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In-Kyu Yoon

Anon Srikiatkhachorn

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Characteristics of Mild Dengue Virus Infection in Thai Children

In-Kyu Yoon, Anon Srikiatkhachorn, Laura Hermann, Darunee Buddhari, Thomas W. Scott, Richard G. Jarman, Jared Aldstadt, Ananda Nisalak, Suwich Thammapalo, Piraya Bhoomiboonchoo, Mammen P. Mammen, Sharone Green, Robert V. Gibbons, Timothy P. Endy, and Alan L. Rothman.

Department of Virology, Armed Forces Research Institute of Medical Sciences, Bangkok, Thailand; University of Massachusetts Medical School, Worcester, Massachusetts; Department of Medicine, University of Toronto, Toronto, Ontario, Canada; Department of Entomology, University of California, Davis, Davis, California; Department of Geography, University at Buffalo, Buffalo, New York; Bureau of Epidemiology, Department of Medical Sciences, Ministry of Public Health, Nonthaburi, Thailand; Department of Infectious Diseases, State University of New York, Syracuse, Syracuse, New York; Institute for Immunology and Informatics, University of Rhode Island, Providence, Rhode Island; Fogarty International Center, National Institutes of Health, Bethesda, Maryland

ABSTRACT

A four-year longitudinal cohort and geographic cluster study in rural Thailand was conducted to characterize the clinical spectrum of dengue virus (DENV) infection. Symptomatic DENV infections in the cohort were detected by active school absence-based surveillance that triggered cluster investigations around ill cohort children. Data from 189 cohort children with

symptomatic DENV infection and 126 contact children in the clusters with DENV infection were analyzed. Of infected contacts, only 19% were asymptomatic; 81% were symptomatic, but only 65.9% reported fever. Symptom-based case definitions were unreliable for diagnosis. Symptomatic infections in contacts were milder with lower DENV RNA levels than the cohort. Infections in contacts with fever history were more likely to have detectable DENV RNA than infections without fever history. Mild infections identified by cluster investigations account for a major proportion of all DENV infections. These findings are relevant for disease burden assessments, transmission modeling, and determination of vaccine impact.

INTRODUCTION

Dengue virus (DENV) causes more human morbidity and mortality globally than any other vector-borne viral disease. Each year, an estimated 390 million people are infected with DENV of which 96 million are clinically apparent.[1] Most of the information about the clinical presentation of dengue illness comes from moderate to severe infections that prompt patients to seek medical care, providing the basis for both the 1997 and 2009 World Health Organization (WHO) guidelines for dengue diagnosis and management.[2-9] The sensitivities and specificities of these clinical indicators in identifying dengue illness are not well established. In particular, clinically mild DENV infections have not been as well described; information that is available has been obtained from prospective cohort studies and not from cluster studies which can potentially detect milder illnesses than cohort studies as well as pre-symptomatic infections.[10-15] Symptomatic DENV infections can be difficult to distinguish from other febrile illnesses using clinical parameters, especially with mild illness and early in the course of infection.[8, 16-18] A better understanding of the clinical and virological characteristics across a wider clinical range of DENV infection is important to more accurately assess the burden of DENV infections and potential for virus transmission, and to make informed assessments of patients suspected of having dengue illness. We, therefore, undertook a combined longitudinal cohort and geographic cluster study in rural Thailand to evaluate the full clinical spectrum of DENV infection, including mild infections detected by cluster investigations. Such mild infections have not been previously well studied.[12, 19, 20]

METHODS

Ethics Statement. The study protocol was approved by the Institutional Review Boards of the Thai Ministry of Public Health (MOPH), Walter Reed Army Institute of Research (WRAIR), University of Massachusetts Medical School (UMMS), University of California, Davis (UCD), and San Diego State University (SDSU). Written informed consent was obtained from the parents of study subjects; assent was obtained from subjects older than seven years.

