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Surveillance of *Aedes* mosquitoes in urban region of Udaipur district of South Rajasthan (India) to monitor potential health risk

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Abstract
Globally, the mosquito borne diseases are considered as the highest public health risk. In humans, dengue fever infection is one of the most threatening arboviral disease transmitted by the *Aedes* mosquito. These *Aedes* mosquitoes can survive almost any climatic conditions and are extremely determined. As a result, in September, 2023 an entomological survey has been undertaken to estimate the risk of vectors of vector borne diseases (VBDs) in Urban Region of Udaipur city of South Rajasthan to diminish potential health risks and prevent from vector borne disease (VBD). The cross-sectional survey was initiated in indoors to collect immature stage in Hiran Magri Sector-3, University College of Science, Ashok Nagar, Madri, Reti Stand, MLSU University Campus and Gyarawas and in around 103 houses of Udaipur city. Sampling of the mosquito larvae was conducted via dipper, pipette, and sieves and it depends on the container types. Morphologically the larvae were predicted and larval indices were also measured. Repeated surveys were undertaken in selected areas of Udaipur Urban. 60 containers were positive for *Aedes* mosquito larvae out of 128 inspected containers. Discarded Sinks, Thermocol Boxes and Disposable Plastic Cups most favorable for breeding whereas coolers were least favorable. Every entomological indices were found to be higher than the critical level for every selected areas according to guidelines given by World Health Organization, (2003; 2011). The larval indices that are House Index (HI), Container Index (CI) and Breteau Index (BI) were found 17.48, 46.88 and 58.25 percent respectively. (80%) *Aedes aegypti* and (20%) *Aedes albopictus* were identified from collected immature stages. To control these mosquitoes (*Aedes*), a cautious and constant surveillance of the study areas is recommended.

Keywords: Dengue, *Aedes* mosquitoes, dengue vector, Breteau Index (BI), House Index (HI), and Container Index (CI)

1. Introduction
Diseases caused by mosquitoes are the greatest and noteworthy public health risks worldwide. Dengue fever is a serious universal threat to the lives of above 2.5 billion people in above 100 countries. Till the mid-20th century, DF (Dengue fever) was limited to distinctive parts of the tropics and subtropics. But in the present day, Dengue Fever is endemic to Africa, Asia, the Caribbean and South America. Approximately 40% of the world's population is now living in countries with a high risk of Dengue Fever transmission. This disease is now speeded into urban and suburban areas across the tropics and subtropics of the world. In recent years, this disease has been specified as an introduced disease in Iran [4].

Dengue fever is one of the most significant arboviral diseases caused in humans and its transmission occurs through *Aedes* mosquitoes. These *Aedes* mosquitoes can survive almost any climatic conditions and are highly persistent [12]. *Aedes aegypti* is the main vector of dengue viruses worldwide. It is strictly related with human inhabitation and can also enter buildings for feeding and resting [3, 16, 11]. Over the last 25 years, there has been observed a significant increment in the distribution of *Aedes aegypti* vectors and the activity of epidemic dengue virus [15].

Aedes aegypti, unlike other mosquito species, is a day-stinging mosquito, and it often feeds on multiple hosts during a single gonotrophic cycle. Adult females particularly feed on humans but can also feed on other vertebrate species, which constitute an insignificant proportion of their blood meals. After feeding, they lay their eggs in manmade or artificial containers such as flower vases, water tanks, discarded tyres, pot plant bases, buckets or other containers; which are characteristically found around or inside the homes [18].

The use of chemicals or insecticides to control insects or arthropods possibly dates back to standard Rome and Greece. In the twentieth century, after the insecticidal potential of DDT, other insecticidal organophosphorous insecticides such as Malathion, Folithion, and Temephos were developed to control vectors [9]. Several Pyrethroids (PYs) such Aspermethrin, Cyphenothrin, Deltamethrin and Lambda-cyhalothrin; organophosphates (OPs) such Aspirimiphos-methyl, Fenitrothion and Malathion are the certain insecticides which were acclaimed by WHO for space spraying [14]. The fenitrothion was applied in space spraying for dengue and malaria mosquito control since 1970s [7, 17, 22].

