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Entomological assessment of malaria outbreak in Bareilly and Budaun districts of Uttar Pradesh, India

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Abstract
Malaria accounts for the major public health global burden caused by the protozoan parasite plasmodium and transmitted by *Anopheles* mosquitoes. Recently Malaria outbreak, particularly by *Plasmodium falciparum, Plasmodium vivax* and both parasites was reported in Bareilly and Budaun districts of Uttar Pradesh during September 2018. This study aimed to have an in-depth understanding of the entomological parameters influencing malaria transmission and guide intervention measures. Malaria endemic villages were selected for entomological investigation from both the districts. An entomological survey revealed the presence of vector species known to be important in transmitting malaria- rural vector, *Anopheles culicifacies* played a major role and was the most common species collected from cattle sheds and human dwellings. Per Man Hour Density (PMHD) of the vector species was 5.5 in Kandharpur village of Bareilly and 7.0 in Bhurinagla village of Budaun District. Per Room Density (PRD) of the vector species were highest with 2.0 in Manona village of Bareilly and 6.0 in Maree village of Budaun District respectively.

Keywords: Malaria, Outbreak investigation, *Anopheles culcifacies*, *Plasmodium falciparum, Plasmodium vivax*

1. Introduction
Almost 85% of all malaria cases globally were in 19 countries: India and 18 African countries [1]. Malaria is mostly present as an endemic disease, but in low transmission areas, it may occur as an outbreak. Malaria outbreaks are among the complex public health challenges attributed to both natural and man-made grounds. Various reports are found on malaria epidemics with the continued localized transmission [2]. In India, the scenario is mostly unstable and outbreaks occur frequently in various parts of the country. The reasons for malaria epidemics and outbreaks are identified as the inadequacy of surveillance, shortage of human resources, residual spray in rural areas, anti-larval measures in urban areas, population migration (in or out of an endemic area), increase in vector breeding sites, and potential vector population, inefficient vector control measures presence of new efficient vectors, inefficient vector control measures, drug resistance in parasite or insecticide resistance in vectors and break down in the control measures, population migration to an endemic area or vice- versa, presence of new efficient vectors, drug or insecticide resistance by the parasite & vectors and break down in the control measures [3, 4].
The malaria outbreak in Bareilly and Budaun districts of Uttar Pradesh (UP) during September 2018 was a challenge to the State Health Department, Government of UP. There have been about 37,387 cases in Bareilly and 20, 289 cases in Budaun district in 2018. The diagnosis of Malaria is carried out by microscopic examination of blood films collected by active and passive agencies. However, due to the large-scale outbreak in the districts, the Health agencies and volunteers treating fever cases in inaccessible areas are being provided with Rapid Diagnostic Test (RDT) kits (Bivalent RDT) for diagnosis of Malaria cases to provide full radical treatment to the confirmed cases. Therefore, based on the request of the State Health Department for technical support, an entomological investigation was made by the team of the National Centre for Disease Control, Delhi comprising of entomology experts.

The investigating team visited the area and after conducting an entomological survey recommended necessary control measures to contain the malaria outbreak. The investigation aimed to investigate the occurrence of malaria outbreak, identify the risk factors, and suggest practical control measures to alleviate the disease burden of the community in Bareilly and Budaun districts, UP.

2. Materials and Methods

2.1 Surveillance Areas

2.1.1 The demographic and meteorological scenario of Bareilly District

Bareilly is a metro city in the northern Indian state of Uttar Pradesh. Standing on the Ramganga river. Bareilly is the 7th largest metropolitan city of Uttar Pradesh and the 50th largest metropolitan city of India. Geographically it forms the outer gateway to enter Uttrakhand State. The Bareilly district is lying between 78.23' longitude East and 28.10' latitude North. An area of about 4120 sq.km has been covered by Bareilly district. The total forest covers an area of 352 hectares in the district. The district is bounded on the North by the Nanital district of Uttranchal, on the South by Badaun district, on the East by Pilibhit &Sahajahanpur districts, and on the West by Rampur district.

The Ramganga River is the main river of the district which makes the natural boundaries with the Badaun district. Bahgul, Shankh, Devrania, Nakatia, Kailasi are some other rivers flowing in the district. Due to the Tarai region, the district has a very pleasant climate with cool and foggy winter and generally hot & humid summer. Here summer starts very early and the temperature of the district varies from 4º C in winter to 44 ºC in summer. Normally, the wet session starts at the end of June month to September. The average rainfall is 1050 mm (Figure 1).

