Occurrence and Correlates of Symptom Persistence Following Acute Dengue Fever in Peru

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Abstract. Dengue virus (DENV) infection causes an acute febrile illness generally considered to result in either complete recovery or death. Some reviews describe persistent symptoms after the febrile phase, although empirical data supporting this phenomenon is scarce. We evaluated symptom persistence in acute febrile DENV-infected and DENV-negative (controls) individuals from Peru. Self-reported solicited symptoms were evaluated at an acute and a follow-up visit, occurring 10–60 days after symptom onset. Rate of persistence of at least one symptom was 7.7% and 10.5% for DENV infected and control subjects, respectively ($P < 0.01$). The DENV-infected individuals had lower rates of persistent respiratory symptoms, gastrointestinal symptoms, headache, and fatigue, but higher rates of persistent rash compared with controls. Older age and female gender were positively associated with symptom persistence. As dengue cases continue to increase annually, even a relatively low frequency of persistent symptoms may represent a considerable worldwide morbidity burden.

INTRODUCTION

The mosquito-borne dengue virus (DENV) impacts human health more than any other arbovirus. Over two-fifths of the world's population is at risk of infection from DENV, and 96 million people experience symptomatic disease annually.^{1,2} Symptomatic DENV infections may result in acute, life-threatening manifestations such as dengue hemorrhagic fever (DHF) or dengue shock syndrome (DSS). The most common presentation, dengue fever, results in a painful, yet non-lethal, febrile illness characterized by headache, malaise, and extreme body aches.

Following the onset of symptoms, the acute febrile phase of dengue fever usually lasts between 3 and 7 days before deferescence, with only a fraction going on to manifest with severe disease around this time.³ Even in those without severe disease, a symptomatic post-febrile phase has been noted by physicians familiar with DENV infection, but rarely described.⁴ A few case studies have detailed persistence of very specific endpoints such as decreased visual acuity, cardiomyopathy, neurological deficits, and elevated liver transaminases weeks to months after DENV infection, and a handful of others have described more general signs and symptoms.^{5–15} However, these prior studies were small in size and only two used a control group of DENV uninfected subjects to better ascribe the source of lingering symptoms to DENV infection.^{13,15}

We collected clinical data from febrile individuals in Peru with or without DENV infection to measure the frequency of persistence of symptoms within 2 months after acute symptom onset and the association of persistence with prior DENV exposure history, DENV serotype, and demographic factors.

MATERIALS AND METHODS

This study used Naval Medical Research Center Detachment (NMRC) protocol 2000.0006, Surveillance and Etiology of Acute Febrile Illnesses in Peru, and was approved by the Naval Medical Research Center (Silver Spring, MD) Insti-

tutional Review Board in compliance with all U.S. Federal regulations governing the protection of human subjects. The study protocol was also reviewed by the Peruvian Ministry of Health. Written consent was obtained from all participating adults and a parent or legal guardian of participants 5–17 years of age. Assent was also obtained from participants between 8 and 17 years of age.

Study site and enrollment. The current study protocol has been used previously to characterize the epidemiology and clinical aspects of dengue and other arboviral diseases in febrile subjects in Peru^{16,17}; this prospective study occurred at 19 healthcare facilities located in six cities throughout Peru (Figure 1): Iquitos, La Merced, Yurimaguas, Puerto Maldonado, Piura, and Tumbes. Participants were enrolled between January 1, 2005 and December 6, 2010. At each site, participants were enrolled with the same inclusion criteria. These were 1) fever $\geq 38.0^{\circ}\text{C}$ at the acute visit or report of recent fever, 2) no obvious source of infection such as cellulitis, dental abscess, or urinary tract infection, and 3) ≥ 5 years of age. Additionally, a follow-up visit occurring between 10 and 60 days after the onset of symptoms was required for this study.

Data and sample collection. Every site used a standardized questionnaire, which collected the demographic and solicited symptom information assessed in this study. If a subject was too young to provide this information, then it was obtained from a parent or guardian. In addition, all participating healthcare personnel received standardized training in data collection. A serum sample was collected at the acute visit and subjects were instructed to return to the healthcare facility for a follow-up visit, usually 1 week or more after the acute visit. At this follow-up visit, symptom information and a serum specimen were once again obtained. Each subject had only one follow-up visit. Samples were stored at -70°C until they were transferred to the Naval Medical Research Unit No. 6 (NAMRU-6) on dry ice.