Prasad and Kumar, (2020) revealed in their study that in Udaipur district of Southern Rajasthan, India Mosquito Larvicidal Oil (M.L.O.), Temephose, Bacticide T.M., Bactiobac liquid, Gambusia fishes and *Bacillus thuringiensis* var. israelensis (Bti.) powder were frequently used insecticides for control of larvae and pupae, while 1,1,1-trichloro-2,2-di (4-chlorophenyl) ethane (D.D.T.), Deltamethrin, Pyrathrum and Alpha-ciphermethrin were frequently used insecticides for control of adult dengue vectors *Aedes aegypti* (L.) in vector control programme[19].

Increase in international travelling systems and urban population, climatic changes, unsuccessful methods of vector control and lack of effective antiviral drug and vaccine are some of the factors which are responsible for the worldwide distribution of Dengue Fever [4]. Recently, due to the rise and expansion of resistance towards insecticides in dengue vectors, vector control programs are facing severe operational challenges. Larval samples of *Aedes aegypti* (L.) were susceptible to two tested larvicides, namely *Bacillus thuringiensis* var. israelensisBti (WP) and Temephos, whereas adults of *Aedes aegypti* (L.) were resistant to DDT, with death rates of less than 80%, and susceptible to Deltamethrin at the given certain diagnostic concentrations. *Aedes aegypti* (L.) respond to Malathion with death rates of 97% (for 24 hours) and 89% knock down (in 1 hour) [20].

Currently, numerous strategies such as source reduction of larvae and pupae are one of the effective and convenient methods to control dengue. According to the behavioral and biological characteristics of *Aedes aegypti* (L.), trapping their eggs or adults is more effective way of identifying the sequential placement of their interferences, which would prevent disease outbreaks eventually [5, 16].

Baruah *et al*., (2021) reported that in 21st century, a major exponential increase of dengue was reported from 2015 in the states of Delhi, West Bengal, Goa, Tamil Nadu, Haryana, Punjab, Gujarat, Pondicherry, Karnataka, Uttar Pradesh, Andhra Pradesh, Rajasthan, Kerela and Maharashtra [1].

Vector borne diseases are much predominant in South Rajasthan, which is tribal conquered hilly zone part of the state. Udaipur district has been also reported as endemic for dengue, with certain incidences recorded yearly. A cross-sectional immature stage survey was conducted indoors in Region-I (Periurban areas), Region-II (Urban areas) and Region-III (Rural areas) of Udaipur in 3645 houses. Larval sampling was done using sieves, dipper or pipette (depending on the container type). Repetitive surveys were done in urban, periurban and rural areas of Udaipur district. A total of 8733 containers were inspected. Among these, 1284 were positive for *Aedes aegypti* (L.) mosquito larvae. It was found that mud pots, plastic drums, coolers, tyres, Flower Pots and discarded buckets were most favorable for their breeding. Morphologically, larvae were identified and larval indices were measured. The larval indices; i.e. House Index (HI), Container Index (CI), Breteau Index (BI) and Pupae Index (PI) varied from 10.20% to 11.76%, from 12.84% to 16.65%, from 31.85 to 41.89 and from 32.42 to 45.02 respectively. As a result, periurban areas were found to be more prone to mosquito breeding as compared to other two areas (i.e. Urban and Rural areas) [12].

The main aim of this study was to analyze the distribution and abundance of dengue vectors at different endemic sites in Urban Region of Udaipur District of South Rajasthan; and to also monitor the susceptibility for dengue and dengue vectors in different study areas of Urban Region. Few studies on distribution and abundance of dengue vectors had also been recorded previously, so our study will be one of those pioneer studies for NVBDCP (National Vector Borne Disease Control Programme) and others to control emergence and spread of dengue vectors.

