

## Eco-bio-social determinants of dengue vector breeding: a multicountry study in urban and periurban Asia

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**Objective** To study dengue vector breeding patterns under a variety of conditions in public and private spaces; to explore the ecological, biological and social (eco-bio-social) factors involved in vector breeding and viral transmission, and to define the main implications for vector control.

**Methods** In each of six Asian cities or periurban areas, a team randomly selected urban clusters for conducting standardized household surveys, neighbourhood background surveys and entomological surveys. They collected information on vector breeding sites, people's knowledge, attitudes and practices surrounding dengue, and the characteristics of the study areas. All premises were inspected; larval indices were used to quantify vector breeding sites, and pupal counts were used to identify productive water container types and as a proxy measure for adult vector abundance.

**Findings** The most productive vector breeding sites were outdoor water containers, particularly if uncovered, beneath shrubbery and unused for at least one week. Peridomestic and intradomestic areas were much more important for pupal production than commercial and public spaces other than schools and religious facilities. A complex but non-significant association was found between water supply and pupal counts, and lack of waste disposal services was associated with higher vector abundance in only one site. Greater knowledge about dengue and its transmission was associated with lower mosquito breeding and production. Vector control measures (mainly larviciding in one site) substantially reduced larval and pupal counts and "pushed" mosquito breeding to alternative containers.

**Conclusion** Vector breeding and the production of adult *Aedes aegypti* are influenced by a complex interplay of factors. Thus, to achieve effective vector management, a public health response beyond routine larviciding or focal spraying is essential.

Une traduction en français de ce résumé figure à la fin de l'article. Al final del artículo se facilita una traducción al español. الترجمة العربية لهذه الخلاصة في نهاية النص الكامل لهذه المقالة.

### Introduction

Dengue, which is the fastest re-emerging arboviral disease in the world, imposes a heavy economic and health burden on countries, families and individual patients.<sup>1,2</sup> In the absence of an effective drug or vaccine, the only strategic options presently available are case management to prevent death and vector control to reduce viral transmission. However, large dengue outbreaks continue to occur every year and the disease is extending to new geographical areas.<sup>3</sup> Integrated vector management can reduce vector densities considerably,<sup>4</sup> but the results of vector control programmes are often far from ideal.<sup>5</sup> Routine interventions against the immature stages of the vector have proved ineffective for a long time,<sup>6</sup> while the results of vertical interventions are often transient.<sup>7</sup> Several user-friendly dengue vector control tools and approaches have become available,<sup>8–12</sup> but questions remain as to their effectiveness, alone or in combination, and their cost-effective delivery by public health services and the private health sector.

Most research on dengue vectors focuses on the biological and behavioural characteristics of the insect,<sup>13,14</sup> the efficacy

and cost of specific interventions,<sup>15</sup> and different delivery strategies for vector management.<sup>16</sup> Although systematic literature reviews and meta-analyses of the results of these "single focus" studies can provide a comprehensive picture of the mix of interventions needed for successful vector control,<sup>5,17</sup> this approach has several limitations. Comparing results is difficult because studies employ different methods and focus on different factors. Furthermore, efficacy trials and effectiveness studies on dengue vector interventions often have questionable outcome measurements. Larval indices (e.g. the house index, the container index, the Breteau index),<sup>18</sup> which are based on the presence or absence of immature forms of the vector in water containers, were useful in eradicating *Aedes aegypti* from the American continent in the late 1940s.<sup>19</sup> However, they are inappropriate for estimating vector densities<sup>20</sup> and of limited use for assessing dengue transmission risk.<sup>21</sup> Work by Focks et al.<sup>22</sup> and subsequent multicentre studies<sup>23–25</sup> have reconfirmed the usefulness of pupal surveys to identify the types of containers that are epidemiologically important and to estimate adult vector abundance. A further limitation is that dengue vector studies usually focus either on house-

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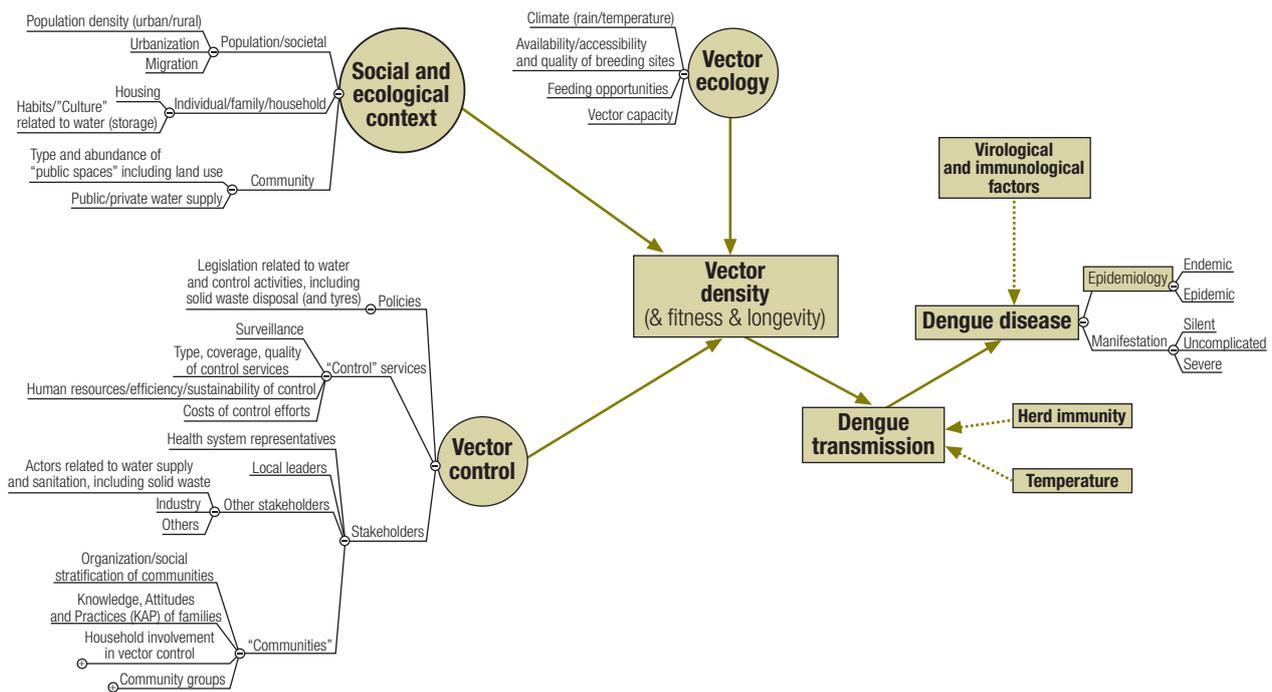
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Fig. 1. Eco-bio-social research on dengue in Asia: a conceptual framework