Prospective Longitudinal Cohort and Geographic Cluster Study. The study was conducted at 11 primary schools and 32 associated villages in rural areas of Muang district, Kamphaeng Phet province in north-central Thailand. The methodology is described elsewhere.[12, 19] From 2004 to 2007, a dynamic prospective longitudinal cohort of approximately 2,000 primary school children aged 4--15 years was monitored by active school absence-based surveillance from June to November each year.[19, 21] An acute blood sample was drawn from cohort children who were absent from school and reported a fever in the previous seven days or had a measured temperature ≥ 38ºC. A convalescent blood sample was drawn 14 days later. A questionnaire assessing 12 specific symptoms was administered during the acute and convalescent visits; these symptoms could have been present at any time from seven days prior to the acute illness visit up to the day of the convalescent visit. Acute blood samples were tested by semi-nested reverse transcriptase polymerase chain reaction (RT-PCR) for detection of DENV RNA as previously described.[22, 23] Paired acute and convalescent blood samples were tested by an in-house DENV/Japanese encephalitis virus (JEV) IgM/IgG capture EIA. JEV, which is endemic in rural Thailand, was included to rule out cross-reactivity with DENV.[24]

Cohort children who were DENV PCR-positive from an acute blood sample collected within three days of illness onset served as an index case for a positive geographic cluster

investigation around the child's house (although the "index" case may not have been the first infection in the cluster). Cohort children who were DENV PCR-negative from an acute blood sample served as an index case for a negative (or control) geographic cluster investigation. Ten to 25 children aged six months to 15 years living within 100 meters of the index case were enrolled in each cluster investigation regardless of presence or absence of symptoms. These contact children were evaluated on the day of enrollment (i.e., day 0), and 5, 10, and 15 days later by temperature measurement and administration of a symptom questionnaire similar to the cohort. Symptoms could have been present at any time from seven days before day 0 up to day 15. Blood samples were collected on days 0 and 15 and tested by both DENV PCR and DENV/JEV IgM/IgG EIA. All DENV PCR-positive acute samples from index cases and contact children also underwent quantitative RT-PCR (qRT-PCR) assay to determine serum viral RNA load at the time of blood collection.[25]

Cohort children additionally underwent scheduled phlebotomy prior to the active surveillance season (i.e., May) and at the end of the surveillance season (i.e., December/January). Paired pre/post-surveillance season blood samples were tested by hemagglutination inhibition (HAI) assay for all four DENV serotypes and JEV.[26] Samples with a four-fold rise in HAI titers were re-tested by serotype-specific plaque reduction neutralization tests (PRNT) for DENV and JEV to confirm DENV seroconversion.[27]

Clinical and Serologic Classification. In cohort children, an acute symptomatic DENV infection was considered to have occurred if a febrile illness was associated with a positive DENV IgM EIA and/or PCR in the acute or convalescent blood sample. DENV IgM-positive cases were considered to be primary infection (i.e., first in that child) if the DENV IgM:IgG ratio

was ≥ 1.8, and secondary infection (i.e., second or more in that child) if the ratio was < 1.8.[24] A cohort child was considered to have clinically "inapparent" DENV infection during a surveillance season if paired pre/post-season blood samples showed a four-fold rise in DENV HAI confirmed by PRNT, but no symptomatic DENV infection was identified during that period.[27]

For contacts in the geographic clusters, an acute DENV infection was considered to have occurred if the day 0 or day 15 blood sample was positive by DENV IgM/IgG EIA and/or PCR. These DENV-infected contacts could have either symptomatic infection (i.e., symptoms detected by questionnaire or fever by temperature measurement) or asymptomatic infection (i.e., no detected symptoms or fever).

For both cohort and contact subjects, symptomatic DENV infections that required hospitalization were classified as dengue fever (DF) or dengue hemorrhagic fever (DHF) according to the 1997 WHO case definitions.[2] Symptomatic DENV infections that did not require hospitalization were considered as non-hospitalized symptomatic DENV infection.

Statistical Analyses. SPSS for Windows version 19 and MedCalc version 12.4 software were used for analyses. Symptoms were compared between the various diagnostic groups using the chi-squared test or Fisher's exact test for categorical variables or *t*-test for continuous variables. Variables significant in univariate analyses were subsequently entered in a logistic binary regression model to identify independent associations. A statistical level of $p<0.05$ was considered significant.