2.1.2 The demographic and meteorological scenario of Budaun District

Budaun is in the Bareilly Division and it is lying between 78.54 and 69.20 longitude east and 28.25 and 29.10 latitude north. The district is bounded on the north by Jyotiphule Nagar, Muradabad, Rampur, and Bareilly district and on the south by Kashiram Nagar and Aligarh, on the east by Shahjahanpur and Farrukhabad District, and the west by Aligarh and Bulandshahar district. The height from sea level is 166.4 meters in the South and 192 meters in North. River Ganga flows through the district which increases the land fertility. The Budaun district covers an area of 5168 sq km. Due to the Tarai region, the district has a very pleasant and moderate climate. The temperature of districts varies from 2 Degree Centigrade in winter to 44 degrees centigrade in summer. The wet season normally starts at the end of June month. The average rainfall is 861mm in Budaun (Figure 1).

3. Methodology

3.1 Entomological Surveillance Areas

An entomological survey was conducted in the affected villages of both districts in September 2018 according to WHO standard techniques for anopheline mosquitoes [5, 6]. The study locations comprise BehtaBujurg, Majgawan, Kandharpur, Manona, Kundarai Khurd, Rampura, and Andupura which are the selected villages of Bareilly district and Maraee, Musajhaag, Mehrauli, Ghidhol and Bhurinagla are the selected villages of Budaun district.

3.2 Adult Anopheline Surveillance

3.2.1 Indoor Hand Collection (HC)

Hand Collection of Indoor resting adult mosquitoes was done from randomly selected households and cattle shades in the morning from 05:00to 09:00 hrs by using an Aspirator and torchlight. Each house was surveyed for 15 minutes. Bedrooms, preferably with complete walls and the highest number of persons slept last night were given priority. The dead and alive mosquitoes were brought to the laboratory for further processing to study various entomological parameters. Per Man Hour Density (PMHD) of each species of mosquitoes were calculated by using the following formula:

PMHD= Total no. of mosquitoes collected /No. of person x Time spent in hours

3.2.2 Spray Sheet Collection (SC)

Adult mosquitoes were collected from indoor surfaces from randomly selected houses per day in the morning hours at 05:00 to 09:00hrs by Spray sheet collection. The Spray sheet mosquito collection method was conducted for the adult mosquito collections [7]. This involved the covering of the floor with white bedsheets. Insecticide was first sprayed from outside of the house onto the eaves, windows, and door before entering the dwelling and spraying the entire inside of the house. All doors and windows remained closed for about 10 min to allow for mosquito knockdown. Collectors then reentered the dwelling and used forceps to collect mosquitoes from the sheets and place them in test tubes. A commercial-grade pyrethroid insecticide (Mortein, active ingredients d-phenothrin, and imiprothrin) was used due to availability and safety. The mosquito collections were placed into test tubes labeled with information that included; location, date, time of collection, number of people per room. The collected mosquito samples were kept for species identification and PRD (Per Room Density) was calculated with the help of the following formula:

PRD = No. of mosquito collected / no of rooms

3.3 Larval AnophelineSurveillance

Entomological investigations based on the larval survey were done to understand the larval species prevalent in the area. Larvae were collected with the help of dippers form the breeding habitats. All kinds of breeding habitats; artificial (overhead tanks, plastic containers, iron/metal drums etc.), and natural (rivers, streams, ponds, wells, rice fields, etc) were searched within one kilometers radius of the affected villages. The community was also analyzed for the examination of waterlogged and storage practices in affected areas. Anopheline mosquito larvae were searched using dippers of 10-centimeter diameter and 300 ml capacity (five dips at each site). Larval samples were brought to the laboratory for rearing till the adult emergence and then the mosquitoes were identified up to species level. Larval densities were expressed per site as the number of larvae per five dips and larval density was calculated. Per Dip Density was calculated with the help of the following formula:

Per Dip Density = No. of Larvae collected / Total no. of Dip

3.4 Mosquito processing and Identification

All mosquitoes collected by HC, SS methods, and the

mosquitoes that emerged from the collected larvae were identified up to species level using an achromatic magnifying lens (×10) and the appropriate taxonomic keys [8].

