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Research article

COVID-19 lockdown: impact assessment on *Aedes* larval indices, breeding habitats, effects on vector control programme and prevention of dengue outbreaks

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ABSTRACT

Aedes aegypti (Linn.) and *Aedes albopictus* (Skuse) are widespread vector mosquitoes responsible for the transmission of various disease-causing viruses to human including dengue virus (DENV). India is endemic for dengue disease and both of these vector mosquitoes are well established throughout India. Since, *Aedes* mosquitoes breeds in containers, WHO recommends to do a regular immature surveillance and implement appropriate control measures. Owing to the current COVID-19 pandemic, most of the countries have implemented continuous shutdown/lockdown, which affected the routine *Aedes* surveillance and vector control measures. In India, the first nation-wide lockdown was implemented on 24th, March 2020. As of now, Government of India has extended the lockdown till 30th, June 2020. In the present study, two rounds of *Aedes* surveillance was carried out in two localities of Bengaluru City (urban) of Karnataka State, India during the COVID-19 lockdown days and results were compared with pre-lockdown surveillance data to assess the impact of lockdown on *Aedes* larval indices, breeding habitats and dengue vector control programme. The recorded house index (HI) and Breteau index (BI) were 6.6 and 9.3 in K.P. Agrahara and 4.0 and 5.3 in Palace Guttahalli during pre-lockdown survey. The house index (HI) and Breteau index (BI) were found to be increased to 26.6 and 34.6 in K.P. Agrahara and 21.3 and 28.0 in Palace Guttahalli during the COVID-19 lockdown second survey. *Aedes* immature density has drastically increased in both the localities due to temporarily discontinued *Aedes* surveillance, larval control activities like source reduction and anti-larval measures during COVID-19 lockdown. The high indices show that the vector is increasing and this may result in higher dengue virus transmission. The results highly recommend to implement the *Aedes* vector control programme with limited health staffs following the physical distance and other protectives measures to prevent dengue outbreaks.

1. Introduction

The sudden report of viral pneumonia (later named as COVID-19) in the late December 2019 from Wuhan city (China), drew attention of whole public health community and affected the smooth functioning of health system (Huang et al., 2020). Currently, COVID-19 has so far affected 235 countries and territories including tropical and subtropical regions of the world (WHO, 2020a), where dengue is a serious public health problem. Dengue is a complex disease involving four genetically different dengue virus (DENV) serotypes called DENV-1, DENV-2, DENV-3 and DENV-4 (WHO, 2020b) and is a major vector borne disease in tropical and sub-tropical countries. In recent years dengue has grown dramatically and according to World Health Organization 100 to 400 million infections are reported each year keeping half of the world's population at risk (WHO, 2020b). *Aedes aegypti* (Linn.) and *Aedes albopictus* (Skuse) are the day-biting vector mosquitoes responsible for

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dengue virus transmission (Reegan et al., 2018). In managing COVID-19 spread, the affected countries have implemented shutdown or lockdown. In India, nation-wide lockdown is being followed from 24th, March 2020.

Entomological surveillance of immature stages of dengue vectors in endemic pockets is an important indicator to determine the density and to implement appropriate control measures in order to minimize the DENV transmission (Bowman et al., 2014). In India, dengue vector control programme mainly involves larval surveillance, source reduction (emptying water holding container), anti-larval spraying and providing health education/awareness (NVBDCP, 2007). World Health Organization recommends sustained *Aedes* surveillance to detect any increase in the vector density and to implement appropriate control measures (WHO, 2009). Due to the lockdown, dengue vector surveillance had to be paused for more than a month in many of the COVID-19 affected countries including India. Further, source reduction and anti-larval measures were also not been implemented during these lockdown days. In India, dengue shows increasing trend, particularly the state of Karnataka reported second highest number (15929 cases) of dengue cases in the country during 2019 (The Hindu, 2019).