RESULTS

Clinical and Virological Features of DENV Infection in the Longitudinal Cohort.

There were 189 symptomatic DENV infections and 346 clinically inapparent DENV infections in the cohort. Twenty additional DENV infections were clinically "unclassified" because they had DENV HAI/PRNT seroconversion between pre-/post-season blood samples but with an acute febrile illness detected during active surveillance that did not have acute/convalescent blood samples collected.[12] General characteristics of dengue EIA-positive cohort children are detailed in Table 1. By study design, all EIA-positive cohort children were symptomatic with a fever history. All four DENV serotypes were detected with a predominance of DENV-1 (46.9%) and DENV-4 (36.7%); secondary infection was much more frequent (93.1%) than primary infection. There were 40 hospitalized DENV infections (31 DF and 9 DHF) accounting for 21.2% of symptomatic DENV infections and 7.2% of all DENV infections (symptomatic plus inapparent/unclassified). By univariate analysis, a measured temperature ≥ 38.0°C, headache, anorexia, muscle/joint pain, rash, drowsiness, abdominal pain, diarrhea and bleeding were significantly more frequent in DENV-infected cohort children with fever history compared with non-DENV infected cohort children with fever history (Table 2). The two symptoms with the highest odds ratios in dengue versus non-dengue febrile illnesses, namely rash and bleeding, occurred infrequently (4.2% and 3.2% of symptomatic DENV infections, respectively). Cough and rhinorrhea were significantly more common in non-dengue illnesses than in dengue illnesses, but still occurred quite frequently with dengue (42.3% with cough and 23.8% with rhinorrhea in dengue). Logistic regression analysis showed that five clinical features (measured temperature ≥ 38.0°C, anorexia, rash, drowsiness, and bleeding) were independently associated with dengue febrile illnesses while cough was associated with non-dengue illnesses (Table 2). No significant differences in symptoms were found between primary and secondary infection (data not shown).

Fever history along with two or more symptoms from the 1997 WHO case definition for suspected DF (i.e., headache, muscle/joint pain, rash and bleeding) had moderate specificity for detecting symptomatic DENV infection (83.7%; 95% CI 82.2--85.2), but low sensitivity (27.5%; 95% CI 21.3--34.5). The positive and negative predictive values for this symptom complex were 11.5% (95% CI 8.7--14.8) and 93.7% (95% CI 92.7--94.7), respectively. These values were similar if symptoms from the 2009 WHO criteria for probable dengue illness were used (i.e., fever with two or more of nausea/vomiting, rash, muscle/joint pain, abdominal pain, drowsiness [used as a surrogate for lethargy] or bleeding). Specificity was 81.2% (95% CI 79.6--82.8), sensitivity was 32.3% (95% CI 25.7--39.4), positive predictive value was 11.7% (95% CI 9.1--14.8), and negative predictive value was 94.0% (95% CI 92.9--95.0).

Some symptom combinations increased the likelihood of a cohort illness being due to DENV infection (e.g., anorexia + bleeding [OR 13.2; 95% CI 3.3--53.3] or rash + absence of cough [OR 8.99; 95% CI 3.1--25.3]). However, these symptom combinations were uncommon (<10% of symptomatic DENV infections). Other symptom combinations including various combinations of headache, anorexia and measured temperature ≥ 38.0°C yielded moderate sensitivities but were common in non-dengue febrile illnesses as well (data not shown).

Drowsiness and bleeding history (e.g., bleeding gums, epistaxis, hematemesis, hematochezia or melena) were the only symptoms individually associated with disease severity. Both symptoms were more common in DHF compared to DF (OR 7.2, 95% CI 1.7--30.2 for drowsiness; OR 29.5, 95% CI 4.9--177.6 for bleeding) and in hospitalized compared to non-hospitalized dengue illnesses (OR 5.0, 95% CI 2.3--10.5; and OR 8.2, 95% CI 1.4--46.3, respectively).

