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Mitigating dengue incidence through advanced *Aedes* larval surveillance and control: A successful experience from Pakistan

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Research Paper

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Abstract

Dengue fever is a viral disease caused by one of four dengue stereotypes (Flavivirus: Flaviviridae) that are primarily transmitted by *Aedes albopictus* (Skuse) and *Aedes aegypti* (L.). To safeguard public health, it is crucial to conduct surveys that examine the factors favouring the presence of these species. Our study surveyed 42 councils across four towns within the Bhakkar district of Punjab Province, by inspecting man-made or natural habitats containing standing water. First, door-to-door surveillance teams from the district health department were assigned to each council to surveillance *Aedes* species and dengue cases. Second, data collection through surveillance efforts, and validation procedures were implemented, and the verified data was uploaded onto the Dengue Tracking System by Third Party Validation teams. Third, data were analysed to identify factors influencing dengue fever cases. The findings demonstrated the following: (1) Predominantly, instances were discerned among individuals who had a documented history of having travelled beyond the confines of the province. (2) Containers associated with evaporative air coolers and tyre shops were responsible for approximately 30% of the *Aedes* developmental sites. (4) Variability in temperature was responsible for approximately 45% of the observed differences in the quantity of recorded *Aedes* mosquito developmental sites. (5) Implementation of dengue prevention initiatives precipitated a 50% reduction in *Aedes*-positive containers, alongside a notable 70% decline in reported cases of dengue fever during the period spanning 2019 to 2020, while the majority of reported cases were of external origin. *Aedes* control measures substantially curtailed mosquito populations and lowered vector-virus interactions. Notably, local dengue transmission was eliminated through advanced and effective *Aedes* control efforts, emphasising the need for persistent surveillance and eradication of larval habitats in affected regions.

Introduction

In recent decades, the global incidence of dengue fever has surged significantly, largely attributed to inadequate surveillance of *Aedes* mosquito breeding sites (Mehlhorn *et al.*, 2005; Buliva *et al.*, 2023). The World Health Organization (WHO) has alarmingly estimated that more than 2.5 billion individuals are at risk of contracting dengue viruses worldwide (Piot *et al.*, 2019). This upsurge in dengue cases has been accompanied by an extensive spread of the virus and its primary mosquito vector, *Aedes* species, which are now prevalent in regions between latitudes 35°N and 35°S across the world (Wichmann *et al.*, 2007; Qualls *et al.*, 2022). Currently, dengue fever is endemic in over 100 countries, spanning tropical and non-tropical regions. Additionally, instances of imported cases have been reported in non-endemic nations (Domingo *et al.*, 2015). The most substantial burden of this disease is borne by regions such as Southeast Asia, the Americas, and the Western Pacific. The first dengue cases and outbreaks in Pakistan were reported in 1960 and 1994, respectively (Rasheed *et al.*, 2013; McKenzie *et al.*, 2019). In 2011 and 2013, the largest (Khan *et al.*, 2016) and second-largest dengue epidemics worldwide occurred in Karachi, Lahore, and Islamabad Regions (Wesolowski *et al.*, 2015; Saeed *et al.*, 2023). The country's climate, urbanisation, and poor sanitation infrastructure made it an ideal site for *Aedes* mosquitoes, leading to a high burden of dengue fever (Muhammad *et al.*, 2023; Saeed *et al.*, 2023). Analysis of these outbreaks showed an increasing trend of dengue occurrence in other zones (Bhakkar, Kallurkot, Darya Khan, and Mankera) with varying degrees of intensity (Khan *et al.*, 2018). In response to these outbreaks, vector control measures, such as larval surveillance and control, have been

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implemented (Bakhsh *et al.*, 2018). The effectiveness of these measures in reducing dengue transmission has yet to be assessed in some areas (Kazi *et al.*, 2020). The escalating incidence of dengue across different regions of Pakistan, combined with the conducive environmental conditions for *Aedes* mosquito proliferation, highlights the urgent need for effective vector control measures.

Weather conditions, such as temperature, relative humidity, and rainfall, significantly influence local dengue transmission (Custódio *et al.*, 2019). Warm, wet climates provide ideal habitats for *Aedes* mosquitoes (Brady *et al.*, 2013). Defining the relationship between surveillance and weather conditions provides an effective model for devising control strategies (Colón-González *et al.*, 2011). Implementing these strategies, we can effectively reduce the larval population and generate a climatogram that serves as an early warning message. This integrated approach plays a pivotal role in effectively managing dengue transmission and mitigating its impact on public health.