In the present study, *Aedes* immature surveillance was carried out in two localities of Bengaluru City (urban) of Karnataka State, India during the COVID-19 lockdown days and the results were compared with the pre-lockdown surveillance data to assess the impact of lockdown on *Aedes* larval indices, breeding habitats and dengue vector control programme.

2. Material and methods

2.1. Study area

The *Aedes* survey was carried out in two endemic localities namely, K.P.Agrahara (12.9679° N, 77.5625° E) and Palace Guttahalli (12.9974° N, 77.5780° E) in Bengaluru City, India. The entomological survey was carried out in March and April 2020 during COVID-19 lockdown with appropriate protection. House to house survey was conducted in all types of wet containers following the standard procedure prescribed by World Health Organization (WHO, 2009) and physical distance was maintained throughout the study. In this survey, 75 houses were covered in each locality and the larval density was denoted as House index (HI), Container Index (CI), Breteau Index (BI) and pupal density was denoted as Pupal Index (PI).

2.2. Aedes larvae and pupae sampling

Aedes larvae and pupae were randomly collected from various breeding habitats like silver containers, drums, barrels, stone grinders, tyres, small stone cistern, scrap tins, tubs, flower pots, tree holes, etc., by standard dipping method (Ravikumar et al., 2013). The immature stages were collected from both indoor and outdoor; transferred to small jars (200ml capacity) from its natural habitat with the help of plastic pipette and transported to the laboratory within 2 h.

The study was conducted in accordance with the ethical norms and it followed the current vector control programme procedure of the country. The study was not conducted on any human subject and did not have any private data.

2.3. Species identification

The field-collected *Aedes* larvae were fed with artificial pellet made of yeast and dog biscuits (40:60 ratio) and reared to adults. The laboratory conditions were 27° ± 2 °C temperature, 48 ± 2% humidity. The adults were sacrificed with chloroform-dipped cotton balls and identified to species following the standard keys (Barraud, 1931, 1934).

2.4. Data processing and analysis

The following *Aedes* immature indices were calculated as per the formula suggested by World Health Organization (WHO, 2009).

House Index (HI): Number of houses found positive/ Number of houses surveyed*100 (1)

Container Index (CI): Number of containers found positive/ Number of wet containers surveyed*100 (2)

Breteau Index (BI): Number of containers found positive/ Number of houses surveyed*100 (3)

Pupal Index (PI): Number of containers found positive for pupae/ Number of wet containers surveyed*100 (4)

Species dominance index (D) was calculated for each breeding containers and study sites using the following May (1975) index formula.

$$D = Y_{max} / Y_t \quad (5)$$

Where, Y_{max} = The number of immatures of the common *Aedes* species (*Ae. aegypti*) collected in the surveyed localities and containers. Y_t = The total number of immatures of all the *Aedes* species. D = Species dominance index.

Breeding preference rate was calculated and bar diagram was plotted using excel spreadsheet. One-way analysis of variance (ANOVA) was performed to determine the significance of containers on species dominance and the data were separated by Tukey's test of multiple comparisons using SPSS program (version 19.0; SPSS Inc., Chicago, IL, USA).

3. Results

3.1. Adult emergence result

A total of 42 immature *Aedes* were collected during pre-lockdown survey (Feb 2020) (Table 1). Similarly, 89 immature stages were collected during COVID-19 lockdown first survey (March 2020). Correspondingly, 129 immature stages (April 2020) were collected during COVID-19 lockdown second survey (Table 1), which was higher than the pre-lockdown collection. *Ae. aegypti* was recorded to be dominant species in both the localities (80.52%) and the *Ae. albopictus* density was found to be less (16.85%). However, the density of both the vector species were noted to be increased during COVID-19 lockdown period in both the localities (Tables 2 and 3). Further, a non-vector *Ae. vittatus* (2.62%) was also recorded during the COVID-19 lockdown second survey (Table 1). The breeding habitat was recorded to be all types of silver containers, plastic drums, barrels, tubs, flower pots, scrap tins, tyres, tree holes, stone cistern and stone grinders (Figure 1).