The proportion of symptomatic DENV infections that were DENV PCR-positive varied with the duration between illness onset and acute blood collection. The PCR-positive rate among symptomatic DENV infections was over 80% when the acute blood sample was collected within three days of illness onset and decreased to 64% after the third day of illness. DENV PCR was positive in 143 (86%) of 166 DENV infections when the acute blood sample was DENV IgM-negative, and in four (22%) of 18 symptomatic DENV infections when the acute blood sample was IgM-positive (Chi-squared, $p<0.001$).

Clinical and Virological Features of DENV Infection in Geographic Clusters. In 50 positive cluster investigations, 119 (14.8%) of 805 contact children had laboratory-confirmed acute DENV infection on day 0. An additional 10 contacts who had DENV infection based solely on day 15 PCR-positive results were not included in further analysis because no clinical information was available after day 15. In 53 negative clusters, seven (0.9%) of 794 contacts had acute DENV infection; an additional two contacts were PCR-positive on day 15, but were not included in further analysis.[12] General characteristics of DENV-infected contacts in the geographic clusters are detailed in Table 1. All four DENV serotypes were recovered with the same two serotypes predominating as in the cohort: DENV-1 (51.1%) and DENV-4 (37.2%); secondary infection (81.7%) was more frequent than primary infection but slightly less so than in the cohort. Asymptomatic DENV infections were detected in the clusters: 24 (19.0%) of 126 DENV infections were asymptomatic while 102 (81.0%) were symptomatic. Of the symptomatic infections, 19 (18.6%) did not have fever history and reported no antipyretic use. Seven DENV-infected contacts were hospitalized with DF or DHF (all from positive clusters) accounting for

7.0% of symptomatic infections and 5.6% of total DENV infections (symptomatic plus asymptomatic).

Clinical symptoms in the 83 DENV-infected contacts who reported fever are detailed in Table 3. The comparison group includes DENV-negative contacts who reported fever in both positive and negative clusters; DENV-negative febrile contacts in positive versus negative clusters had no significant differences in symptoms (data not shown). Univariate analysis showed that symptoms of headache, anorexia, nausea/vomiting, muscle/joint pain, rash, abdominal pain and bleeding were more frequent in DENV-infected febrile contacts compared with DENV-negative febrile contacts. Logistic regression analysis showed that headache, muscle/joint pain and rash were independently associated with DENV infection (Table 3). Rash had the strongest association (AOR 7.6; 95% CI 3.0--19.8), but was present in only 15.7% of DENV-infected febrile contacts. Comparing primary with secondary symptomatic DENV infections, nausea and rash were more common in primary infection (AOR 18.2; 95% CI 1.8--186.7, and AOR 13.9; 95% CI 2.0--97.5, respectively) whereas headache was less common in primary infection (AOR 0.2; 95% CI 0.01--0.3). Comparing symptomatic DENV-infected contacts with and without fever history, those without fever had fewer symptoms (not including fever) than those with fever (Table 4). Headache and nausea/vomiting were significantly more common in DENV-infected contacts with fever compared to symptomatic DENV-infected contacts without fever. None of the 19 symptomatic DENV-infected contacts without fever reported any symptoms of nausea/vomiting, muscle/joint pain, drowsiness, bleeding or diarrhea.

Although we identified individual symptoms that distinguished dengue from non-dengue febrile illnesses in contacts, no combination of symptoms was able to distinguish between them. We were also unable to determine any symptom or symptom combination associated with

dengue severity, although our analysis was limited by the small number of contacts with severe disease. Symptoms from the 1997 WHO case definition for suspected DF had high specificity for detecting febrile symptomatic DENV infection (94.7%, CI 92.3--96.6), but low sensitivity (27.7%, CI 18.5--38.6). Positive and negative predictive values were 48.9% (95% CI 34.1--63.9) and 87.8% (95% CI 84.6--90.5), respectively. Values were similar using symptoms from the 2009 WHO criteria for probable dengue illness with high specificity (91.7%, 95% CI 88.7--94.0) and low sensitivity (28.9%, 95% CI 19.5--39.9); positive and negative predictive values were 38.7 (95% CI 26.6--51.9) and 87.6% (95% CI 84.3--90.4), respectively.