2. Material methods

2.1 Study Area

The present entomological survey was undertaken in and around Urban Region of Udaipur District of South Rajasthan in September, 2023. Among the 33 districts of Rajasthan in India, Udaipur district is one of them and it lies between the latitudes 23°46’ and 25°5’ north latitudes and 73°9’ and 74°35’ east longitude with a usual altitude of 598.00 meter (1,962 feet) and total area 64 km^2. Udaipur district is generally hilly.

The entomological survey was carried out door-to-door in peri-domestic and human dwelling areas to recognize the mosquito larval breeding sites with a view to study the level of invasion of location with the larvae of *Aedes* (Figure 1). The climate of that particular study area is hot and clammy throughout the sunlight hours and hours of darkness.

2.2 Entomological Surveillance

Tun-Lin *et al*., (1996); Focks, (2003) reported that the entomological survey of the mosquito *Aedes* has been standardized on unusual indices based on the determination of occurrence or deficiency of larvae of *Aedes* either in different water containers or container of the houses. Percentage of houses positive for immature stages (Larvae and Pupae) is called House Index (HI), percentage of containers positive for larvae and pupae called Container Index (CI), and number of positive containers per 100 houses were calculated as Breteau Index (BI) [6, 27].

Surveillance was conducted in September, 2023 in Monsoon season in Urban Region of Udaipur city in Hiran Magri Sector-3, University College of Science, Ashok Nagar, Madri, Reti Stand, MLSU University Campus and Gyarawas. Larval and pupal collection was carried out three times at each selected area with the help of trays, droppers, plastic cups, pipettes, dippers, and sieves. Every container with water was

examined. The name or type of the containers specifically as Cattle Drinking Tanks, Bird Drinking Bowls, Plastic Drums, Discarded Sinks, Disposable Plastic Cups, Thermocol Boxes, Water Storage Plastic Tanks, Discarded Buckets and Coolers. The water of narrow mouth was sucked up with the pipette. Small containers (Less than 20 liter capacity) were fully drained all the way through a strainer towards the larval sampling tray to gather pupae and larvae. 250 ml larval dipper should be used to sample in bigger containers. From the surface water three dips was taken. Some tiny plastic bottles (wide-mouthed) labeled with type of water bodies, places and date were used to bring larvae and pupae to insectary of Laboratory of Public Health Entomology, Department of Zoology, Mohanlal Sukhadia University, Udaipur, Rajasthan, India for rearing and adult emergence. These bottles having air in the top 1-2 cm. The adults who emerged out it then cautiously recognized beneath a microscope, using taxonomic key of "Pictorial Identification key of Important Disease Vectors in the WHO South East Asia Reason (2020) [30].

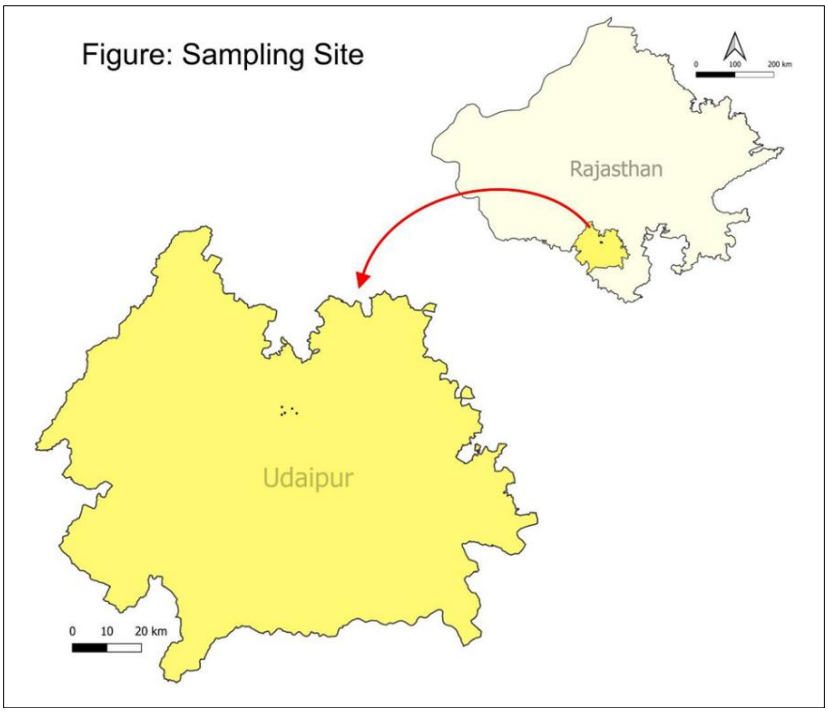


Fig 1: Sample Collection Areas of Urban Region of Udaipur District of South Rajasthan