holds or on defined public spaces<sup>26,27</sup> and therefore lack the analysis of vector production in defined geographical areas (spatial focus). Finally, even though the factors influencing dengue vector densities and ultimately viral transmission are ecological, biological and social (eco-bio-social), as illustrated in Fig. 1, multivariate analyses comprising a combination of these factors have, to our knowledge, not been conducted on a large scale.

For all the reasons cited, we performed a multicountry study focused on geographical areas, including private and commercial premises as well as public spaces and buildings, in six large and middle-sized Asian cities. Its purpose was to answer the following research questions: (i) What is the relative importance of domestic, peridomestic and public spaces for the production of dengue vectors? (ii) What ecological, biological and social factors determine dengue vector densities and contribute to viral transmission? (iii) What are the main implications for vector control services?

## Methods

### Study period and sites

The study was designed in Bangkok in 2006 during a protocol development workshop that was attended by all

principal investigators. It was to be conducted in two phases: Phase 1 was the situational analysis that is described in this paper (field studies were carried out in 2007–2008 and the data were analysed in 2008–2009); phase 2 was designed as an intervention study in six sites, with some intervention and some control neighbourhoods and a cluster randomized study design in the other three sites (phase 2 studies started in 2009). The following six study sites were chosen on the basis of their dengue case load over the preceding three years and their accessibility to the research teams: large cities in India (Chennai), Indonesia (Yogyakarta), Myanmar (Yangon) and the Philippines (Muntinlupa City), and middle-sized provincial towns and their periurban areas in Sri Lanka (Gampaha district) and Thailand (Chachoengsao province).

### Sampling

To obtain a representative sample from each urban or periurban area for conducting household surveys, background surveys and entomological surveys, all study sites followed a joint protocol based on area clusters. A cluster was defined as a neighbourhood of around 100 buildings, including private households, commercial buildings or restau-

rants, with public spaces between them or around them. Public spaces in this study were defined as public streets or pathways, green areas for leisure (parks) or religious worship, abandoned areas and dumping grounds, public buildings like schools or hospitals, religious buildings such as temples, churches or mosques, or private businesses.

To obtain a sample of clusters, we created a map of each study site using Google Earth software (Google Inc., Mountain View, CA, United States of America)<sup>28</sup> and placed a grid on it with 200 squares. We then numbered the squares and used simple random numbers to select 20 in India, Myanmar and Sri Lanka and 12 in Indonesia, the Philippines and Thailand. The sample size in each site was calculated as required for the cluster randomized intervention studies to be conducted during phase 2 of this research project. It was based on a post-intervention cross-sectional comparison of the number of pupae per person in the intervention and control clusters using a two-level hierarchical model with clustering at the neighbourhood level. The sample size reflected a desired power of 80% with the significance level set at 5%. The mean number of pupae per person in control and intervention clusters was assumed to be 3.0 and 0.3, respectively, based on previous studies.<sup>11</sup> For a negative

binomial distribution with a dispersion coefficient of 0.02 and an intra-cluster coefficient of 0.05, 8.9 clusters with 100 households per cluster were needed per study arm, so the number was increased to 10 per study arm (i.e. 20 clusters per study site). We assumed a negative binomial distribution to ensure a large enough sample, even if it was not clearly needed. However, in those sites in the second phase of the study in which a case-study design was to be used to analyse the processes and outcomes of policy interventions, a sample of 12 clusters per site was deemed sufficient. For analysis at the household level, this sample size would yield short 95% confidence intervals (CIs).

### Cluster definition

On the grid we identified the south-eastern corner of each of the selected squares and physically located this point in the city using a global positioning system. We then located the street intersection nearest to this point and made the intersection the bottom left hand corner of a square or rectangle containing the desired sample of approximately 100 buildings. Starting from the intersection, a researcher identified the closest crossing of two streets, one of them representing the vertical line of the square on the map and the other the horizontal line. A researcher then walked roughly 100 metres (m) along the horizontal line or street, turned left and, looking into the “vertical” direction, identified a street parallel to the first vertical street, thereby obtaining a U-shaped form. The researcher then looked for 100 buildings (houses, flats, small business units) within the U-shaped area and, once s/he had found all 100 of them, closed the U and bordered the cluster on the map. A simple map was drawn for orientation. If the square fell over a football ground, large park or any open public space, the next corner of an intersection was used to construct the U. All houses as well as public and private open spaces were included in the cluster analysis.