For those symptomatic DENV infections among contacts in which day 0 blood samples were collected within three days of illness onset, 18 (58.1%) of 31 blood samples were PCR-positive. DENV PCR was positive in 33 (60.0%) of 55 DENV infections when the day 0 blood sample was DENV IgM-negative, but in only three (15.0%) of 20 DENV infections when the day 0 sample was IgM-positive (Chi-squared, $p<0.001$).

Dengue Virus RNA Levels in Cohort and Contact Children. Comparing symptoms between DENV-infected cohort and contact children, cohort children were more likely to have a measured temperature $\geq 38°C$, headache, muscle/joint pain, rash and drowsiness whereas contacts were more likely to have rhinorrhea and cough (Table 5). Per study design, all 50 index cases in positive clusters were DENV PCR-positive from their acute blood samples within three days of illness onset; three of these developed DHF. Among contacts, 40 PCR-positive DENV infections were detected from day 0 blood samples, of which 18 were collected within three days of illness onset. All 18 of these contacts were symptomatic with fever; none had DHF. Comparing the 47 non-DHF dengue index cases with these 18 DENV PCR-positive contacts, the

mean quantity of DENV RNA was 2.3 X 10^7 (range 9.9 X 10^2--2.14 X 10^8) copies/mL in index cases versus 7.1 X 10^6 (range 1.9 X 10^2--4.3 X 10^7) copies/mL in contacts (*t*-test, p = 0.03). Symptoms were not significantly different between these two groups (data not shown).

DISCUSSION

Our combined cohort and cluster study demonstrates characteristics of DENV infection in children across a wide clinical spectrum of disease including mild infections identified from cluster investigations that have not been previously well described. Our results are applicable for disease burden assessments in endemic areas, virus transmission dynamics, clinical diagnosis, and disease pathophysiology. In our study population, mild dengue illness accounted for a majority of symptomatic DENV infections across all four serotypes.

Symptomatic DENV infections accounted for 81.0% of all DENV infections in contact children, but only 35.3% in cohort children. However, symptomatic infections seen in contacts were milder than in cohort children with fewer symptoms being reported. This may be explained by the difference in surveillance methods between the two groups. Contacts were evaluated for acute infection without regard to their clinical or functional status whereas cohort children were only evaluated for acute infection if they missed school due to a febrile illness. Hospitalized DENV infections accounted for a similar percentage of all DENV infections in both cohort and contact children (7.2% and 5.6%, respectively). Since it is likely that hospitalized infections would be detected no matter what the surveillance method, the fact these percentages were similar suggests that most DENV infections were detected in both the cohort and clusters although the sensitivity for detecting symptoms varied with the surveillance method. These findings demonstrate that mildly symptomatic DENV infections, including afebrile illnesses and

mild febrile illnesses identified only by cluster investigations, constitute a previously uncharacterized spectrum of symptomatic disease. This highlights the importance of the surveillance method in defining illness for purposes such as disease burden assessments, transmission modeling, and determination of vaccine impact.

Certain symptoms and symptom combinations were able to distinguish dengue from non-dengue illnesses. However, these symptoms or symptom combinations occurred infrequently in DENV infections. Cough was able to distinguish non-dengue from dengue illnesses, but occurred too frequently in dengue illness to be used to exclude the diagnosis. Abdominal pain is a "warning sign" from the 2009 WHO dengue guidelines requiring close observation and medical intervention, although these "warning signs" have not been validated with respect to sensitivity, specificity, positive and negative predictive values. We noted no significant difference in abdominal pain between DENV-infected and uninfected symptomatic children in both cohort and contact children. Abdominal pain was, in fact, not infrequent in mild DENV infections. Symptoms from the 1997 and 2009 WHO case definitions for suspected or probable DF had moderate to high specificity and negative predictive value in distinguishing mild dengue illness from non-dengue illnesses, although the sensitivity and positive predictive value were low. In hyperendemic regions of Asia, these symptoms, therefore, may have some limited utility in excluding DENV infection. This finding would depend, however, on the identity and attack rates of other co-circulating pathogens such as influenza, chikungunya, or leptospirosis. Our study did not determine the specific etiologies of non-dengue illnesses.