Fig 2: *Aedes* mosquito larvae and pupae breeding in various containers: (A) Cattle Drinking Tanks, (B) Bird Drinking Pots, (C) Discarded Sinks, (D) Disposable Plastic Cups, (E) Thermocol Boxes, (F) Water Storage Plastic Tanks, (G) Coolers

2.3 Data Analysis

For calculation of risk factors of an area following calculations were done for the collected Surveillance data-

$$\text{House Index (HI)} = \frac{\text{No.of house positive with aedes larvae and pupae}}{\text{Total no.of house searched}} \text{X100}$$

$$\text{Container index (CI)} = \frac{\text{No.of container found positive with Aedes larvae and pupae}}{\text{Total no.of container searched}} \text{X100}$$

$$\text{Breteau Index (BI)} = \frac{\text{No.of container found positive with Aedes larvae and pupae}}{\text{Total no.of house searched}} \text{X100}$$

2.4 Statistical Analysis

The immature stages survey data recorded from different breeding areas based on name or type of the containers (specifically, Hiran Magri Sector-3, University College of Science, Ashok Nagar, Madri, Reti Stand, MLSU University Campus and Gyarawas) was analyzed. Plain vanilla probabilistic test to examine the importance of getting positive containers (that is containers which are having single larvae of *Aedes*) in studied areas were applied. For different areas entomological indices (specifically as HI, BI, and CI) were considered.

3. Results

In September, 2023, a survey was conducted in 103 houses of selected areas of Urban Region of Udaipur District of South Rajasthan to check the presence of *Aedes* mosquitoes in artificial breeding containers. Among these, 18 houses were reported with the presence of positive containers. Total 60 positive containers were reported among 128 artificial containers (Graph: 1). Surveillance was done in various artificial breeding containers such as Cattle Drinking Tanks, Bird Drinking Bowls, Plastic Drums, Discarded Sinks, Disposable Plastic Cups, Thermocol Boxes, Water Storage Plastic Tanks, Discarded Buckets and Coolers. Percentage

positivity in different artificial containers in study areas were depicted in Table: 1 & Figure: 2. among these different artificial breeding containers, Discarded Sinks have been reported with the most favorable breeding sites for *Aedes* mosquitoes with probability of 100%, while Coolers have been reported with the least favorable conditions for breeding with the probability of 25% (Table 1). Although other containers such as Cattle Drinking Tanks, Disposable Plastic Cups, Bird Drinking Bowls, Thermocol Boxes and Water Storage Plastic Tanks were also reported as favorable for the breeding of *Aedes* mosquitoes. The outcomes of the larval indices House Index (HI), Container Index (CI) and Breteau Index (BI) are depicted in Table: 2. HI, CI and BI were found 17.48%, 46.88% and 58.25% respectively, at study areas in the Udaipur district. These indices depicted the high invasion of the immature stages of *Aedes* mosquitoes in artificial water containers, which might be cause of an occurrence of dengue fever (Table: 2). 1652 (80%) *Aedes aegypti* and 413 (20%) *Aedes albopictus* were identified out of 2065 collected immature stages (Larvae and Pupae).