## Surveys

### Household survey

A demographic and knowledge, attitude and practice survey was carried out together with a larval/pupal (entomological) survey, usually simultaneously

but in some cases with an intervening short time interval. After pilot testing the jointly developed questionnaire in each site, the corrected and agreed on final version was administered to the most senior household member in 6000 households in India, Myanmar and Sri Lanka and in 3391 households in Indonesia, the Philippines and Thailand by trained interviewers from universities or research institutions (4 to 8 per site). The interviewers used the structured questionnaire to obtain information on interviewees’ demographic characteristics, their knowledge about dengue and its prevention, and their perceptions of and attitudes towards dengue risk and current dengue prevention efforts. There were also questions about housing conditions (purpose of building, number of floors, construction material, protection of windows, characteristics of the peridomestic area; water supply and storage, container management, toilets, waste disposal) and other environmental factors (trees or bushes around the house). An observational checklist was used to gather additional information.

### Cluster background survey

A cluster (neighbourhood) background survey instrument was developed, pilot tested in each site and subsequently used to gather detailed information on the selected clusters and adjacent areas. Team members recorded cluster size in square metres (m<sup>2</sup>) using hand-held global positioning system (GPS) devices, as well as human population density (through the household survey), infrastructure (water, electricity, construction materials of houses and roads), distribution of public and residential areas and of sunny and shaded places, and other contextual factors, such as the characteristics and purpose of green areas, religious buildings, market places, schools, hospitals and other public spaces. They also recorded the source of water supply; the existence of sanitation facilities in and around the house; the presence near the house of solid waste that could collect rainwater; other potential *Aedes* breeding/resting places inside and outside the house; and the distance between the house and the nearest source of water (if available). A GPS was used to determine the location of the houses, public spaces and water collection areas.

## Entomological survey

During the wet season larval/pupal surveys were conducted according to standard practice by 2 to 6 university or vector control staff members who were trained in the use of the common pilot tested data collection instrument. In each cluster, intradomestic and peridomestic spaces as well as public (non-household) spaces were inspected. Containers were classified according to type, source of water, capacity, presence of a proper lid, proximity to shrubbery, and presence of larval control measures. Only containers with water were examined. The surveyor determined the presence or absence of *Aedes* larvae in each container and counted all the pupae. In a few sites with large water containers or wells, either the sweeping method or the funnel technique<sup>29</sup> was employed for estimating the number of pupae and a correction factor<sup>30</sup> was sometimes applied to improve the estimated total pupal counts. The sweeping method, in which the larvae are caught with a sweeping net, was used specifically in water drums, whereas the funnel technique, in which a weighted funnel and bottle inverts on entering or exiting the water surface to retain surfacing pupae and larvae, was used in larger containers. A sample of the pupae thus obtained was examined in the laboratory and left to develop into adult mosquitoes, which were then identified by species and sex.

The total number of *A. aegypti* pupae<sup>22</sup> was used as a proxy indicator for adult dengue vectors; the pupae per hectare index (PHI) as an indicator of pupal production per area (as a proxy for adult mosquito production per area,<sup>27</sup> and the pupae per container index (PCI) as an indicator of the infestation levels of different container types. In contrast, larval indices were used to analyse preferred breeding places.

## Survey data analysis

All data were double checked by field supervisors before being entered twice, for quality assurance, into EpiData 2.0 (EpiData Association, Odense, Denmark) by trained personnel. All data files were checked and cleaned by data entry supervisors. The data files of all study sites were merged and analysed jointly for different units of analysis: containers (positivity for pupae/larvae, pupal counts, PCI), and study

Table 1. Characteristics of the study sample<sup>a</sup> in a study of risk factors for dengue vector breeding in six Asian sites, 2006–2009

Characteristic	India	Indonesia	Myanmar	Philippines	Sri Lanka	Thailand	Total
	Chennai	Yogyakarta	Yangon	Muntinlupa	Gampaha district	Chachoengsao province	
No. of clusters studied	20	12	20	12	20	12	86
No. of study households visited	2000	1047	2000	1144	2000	1200	9391
No. of cluster dwellers interviewed	9048	4744	10 488	5672	8241	4168	42 361
Average no. of dwellers per household	4.5	4.5	5.2	5.0	4.1	3.5	4.5
Per cent of household heads unemployed or unable to work	18.7	12.9	36.3 <sup>b</sup>	13.1	20.0	7.2	19.7
Per cent of household heads self-employed (mainly commercial)	17.0	22.2	22.1	7.5	7.1	43.0	18.7
Dwellers aged < 15 years (%)	1732 (19.1)	766 (16.1)	1948 (18.6)	1511 (26.8)	1839 (22.3)	730 (17.5)	8566 (20.1)
Most frequent religion (%)	Hindu (85.3)	Muslim (84.0)	Buddhist (84.3)	Christian (96.9)	Buddhist (53.4)	Buddhist (98.4)	–

<sup>a</sup> Data collected through household survey.

<sup>b</sup> In Myanmar, most respondents were women.

clusters (house index, Breteau index, PHI). Multivariate regression analysis was performed to assess the association between several covariates and the number of pupae per container in the households. Negative binomial regression with a sandwich estimator allowing for clustering at the study cluster level was used. Backward elimination based on significance level was used to select a final model based on a set of potentially important covariates. STATA version 10.1 (StataCorp LP, College Station, TX, USA) was used in the regression analysis.

## Results

### Study sample and cluster characteristics

Table 1 presents the characteristics of the study sample, as revealed by the household survey. As shown, 9391 buildings were visited (93.1% of them being private households, 5.8% small private businesses and 1.1% small restaurants), and 42 361 cluster dwellers were interviewed in total. Of respondents in all sites, 88.9% were older than 25 years and 65.7% were females. The occupational profile of the heads of households and the religious affiliations of the families in the six study sites are

also shown in Table 1. Most families lived in crowded conditions, particularly in Yangon (Myanmar).

