in the community etc. Mosquito density is influenced by various climatic factors such as temperature, humidity and rainfall [1]. Which significantly affect the distribution, abundance and developmental cycle of the mosquito [2-3]. Abundance of mosquito is associated with amount of rainfall and number of aquatic habitats.

Rainfall is an important abiotic factor that affects the development of immature stages of mosquito. Moderate rainfall creates new breeding sites and provides humidity for the better survival of adult mosquito. Malaria vectors have been found to breed in a variety of aquatic habitats [4-5]. Aquatic habitats are important part where mosquitoes lay eggs, hatch to larvae and mature pupae developed in to adults but is not only required for egg laying but are also essential for the growth and development of immature stages of mosquito [6-12]. Anopheline larvae have been found in both permanent and temporary water bodies [13-14]. Shallow water bodies such as pools, pits, ditches and roadside puddles constitute some of the favorable breeding sites for mosquito larvae formed due to rainfall [15-19]. Such breeding habitats support abundance and population dynamics of malaria vectors [20]. It has been observed that malaria transmission occurs within certain radius from the breeding habitats [21].

Therefore, breeding habitats are an important part of the transmission dynamics, where *Anopheles* mosquito larvae develop. While moderate rainfall favors transmission, heavy downpour can be detrimental for the aquatic stages of mosquitoes. The flow of running water after a rainfall event can flush off mosquito larvae, leading to noticeable mortality in aquatic populations of mosquitoes living usually in stagnant water [22]. Also, direct hit of raindrops on anopheline larvae exposed to rain shower may promote larval mortality. However, in this case the direct damage may be dependent on the size of the raindrop [23]. Tuno *et al* [24] also observed a high larval mortality in open habitats in the western Kenya highlands and suggested a damaging effect of raindrops on larvae. Thus, precipitation can dramatically alter the mosquito population dynamics by promoting creation of breeding habitats, flushing off larvae out of their habitats or by directly damaging the resident larvae [23, 26]. Here, we explore the impact of rainfall on larval density of malaria vectors in permanent and temporary breeding habitats.

MATERIALS & METHODS

Study area: The present study was carried out in district Baghpat, Uttar Pradesh, India. It is geographically extended between 28˚57' North latitude, 77˚13' East longitudes. The average annual rainfall is 300-500 mm. The peak months of rainfall are from July to September. The study sites Katha, Khekra, Baragaon, and Baghpat were selected based on the high malaria cases and availability of breeding habitats (Figure 1).

Figure 1: Map showing the location of study sites (mark with stars) in district Baghpat, Uttar Pradesh, India.

Larval sampling: Anopheline larval surveys were conducted fortnightly from July 2014 to June 2015. To ascertain the impact of rainfall on breeding habitats of malaria vectors, larval sampling was done before and after rainfall within a period of 2-3 days. The larval survey was carried out in habitats present within one-kilometer radius of each locality. These habitats were observed for the presence or absence of anopheline larvae. Habitats surveyed were classified as permanent and temporary habitats such as ponds and ditches respectively.

Larvae were collected using a 350 ml dipper as per the dipping method WHO [27] from various breeding habitats and transported in plastic containers. Larvae collected from the field were reared in the laboratory for development of adult stage and their identification.

Larval density: Anopheline larval density was determined in different type of habitats before and after rainfall. Larval density per dip in a particular habitat was calculated using the following formula:

$$\text{Larval density per dip (LDPD)} = \frac{\text{Total number of larvae collected}}{\text{Total number of dips}}$$

Emergence and identification: Larvae collected from study sites were reared for further development at optimum temperature (26-28°C) and humidity (50-60%) in the insectary maintained at National Institute of Malaria Research, New Delhi. The mixture of dog biscuit powder and yeast extract (60:40) was used as larval diet. Pupae were collected in small plastic bowl filled with water and placed in emergence cages. Newly emerged adult mosquitoes were captured using an aspirator (suction tube), transferred into test tubes and anesthetized using ether (Spectrochem Pharma). Anesthetized mosquitoes were examined under a dissecting microscope and morphologically identified with the help of standard identification key for Indian Anopheline [28].