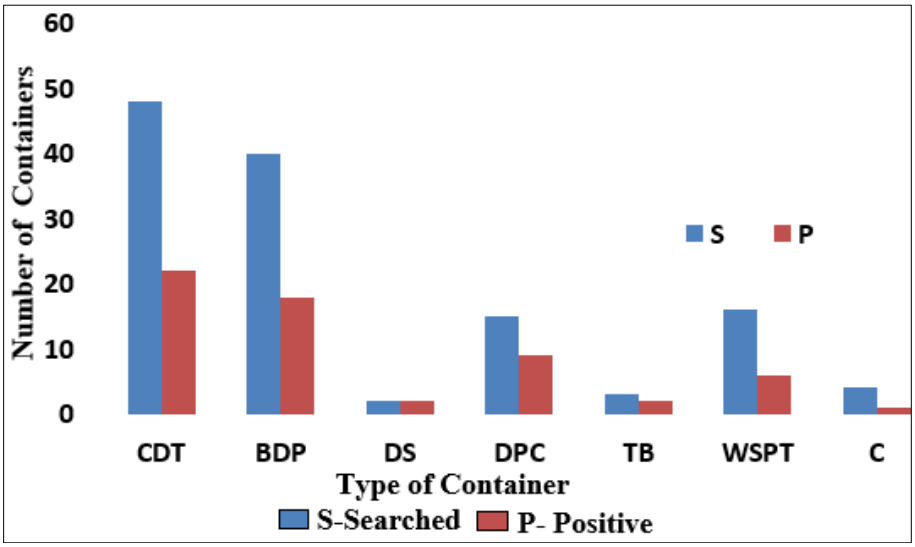
Table 1: Percentage positivity showing positive mosquito (larvae and pupae) breeding among different types of containers in Urban Region of Udaipur District of South Rajasthan during September, 2023

Area/ Containers type	CDT	BDP	DS	DPC	TB	WSPT	C	Total
Urban Region of Udaipur District	46%	45%	100%	60%	66%	38%	25%	85%

*CDT-Cattle Drinking Tanks, BDP-Bird Drinking Pots, DS-Discarded Sinks, DPC-Disposable Plastic Cups, TB-Thermocol Box, WSPT-Water Storage Plastic Tanks, C-Coolers

Table 2: Entomological Indices of Urban Region of Udaipur District of South Rajasthan during September, 2023

Region (Area)	House Index (HI)	Container Index (CI)	Breteau Index (BI)
Urban Region of Udaipur District	17.48%	46.88%	58.25%



* CDT-Cattle Drinking Tanks, BDP-Bird Drinking Pots, DS-Discarded Sinks, DPC-Disposable Plastic Cups, TB-Thermocol Box, WSPT-Water Storage Plastic Tanks, C-Coolers

Graph 1: Number of Containers Searched and Found Positive for *Aedes* Breeding in Urban Region of Udaipur District of South Rajasthan

4. Discussions

Our present study depicted that most commonly preferred breeding habitats for *Aedes* mosquitoes in study areas (Urban Region of Udaipur City) were Cattle Drinking Tanks, Bird Drinking Bowls, Discarded Sinks, Disposable Plastic Cups, Thermocol Boxes, Water Storage Plastic Tanks and Coolers. Discarded Sinks were most favourable breeding containers with 100% probability whereas Coolers were least favourable with 25% probability among all surveyed containers in study area. The majority of the residents in Udaipur district of South Rajasthan store rain water and tap water in domestic use containers. Tap water and rain water storage is one of the most common practices in these study areas. According to the reports of Kumar and Prasad, (2021) the common breeding habitats for *Aedes aegypti* (L.) in the observed study areas were found to be Coolers, Plastic Drums, Discarded Buckets, Underground Cemented Tanks, Cemented Tanks, Cattle Drinking Tanks, Mud Pots, Flower Pots, Bird Drinking Bowls, Plastic Bowls, Metallic Bowls and Tires. Among these artificial breeding containers; Coolers, Tires, Discarded Buckets, Flower Pots, Mud Pots and Plastic drums were found to be the most favorable [12]. *Aedes aegypti* are adaptive towards human residential areas and they frequently prefer urban areas, on the other hand, *Aedes albopictus* frequently prefer vegetation areas [4, 10, 26].