Dengue virus (DENV) is transmitted in Pakistan and worldwide primarily by *Aedes albopictus* (Skuse) and *Aedes aegypti* (L.) (Khan *et al.*, 2018; Brady and Hay, 2020). *Ae. albopictus* also serves as a secondary vector for viruses (Gardner *et al.*, 2017), such as West Nile virus, yellow fever virus, and Zika virus (Benelli and Romano 2017; Leta *et al.*, 2018). *Aedes* larval surveillance and control, which involves enhanced monitoring of mosquito development sites and the application of larvicides to control mosquito populations, have reduced dengue fever incidence in other countries (Nguyen *et al.*, 2022). However, the implementation and effectiveness of these measures in Pakistan have not been evaluated (Khan *et al.*, 2018). In 2019–2020, the Punjab government developed an online electronic-based Dengue Tracking System (DTS) (Mahroof and Haider, 2022). Using this data management system, we collected data for analysing the relationship between environmental factors and dengue fever occurrence. The aim of this study was to implement an integrated approach to prevent the onset of a dengue epidemic by effectively managing the distribution and abundance of vectors. The specific objectives were as follows: (1) Reduction of vectors population across the entire area by identifying and analysing the abundance of *Aedes* species and eliminating their immature developmental sites (both indoor and outdoor). (2) Cross-validate and eliminate immature developmental sites through Third-Party Validation (TPV), employing both mechanical and chemical larviciding methods. (3) Investigate the association between *Aedes* population incidence and key weather factors. (4) Assess the effectiveness of an Anti-dengue mobile application in enhancing *Aedes* surveillance and monitoring dengue occurrence trends.

Materials and methods

Aedes *surveillance*

The entomological survey was conducted in four towns (locally called tehsil) in Bhakkar District (Bhakkar, Kallurkot, Darya Khan, and Mankera Tehsil) during 2019 and 2020. Every town was surveyed weekly, as it took a week to cover each district. A total 48 surveys were conducted each year. The district's health department assigned two indoor teams and one outdoor team to each of the district's 42 Union Councils (UC) to conduct door-to-door surveillance for the presence of *Aedes* larvae within natural and artificial containers (Table 1). Every surveillance team had an electronic device containing the Anti-dengue Android mobile application to report containers with confirmed *Aedes* larvae. Surveillance was monitored by the online Dengue Tracking System (DTS) developed by PITB (Punjab Information Technology Board). The teams uploaded data to the online dengue dashboard that tracked all the reported *Aedes* positive containers, enabling the implementation of control measures and the monitoring of positive containers.

Table 1. Indoor and outdoor containers in 2019–2020 with positive immature in District Bhakkar

Indoor sampling containers	Outdoor sampling containers
House refrigerator trays	Cemeteries
Water bath/tub	Junkyards
Bird baths	Public parks and hotels
Evaporative air conditioners	Tyre shops
Small water reservoir at home	Factories
Tyres at home	Nurseries
Discarded containers on the rooftops	Workshops
Small pits in house floor/corners	Flower pots
Plant vessels	Swimming pools
Water drums	Tubewell
Earthen pots	Animal zoo area
Flower pots	Tree holes

Most venerable containers for mosquito immature development (considered containers with larvae only).

Responsibilities of third-party validation and his role in dengue vector management

Larval *Aedes* and DENV case surveillance were conducted by district health department teams. TPV team's role was the identification and elimination of larval species through field inspection to ensure the precise allocation of resources and to avoid wasting time and effort. TPV teams conducted field inspections to validate data collection, assure accuracy, and evaluate the authenticity of the mosquito control activities and dengue cases after laboratory confirmation until sites were completely clear from the vectors. The third party here consisted of a paid independent analyst, which governmental organisations rely on to confirm performance (Abramson *et al.*, 1995). TPV confirmed the location, larval presence, and species identification and suggested control strategy according to the situation (Mintz, 2014; Rentz *et al.*, 2021). Subsequent visits were continued until the area was entirely free from *Aedes* larvae.