Rainfall data: Rainfall data was recorded from July 2014 to June 2015. The quantity of rainfall (mm) was measured using a rain gauge installed at Khekra study site of district Baghpat. All the rainfall events in a month were summated to give the monthly rainfall. Monthly rainfall data was correlated with anopheline larval density in different permanent and temporary breeding habitats. All the analysis was done using Excel MS Excel 2007.

RESULTS

Table-1 summarizes total monthly rainfall which ranged from 7.4 to 88.6mm but the highest rainfall 67.4mm was observed in March, 2015 and the effect of rainfall on the population dynamics of mosquito larvae in various aquatic habitats across the four study sites district Baghpat, Uttar Pradesh, India. The breeding of anopheline larvae was found in various habitats i.e., agriculture channels, ponds, pools, pits, ditches, and rice fields.

Table: 1 Impact of rainfall on anopheline larval density per dip in permanent and temporary breeding habitats of the district Baghpat.

Month	Rainfall (mm)	LDPD in permanent habitat	LDPD in permanent habitat	LDPD in temporary habitat	LDPD in temporary habitat
		Before rainfall	After rainfall	Before rainfall	After rainfall
Jul-14	12.1	16.25	18.85	1.22	1.56
Aug-14	21.1	15.49	16.74	1.62	1.80
Sep-14	45.6	17.52	9.2	4.02	1.86
Oct-14	0	0.33	3.23	0	0.06
Nov-14	0	1.51	0.12	0	0.04
Dec-14	7.4	0.2	0	0	0
Jan-15	9.1	0.4	0	0	0
Feb-15	0	0.68	0.1	0	0
Mar-15	67.4	3.36	0.41	0.85	0.17
Apr-15	10.2	0.86	1.97	1.4	2.24
May-15	29.2	5.15	6.74	0.15	0.78
Jun-15	11.3	4.03	4.11	0.98	1.58

The results clearly showed a rainfall dependent growth of anopheline larvae. Before rainfall, the maximum larval density in permanent habitats was found to be 17.52 (larval density/dip), but after rainfall (45.6mm) it was reduced to 9.2 (larval density/dip.) In temporary habitats the larval density was reduced from 4.02 (larvae density/dip) to (1.86 larvae/ dip). It was observed that the average rainfall of 45.6mm causes reduction in larval density to 8.32 and 2.16 (larvae density/dip) in permanent and temporary breeding habitats, respectively. Larval density was found reduced in both temporary and permanent breeding habitats following average rainfall of 45.6mm in the month of September.

The annual rainfall (mm) ranged from 7.4 to 67.4 with no rainfall in the month of October, November and February but larval density was recorded 3.23, 0.12 and 0.1 (density/dip) respectively, in permanent habitats. After rainfall, in the month of October, November and February larval density was recorded as 0.06, 0.04 and 0 (density/dip) in temporary habitats (Table 1).

This reduction in the larval density may be caused by flushing out of larvae from their aquatic habitats. Larval density was observed to be very low from December to February (0.2-0.68 larval density/dip) which may be due to low temperature as well as very low rainfall. Average amount of rainfall lower than 45.6mm was found to promote larval development in all habitats. Thus, the rainfall required for flushing of larvae out of their aquatic habitats was found to be 45.6mm. The results of correlation analysis showed positive correlation between larval density and after rainfall (r=0.20) and (r = 0.31) in the permanent and temporary habitats, respectively (Table 1).

Table 2: Monthly occurrence of anopheline larvae recorded in different types of habitats during 2014-15 in district Baghpat, Uttar Pradesh.