Considering only those symptomatic DENV infections not resulting in DHF, serum DENV RNA levels were significantly higher in DENV PCR-positive index cases than in PCR-positive contacts from blood samples collected within three days of illness onset. Infections in

contacts were generally milder than in cohort children with fewer reported symptoms. In addition, DENV infections in contacts with fever history were more likely to be PCR-positive than those without fever history. The 19 DENV infections without fever history described in Table 4 were, for the most part, diagnosed by serology alone. These were, nevertheless, still probably acute infections since other symptoms besides fever were present and given that very few infections were comparably detected by serology in the negative clusters. Other studies have reported higher viral loads in DHF than in DF.[28, 29] Our results suggest that serum viral load differs as well with more subtle clinical differences in symptomatic outpatient dengue. Differing viral loads in outpatients has implications for the differential ability of infected humans to transmit virus to mosquitoes. The severity of disease across the entire clinical spectrum should be factored into models that seek to predict patterns in DENV transmission.

Our study was limited to children in a dengue hyperendemic area of rural Thailand. We do not know, therefore, how well these findings apply to other regions with different dengue epidemiology or to adult populations. In addition, our study design did not allow us to conduct detailed analyses of changes in clinical features during the course of dengue illness as has been presented from a Nicaraguan longitudinal cohort,[16] and our use of a symptom questionnaire at intervals separated by several days to two weeks may have led to some recall bias. We also cannot exclude the possibility of enrollment bias in the cluster investigations, for example, if healthy children were less inclined to participate in the study than sick children. Nevertheless, data obtained from DENV-infected children from our cluster investigations provides unique information on mild illness that has largely been unavailable previously.

By combining data from a longitudinal cohort with cluster investigations, we show a wide clinical spectrum of DENV infection in children that includes mild illnesses. While mildly

symptomatic DENV infection is difficult to distinguish from other febrile illnesses, it is quite common and these mild cases should, therefore, be considered when characterizing DENV infection and transmission. In addition, detection of DENV infection through cluster studies may be useful in addressing pathophysiological, immunologic and clinical aspects of disease progression from very early in the course of infection.

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AUTHORS' CURRENT ADDRESSES

Corresponding Author: In-Kyu Yoon, M.D.

Mailing Address: USAMC-AFRIMS, APO, AP, USA 96546

Physical Address: 315/6 Rajvithi Rd., Bangkok, 10400 Thailand

Tel: +66-2-696-2752

Fax: +66-2-644-4760

Email: Yooni@afrims.org

Anon Srikiatkhachorn

University of Massachusetts Medical School, Worcester, Massachusetts, USA 01655

Email: AnonS@afrims.org

Laura Hermann

Department of Virology, Armed Forces Research Institute of Medical Sciences, 315/6 Rajvithi Rd., Bangkok, 10400 Thailand

Email: laura.hermann@afrims.org

Darunee Buddhari

Kamphaeng Phet - AFRIMS Virology Research Unit (KAVRU), 382 Rajdamnern Rd., Muang District, Kamphaeng Phet, 62000 Thailand

Email: daruneet@afrims.org

Thomas W. Scott

Department of Entomology, University of California, Davis, Davis, California, USA, 95616-5270

Email: twscott@ucdavis.edu

Richard G. Jarman

Viral Diseases Branch, Walter Reed Army Institute of Research, Silver Spring, Maryland, USA 20910

Email: richard.g.jarman.mil@mail.mil

Jared Aldstadt

Department of Geography, University at Buffalo, Buffalo, New York, USA 14261-0055

Email: geojared@buffalo.edu

Ananda Nisalak

Department of Virology, Armed Forces Research Institute of Medical Sciences, 315/6 Rajvithi Rd., Bangkok, 10400 Thailand

Email: AnandaN@afrims.org

Suwich Thammapalo

Bureau of Epidemiology, Department of Medical Sciences, Ministry of Public Health, Nonthaburi, 11000 Thailand