Table 1. Adult emergence report of *Aedes* mosquitoes collected from Bengaluru City, India.

Locality (Bengaluru City)	22,23, Feb 2020		28,29, March 2020		25,26, April 2020		
	Ae. aegypti	*Ae. albopictus*	*Ae. aegypti*	*Ae. albopictus*	*Ae. aegypti*	*Ae. albopictus*	*Ae. vittatus*
K.P. Agrahara	18	5	41	3	55	9	0
Palace Guttahalli	16	3	38	7	47	18	7
Total	34	8	79	10	102	27	7

Table 2. Immature Indices of *Aedes* vectors during pre-lockdown survey.

Date of survey	Study site (Bengaluru City)	No. of houses surveyed (found positive)	No. of wet containers surveyed (found positive)	Indices			
				HI[a]	CI[b]	BI[c]	PI[d]
22.02.20 and 23.02.20	K.P. Agrahara	75(5)	121(7)	6.6	5.7	9.3	0.8
	Palace Guttahalli	75(3)	82(4)	4.0	4.8	5.3	0.0

[a] House Index: The % of houses positive for *Aedes* larvae.
[b] Container Index: The % of containers positive for *Aedes* larvae.
[c] Breteau Index: The number of positive containers per 75 houses.
[d] Pupal Index: The % of containers positive for *Aedes* pupae.

Table 3. Immature Indices of *Aedes* vectors during COVID-19 lockdown survey.

Date of survey	Study site (Bengaluru City)	No. of houses surveyed (found positive)	No. of wet containers surveyed (found positive)	Indices			
				HI[a]	CI[b]	BI[c]	PI[d]
28.03.20 and 29.03.20	K.P. Agrahara	75(7)	143(10)	9.3	6.9	13.3	2.1
	Palace Guttahalli	75(9)	102(12)	12.0	11.7	16.0	2.9
25.04.20 and 26.04.20	K.P. Agrahara	75(20)	140(26)	26.6	18.5	34.6	7.1
	Palace Guttahalli	75(16)	118(21)	21.3	17.7	28.0	6.7

[a] House Index: The % of houses positive for *Aedes* larvae.
[b] Container Index: The % of containers positive for *Aedes* larvae.
[c] Breteau Index: The number of positive containers per 75 houses.
[d] Pupal Index: The % of containers positive for *Aedes* pupae.

3.2. Impact on larval indices

The calculated *Aedes* house index (HI) during pre-lockdown survey (Feb 2020) were 6.6 in K.P. Agrahara and 4.0 in Palace Guttahalli. Similarly, the CI, BI, PI were 5.7, 9.3, 0.8 in K.P. Agrahara and 4.8, 5.3, 0.0 in Palace Guttahalli during pre-lockdown survey (Table 2). It was clear from our survey, the *Aedes* larval breeding was found to be increased during the COVID-19 lockdown period. The *Aedes* house index (HI) during COVID-19 lockdown second survey (April 2020) was 26.6 in K.P. Agrahara and 21.3 in Palace Guttahalli (Table 3). Similarly, the CI, BI, PI were increased to 18.5, 34.6, 7.1 in K.P. Agrahara and 17.7, 28.0, 6.7 in Palace Guttahalli during COVID-19 lockdown second survey (Table 3). The historical data of *Aedes* indices during the same study period from 2017 to 2019 are given in Table 4, which shows that the *Aedes* immature indices are below the threshold level (Table 4). The high indices on COVID-19 lockdown days survey show that the dengue vector breeding has increased significantly and reached above threshold level to transmit dengue virus to human.