The Punjab government selected a scientific officer from the Arid Zone Research Institute, Bhakkar (ARZI), as the TPV agency to determine the accuracy of anti-dengue activity data within district Bhakkar. TPV of dengue vector surveillance (larval surveys) was performed through an Android mobile phone Anti-dengue TPV application, which was registered by PITB to use for the data management system DTS (fig. 1). Once a potential positive container was identified by Health Department surveillance teams, three types of surveillance were done on the android-based application; namely, audit larvae, audit patients, and audit activities (fig. 2) were uploaded via the Anti-dengue TPV application by the ARZI teams. At the same time, a photograph was taken of

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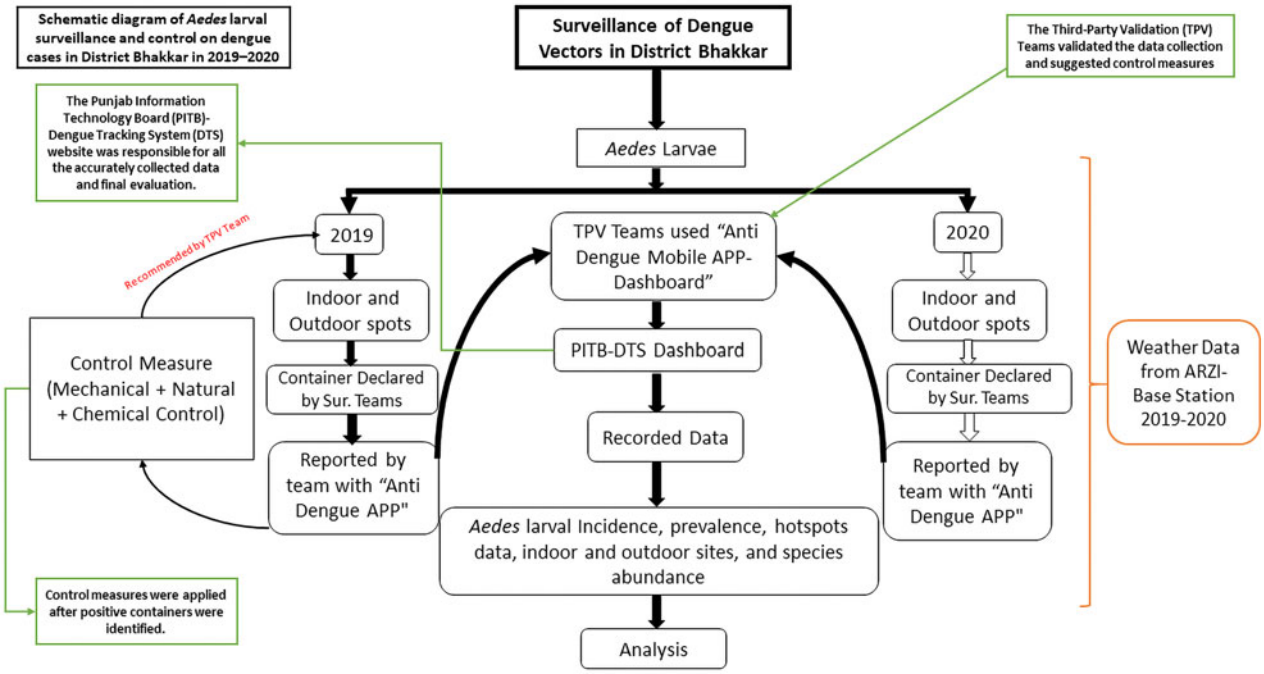


Figure 1. Schematic diagram of *Aedes* larval surveillance and control on dengue cases in District Bhakkar in 2019–2020.

each larval habitat and uploaded. ARZI teams then performed the following tasks: (1) Checked the containers based on audit larvae and the photograph. (2) Validated previous and current dengue fever cases based on blood test reports. (3) Suggest chemical control measures (larvicides: Temephos 50EC @ 1 ml/liter water; adulticides: Pirimiphos-methyl CS applied IRS @ 1000 mg/m^2 (Actellic® 300CS, Syngenta) with hand compression sprayer and Deltamethrin and Cypermethrin (2 ml/Liter water) spray applied at 7-day intervals with ULV fogger machine sprayer (ULV application rate: 0.5-1Liter/Hectare; Wall spray: 75 ml/10-Liter water). (4) *Aedes* larvae were collected from each sampling site and identified for analysing *Aedes* species abundance – larvae were morphologically identified using taxonomic keys (Rueda, 2004). (5) Finally, the teams validated the door-to-door visits based on audit activities and the GPS location of past activities.