According to Sharma and Chharang, (2016) people are not alert about the breeding of dengue vector and approaches for prevention against them in Alwar city of Rajasthan. It is reported that the long term storage of water in containers has led to water supply problems. People were not much alert and apprehensive about dengue and its prevention. There was found that there was no alertness in the community about dengue vector breeding and its prevention. Therefore, it is important to strengthen Information Education and Communication (IEC) activities in the community to create alertness of dengue vector breeding and its control [24]. In another study done in Alwar, Rajasthan; it was reported that there was no continuous water supply even though pipelines

were present, which had led to water storage in different containers. Almost every house had cemented tanks to store water for a long time. Cemented tanks were acknowledged as principle containers which contribute for breeding of *Aedes aegypti* during the month of March, 2008 [23].

Getachew *et al*., (2015) reported that barrels (16.04%), tires (33.33%), Jericans (19.01%) and plastic drums (24.19%) were found to be most favorable for breeding of *Aedes* mosquitoes in East Ethopia and Dire Dawa, as compared to other examined containers in the entomological surveillance conducted from May-June to September-October, 2014 because of the difficulty to store rain water and tap water in containers for daily use [8].

The present study also revealed that the larval indices i.e. House Index (HI), Container Index (CI) and Breteau Index (BI) were found to be 17.48%, 46.88% and 58.25% respectively, at different locations in Urban Region of Udaipur District of South Rajasthan. According to these indices, there was a high invasion of artificial water containers by immature stages of mosquitoes (larvae and pupae) which may cause an outburst of dengue. Overall, 2065 adults had emerged out from collected immature stages (Larvae and Pupae) among which maximum were *Aedes aegypti* 1652 (80%) and *Aedes albopictus* were 413 (20%). All indices were at their critical levels. House Index, Container Index, Pupal Index and Breteau Index were found to be 58.62%, 12.44%, 141.38% and 64.66% respectively in and around different locations in Kabir Nagar, Alwar, Rajasthan during March 2016 [13].

Prasad *et al*., (2022) conducted door to door survey in 185 houses in various places of Jaipur district named as, Mahal Road, Jagatpura, Chhoti Chopad, Chand Pole, and Chaksu (Kothun) to find out the breeding preferences, relative abundance and larval density of *Aedes* and *Anopheles* mosquitoes during the month of October, 2021. 140 houses were reported positive out of 185 houses and House Index (HI), Container Index (CI), Breteau Index (BI) and Pupae Index (PI) were found as 75.67%, 46.89%, 142.70 and 25.94 respectively. Cattle drinking tanks were more favored by mosquitoes with 82.75% positivity [21].

A surveillance study was also conducted at different airports-Dum Dum of Calcutta, Santa Cruz of Bombay and Palam of Delhi. Container Index (CI) ranged from 1.6% to 86.7% at the Delhi Airport, between 5.7% and 11.2% at Vizag and between 23.4% & 30.7% at Cochin. Highest Breteau Index was recorded at old Chennai airport as 185.7, and it ranged from 1.8% to 33.3% at Delhi [25].

5. Conclusion

In the Udaipur district, the community store water indifferent domestic containers for long period of time for the daily use. In calculation to household containers, different discarded containers namely as Discarded Sinks, Disposable Plastic Cups, Thermocol Boxes, Discarded Buckets and Tyres hold rain water for long period time. This enables *Aedes* to breed in these containers. As our present study showed, mainly of the containers were infested with *Aedes* mosquitoes which may serve as vector of dengue disease. From this investigation, it is clear that there are many chances of dengue disease transmission in the sampling areas. However, to conclude whether this mosquito is transmitting ailment or not by looking for the virus in the mosquitoes needs additional investigation. There has to be a viral separation through collecting the adult females to look if they harbor the dengue disease pathogen. It also requires awareness creation of the people not to be affected by the disease in case epidemic may occur. Since this study was only in Urban Region of Udaipur District of South Rajasthan, it should also be in the whole South Rajasthan to identify the foci of the disease. In containers containing only tap water, *Aedes* mosquito larvae were not abundant and were found greatly in tap water mixed with rain water. This showed that the requirement to study water chemistry to know the motive behind the fact.

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7. Conflict of Interests

Authors do not have any conflict of interests.

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