Table 2 shows the overall infrastructural, socioeconomic and spatial characteristics of the study clusters, as determined by the cluster background survey.

### Knowledge, practices and vector control measures

#### Knowledge and practices

Respondents' knowledge about dengue and how it is transmitted was generally very good. As shown in Table 3, all respondents had heard of the disease, except for a few in the Philippines, and the majority knew that dengue was a serious but preventable illness transmitted by mosquitoes. Table 3 provides details on people's knowledge about dengue and how to protect themselves from mosquito bites and keep mosquitoes from breeding in and around houses.

#### Vector control and peoples' expectations

In all study sites except Myanmar, the main government action against dengue vectors was reportedly fogging (space spraying) with insecticides, followed in frequency by water treatment (Table 3). Container checking

and health education were much less frequent. Details on visits by inspectors from the vector control office and people's expectations and suggestions for improving government vector control are provided in Table 3.

### Vector abundance and rainfall

In this study, the most common dengue vector was *A. aegypti*. *A. albopictus* was identified in Sri Lanka, but only in very small numbers. There was a positive temporal association between rainfall and the number of laboratory confirmed dengue cases reported in all six study sites, but it was less obvious in Gampaha district, Sri Lanka, whose bimodal rainfall pattern made for a more complex association.

### Vector breeding places

A total of 46 627 containers holding water were identified during the rainy season in all study sites. Their characteristics in each site are shown in Table 4. The factors associated with increased vector breeding and/or pupal production showed a consistent pattern across sites. On multivariate regression analysis, the number of pupae in household containers showed a strong positive association with the presence of shrubbery above the container; the lack of use

Table 2. **Infrastructural and socioeconomic characteristics of clusters<sup>a</sup> included in a study of risk factors for dengue vector breeding in six Asian sites, 2006–2009**

Characteristic	India (n <sup>b</sup> = 20)	Indonesia (n = 12)	Myanmar (n = 20)	Philippines (n = 12)	Sri Lanka (n = 20)	Thailand (n = 12)
<b>Infrastructural</b>						
<b>Per cent of clusters</b>						
with electricity	100	100	100	100	100	100
with paved streets	100	100	75	91	67	100
with areas for leisure activities	35	58	20	53	25	42
with green areas	30	50	75	100	70	67
with marketplaces	47	17	45	42	25	17
with cemeteries	20	42	0	0	10	0
with religious sites	100	100	65	83	20	17
with schools	80	42	53	100	13	17
<b>Average no. of school premises per cluster</b>	1	1	1	2	0.4	0.2
<b>Socioeconomic</b>						
<b>Per cent of clusters</b>						
that were entirely residential	70	50	75	100	30	25
that were mixed residential/commercial	30	50	25	0	70	75
composed of upper middle class	45	42	20	50	10	25
with a mix of middle and lower class	35	58	20	0	80	42
composed of lower class	20	0	60	50	10	33
with good/satisfactory housing conditions	80	100	95	100	95	83
with poor housing conditions	20	0	5	0	5	17
with mostly 1-storey buildings	40	100	70	58	100	8
with mostly 2–5 storey buildings	60	0	30	42	0	92
where most buildings had patios/gardens	32	63	34	36	92	69
where most buildings had trees/high bushes	37	71	26	26	82	55
with piped water <sup>c</sup>	72	32	11	26	43	90
with well	2	72	86	62	56	4
with indoor toilet/latrine <sup>c</sup>	75	89	10	90	43	92
with solid waste collection at least once a week	80	100	100	83	40	100
with visible garbage dumps	35	42	70	33	70	27
with tyre capping facilities	25	100	35	25	15	0
with visible open water pools	30	0	40	50	35	33
<b>Mean distance between buildings (metres)</b>	0	2.4	7.6	3.3	10.2	9.0

<sup>a</sup> Data collected through cluster background survey unless otherwise indicated.

<sup>b</sup> *n* is the number of clusters studied.

<sup>c</sup> Data collected through household survey.

of the container for the previous 7 days or more, and the complete or partial absence of a container cover (Table 5). Across all sites the pupal production was considerably higher in rainwater and outdoor containers compared to tap water and indoor containers, but in the regression analysis the type of water (rain or tap) and the location of the container (indoors or outdoors) were no longer significantly associated with the number of pupae per container.

#### Container treatment

In Thailand, 61.8% of 7802 water containers were treated with *Bacillus thuringiensis israelensis* and Temephos

just before the entomological survey to reduce larval/pupal infestation and pupal counts. The measure was highly effective, as evidenced by the results: 3.7% of treated containers versus 13.8% of untreated containers were positive for pupae and/or larvae ( $P < 0.001$ ). In the other five study sites, water containers had not been treated by government vector control services in the recent past.

#### Productive containers in public versus private spaces

Of the 1982 public spaces in the study clusters, most were public or religious buildings (Indonesia, Myanmar, the

Philippines and Thailand), private businesses (India) or dumping grounds, and other abandoned areas (Sri Lanka) (Table 6).