3.3. Impact on breeding habitats and species dominance

The percent container positive was drastically increased after a month of implementation of lockdown (12.81% has increased). *Ae. albopictus* was recorded for the first time from Palace Guttahalli locality of Bengaluru City. The proportion percent of *Ae. albopictus* was 2.62% in K.P. Agrahara and 3.37% in Palace Guttahalli. Among the several containers surveyed, plastic drums were predominantly positive (29.7%) for *Ae. aegypti* larvae and pupae in both the localities (Figure 2). Similarly,

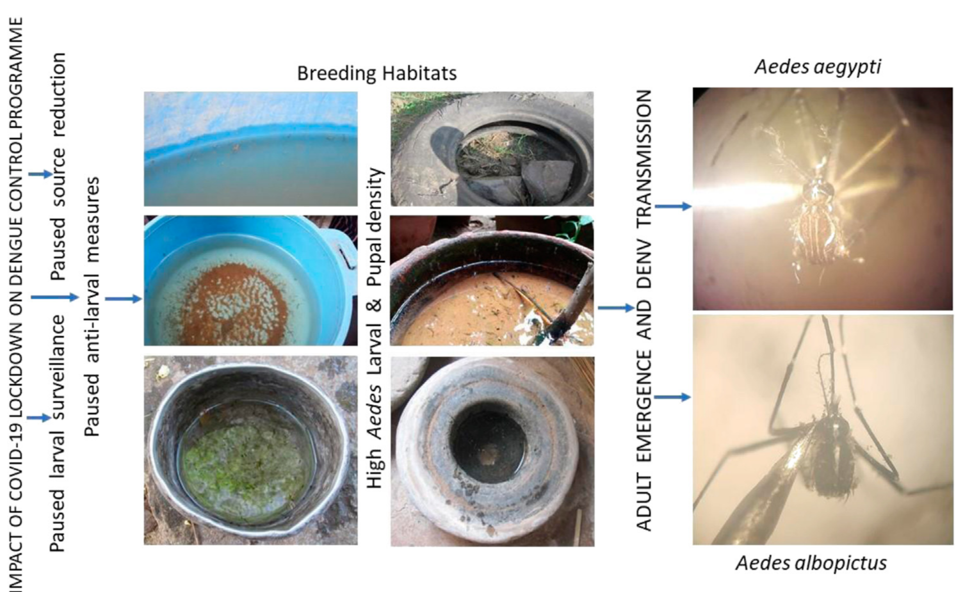

Figure 1. Figure shows the impact of COVID-19 lockdown on *Aedes* vector control programme. *Aedes* positive breeding containers and adult vectors emerged (*Ae. aegypti* & *Ae. albopictus*) from the collected samples are given.

Table 4. Historical data of *Aedes* immature indices from the same study localities (Bengaluru, India).

Date of survey	Study site (Bengaluru City)	No. of houses surveyed (found positive)	No. of wet containers surveyed (found positive)	Indices			
				HI[a]	CI[b]	BI[c]	PI[d]
March 2017	K.P. Agrahara	75(3)	161(3)	4.0	1.8	4.0	0.6
April 2017	Palace Guttahalli	75(2)	107(2)	2.6	1.8	2.6	0.0
March 2018	K.P. Agrahara	75(4)	204(4)	5.3	1.9	5.3	0.4
April 2018	Palace Guttahalli	75(5)	94(5)	6.6	5.3	6.6	1.0
March 2019	K.P. Agrahara	75(6)	179(7)	8.0	3.9	8.0	1.1
April 2019	Palace Guttahalli	75(4)	126(4)	5.0	3.1	5.3	0.7

[a] House Index: The % of houses positive for *Aedes* larvae.
[b] Container Index: The % of containers positive for *Aedes* larvae.
[c] Breteau Index: The number of positive containers per 75 houses.
[d] Pupal Index: The % of containers positive for *Aedes* pupae.

flower pot were predominantly positive (62.5%) for *Ae. albopictus* larvae and pupae in both the localities (Figure 2). Tables 5 and 6 illustrate the species dominance index for each study site and containers, respectively. The present study showed that *Ae. aegypti* was the dominant species (>70%) in both the study sites (Table 5). Dragonfly naiads, a natural predator was observed only in the stone cistern during the present survey.