4. Results

4.1 Epidemiological situation and indicators for malaria (2017–2018)

The districts are highly endemic for malaria in 2018 (annual parasitic index>1). The API increased in 2018 and reach 7.32 in Bareilly and 5.54 in Budaun district. However, API is low in 2017 with 0.06 in Bareilly and 0.42 in Budaun district in 2017. The Annual Parasite Incidence (API) depends upon the ABER whereas the level of ABER depends on the number of fever cases reported in the community. However, the Slide Positivity Rate (SPR) among the blood smears collected through surveillance gives more accurate information on the distribution of malaria infection in the community over a while. The slide positivity rate (SPR) encountered higher in 2018 for both districts with 14.94 in Bareilly and 12.72 in Budaun respectively. (Table 1).

4.2 Entomological observation

Mosquitoes collected during the survey belong to three genera viz. *Anopheles*, *Culex,* and *Armigers*. Among the total collection, Anophelines were the most dominant genus comprising and the species were *An. culicifacies*, *An. annularis*, *Anopheles subpictus*, *Anopheles maculates*, and *Anopheles barbirostris. Anopheles subpictus* was the most dominant species followed by *An. vagus*, *An. culicifacies* and *An. annularis*. The PMHD of the primary vector *An. culicifacies* was 5.5 in Kandharpur followed by Manona with 5, Andupura with 5, BehtaBujurg with 3 and Kundarai Khurd with 2 of Bareilly district. However, the primary vector was not found in the Majgawan and Rampura villages of Bareilly. Whereas in PMHD in affected villages of Budaun district shows PMHD of 7.0 in Bhurinagla of which was higher than the critical density (3.3) followed by Maraee with 3.5, Musajhaag with 2, Mehrauli with 1, and no vector density was found in Gidhool village of Budaun district. The PRD was found highest with 2.0 in Manona village of Bareilly and 6.0 in Maraee village of Budaun district. The PMHD and PRD of all other collected mosquito species were given in table 2 and Figure 1, 2, 3, 4 showing PMHD and PRD of anopheline species collected in both districts.

4.3 Larval survey

During the larval investigation, the anopheline breeding was surveyed in various places like paddy fields, ponds, rainwater pools, mud pools, and pits, etc. at affected villages of both the districts. The details of *An. culicifacies*, *An. subpictus*, breeding in the above habitat are presented in Table 3. Both species of anophelines, *An. culicifacies*, *An. subpictus* were found in clean stagnant water, Pools, pits. Anopheline larval density per dip revealed the highest density with 10 in clean stagnant water at Bareilly district and 25 in Stagnant water at Budaun district. The high larval index was found in all potential breeding sources favoring disease transmission. The result of the larval survey undertaken is given in Table 3.

5. Discussion

Presently, Malaria is an endemic disease but in low transmission areas, it may perhaps occur as an outbreak. Malaria outbreaks are the complex public health challenges attributed to both natural and man-made causes [9]. Several reports are available on small malaria outbreaks with the continued localized transmission. A study reported a focal outbreak of malaria at Bonta sub-center of PHC Kilvani, the UT of Dadra & Nagar Haveli during August 2014 revealing the presence of three known malaria vectors viz. *Anopheles culicifacies*, *A. stephensi*, and *A. subpictus* [10].

Recently, various outbreaks of Malaria had been reported from different states of India, such as Orissa [11], Haryana [12], Chhattisgarh [13, 14], Assam [15-18] Bihar [19, 20], UP [21] , Dadra & Nagar Haveli [22] Kerala [9, 23]. Several reports are available on the malaria outbreak in the non-endemic area also. A study reported that climate change affects improving conditions for competent Anopheline vectors in non-endemic areas, resulting in outbreaks due to *P. falciparum* infection [2]. During the 2009 Malaria outbreak in March in district Korea of Chhattisgarh found with SPR 22.2% and showed the average PMHD of *An. culicifacies* (5.5) followed by *An. Subpictus* (2.0) and *An. fluviatilis* (1.5) [14]. The entomological survey revealed that all selected villages of both districts with poor living conditions, low socioeconomic conditions (education, occupation & income), lots of animal shelters (Cattle sheds), and poor drainage system which leads to waterlogging. These conditions increase mosquito density.