Association of environmental data with vector population

Environmental data on temperature, rainfall, and relative humidity were obtained from regional weather stations (Arid Zone Research Institute, Bhakkar) and evaluated for their role in mosquito population dynamics. The average distance between the surveillance sites and weather stations was approximately 10–20 kilometres. Data were analysed using yearly data combined from all containers.

Statistical analysis

Data on total visited spots (sites visited for surveillance), number of positive containers (confirmed *Aedes*-larvae number ≥1 for 30 days), active patients (number of DEN positive fever cases at each location), incidence per village (new DEN cases per unit of population per time interval), and weather were derived through the DTS system and processed in two ways. (1) Correlation analysis: The incidence of positive containers per month was correlated with temperature (°C), rainfall (mm), and relative humidity (%) on monthly basis (all the variables were assumed to be random and distributed normally). (2) Multiple linear regression analysis: This was used to calculate the relationship in the form of an equation (the incidence of positive containers was considered random and distributed normally). Descriptive statistics were used to analyse *Aedes* species abundance and the indoor and outdoor container contribution to *Aedes* immature development, i.e. the number of positive containers (n) and percentage (%). The coefficient of determination (R^2) for the regression models and impact (%) were calculated using statistical software Origin 2023b. All graphical representations were made in Origin 2023b.

The prevalence of vectors in different towns was compared using the following indices.

$$\text{House Index (HI)} = \frac{\textit{Infested Houses}}{\textit{Total Houses Inspected}} \times 100$$

$$\text{Container Index (CI)} = \frac{\textit{Larvae Positive Containers}}{\textit{Total Containers Inspected}} \times 100$$

$$\text{Breteau Index (BI)} = \frac{\textit{No of Positive Containers}}{\textit{Houses Inspected}} \times 100$$

Results

Indoor surveillance indicated that evaporative air coolers (air water coolers), discarded tyres, tyre shops, discarded house freezer trays, bird baths (pans containing drinking water for flying birds), and water storage drums were the containers found most frequently positive for larvae in 2019 and 2020. Containers most frequently associated with evaporative air conditioners contributed

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Fig.1. Hotspots investigation and system information questionnaire
(Fig.1. First section displays different larval investigation hotspots, and its second section displays an information questionnaire on the Anti-Dengue Android Application System)

Figure 2. Investigation and system information questionnaire. The first section displays different larval investigation containers, while the second section presents an information questionnaire on the anti-dengue android application system; (1) Dengue mobile app dashboard: Offering various larval audit options such as Audit larvae, Audit patient, and Audit activities, teams selected audit options based on the situation of surveillance containers. (2) Audit activities: For example, the TPV Team selected this option and encountered this interface, where they input the required information and submitted it. (3) Audit patient: Patients were observed during surveillance, and their data were submitted to maintain a history. (4) Audit larvae: The TPV team had to submit information on larvae observed during surveillance.

30% and 33% of positive larvae in 2019 and 2020, respectively. Outdoor surveillance found containers positive for larvae at tyre shops, nurseries, and cemeteries, as well as standing water in tube wells in 2019 and 2020. Tyre shops were the most frequently positive containers, contributing 30% and 32% in 2019 and 2020, respectively (Table 2).

The *Aedes* species abundance from the mean number of immatures collected from indoor and outdoor positive containers in each town of Bhakkar district was determined by identifying larval populations in a laboratory (Table 3). The average number of *Aedes* larvae that were collected during surveillance revealed that *Ae. albopictus* was more abundant species as compared to

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Table 2. Contribution of indoor and outdoor positive containers to immature development

	Indoor container			Outdoor containers	
Positive containers	2019 ($n = 76$)	2020 ($n = 48$)	Positive containers	2019 ($n = 165$)	2020 ($n = 72$)
Bird pots	10 (13%)	6 (13%)	Cemeteries	24 (15%)	11 (16%)
Freezer tray	17 (23%)	13 (27%)	Tyre shops	49 (30%)	23 (32%)
Evaporative air coolers	23 (30%)	16 (33%)	Nurseries	32 (19%)	13 (18%)
Discarded tyres at home	7 (9%)	2 (4%)	Public parks	14 (8%)	6 (8%)
Discarded cups	9 (12%)	5 (10%)	Junkyards	18 (11%)	7 (9%)
Water drums	10 (13%)	6 (13%)	Tube well	28 (17%)	12 (17%)

N, number of positive *Aedes* containers; % = Container contributed to *Aedes* immature development.