3.4. Effects on vector control programme

As per the programme guideline, *Aedes* immature surveillance and control activities are implemented once on every six days in the given area or locality by health staffs. All these activities have paused due to COVID-19 lockdown. Further, awareness building on dengue control among public has taken a backseat. These prevailing circumstances have badly affected the vector control programme. Thus, the increase of *Aedes* mosquito population could lead to the loss of previous gains in vector control and result in high level of dengue virus transmission in the coming days.

4. Discussion

Aedes mosquitoes profusely breeds in all types of water storing containers. The paused *Aedes* surveillance and control activities during this COVID-19 lockdown showed its effect on *Aedes* mosquito breeding density. Additionally, the recent sporadic rain in many places of Karnataka including Bengaluru City (The New Indian Express, 2020; The Times of India, 2020; The Hindu, 2020) fuelled the raise of *Aedes* mosquito population and hence the number of containers found positive for larvae/pupae was high in our second survey. The increase of dengue vector density and the involvement of transovarial dengue-virus transmission (Soni and Sharma, 2017) will result in fast DENV transmission and lead to massive increase of dengue cases.

There are growing warnings from the experts on other health threats due to the continuous lockdown (Wang et al., 2020; Xu and Li, 2020). World Health Organization has warned that the number of deaths from malaria in Sub-Saharan Africa this year could double due to not carrying out malaria control activities during COVID-19 lockdown and recommended to implement indoor residual spray (IRS) and few other malaria control activities as per the plan (WHO, 2020c). Similarly, the dengue transmission can be contained with innovative and skilful approach. Few health staffs can be permitted to carryout *Aedes* surveillance, particularly in endemic pockets by following physical distance and other safety measures. Breeding sources can be monitored and insecticides can be sprayed with the help of drones (unpiloted small aircraft). Public should be sensitised on dengue control through television, radio, local newspaper and social media to take dengue preventive measures like reducing the breeding sources in their house premises or backyards and wearing

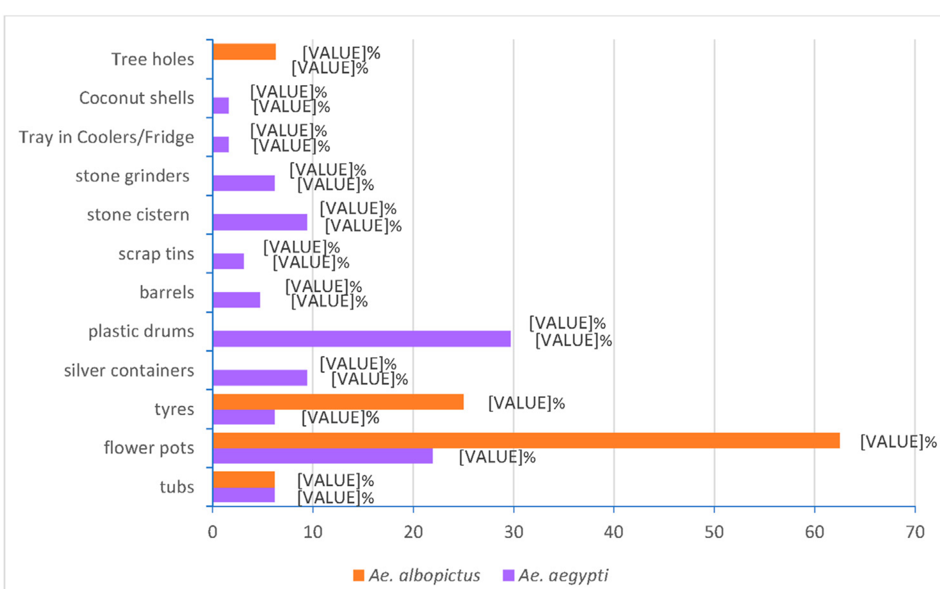

Figure 2. Breeding preference rate (in %) of *Ae. aegypti* and *Ae. albopictus* with respect to different breeding habitats.