Primary Vector, *Anopheles culcifacies* were found have predominant breeding sites in stagnant water, Pools, and pits which were near to the malaria cases. In response to this outbreak, indoor residual spraying (IRS), larvicidal spraying, space spray, and fogging were done in the affected areas. The area has the potential for malaria to re-emerge as there is the presence of environmental and climatic conditions that are favorable towards mosquitoes of the genus *Anopheles* and vulnerability, through the constant presence of infected individuals coming from endemic areas and due to the presence of a large number of migrant laborers. The current outbreak demonstrates the potential for the reintroduction of malaria in the area. Soon before the outbreak, there was heavy rainfall in July-August. That might have caused increased humidity in the area which was suitable for the vector to survive long and parasite to grow inside the vector. As the proportion of humans: cattle were very low, man vector contact increased and transmission of malaria was high, resulting in the outbreak.

6. Recommendations

Since mosquito control was highly essential now. The outbreak could be controlled by taking immediate action by the state health department as per the recommendation given by the investigating team. The reason for the outbreak in most of the studies reported was due to breakdown of surveillance, consistently low API, villages not covered under the indoor residual spray and favorable weather conditions for vectors to grow rapidly. But from our investigation the possible reasons for this outbreak were i) abnormal and sudden climatic change resulting in a sudden increase of temperature to 42ºC and unusual rain favoring the breeding of the vector, ii) population migration from the non-endemic area to an endemic area, iii) area not covered under IRS programme.

With the experience from this study, the following recommendations were made to avert the malaria outbreak in the future.

i. Long-term and systematic monitoring of environmental risk factors, Strengthening of the vector surveillance for

the early prediction of the outbreak using the vector surveillance data,

ii. Promotion of the use of personal protection measures with long-lasting insecticide-treated bed nets(LLIN) and implementation of IRS

iii. Malathion fogging spray in areas having a greater concentration of cases must be undertaken on a priority basis.

iv. Anti-larval measures with Temephos (Abate) may be undertaken for larval control. Besides, the use of kerosene/used diesel oil in stagnant water bodies may also be undertaken.

v. Vector & larval surveillance should be carried out throughout the year to map the vector density & larval breeding sites through Health Workers/Surveillance Workers.

vi. Availability of drugs and rapid test kits should be ensured at all the hospitals for preliminary screening of cases and their prompt treatment.

vii. Waste management should be properly planned by District Health Authorities

viii. Awareness of Community through IEC & BCC must be done for the success of intervention methods. This should cover the following aspects:

a) Cause and transmission of malaria, about the vector breeding places, breeding, and biting habits, etc, symptoms of the disease, management including treatment of the cases, and community measures for prevention of breeding and to prevent man-mosquito contact.

b) Spread of awareness on preventive measures like wearing full-sleeved clothes, using insect repellents, mosquito nets, fumigation, taking additional care of children, elderly and pregnant women, and undertaking source reduction activities.

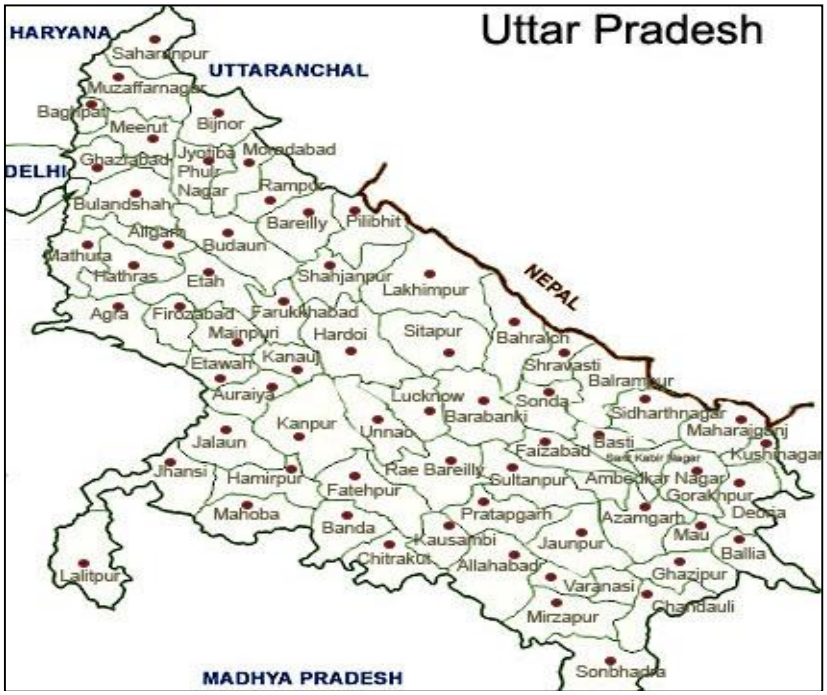

Fig 1: Map of Uttar Pradesh state showing the location of Bareilly and Budaun districts