Roughly half of the water containers in public spaces were indoors; 65.7% were filled with tap water and the remainder, with rainwater. Public spaces had much fewer water-filled containers than private spaces (1982 versus 46 627), but the container index (per cent of water containers with *Aedes* larvae) was similar, or even somewhat higher in public spaces (in India and the Philippines). However, overall pupal production (as an indicator of vector abundance or density) was much higher

Table 3. Knowledge and practices surrounding dengue and dengue prevention<sup>a</sup> in a study of risk factors for dengue vector breeding in six Asian sites, 2006–2009

	Site						Total
	India (n <sup>b</sup> = 20)	Indonesia (n = 12)	Myanmar (n = 20)	Philippines (n = 12)	Sri Lanka (n = 20)	Thailand (n = 12)	
<b>Knowledge</b>							
<b>Per cent of cluster dwellers who</b>							
had heard about dengue	94	98	100	66	99	100	93
considered dengue serious	63	97	80	98	97	94	88
knew that mosquitoes transmit dengue	55	90	79	52	93	90	77
knew that dengue is preventable	55	93	93	97	92	96	88
knew that mosquitoes develop from eggs/ larvae	33	86	89	79	91	79	76
had seen mosquito larvae	71	87	87	68	47	97	76
<b>Practices</b>							
<b>Per cent of cluster dwellers recommending</b>							
doing nothing	6	5	0	0.6	5	1	3
indoor spraying	4	12	13	18	2	19	11
cleaning rubbish	15	27	32	26	17	22	23
covering water containers	9	8	16	22	0.8	16	12
putting chemicals in water	3	10	1	2	0.3	16	5
putting fish in water	0.5	8	5	0.8	2	7	4
using mosquito coils and other chemicals	33	13	30	17	25	15	22
using insect repellents	30	17	4	14	48	4	19
<b>Per cent of clusters where the government</b>							
checked containers	2	18	27	16	13	9	14
added chemicals to water	26	10	0.5	3	31	29	17
sprayed houses	12	10	15	10	4	4	9
educated the people	1	12	9	17	15	13	11
supplied lids for outdoor containers	0.1	2	0	7	0.4	0.9	2
provided fish to put into water	0.4	1	0.4	0.4	0.4	0.4	1
practised fogging with insecticides	44	41	6	29	33	40	32
cut the plants	2	2	2	13	1	0.8	3
<b>Per cent of clusters last visited by a health inspector</b>							
during the past month	22	48	27	10	3	31	24
during the past 2–6 months	6	25	14	8	6	31	15
more than 6 months before	6	10	8	8	6	9	8
never/could not remember	67	16	51	73	85	29	54
<b>Per cent of cluster dwellers recommending that the government</b>							
conduct fogging	31	34	14	17	24	35	26
put chemicals in water	17	6	2	8	19	25	13
conduct residual spraying indoors	9	26	24	13	3	4	13
check water containers	5	14	7	17	6	9	10

<sup>a</sup> Data collected through household survey.

<sup>b</sup> *n* is the number of clusters studied.

in private than in public spaces, although more pupae per container were found in public spaces than in private ones. Fewer types of containers were found in public spaces than in private spaces. In Indonesia, Myanmar and the Philippines, large tanks or ceramic jars in public or religious buildings harboured more than 70% of all the

pupae found in public spaces; in India, most pupae were found in tyres and tins or bottles in private businesses, and in Thailand, in small bowls in religious or public buildings, as well as in tyres in small businesses.

Regression analysis of container data for public spaces identified rain-water and being under shrubbery as the

only statistically significant explanatory variables for pupal production (PCI).

### Interface of ecological, biological and social variables

In private as well as in public spaces, the PHI was significantly higher in clusters with a high population density (74.6; 95% CI: 46.3–102.9) than in

Table 4. Vector breeding places and measures of vector production in buildings<sup>a</sup> in a study of risk factors for dengue vector breeding in six Asian sites, 2006–2009

Parameter	Site					
	India (n <sup>b</sup> = 20)	Indonesia (n = 12)	Myanmar (n = 20)	Philippines (n = 12)	Sri Lanka (n = 20)	Thailand (n = 12)
Container index <sup>c</sup>	5.4	10.7	7.1	12.9	11.1	7.6
House index <sup>d</sup>	19.4	33.1	36.3	16.6	9.1	30.2
Breteau index <sup>e</sup>	28.1	55.3	65.9	24.1	11.3	48.8
Total no. of water containers	10 511	5420	18 510	2319	2063	7804
Per cent of all containers located indoors	83.4	51.5	37.5	42.8	7.1	62.2
Per cent of all containers filled with tap water	95.0	77.6	81.6	86.4	53.6	67.0
Most frequent container types (% of all containers)	Plastic pot (45.4)	Bucket (26.0)	Flower vase (48.7)	Drum/barrel (38.7)	Tin/bottle (27.1)	Ceramic jar (50)
	Metal container (21.5)	Cement tank (25.7)	Cement tank (14.3)	Ceramic jar (32.5)	Bowl (16.2)	Cement tank (13.7)
	Drum/barrel (10.5)	Tin/bottle (6.4)	Drum (12.4)	Coconut (16.17)	Plant axil (11.7)	Bucket (9.9)
Total no. of pupae in all containers	1652	2324	2155	1478	543	453
Most productive container types (% of all pupae)	Cement tank (39.9)	Cement tank (42.8)	Spiritual flower bowl (51.7)	Drum/barrel (49.2)	Bowl (41.6)	Bucket/bowl (38.9)
	Drum/barrel (14.0)	Drum/barrel (13.8)	Cement tank (19.5)	Coconut (18.8)	Tin/bottle (38.6)	Tyres (14.6)
	Grinding stone (13.4)	Flower vase (12.5)	Flower vase (7.2)	Ceramic jar (9.8)	Cement tank (5.7)	Tins/bottles (10.8)

<sup>a</sup> Data collected through entomological survey, wet season only.

<sup>b</sup> n is the number of clusters studied.