Laboratory assays. At NAMRU-6, acute samples were initially evaluated for DENV using either reverse transcription-polymerase chain reaction (RT-PCR) or culture on Vero and C6/36 cells with standard indirect immunofluorescence assay, as previously described.^{17,18} Specimens found to have DENV by culture using screening polyclonal antisera were grouped into serotypes (DENV-1, DENV-2, DENV-3, or DENV-4) using serotype-specific monoclonal antibodies. The RT-PCR

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FIGURE 1. The six recruitment sites in Peru are denoted with black circles. The capital of Peru, Lima, is denoted with a star and was not a recruitment site.

also allowed for identification of the infecting DENV serotype.¹⁸ Enzyme-linked immunosorbent assay (ELISA) for immunoglobulin M (IgM) against DENV was performed on both the acute and follow-up samples, as previously described.¹⁷ Primary versus secondary DENV infection was determined using an IgM/IgG ratio on the acute sample, similar to an approach used by others.¹⁹ Culture, RT-PCR, and ELISA were also used to identify non-DENV endemic arboviruses: Mayaro virus (MAYV), Venezuelan equine encephalitis virus (VEEV), Oropouche virus (OROV), Guaroa virus (GUAV), and the Group C orthobunyaviruses (GRCV).¹⁷ If positive results were available at the follow-up visit, they were conveyed to the subject. Persistent symptoms at the follow-up visit were not typically evaluated for a new process.

Definition of terms. The presence or absence of 17 symptoms, collated into six symptom categories, was assessed in this study. The six symptom categories and their corresponding symptoms were “fatigue” (malaise and/or asthenia), “rash” (truncal rash, extremity rash, and/or facial rash either reported by the subject or noted by healthcare personnel),

“respiratory” (cough, rhinorrhea, and/or expectoration), “body pain” (myalgia, bone pain, and/or joint pain), “gastro-intestinal” (decreased appetite, abdominal pain, diarrhea, nausea, and/or vomiting), and “headache” (headache). A symptom category was considered positive if one or more of the symptoms in that category was present. Finally, an “at least one symptom” group was defined based on the presence of any of the 17 symptoms.

The primary outcome of this study was symptom “persistence.” “Persistence” was defined as the repeat detection of at least one symptom within a given symptom category at both the acute and follow-up visits. Two main subject groups were compared throughout the study, termed “DENV infected” and “febrile controls” (Figure 2). A subject was considered “DENV infected” if a) culture or RT-PCR of the acute sample identified DENV or b) DENV IgM ELISA at either the acute or follow-up visit was positive (and all assays were negative for other endemic arboviruses: MAYV, VEEV, OROV, GUAV, and GRCV). A subject was determined to be a “febrile control” if a) culture or RT-PCR of the acute sample

FIGURE 2. The number of subjects enrolled, excluded, and included in the febrile control and dengue virus (DENV) infected groups based on laboratory diagnosis. *Cross-reactivity was defined as immunoglobulin M (IgM) serologic positivity for both DENV and a non-DENV arbovirus. †Removed from analysis.

identified a non-DENV arbovirus or b) all assays were negative for DENV. Subjects with ELISA IgM results positive for both DENV and another endemic arbovirus were excluded from the analysis.

Statistical analyses. The rate of persistence was estimated and compared between DENV infected versus febrile controls and by time interval from symptom onset to the follow-up visit, defined as 10–20 days, 21–30 days, or 31–60 days, using a χ^2 test. Odds ratios (ORs) (95% confidence interval [CI]) were estimated using logistic regression to estimate the association of gender (male versus female), age (5–10 years, 11–20 years, 21–30 years, 31–40 years, 41–50 years, and ≥ 51 years) and DENV-related factors (serotype, primary versus secondary infection) with symptom persistence. Separate regression models were fit for the DENV infected and febrile control groups. Models fit among febrile control individuals were mutually adjusted for gender, age, city of residence (Iquitos, La Merced, Yurimaguas, Puerto Maldonado, Piura, Tumbes), year of diagnosis (2005, 2006, 2007, 2008, 2009, 2010), time from symptom onset to the follow-up visit, and time between acute and follow-up visits. Models fit among DENV-infected individuals were further adjusted for DENV serotype and prior DENV infection status. The difference in the ORs estimating the association of risk factors with persistence of at least one symptom by group (DENV infected versus febrile control) was assessed through inclusion of a product term in the multivariate logistic regression model. A *P* value for trends in the OR was estimated through inclusion in the regression model of the collapsed ordinal variable. Differences in the associations of risk factors with at least one persistent symptom by time interval from symptom onset to the follow-up visit were assessed through inclusion of a product term in the multivariate regression model. A *P* value of < 0.05 was considered statistically significant for

all comparisons. All analyses were performed using STATA 12.1 (STATA Corp.; College Station, TX).

RESULTS

Study sample. Nine thousand three hundred and twenty-seven individuals from Peru were enrolled in the study (Figure 2). Four subjects had incomplete age or gender information and were excluded from analysis. Subjects not tested with culture or RT-PCR ($N = 30$), with negative culture/RT-PCR results and no serology testing ($N = 122$), or with ELISA IgM results suggesting infection with DENV and another arbovirus ($N = 104$) were also excluded. In total, 9,067 subjects were included in the analysis, 3,659 DENV infected and 5,408 febrile control subjects.