Table 3. Abundance of *Aedes* mosquito species in each sampling towns in District Bhakkar Punjab, Pakistan

		Aedes albopictus (larvae)		*Aedes aegypti* (larvae)	
Towns	Positive containers	2019	2020	2019	2020
Bhakkar	Indoor	597 ± 2.34	241 ± 2.87	453 ± 3.56	165 ± 2.45
	Outdoor	621 ± 3.86	289 ± 2.77	489 ± 3.84	293 ± 2.56
Kallur Kot	Indoor	485 ± 4.53	196 ± 1.67	204 ± 2.76	92 ± 1.87
	Outdoor	533 ± 2.73	202 ± 2.97	291 ± 0.95	174 ± 2.87
Darya Khan	Indoor	580 ± 3.85	218 ± 2.44	378 ± 3.76	104 ± 3.77
	Outdoor	617 ± 1.65	279 ± 3.54	385 ± 2.96	119 ± 3.67
Mankera	Indoor	197 ± 3.34	98 ± 5.65	108 ± 3.78	47 ± 4.78
	Outdoor	239 ± 4.76	121 ± 4.54	117 ± 5.65	58 ± 5.12

Mean values with standard error of immature collected from indoor and outdoor positive *Aedes* containers.

Table 4. Indoor and outdoor surveillance activities in Bhakkar District, Pakistan during 2019–2020.

	2019			2020			
	Indoor	Outdoor	Total	Indoor	Outdoor	Total	% Change
Visited containers	732,669	215,202	947,871	1,173,218	341,665	1,514,883	+37.55
Positive containers	76	165	241	48	72	120	−50.20
TPV conducted	315	457	772	402	596	998	+22.64
Active febrile patients		102			28		−70.47
TPV satisfactory (%)		90			99		+9.09

Visited containers: Sites visited to identify *Aedes* larvae.
Positive spot: Visited containers where *Aedes* larvae were confirmed.
TPV conducted: The follow-up surveillance of the visited containers by TPV.
Active febrile patients: Confirmed patients who were carrying the dengue virus.
TPV satisfactory (%): Successfully treated containers that had been identified to have *Aedes* mosquito.

Ae. aegypti from 2019 to 2020. Specifically, more *Ae. albopictus* larvae were collected from the field than *Ae. aegypti*. Among the sampling locations, Bhakkar and Darya Khan had the highest abundance of *Ae. albopictus* larvae confirmed after identification.

In areas where surveillance was increased to control the development of *Aedes* by eliminating stagnant water and other potential developmental sites, there was a 50.20% reduction in the number of positive mosquito larval sites following an increased TPV (the follow-up surveillance of the visited spots by TPV) of 22.64% (Table 4). Additionally, there was a 70.47% decrease in the number of active febrile patients, with the rate of TPV satisfactorily (successfully treated spots that had been identified to have *Aedes* mosquito) conducted at 9.09% higher in 2020 compared to 2019. These mosquito control activities were carried out throughout the province, which has 36 districts. This 70.47% reduction in patients reflects the overall impact of all surveillance and control activities in district Bhakkar only (Table 4). All cases were individuals with a confirmed history of travel outside of the province.

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Table 5. Incidence and correlation of containers with weather factors during 2019–2020 year.

		2019 year				2020 year			
		Containers and (%) incidence	Average temp. (°C)	R.H. (%)	Rainfall (mm)	Containers and (%) incidence	Average temp. (°C)	R.H. (%)	Rainfall (mm)
Investigation date	January	1.0 (0.16)	8.34	55.36	2.54	0.0 (0.00)	10.65	55.36	0
	February	17 (2.64)	10.29	53.56	0.58	15 (1.63)	11.45	53.56	2.61
	March	31 (4.82)	14.95	54.84	0	42 (4.57)	15.62	54.84	0.23
	April	98 (15.24)	18.94	46.12	3.61	159 (18.37)	20.45	46.12	0
	May	109 (16.95)	23.75	38.44	0	149 (16.20)	25.75	38.44	1.69
	June	44 (6.84)	27.64	40.96	0	68 (7.39)	26.94	40.96	13.65
	July	37 (5.75)	29.48	61.42	10.2	54 (5.87)	30.43	61.42	0
	August	119 (18.51)	25.73	67.56	0.87	195 (21.20)	27.52	67.56	6.15
	September	127 (19.75)	23.43	59.8	0	173 (18.80)	25.61	59.84	0
	October	41 (6.38)	17.64	45.31	0	39 (4.24)	19.37	45.31	0
	November	17 (2.64)	11.73	53.73	0	16 (1.74)	13.46	53.73	0
	December	2.0 (0.31)	7.36	58.38	0	0.0 (0.00)	8.47	58.38	4.56
Total containers		643 (100%)				920 (100%)			
Correlation of containers			0.660*	−0.018^{ns}	0.562^{ns}		0.700*	0.106^{ns}	0.485^{ns}
P value			(0.01)	(0.956)	(0.124)		(0.01)	(0.743)	(0.09)
DF			11	11	11		11	11	11