Email: sthammapalo@yahoo.com

Piraya Bhoomiboonchoo

Department of Virology, Armed Forces Research Institute of Medical Sciences, 315/6 Rajvithi Rd., Bangkok, 10400 Thailand

Email: PirayaB@afrims.org

Mammen P. Mammen

Vical Incorporated, 10390 Pacific Center Court, San Diego, California, USA 92121-4340

Email: Mammen.Mammen@vical.com

Sharone Green

University of Massachusetts Medical School, Worcester, Massachusetts, USA 01655

Email: Sharone.Green@umassmed.edu

Robert V. Gibbons

Department of Virology, Armed Forces Research Institute of Medical Sciences, 315/6 Rajvithi Rd., Bangkok, 10400 Thailand

Email: Robert.Gibbons@afrims.org

Timothy P. Endy

Department of Infectious Diseases, State University of New York, Syracuse, Syracuse, New York, USA 13210

Email: EndyT@upstate.edu

Alan L. Rothman

Institute for Immunology and Informatics, University of Rhode Island, Providence, Rhode Island, USA 02881

Email: alan_rothman@mail.uri.edu

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Description	**DENV-infected cohort children; N = 189**	**DENV-infected contacts; N = 126***
Median age (range)	9 (5-13)	9 (0-15)
Sex		
Male	92 (48.7)	68 (54.0)
Female	97 (51.3)	58 (46.0)
Fever history	189 (100)	83 (65.9)
Serological category		
Primary	13 (6.9)	23 (18.3)**
Secondary	176 (93.1)	103 (81.7)**
Dengue PCR positive	147 (77.8)	43 (34.1)
Serotype		
DENV-1	69 (46.9)	22 (51.1)
DENV-2	20 (13.6)	3 (7.0)
DENV-3	4 (2.7)	2 (4.7)
DENV-4	54 (36.7)	16 (37.2)

Table 1: Characteristics of DENV-infected cohort and contact children; *includes 119 contacts from positive clusters and 7 from negative clusters; **variable timing of blood collection after infection in contacts makes this categorization less certain.

Symptom	DENV-infected with fever history (%); N = 189	Non-DENV infected with fever history (%); N = 2449	Odds ratio (95% CI)	Adjusted odds ratio (95% CI)	p-value for adjusted odds ratio
Temp ≥ 38°C	125 (66.1)	1374 (56.1)	1.5 (1.1-2.1)	1.6 (1.1-2.2)	0.007
Headache	160 (84.7)	1872 (76.4)	1.7 (1.1-2.6)	-	NS
Anorexia	61 (32.3)	473 (19.3)	2.0 (1.4-2.8)	1.7 (1.2-2.5)	0.002
Nausea/Vomiting	61 (32.3)	673 (27.5)	1.3 (0.9-1.7)	-	NS
Muscle/Joint pain	49 (25.9)	446 (18.2)	1.6 (1.1-2.2)	-	NS
Rash	8 (4.2)	19 (0.8)	5.7 (2.4-13.1)	4.3 (1.7-10.4)	0.002
Rhinorrhea	58 (30.7)	1163 (47.5)	0.5 (0.4-0.7)	-	NS
Cough	80 (42.3)	1637 (66.8)	0.4 (0.3-0.5)	0.4 (0.3-0.5)	<0.001
Drowsiness	45 (23.8)	278 (11.4)	2.4 (1.7-3.5)	2.0 (1.4-2.9)	<0.001
Abdominal pain	46 (24.3)	377 (15.4)	1.8 (1.3-2.5)	-	NS
Diarrhea	14 (7.4)	92 (3.8)	2.1 (1.1-3.7)	-	NS
Bleeding (any site)	6 (3.2)	15 (0.6)	5.3 (2.0-13.9)	4.5 (1.6-12.4)	0.004

Table 2: Comparison of symptoms in DENV-infected and non-infected cohort children with fever history by univariate analysis (odds ratio) and binary logistic regression (adjusted odds ratio); NS = not significant