<sup>c</sup> Per cent of water containers positive for immature forms of *Aedes*.

<sup>d</sup> Per cent of inspected houses with at least one container positive for immature forms of *Aedes*.

<sup>e</sup> Number of containers positive for immature forms of *Aedes* per 100 inspected houses.

those with a low one (11.0; 95% CI: 7.8–14.1); in clusters with schools (42.7; 95% CI: 25.21–60.3) than in those without schools (14.4; 95% CI: 7.7–21.2); in clusters with religious sites (38.4; 95% CI: 23.8–52.9) than in those without them (11.8; 95% CI: 3.2–20.4); in clusters with houses separated from each other by an average distance of > 4 m (35.4; 95% CI: 19.7–51.1) than in those separated by ≤ 4 m (11.6; 95% CI: 5.4–17.8). Across all study sites, people's knowledge about the dengue vectors was negatively correlated with the PHI (overall correlation coefficient: –0.6).

Other variables associated with a higher PHI but not significantly were middle or lower socioeconomic stratum; poor housing conditions; house with garden; residential area (as opposed to commercial area); presence of cemetery or garbage dump in the neighbourhood; availability of abundant piped water (the only exception being Myanmar); and the absence of vector control interventions.

## Discussion

### Factors determining dengue vector densities

Our spatial analysis of dengue vector abundance and its determining factors in randomly selected geographical units (clusters, neighbourhoods) has provided a more comprehensive understanding of vector ecology, specifically how it can vary and what are its common elements. Scholars and dengue programme managers are already familiar with some of the factors associated with high vector abundance, but they do not fully understand their relative importance and interaction. Key explanatory variables for dengue vector abundance were identified in our multicentre study and analysed in light of their relevance for control services.

The importance of climate (rainy season) for dengue virus transmission was obvious in all study sites: The positive temporal association between dengue incidence and rainfall (“dengue

season”)<sup>31–33</sup> underlines the association between vector density and viral transmission. Dengue morbidity is positively associated with rainfall because the dengue vector proliferates more during the rainy season, when the relative humidity is high, even if water containers in and around households are not exposed to rainfall. Two vector-related groups of factors were important: accessibility of appropriate water sources for breeding and accessibility of human blood for feeding.

### Water sources

Across all study sites, unused and unprotected outdoor containers in shaded areas were the highest contributors to pupal production.<sup>26,34,35</sup> They therefore require special attention by control services. Such containers were particularly accessible to vectors, as shown in our study by an increased PHI, where buildings were widely separated from each other, particularly by shaded areas. This implies that the higher social strata

may be at greater risk of viral transmission, particularly when not protected by air conditioning, fully glazed or screened windows, or locked doors, none of which was found in our study sites. Indoor containers outnumbered outdoor containers in our study (63.3% versus 36.7% of all water containers, respectively), yet they, along with containers that were filled with tap water, were less important sites for breeding and pupal production. This suggests that the vector prefers “natural”, untreated water and reconfirms reports that rainwater-filled containers appeal to *A. aegypti* for breeding, even if they are indoors.<sup>26,27</sup> In the site in Sri Lanka small discarded containers were the main breeding places and the most productive for pupal development because they were seldom removed by infrequent waste disposal services and people did not commonly use larger water containers. The relationship between domestic water supply and pupal production is complex, since both an irregular supply of water and the absence of piped water can lead to greater water storage. Study sites with an irregular supply of piped water and sites without piped water (Myanmar) were had a higher PHI than other sites, although the differences not statistically significant. In general, public spaces contributed to pupal production

Table 5. Container characteristics significantly associated<sup>a</sup> with the number of pupae per container in a study of risk factors for dengue vector breeding in private spaces in six Asian sites, 2006–2009

Container	Incidence rate ratio <sup>b</sup>	95% CI	P-value
not under shrubbery	Reference		
fully or partially under shrubbery	0.51	0.33–0.78	0.002
used during past 7 days	Reference		
not used during past 7 days	6.74	4.37–10.37	< 0.001
fully covered	Reference		
partially covered	3.79	1.53–9.34	0.004
not covered	2.58	1.27–5.21	0.008

CI, confidence interval.

<sup>a</sup> Results of negative binomial regression with clustering at the study cluster level.

<sup>b</sup> Example: the expected pupal count for containers not used in the past 7 days is 6.74 times higher than that for containers used in the past 7 days.

much less than domestic and peridomestic spaces, but schools and religious places provided many breeding opportunities for dengue vectors. Since only a few study clusters had cemeteries, their role in pupal production<sup>36,37</sup> could not be explored.

### Vector feeding opportunities

The higher the population density in our study sites, the more the opportunities for feeding that mosquitoes had and the higher the vector abundance. Thus, control operations should target neighbourhoods endemic for dengue

with high population densities and crowded living conditions.

Two groups of factors were protective against high vector densities: people’s knowledge and awareness of dengue and vector control activities.