Habitat	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	Jun.
Pit	-	+	+	+	-	-	-	-	-	+	-	-
Ditch	+	+	+	+	+	-	-	+	+	+	+	+
Drain	-	-	+	+	-	-	-	-	-		-	-
Pool	+	+	+	+	+	+	-	-	-	+	+	+
Pond	+	+	+	+	+	+	+	+	+	+	+	+
Agriculture channel	+	+	+	+	+	+	+	+	+	+	+	+
Cemented channel	+	+	+	+	+	+	-	-	+	+	-	+
Rice field	-	+	+	+	+	-	-	-	-	-	-	-

(+) Presence of anopheline larvae, (-) Absence of anopheline larvae

Table 3: Anopheline larval density per dip (LDPD) before and after rainfall in localities of the district Baghpat, Uttar Pradesh

Date of collection	Rainfall (mm)	LDPD in Baghpat		LDPD in Katha		LDPD in Khekra		LDPD in Baragaon	
		before rainfall	**after rainfall**	**before rainfall**	**after rainfall**	**before rainfall**	**after rainfall**	**before rainfall**	**after rainfall**
Jul, 14	12.1	5.17	5.62	3.8	4.24	3.6	4.35	4.9	6.2
Aug, 14	21.1	4.25	6	3.69	3.5	5.96	5.6	3.21	3.44
Sep, 14	45.6	5.6	3.8	3.8	2.42	5.2	2.2	6.94	2.64
Dec, 14	7.4	0.2	0	0	0	0	0	0	0
Jan, 15	9.1	0.2	0	0	0.01	0	0	0.2	0
Mar, 15	67.4	0.6	0.08	1.51	0.12	1	0.1	1.1	0.28
Apr, 15	10.2	0.4	1.6	0.3	0.11	0.85	1.5	0.71	1
May, 15	29.2	1.3	1.16	1	2.3	1.58	3.66	1.42	0.4
Jun, 15	11.3	1.6	1.23	0.8	2.4	1.7	0.63	0.91	1.43

Further, Table 2 shows the monthly occurrence of anopheline larvae in different types of habitats. The presence of larvae was recorded in agriculture channels and ponds throughout the year, whereas in ditch type of habitat, the larvae were observed from July to November and February to June. In pits, the larval presence was recorded from August to October, whereas in drains, the larval presence was obtained in September and October. In pools, the larvae were found from July to December and April to June, whereas in rice field the presence of larvae was recorded from August to November. Highest larval density was observed in ditch, agriculture channel and cemented channel. After rainfall of 12.1mm, the maximum larvae density of 6.2 (density/dip) was obtained in July and minimum density of 0.4was recorded at the rainfall of 29.2mm in May. In December to January, 0 (density/dip) was observed in Baragaon. In Katha, the maximum larval density/dip of 4.24 was recorded in July at the rainfall of 12.1mm and minimum density of 0.01 at the rainfall of 9.1mm in January was recorded. Maximum density/dip of 4.35 was observed after rainfall of 12.1mm in July, whereas 0.1 minimum (density/dip) was obtained after the rainfall of 67.4mm in March at Khekra site. In Baragaon study site, the maximum density/dip was 6.2 after rainfall of 12.1mm in July. At a rainfall of 67.4 mm, the minimum density/dip was found to be 0.4 in May (Table 3). Finally, the rainfall of 45.6mm was found induce the reduction of larvae due to flushing off the breeding habitats.

Larvae emerged in adult from the selected locality composed of different anopheline species i.e., Anopheles culicifacies, An. stephensi, An. subpictus, An. pulcherrimus, and An. vagus with An. culicifacies and An. stephensi being the predominant vectors.

DISCUSSION

Rainfall plays an important role in malaria transmission because water not only provides the medium for the development of aquatic stages of mosquito but also maintains the relative humidity required for the survival of adult stage of mosquito [29]. Our study revealed the direct relationship between larval density of anopheline species and intensity of rainfall. On the basis of correlation analysis, the positive correlation was obtained between larval density and rainfall in both permanent and temporary habitats. As the rainfall increase, the increase in the number of mosquito breeding sites was observed. We have found that an average rainfall of 45.6mm can induce the reduction in the larval density and flushed out larvae and pupae from their breeding habitats. Rainfall may prove beneficial to mosquito breeding if moderate, but it may destroy breeding sites and flush out the mosquito larvae when it is excessive [30-30]. Grover-Kopec *et al* [31] reported that continuous and heavy rainfall flushed off the sites of vector breeding.