Note: Containers = numbers of positive containers × numbers of valid investigation; valid investigation means investigation of at least one dengue larva being identified.

The number of containers positive was highest in September 2019, and August 2020 was significantly associated with temperature, whereas there was no significant correlation with relative humidity and rainfall. During the peak months of April, May, August, and September in 2019 and 2020, positive containers included newly positive and repeated positives in surveys to eradicate the *Aedes* immatures populations until these containers had 0% infestation after repeated surveys (Table 5). A regression equation showed the impact of these factors on per-unit change in container frequency. Temperature was accounted for 43.3 and 48.9% of the variance in the number of *Aedes* positive developmental sites recorded during the years 2019–20, respectively, whereas relative humidity was responsible for 0.1% and 0.8% during 2019 and 2020, respectively (Table 6).

The prevalence of *Aedes* presence in towns was compared by calculating the House (HI), Container (CI), and Breteau Indices (BI) (fig. 3) from indoor and outdoor positive containers. The Bhakkar township had the highest HI and BI indexes, followed by Kallurkot, Darya Khan, and Mankera in both 2019 and 2020. Different types of standing water had varied impacts on container prevalence and index calculations. The prevalence index refers to a measure of the abundance of *Ae. albopictus* and *Ae. aegypti* mosquitos in a given area. Evaporative air coolers, discarded house refrigerator trays, bird baths, water drums, tyre shops, nurseries, standing water in tube wells, and cemeteries all contributed to *Aedes* developmental sites. Maximum HI (%) index of 15.2 and 14.9%, CI (%) index of 25.3 and 21.1%, and BI (%) index of 10.8 and 7.5% during 2019–20 were recorded at tehsil Bhakkar while the lowest HI (%) index, CI (%) index, and BI (%) index were recording at Mankera, respectively.

Discussion and conclusion

Dengue fever is a serious problem in Pakistan but can be controlled through the validation of potential developmental sites and well-managed surveillance of *Aedes*-positive containers, which can result in a decrease in the number of mosquito developmental sites (Mohamud *et al*., 2020). Surveillance on the cross-validation and management of developmental sites of *Ae. albopictus* and *Ae. aegypti* mosquitoes using Anti-dengue mobile application technology was implemented, coupled with TPV control operations (natural, mechanical, and chemical control), to stop the local transmission of the dengue virus from imported cases in the Bhakkar district (Table 2; Fig. 2). People traveling to other areas, including those within Pakistan, were the main reason for the dengue cases detected in the province. If local *Aedes* mosquitoes bite these individuals, they may become virus carriers, leading to locally-acquired dengue cases (Rimal *et al*., 2023). Studies have shown that most of Pakistan's affected areas include cases with a travel history, which aligns with our findings (Ahmad *et al*., 2020; Rehman *et al*., 2020). The reduction of a significant number of positive containers of *Aedes* mosquito control resulted in no local transmission from imported cases from 2019 to 2020.

Ae. albopictus predominated over *Ae. aegypti* in all the zones of District Bhakkar, although declining gradually in 2020. In agreement, immatures of the were more abundant in Punjab than those of the *Ae. aegypti* (Saleem *et al*., 2014; Akhtar *et al*., 2022). These findings were in accordance with *Ae. albopictus* immatures found more abundant species in Punjab province as compared to *Ae. aegypti* (Wint *et al*., 2022; Ullah *et al*., 2023).

Our enhanced surveillance for *Aedes* developmental sites identified peak periods in April, May, August, and September for positive indoor and outdoor containers, indicating increased

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Table 6. Regression equation with the impact of different environmental factors on containers

Year	Regression equation	R^2 (%)	Impact (%)	SE coefficient	*F* value	*P* value
2019	X = −18.9 + 3.96 Y*	43.6	43.6	1.42	7.73	0.01
	X = −27.3 + 3.98 Y* + 0.16 Z	43.7	0.1	1.34	3.49	0.09
2020	X = −56.6 + 6.78 Y*	48.9	48.9	2.19	9.59	0.01
	X = −90 + 6.76 Y* + 0.62 Z	49.7	0.8	1.70	4.44	0.72

X, Containers; Y, Average temperature; Z, Relative humidity; R, Coefficient of determination.
* = Significant, ^{ns} = non-significant.