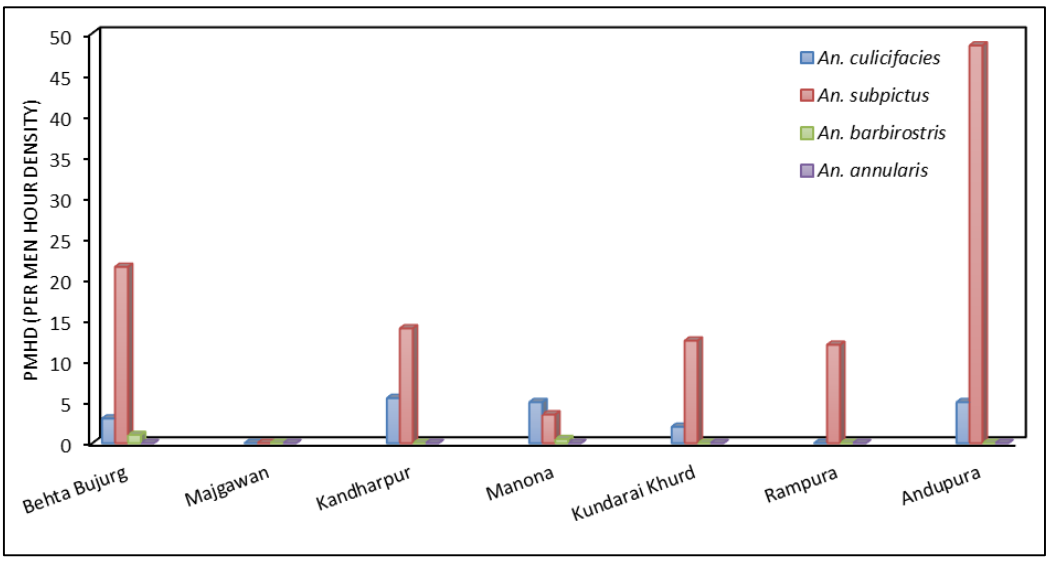

Fig 2: Comparative PMHD of different Anopheline species collected from affected villages of Bareilly district

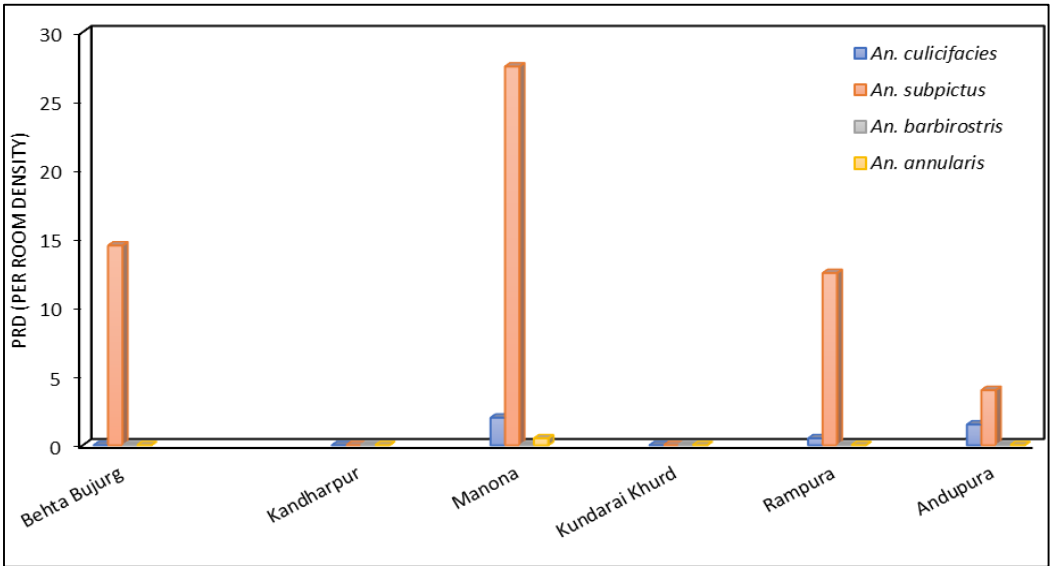

Fig 3: Comparative PRD of different Anopheline species collected from affected villages of Bareilly district