Paaijmans *et* al [32] and Chandrasekar *et al* [33] also investigated the effect of rainfall on various habitats and reported that heavy rainfall kills *Anopheles* larvae and flushed them out of their habitats without relating the value of rainfall. On the contrary, Bayoh [34] mentioned that pathogens or predators of mosquito larvae were sometime washed away with heavy rainfall, which increases the larval densities of various mosquito species in habitats. Zulueta *et al* [35] highlighted the rise in malaria after heavy rainfall, but also pointed out that rainfall alone is not responsible for malarial epidemics. Markl and Hauff [36] reported that the rainfall generates breeding grounds for female mosquitoes and the vibrations of rain drop on water surface acts as a driving force for larval growth, whereas Roberts [37] found that raindrops provide stimulation for hatching of larvae from eggs. Based on the above literature studies, it can be postulated that rainfall helps in the development of new habitats, whereas heavy rainfall sometimes disturb the existing habitats and washed-off the larvae, thereby decreases the density of mosquitoes [38-40].

Rainfall is a key factor affecting the survival of immature stages of vector besides other abiotic factors such as temperature and humidity also plays important role with rainfall. Russell *et al* [23] reported that the size of rainfall drop is also important for larval growth. Larval survival and development is also associated with optimum temperature of water in the breeding habitats which may be altered due to rainfall. In the present study we observed that larval density was moderate before rainfall, minimum after heavy rainfall and very low in winter season due to low temperature as well as very low rainfall. Flushing off mosquito larvae also depends on the type of habitat. Temporary habitats such as agriculture channels, ditches and pits were mostly affected by heavy rainfall in comparison to permanent habitats. Thus, rainfall is an important factor influencing larval density in natural habitats. Rainfall is a direct mortality factor for immature stages of vectors because it damages and may deform flora of breeding habitats. Larval survival and development is also associated with optimum water temperature of breeding habitats which may get disturbed by the rainfall.

CONCLUSION

The study demonstrated impact of rainfall on larval density of malaria vectors in permanent and temporary breeding habitats. An average rainfall of 45.6mm and above flushed out larvae and pupae from their breeding habitats resulting in the reduction of larval density. The study showed that *An. culicifacies, An. stephensi, An. subpictus, An. pulcherrimus,* and *An. vagus* are common anopheline species in selected sites with *An. culicifacies and An. stephensi* being the predominant vectors in the area. Further studies from different geographical areas are required to find out the cutoff value of rainfall for flushing out the mosquito larvae.