Symptom	DENV-infected with fever history (%); N = 83	Non-DENV infected with fever history (%); N = 455	Odds ratio (95% CI)	Adjusted odds ratio (95% CI)	p-value for adjusted odds ratio
Temp ≥ 38°C	29 (34.9)	165 (36.3)	0.9 (0.6-1.5)	-	NS
Headache	50 (60.2)	159 (34.9)	2.9 (1.8-4.6)	2.1 (1.3-3.5)	0.004
Anorexia	20 (24.1)	52 (11.4)	2.5 (1.4-4.4)	-	NS
Nausea/Vomiting	26 (31.3)	65 (14.3)	2.7 (1.6-4.7)	-	NS
Muscle/Joint pain	13 (15.7)	18 (4.0)	4.5 (2.1-9.6)	2.8 (1.2-6.5)	0.01
Rash	13 (15.7)	8 (1.8)	10.4 (4.2-25.9)	7.6 (3.0-19.8)	<0.001
Rhinorrhea	40 (48.2)	239 (52.5)	0.8 (0.5-1.3)	-	NS
Cough	44 (53.0)	232 (51.0)	1.1 (0.7-1.7)	-	NS
Drowsiness	9 (10.8)	25 (5.5)	2.1 (0.9-4.7)	-	NS
Abdominal pain	17 (20.5)	42 (9.2)	2.5 (1.4-4.7)	-	NS
Diarrhea	5 (6.0)	25 (5.5)	1.1 (0.4-3.0)	-	NS
Bleeding (any site)	6 (7.2)	10 (2.2)	3.5 (1.2-9.8)	-	NS

Table 3: Comparison of symptoms in DENV-infected and non-infected contacts with fever history by univariate analysis (odds ratio) and binary logistic regression (adjusted odds ratio); NS = not significant.

Symptom	Symptomatic DENV-infected with fever history (%); N = 83*	Symptomatic DENV-infected without fever history (%); N = 19*	p-value
Temp ≥ 38°C	29 (34.9)	0 (0.0)	0.001
Headache	50 (60.2)	3 (15.8)	<0.001
Anorexia	20 (24.1)	2 (10.5)	NS
Nausea/Vomiting	26 (31.3)	0 (0.0)	0.003
Muscle/Joint pain	13 (15.7)	0 (0.0)	NS
Rash	13 (15.7)	1 (5.3)	NS
Rhinorrhea	40 (48.2)	9 (47.4)	NS
Cough	44 (53.0)	8 (42.1)	NS
Drowsiness	9 (10.8)	0 (0.0)	NS
Abdominal pain	17 (20.5)	4 (21.1)	NS
Diarrhea	5 (6.0)	0 (0.0)	NS
Bleeding (any site)	6 (7.2)	0 (0.0)	NS

Table 4: Comparison of symptoms between DENV-infected contacts with and without fever history; *38/83 with fever history and 2/19 without fever history were DENV PCR-positive (Fisher's exact, p=0.004); NS = not significant

Symptom	Symptomatic DENV-infected cohort children (%); N = 189	Symptomatic DENV-infected contacts (%); N = 102	p-value
Fever history	189 (100.0)	83 (81.4)	0.0001
Temp ≥ 38°C	125 (66.1)	29 (28.4)	0.0001
Headache	160 (84.7)	53 (52.0)	0.0001
Anorexia	61 (32.3)	22 (21.6)	NS
Nausea/Vomiting	61 (32.3)	26 (25.5)	NS
Muscle/Joint pain	49 (25.9)	13 (12.7)	0.01
Rash	8 (4.2)	14 (13.7)	0.005
Rhinorrhea	58 (30.7)	49 (48.0)	0.005
Cough	80 (42.3)	52 (51.0)	0.0002
Drowsiness	45 (23.8)	9 (8.8)	0.002
Abdominal pain	46 (24.3)	21 (20.6)	NS
Diarrhea	14 (7.4)	5 (4.9)	NS
Bleeding (any site)	6 (3.2)	6 (5.9)	NS

Table 5: Comparison of symptoms between symptomatic DENV-infected cohort and contact children; NS = not significant