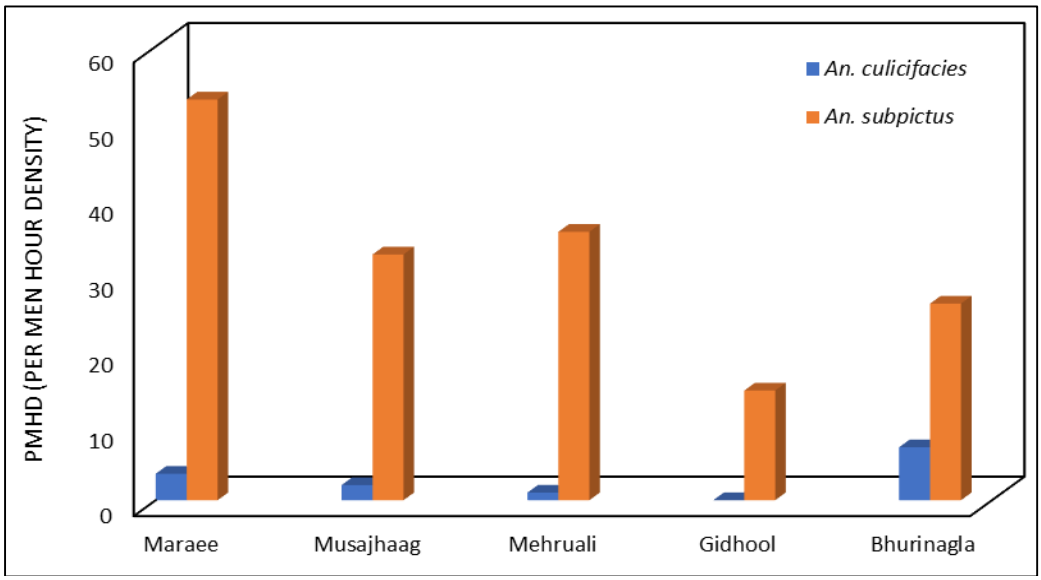

Fig 4: Comparative PMHD of different Anopheline species collected from affected villages of Budaun district

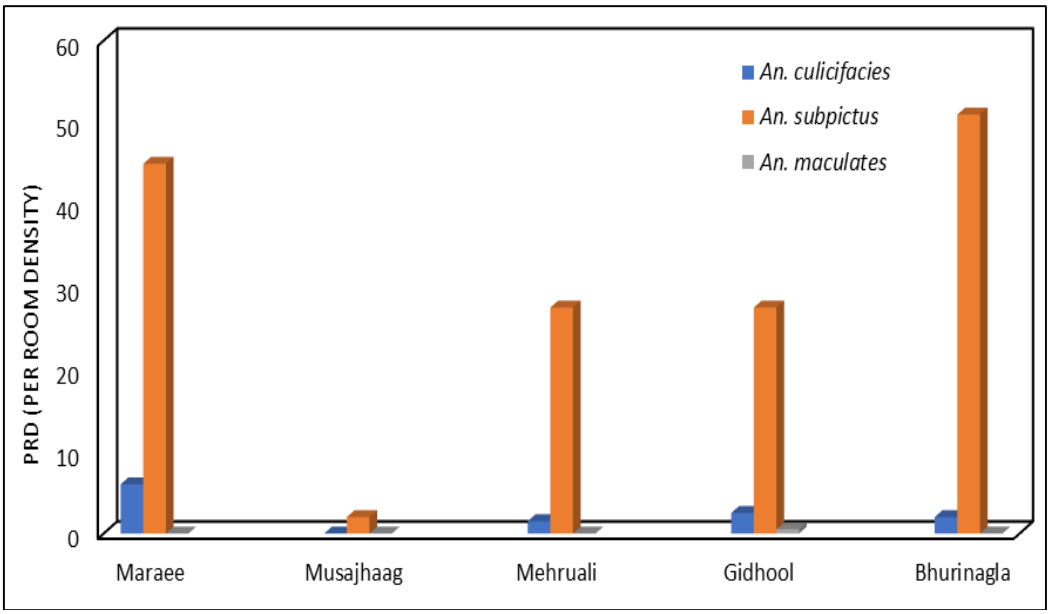

Fig 5: Comparative PRD of different Anopheline species collected from affected villages of Budaun district

Table 1: Malaria epidemiological data of Bareilly and Budaun district for 2017 and 2018 (till Sept.)