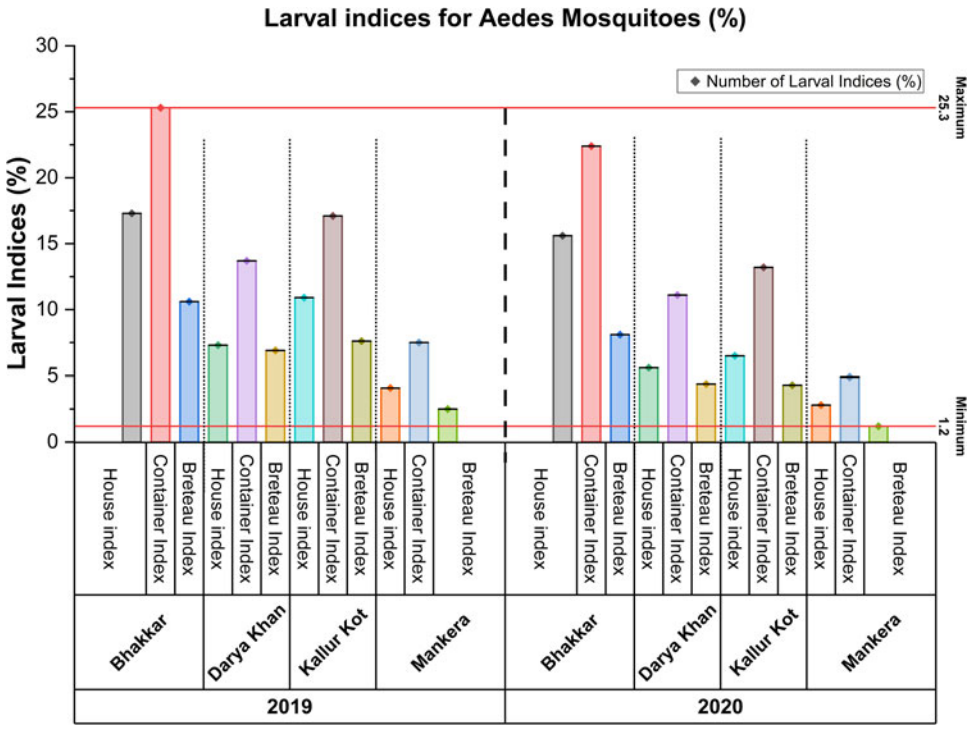


Figure 3. Different indices of four Tehsil towns of District Bhakkar in 2019–2020.

immature *Aedes* incidence due to favourable environmental conditions. The months of August, September, and October in Pakistan have the most significant number of *Aedes* larval developmental sites (Table 5). The implications of identifying peak periods for *Aedes* larval activity were profound, as they hold crucial significance for public health and vector control efforts (Patil *et al.*, 2021). Many studies are completely aligned with our findings that continuous anti-dengue activities (surveillance and control measures) posed direct reflection on the reduction of vector population and induced firm grip on elimination of local transmission (Sharmin *et al.*, 2018; Xia *et al.*, 2021). Our study supports continuous vectors management strategies using mobile technology (reduction in larval developmental sites) during the identified peak months, as this strategy disrupts the mosquito developmental cycle (egg, larvae, and pupae) and inhibits the *Aedes* adult emergence, thereby reduces local transmission (Hafeez *et al.*, 2011; Faraji and Unlu, 2016). Integrating our findings into existing public health frameworks enhances preparedness and responsiveness to potential *Aedes* developmental sites. As we move forward, continued research and surveillance efforts using mobile-based technology will be instrumental in refining vector control strategies and safeguarding communities against the threats posed by *Aedes* mosquitoes.