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REFERENCES

[1] Warrell D, Gilles H. Essential Malariology. London fourth edition, Arnold, 2002.
[2] Pampana E. A text book of Malaria Eradication. London, Oxford University Press 1969.
[3] Dhiman RC, Pahwa S, Dash AP. Reg Hlth For 2008; 12: 27-31.
[4] Sattler M, Mtasiwa D, Kiama M, Premji Z, Tanner M, Killeen G, Lengeler C. Malar J 2005; 4: 4.
[5] Muturi E, Mwangangi J, Shililu J, Muriu S, Jacob B. J Med Entomol 2007; 44: 336-344.
[6] Hennessy KJ, Whetton P. Australian Medical Association and Greenpeace International, Sydney 1997; 7-18.
[7] Tong SL, Hu W. Envion Hlth Persp 2001; 109: 1271-1273.
[8] Tong SL, Hu W, Mc Michael AJ. Trop Med Int Hlth 2004; 9: 298-304.
[9] Fillinger U, Sonye G, Killeen G, Knols BGJ, Becker N. Trop Med and Int Hlth 2004; 9: 1274-1289.
[10] Koenraadt CJ, Githeko AK, Takken W. Acta Trop 2004; 90:141-153.
[11] Munga S, Minakawa N, Zhou G, Mushinzimana E, Barrack O, Githeko A, Yan G. Am J Trop Med Hyg 2006; 74: 69-75.
[12] Mutuku FM, Alaii JA, Bayoh MN, Gimnig JE, Vulule JM, Walker ED, Kabiru E, Hawley WA. Am J Trop Med Hyg 2006a; 74: 44-53.
[13] Gillies MT, Coetzee M. The South African Institute for Medical Research, Pub. No.55, Johannesburg 1987: 143.
[14] Paaijmans KP, Wandago MO, Githeko AK, Takken W. PLoS One 2007; 2: e1146.
[15] Gillies MT, De Meillon B. The South African Institute for Medical Research, Pub. No.54, Johannesburg 1968: 343.
[16] White GB. Trans R Soc Trop Med Hyg 1972; 66: 572-581.
[17] Minakawa N, Mutero CM, Githure JI, Beier JC, Yan G. Am J Trop Med Hyg 1999; 61:1010-1016.
[18] Gimnig JE, Ombok M, Kamau L, Hawley WA. J Med Entomol 2001; 38: 282-288.
[19] Mutuku F, Bayoh M, Gimnig J, Vulule J, Kamau L, Walker E, Kabiru E, Hawley W. Am J Trop Med Hyg 2006b; 74: 54-61.
[20] Ndenga B, Githeko A, Omukunda E, Munyekenye G, Atieli H, Wamai P, Mbogo C, Minakawa N, Zhou G, Yan G. J Med Entomol 2006; 43: 200-206.
[21] Carter R, Mendis KN, Robert SD. Bull World Health Organ 2000; 78 (12): 246-50.
[22] Reisen WK, Meyer P, Shields J, Arbolante C. J Med Entomol, 1989; 26: 10-22.
[23] Russell PF, West LS, Manwell RD. Practical Malariology, Philadelphia, WB Saunders Company 1946; 555.
[24] Tuno N, Okeka W, Minakawa N, Takagi M, Yan G. J Med Entomol 2005; 42: 270–277.
[25] Jepson WF, Moutia A, Courtois C. Bull Entomol Res 1947; 38: 177-208.
[26] Edillo FE, Toure YT, Lanzaro GC, Dolo G, Taylor CE. J Med Entomol 2004; 41: 333-9.
[27] World Health Organisation (WHO). Manual on Practical Entomology in Malaria. WHO Division of Malaria and other Parasitic Disease, Part II Methods and Techniques, 1975; 58-62.

[28] Nagpal BN, Sharma VP. Indian Anophelines, Oxford and IBH Publishing Co. Pvt. Ltd. New Delhi 1995; 1-416
[29] McMichael AJ, Marten WJM. Ecos Hlth 1995; 1**:** 23-33.
[30] Gill CA. J Med Res 1921; 8: 633-693.
[31] Grover-Kopec E, Kawano M, Klaver RW, Blumenthal B, Ceccato P, Connor SJ. Malar J. 2005 Jan 21; 4(1): 6.
[32] Paaijmans KP, Wandago MO, Githeko AK, Takken W. PLoS One 2007; 2: e1146.
[33] Chandrasekar S, Sundaravadivelan C, Sevarkodiyone SP, Meriselvan M, Ramesh U. Int J Env Sc. 2012; 2(4): 2167-2172.
[34] Bayoh MN. Durham University of Durham, 2001: 134.
[35] Zulueta JDE, Mujtaba SM, Shah IM. Trans R Soc Trop Med Hyg 1980; 74: 624-632.
[36] Markl H, Hauff J. Naturwissenschaften 1973; 60: 432-433.
[37] Roberts D. Med Vet Entomol, 2001; 15: 215-218.
[38] Gubler DJ, Reiter P, Ebi K, Yap W, Nasci R, Patz J. Environ Health Perspect 2001; 109: 223–233.
[39] Woodruff RE, Guest CS, Garner MG, Becker N, Lindesay J, Carvan T, Ebi K. Epidemiology 2002; 13*:* 384–393.
[40] Kelly-Hope LA, Purdie DM, Kay BH. J Med Entomol 2004; 41: 133–150.