Year	District	A.B.E.R	A.P.I	S.P.R.	Total cases	Deaths
2017	Bareilly	2.02	0.06	0.29	274	0
	Budaun	2.10	0.42	2.0	1325	0
2018	Bareilly	4.90	7.32	14.94	16,802	48
	Budaun	4.35	5.54	12.72	9276	119

ABER– Annual Blood Examination Rate; API– Annual Parasite Incidence; SPR– Slide Positivity Rate

Table 2: Results of the adult Mosquito species prevalence in the affected village of both districts

Districts	Name of Village	Species	PMHD	PRD
Bareilly	Behta Bujurg	*An. culicifacies*	3.0	-
		An. subpictus	21.5	14.5
		An. barbirostris	1.0	-
		Armiger sps	1.0	0.5
		Cx. vishnui	1.0	-
		Cx. quinquefasciatus	-	0.5
	Majgawan	*Cx. quinquefasciatus*	2.5	-
	Kandharpur	*An. culicifacies*	5.5	-
		An. subpictus	14	-
	Manona	*An. culicifacies*	5.0	2.0
		An. subpictus	3.5	27.5
		An. barbirostris	0.5	-
		An. annularis	-	0.5
		Cx. quinquefasciatus	1.0	7.0
		Cx. tritaeniorhynchus	-	0.5
	Kundarai Khurd	*An. culicifacies*	2.0	-
		An. subpictus	12.5	-
	Rampura	*An. culicifacies*	-	0.5
		An. subpictus	12.0	12.5
		Ae. aegypti	2.0	2.0
	Andupura	*An. culicifacies*	5.0	1.5
		An. subpictus	48.5	4.0
		Cx. quinquefasciatus	2.0	0.5
		Cx. vishnui	0.5	-
Budaun	Maraee	*An. culicifacies*	3.5	6.0
		An. subpictus	53	45.0
		Cx.quinquefasciatus	2.5	2.0
	Musajhaag	*An. culicifacies*	2.0	-
		An. subpictus	32.5	2.0
		Cx. quinquefasciatus	1.0	-
		Cx. vishnui	0.5	-
	Mehrauli	*An. culicifacies*	1.0	1.5
		An. subpictus	35.5	27.5
		Cxquinquefasciatus	1.0	0.5
		Cxtritaeniorhynchus	0.5	0.5
	Gidhool	*An. culicifacies*	-	2.5
		An. subpictus	14.5	27.5
		An. maculates	-	0.5
		Cx. tritaeniorhynchus	0.5	-
	Bhurinagla	*An. culicifacies*	7.0	2.0
		An. subpictus	26.0	51.0
		Cx. quinquefasciatus	3.0	0.5
		Cx. tritaeniorhynchus	-	1.0
		Armiger sps	1.0	-

*PMHD=Per Men Hour Density; PRD=Per Room Density

Table 3: Showing the larval survey carried out in the affected villages during the malaria outbreak

Districts	Area Surveyed	Species Identified	Larval Density/Dip	Type of Breeding site
Bareilly	Behta Bujurg	*Cx. vishnui* *Cx. tritaeniorhynchus*	5	Pond and rice fields
	Kandharpur	*Anopheles culicifacies*	6	Rainwater collection
	Kundarai Khurd	*Anopheles culicifacies*	10	Clean stagnant water
	Andupura	*Anopheles culicifacies* *Cx. tritaeniorhynchus*	5 5	Pit Rice field
Budaun	Maraee	*Cx. vishnui* *Cx. tritaeniorhynchus*	5	Pits Drainage
	Ghidhol	*Anopheles culicifacies* *Cx. tritaeniorhynchus*	25 4	Clean Stagnant water Pools, pits
	Bhurinagla	*Anopheles culicifacies*	5	Clean Stagnant water Pools, pits

7. Conclusion

The outbreak of malaria is caused due to several factors as like climatic and non-climatic factors. Temperature, rainfall, and relative humidity are the climatic variables. Parasites, vectors, human host factors, population movement or migration, deforestation, urbanization, and interruption of control, and preventive measures are the non-climatic factors. The non-climatic factor as like vectors, human host factors may cause an outbreak during the non-transmission period. It can also be assumed that high temperature and humidity followed by spells of rain may be favoring behavioral changes in vector leading to increased parasitic load and its transmission, thus resulting in the present outbreak.

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