Table 5. Species dominance index in the surveyed localities.

Surveyed area	Y_{max}	Y_t	D
K.P. Agrahara	114	131	0.87
Palace Guttahalli	101	136	0.74
Combined	215	267	0.80

Y_{max} = The number of the common *Aedes* species (*Ae. aegypti*) identified in the surveyed localities. Y_t = The total number of all the *Aedes* species. D = Species dominance index.

Table 6. Species dominance index in different breeding containers.

Surveyed Containers	Y_{max}	Y_t	D
Silver containers	30^{bc}	35^{b}	0.85^{b}
Plastic drums	57^{a}	57^{ab}	1.0^{a}
Barrels	19^{c}	19^{cd}	1.0^{a}
Tubs	18^{cd}	26^{c}	0.69^{c}
Flower pots	33^{b}	62^{a}	0.53^{d}
Scrap tins	13^{cde}	13^{def}	1.0^{a}
Tyres	9^{de}	15^{de}	0.60^{cd}
Stone cistern	15^{cd}	15^{de}	1.0^{a}
Stone grinders	16^{cd}	17^{d}	0.94^{ab}
Tree holes	0^{f}	3^{ef}	0.0^{e}
Tray in Coolers/Fridge	3^{e}	3^{ef}	1.0^{a}
Coconut shells	2^{ef}	2^{f}	1.0^{a}

Y_{max} = The number of the common *Aedes* species (*Ae. aegypti*) identified from different breeding containers. Y_t = The total number of all the *Aedes* species. D = Species dominance index. Data were separated by Tukey's test of multiple comparisons, one-way analysis of variance (ANOVA). $p \leq 0.5$, level of significance. Results with same letters in the column are not significantly different.

protective cloths, to use mosquito repellents and to use bed-nets during day time nap.

At present, it is difficult to predict the end of COVID-19 pandemic. However, we are learning lessons to strengthen our health system to manage dengue and other disease control activities during a pandemic situation like COVID-19. Hence it is essential to ensure continued dengue vector surveillance and strengthen vector control interventions during a pandemic with an improved method. Especially, field-level health staffs need to be trained appropriately and provided with personal protective equipment (PPE) to confirm their safety, to make efforts to forecast epidemic and minimize the dengue transmission and any outbreaks in the coming days.

5. Conclusion

This study evaluated the impact of COVID-19 lockdown on the dengue vector control programme. *Aedes* larval surveillance were carried out during COVID-19 lockdown days and the *Aedes* indices were compared with pre-lockdown data. The immature density of *Aedes* mosquito was found to be drastically increased during the lockdown survey due to the paused dengue vector control programme. This study observed high larval and pupal indices after a month of lockdown than pre-lockdown indices in both the surveyed areas in Bengaluru City of India. The *Aedes* HI were observed to be slightly increased during the COVID-19 lockdown first survey (after a week of lockdown). Correspondingly, *Aedes* HI was highly increased during the COVID-19 lockdown second survey (after a month of lockdown). Similarly, the Breteau Index (BI) and Pupal Index (PI) were also highly increased after a month of lockdown. This shows that the DENV transmission may increase in the coming days. Hence, it is highly recommended to adapt innovative methods like finding breeding grounds and larviciding with insecticides using drones. Limited trained staffs can be permitted with all protective measures and awareness should be given to public simultaneously through television, radio, local newspaper and social media to prevent any future dengue outbreaks.

Declarations

Author contribution statement

Appadurai Daniel Reegan: Conceived and designed the experiments; Performed the investigation; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Munusamy Rajiv Gandhi: Conceived and designed the experiments; Performed the investigation; Analyzed and interpreted the data.

Antony Cruz Asharaja: Conceived and designed the experiments; Analyzed and interpreted the data.

Chitra Devi: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data.

Shanmugam Perumal Shanthakumar: Conceived and designed the experiments; Analyzed and interpreted the data; Wrote the paper.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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