The negative association between knowledge about the dengue vector and pupal counts is mediated by behaviour change,<sup>38–41</sup> but the exact mechanisms leading from knowledge to such change and to reduced mosquito densities have yet to be explored. In our study, the use of mosquito coils and other domestic protective methods was frequent in

Table 6. Vector breeding and production in public spaces during the wet season in a study of risk factors for dengue vector breeding in six Asian sites, 2006–2009

Parameter	Site					
	India (n <sup>a</sup> = 20)	Indonesia (n = 12)	Myanmar (n = 20)	Philippines (n = 12)	Sri Lanka (n = 20)	Thailand (n = 12)
Most frequent type of public space (per cent of all public spaces)	Business area (48.9)	Public building (48.1)	Religious building (57.0)	Public building (52.4)	Abandoned/dumping area (69.1)	Religious and public building (55.5)
Container index <sup>b</sup>	11.3	10.9	6.2	43.6	7.3	13.3
Total no. of water containers	354	457	453	133	68	517
Most frequent container type (per cent of all container types)	Ceramic jar (29.1)	Cement tank (32.0)	Flower vase (41.5)	Flower vase (32.3)	Tin/bottle (29.4)	Cement tank (22.4)
	Drum (16.4)	Bucket (25.4)	Cement tank (25.8)	Coconut (12.8)	Tyres (17.6)	Ceramic jar (17.8)
	Bucket (11.0)	Tin/bottle (14.0)	Miscellaneous <sup>c</sup>	Miscellaneous <sup>c</sup>	Miscellaneous <sup>c</sup>	Miscellaneous
Total no. of pupae	264	145	69	412	15	95
Most productive container type (per cent of all pupae)	Tyre (43.6)	Cement tank (70.3)	Cement tank (81.1)	Ceramic jar (52.7)	Well (100)	Bowl (30.5)
	Tin/bottle (21.6)	Tyre (18.6)	Miscellaneous	Coconut (14.6)	–	Tyre (28.4)

<sup>a</sup> n is the number of clusters studied.

<sup>b</sup> Per cent of water containers positive for immature forms of *Aedes*.

<sup>c</sup> Small containers of different shapes.

more prosperous neighbourhoods in India and Sri Lanka, whose dwellers knew more about dengue than those in poorer areas, and other studies have shown similar findings.<sup>42–44</sup>

Vector control to reduce the availability of appropriate breeding sites by chemical or non-chemical interventions could not be analysed anywhere in our study except in Thailand, where recent larviciding had reduced vector breeding (as shown by the low larval indices), nearly eliminated pupal development, and, by inhibiting the development of pupae in the “usual” containers, caused adult vector production to shift to alternative ones (located indoors, filled with tap water and covered).

## Conclusion

The variables that influence vector breeding and the production of adult *Aedes* mosquitoes are many and complex, and the public health response should extend beyond larviciding or focal spraying.<sup>5</sup> A change of paradigm in vector management seems essential. Traditionally, communities have looked to public vector control services to carry out the job, normally through insecticide fogging, but such services tend to apply a one-size-fits-all approach. For integrated vector management to succeed, ways must be found to stimulate communities, as well as their political and religious leaders, to join the battle against dengue. Close interaction between communities and municipal vector control services is critical for the success of dengue vector control.

Several specific messages for vector control programmes can be derived from this study. Productive container types have to be identified and targeted in each setting,<sup>8</sup> with special attention to those that are outdoors, unused, uncovered and in shade. In premises whose local dwellers do not allow control programme inspectors to enter their houses, eliminating or treating unprotected and abandoned outdoor containers can still make a big difference. Covering water containers is effective against vector breeding only if the cover offers full protection. This is only possible, however, with the use of certain modern synthetic water deposits that can be sealed off or of insecticide treated materials.<sup>4,11</sup> Public spaces and commercial areas are important contributors to dengue vector production, although less so than domestic and peridomestic spaces. Schools, places of worship and potentially cemeteries (not included in this study) must also be monitored carefully for *Aedes* breeding. ■

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**Competing interests:** None declared.

## Résumé

### Déterminants écologiques, biologiques et sociaux conditionnant la reproduction des vecteurs de la dengue : étude menée en zone urbaine et périurbaine dans plusieurs pays d’Asie

**Objectif** Étudier les schémas de reproduction des vecteurs de la dengue dans diverses conditions et dans des espaces publics et privés, rechercher les facteurs écologiques, biologiques et sociaux impliqués dans la reproduction de ces vecteurs et la transmission virale et déterminer les principales implications pour la lutte antivectorielle.

**Méthodes** Dans six grandes villes ou zones périurbaines d’Asie, une équipe a sélectionné au hasard des groupes urbains pour mener des enquêtes auprès des ménages standardisées, des enquêtes de voisinage et des enquêtes entomologiques. Les équipes ont recueilli des informations sur les sites de reproduction des vecteurs, les connaissances des habitants, les attitudes et les pratiques à propos de la dengue et les caractéristiques des zones étudiées. Tous les lieux

ont été inspectés ; les équipes ont utilisé les indices larvaires pour évaluer sur le plan quantitatif les sites de reproduction vectorielle et le décompte des pupes pour identifier les types d’objets renfermant de l’eau les plus productifs et pour servir de mesure indirecte de l’abondance des vecteurs adultes.

**Résultats** Les sites de reproduction les plus productifs étaient les objets renfermant de l’eau situés à l’extérieur, en particulier lorsqu’ils étaient dépourvus de couvercle, sous des broussailles et inutilisés pendant au moins une semaine. Les zones péri-domestiques et intradomestiques jouaient un rôle beaucoup plus important dans la production de pupes que les espaces commerciaux et publics autres que les écoles et les édifices religieux. Une association complexe, mais non significative, a été relevée entre

l'approvisionnement en eau et le nombre de pupes décomptées et on a constaté un lien entre le manque de services d'élimination des déchets et une plus grande abondance des vecteurs sur un site seulement. La présence de meilleures connaissances sur la dengue et sa transmission chez les habitants était également associée à une reproduction et à une production plus limitées des vecteurs. Les mesures de lutte antivectorielle (principalement l'application de larvicide sur le site) ont permis de réduire substantiellement les nin de larves et de pupes et a « déplacé » la reproduction des

moustiques vers d'autres récipients.