The temperature was responsible for the variation in the number of positive *Aedes* developmental sites recorded during the years 2019 and 2020, respectively (Table 6). *Aedes* mosquitoes thrive in warm and humid environments, with temperatures ranging from 25–28 °C being optimal for their growth and reproduction (Aryaprema *et al.*, 2023). When temperatures rise above this range, mosquito populations can decrease rapidly because of the reduction of *Aedes* developmental sites. Temperature is a fundamental driver for the *Aedes* physiology and developmental periods (Ware-Gilmore *et al.*, 2021). Our results agree with the study that temperature exerted a significant impact, while relative humidity and rainfall had limited contributions to the transmission of dengue fever (Ware-Gilmore *et al.*, 2021). Our results on rainfall agree with the study that rainfall had a positive but non-significant correlation with the positive containers, whereas average temperature exhibited considerable influence on containers (Naqvi *et al.*, 2021; de Souza *et al.*, 2022). The incidence and spatial distribution of *Aedes* larvae were also found to be influenced by temperature (Couret *et al.*, 2014; de Oliveira Lemos *et al.*,

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2021). Surveillance of *Aedes* developmental sites during these particular environmental conditions will be effective.

Different sources of standing water had varying impacts on container and frequency index calculations here and in previous studies. Standing water sources such as evaporative air coolers, discarded house refrigerator trays, bird pots, water drums, tyre shops, nurseries, tube wells, and cemeteries had maximum containers and contribution for the *Aedes* developmental sites (Table 2) with the average house index (HI), container index (CI), and breteau index (BI) of 8.75, 14.0 and 5.6% in different towns of Bhakkar (fig. 3). This agreed with previous findings that water tanks, evaporative air coolers, discarded tyres, drums, tree holes, pots, flower vessels, and discarded cups were the most common *Aedes* developmental sites (Kesetyaningsih *et al*., 2018; Fatima, 2019; Khan 2021; Hafeez *et al*., 2022). A study found the highest number of dengue mosquito larvae and pupa found in plastic boxes, grinding stones, discarded cups, earthen pots, and discarded tyres, with nearly identical trends of HI, CI, and BI, as reported in our study (Saleem *et al*., 2014). Interestingly, few researchers reported high values of HI, CI, and BI because the Bhakkar district largely consists of urban areas with relatively low intensity of dengue virus as compared to other larger, more-populated cities. In our study, dengue fever cases were successfully prevented from local transmission from imported cases as a result of vector prevention efforts, and there were no cases of locally transmitted dengue fever reported because of the elimination of developmental sites, insecticide spraying, and public education campaigns. The management of any disease and its vector, particularly dengue and *Aedes* mosquitoes, depends critically on public awareness and education of *Aedes* developmental sites.

The study on enhanced larval surveillance of *Aedes* mosquitoes conducted in Bhakkar provided positive results which demonstrated the potential of technology in improving dengue control efforts. Anti-dengue mobile application and online integrated PITB-DTS have been successfully implemented in district Bhakkar, demonstrated the significance of technology in advancing public health (fig. 2). The dynamic and complex nature of dengue transmission and control was enhanced through the use of smartphones and mobile apps (Regis *et al*., 2008). These innovative ways provided real-time information on dengue cases, vector presence, and control activities, enabling users to take preventive measures and avoid potential exposure to the disease (Orong *et al*., 2015; Razaq *et al*., 2016). It facilitated coordination and collaboration between various stakeholders involved in dengue control, including public health authorities, healthcare providers, community leaders, and the general public (Pley *et al*., 2021). Further research and implementation of these technologies to combat other vector-borne diseases and public health issues are crucial.

In conclusion, this study provided valuable insights into the factors contributing to the proliferation of *Aedes* developmental sites and highlighted the potential of technology-based solutions for disease surveillance and control. This study has identified optimal temperature conditions (23–28°C) and key months (April, May, August, and September) conducive to the development of the dengue vector. By emphasising the critical need to eliminate standing water sources such as evaporative air coolers and tyre shops, our findings offer enhanced strategies for more effective dengue prevention and control. The effectiveness demonstrated by the online PITB-DTS configured system and Anti-Dengue mobile application in District Bhakkar highlights the significance of harnessing technology to enhance public health outcomes. This is particularly crucial in resource-limited settings, where traditional disease surveillance and control methods may prove insufficient. The findings of this study can serve as a model for other regions with similar characteristics, providing valuable insights into the development of effective and sustainable dengue control programs. Moreover, this study contributes to the broader field of vector-borne disease control by confirming the importance of taking a comprehensive approach to disease prevention and control. Overall, this study contributes significantly to our comprehension of dengue transmission and control, providing valuable insights that can guide the formulation of more efficacious prevention and control strategies in regions affected by the disease.

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