**Conclusion** Un ensemble interactif complexe de facteurs influe sur la reproduction et la production de moustiques *Aedes aegypti* adultes. Ainsi, pour gérer efficacement les populations vectorielles, une réponse de santé publique globale, allant au-delà des traitements larvicides de routine ou de la pulvérisation focale, est indispensable.

## Resumen

### Determinantes ecobiosociales de la reproducción del vector del dengue: estudio multipaís en zonas urbanas y semiurbanas de Asia

**Objetivo** Estudiar las características de la reproducción del vector del dengue en diversas condiciones en espacios públicos y privados; investigar los aspectos ecológicos, biológicos y sociales (ecobiosociales) que intervienen en la reproducción del vector y la transmisión del virus, y determinar las principales implicaciones para la lucha antivectorial.

**Métodos** En cada una de las seis zonas urbanas o periurbanas de Asia estudiadas, un equipo seleccionó al azar conglomerados urbanos para realizar encuestas de hogares normalizadas, encuestas basales del vecindario y estudios entomológicos. Reunieron información sobre los criaderos de vectores, los conocimientos de la gente al respecto, las actitudes y prácticas relacionadas con el dengue, y las características de las zonas estudiadas. Se inspeccionaron todos los locales, y se usaron los índices larvarios para cuantificar los criaderos del vector, y el número de pupas para distinguir el tipo de contenedores de agua productivos y como indicador sustitutivo de la abundancia de vectores adultos.

**Resultados** Los criaderos de vectores más productivos fueron los contenedores de agua situados al aire libre, sobre todo los que

estaban sin cubrir o debajo de arbustos, y los que no habían sido utilizados por lo menos en una semana. Las áreas peridomésticas e intradomésticas contribuían a la producción de pupas mucho más que los espacios comerciales y públicos, exceptuando las escuelas y los centros religiosos. Se observó una relación compleja aunque no significativa entre el suministro de agua y el número de pupas, y la falta de servicios de evacuación de desechos se asoció a una mayor abundancia de vectores en un solo sitio. La posesión de mayores conocimientos sobre el dengue y su transmisión se asoció a una menor reproducción y producción de mosquitos. Las medidas de lucha antivectorial (principalmente la aplicación de larvicidas en un sitio) lograron reducir sustancialmente el número de larvas y pupas y « empujaron » los criaderos de mosquitos hacia otros contenedores.

**Conclusión** La reproducción del vector y la producción de *Aedes aegypti* adulto dependen de una compleja interacción de factores. En consecuencia, para lograr controlar eficazmente los vectores, es fundamental articular una respuesta de salud pública que no se limite a la aplicación de larvicidas o el rociamiento focalizado.

## ملخص

### المحددات الإيكولوجية والحيوية والاجتماعية لتكاثر نواقل الدنك: دراسة لعدد من البلدان في المدن والمناطق المحيطة بالمدن في آسيا

تستخدم لمدة لا تقل عن أسبوع. أما الأماكن المحيطة بالمنازل ودخلها فكانت لها أهمية أكبر بالنسبة لإنتاج الخادرات من الأماكن التجارية أو الأماكن العامة باستثناء المدارس والمرافق الدينية. ووجدت علاقة معقدة لكن لا يُعتد بها إحصائياً بين إمداد المياه والعدّ الخادري، وارتبطت قصور خدمات التخلص من الفضلات مع المعدلات العالية لكثرة عدد النواقل في موقع واحد فقط. وارتبطت وفرة المعارف حول الدنك وانتقال العدوى به مع انخفاض معدلات تكاثر وإنتاج البعوض. وقد أدت إجراءات مكافحة النواقل (وبدرجة رئيسية باستخدام مبيدات اليرقات في موقع واحد) إلى انخفاض ملموس لليرقات ونتائج العدّ الخادري، مما "دفع" بعملية تكاثر البعوض إلى الانتقال إلى أوعية بديلة أخرى.

**الاستنتاج:** خلصت الدراسة إلى أن تكاثر النواقل وإنتاج النواقل البالغة من الزاعجة المصرية يتأثران بعوامل معقدة ومتشابهة، ولذا كان من الضروري لتحقيق مكافحة فعالة للنواقل، أن يكون هناك استجابة صحية عمومية تتعدى الاستخدام الروتيني لمبيدات اليرقات أو الرش البؤري.

**الغرض:** دراسة أمط تكاثر نواقل الدنك في مختلف الظروف في الأماكن العامة والخاصة، والكشف عن العوامل الإيكولوجية والحيوية والاجتماعية المؤثرة في تكاثر النواقل وانتقال العدوى بفيروس هذا المرض، وتحديد النتائج الرئيسية لمكافحة النواقل.

**الطريقة:** في ست مدن آسيوية، انتقى فريق البحث عشوائياً مناطق من المدن والأماكن المحيطة بها لإجراء مسوحات منزلية معيارية، ومسوحات عن خلفية المناطق المجاورة لها، إضافة إلى إجراء مسوحات الحشرات. وجمع فريق الباحثين معلومات حول مناطق تكاثر النواقل، ومعارف ومواقف وممارسات الناس المتعلقة بالدنك، وما تتميز به المناطق التي خضعت للدراسة. وفحصت جميع الأماكن؛ واستخدمت مناسب اليرقات للتحديد الكمي لأماكن تكاثر النواقل، والتعداد الخادري لتحديد أمط أوعية المياه المنتجة لهذه النواقل، وكطريقة بديلة لقياس وفرة النواقل البالغة.

**الموجودات:** كانت أكثر مواقع تكاثر النواقل هي أوعية المياه الموضوعة خارج المنازل، ولاسيما الأوعية غير المغطاة، أو الموضوعة تحت الأشجار، أو التي لم

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