Demographic information about the participants and DENV-specific information (serotype, primary versus secondary infection) is provided in Table 1. Overall, the mean age of the study sample was 28 years (SD: 14 years), 51.9% were female, 63.1% were recruited from Iquitos, and 59.2% had secondary DENV infection. Among those with a known DENV serotype, 51.1% had DENV-3. The mean time from symptom onset to the follow-up visit was 22 days (SD: 8 days).

Rate of symptom persistence in DENV infected and febrile control participants. The overall rate of persistence of at least one symptom was 9.3%. Individuals who were DENV infected were less likely to have persistence of at least one symptom compared with febrile controls (7.7% versus 10.5%; $P < 0.01$; Table 2). Similarly, individuals with DENV infection had lower rates of persistence of respiratory symptoms (0.6% versus 2.0%; $P < 0.01$), gastrointestinal symptoms (1.3% versus 1.9%; $P = 0.02$), headache (5.6% versus 6.8%; $P = 0.02$), and fatigue (2.7% versus 3.8%; $P < 0.01$) compared with febrile controls. Those with DENV had a higher rate of

TABLE 1
Characteristics of the study sample

	Total (N = 9,067)	DENV infected n (%) (N = 3,659)	Febrile controls n (%) (N = 5,408)
Gender			
Male	4,360 (48.1)	1,740 (47.5)	2,620 (48.5)
Female	4,707 (51.9)	1,919 (52.5)	2,788 (51.5)
Age			
5–10	639 (7.1)	183 (5.0)	456 (8.4)
11–20	2,629 (29.0)	1,076 (29.4)	1,553 (28.7)
21–30	2,488 (27.4)	1,046 (28.6)	1,442 (26.7)
31–40	1,536 (16.9)	640 (17.5)	896 (16.6)
41–50	1,024 (11.3)	440 (12.0)	584 (10.8)
≥ 51	751 (8.3)	274 (7.5)	477 (8.8)
Year of diagnosis			
2005	1,666 (18.4)	526 (14.4)	1,140 (21.1)
2006	1,737 (19.2)	542 (14.8)	1,195 (22.1)
2007	1,417 (15.6)	448 (12.3)	969 (17.8)
2008	1,550 (17.1)	897 (24.5)	653 (12.1)
2009	1,245 (13.7)	576 (15.7)	669 (12.4)
2010	1,452 (16.0)	670 (18.3)	782 (14.5)
City			
Iquitos	5,724 (63.1)	2,462 (67.3)	3,262 (60.3)
La Merced	281 (3.1)	116 (3.2)	165 (3.1)
Yurimaguas	1,013 (11.2)	452 (12.4)	561 (10.4)
Puerto Maldonado	773 (8.5)	235 (6.4)	538 (9.8)
Piura	236 (2.6)	65 (1.8)	171 (3.2)
Tumbes	1,040 (11.5)	329 (8.9)	711 (13.2)
DENV serotype			
DENV-1		219 (10.7)	
DENV-2		34 (1.6)	
DENV-3		1,048 (51.1)	
DENV-4		751 (36.6)	
Prior DENV infection status			
Primary		1,449 (39.6)	
Secondary		2,165 (59.2)	
Indeterminate		45 (1.2)	
Time from symptom onset to follow-up visit (days)			
10–20	4,959 (54.7)	2,026 (55.3)	2,933 (54.2)
21–30	2,887 (31.8)	1,144 (31.3)	1,743 (32.3)
31–60	1,221 (13.5)	489 (13.4)	732 (13.5)
Time from acute visit to follow-up visit (days)			
≤ 7 days	89 (0.9)	30 (0.8)	59 (1.2)
8–14 days	2,755 (30.4)	1,134 (30.9)	1,621 (29.9)
15–30 days	5,421 (59.8)	2,180 (59.7)	3,241 (59.9)
31–60 days	802 (8.9)	315 (8.6)	487 (9.0)

DENV = dengue virus.

persistent rash (0.6% versus 0.1%; $P < 0.01$) compared with febrile controls. The frequency of body pain (2.4%) was the same for the two groups.

The lower frequency of symptom persistence among DENV-infected subjects as compared with febrile controls was consistent across each time interval between symptom onset to the follow-up visit (Figure 3). The rate of symptom persistence declined significantly with an increasing time

interval between symptom onset and follow-up for both DENV infected and febrile controls (p_{trend} for both < 0.05). Among those who were DENV infected, there was a significant decline in persistence of headache ($p_{\text{trend}} < 0.05$) over time (Figure 4). In addition, marginal declines in persistence with increasing time from symptom onset to follow-up were also observed for body pain and fatigue ($p_{\text{trend}} = 0.05$ for both). Among febrile controls, significant declines in

TABLE 2
Frequency and distribution of symptom persistence in subjects with DENV infection and febrile controls

Variable	Total (N = 9,067) n (%)	DENV infected (N = 3,659) n (%)	Febrile control (N = 5,408) n (%)	P value
At least one symptom	847 (9.3)	281 (7.7)	566 (10.5)	< 0.01
Respiratory	129 (1.4)	21 (0.6)	108 (2.0)	< 0.01
Gastrointestinal	149 (1.6)	46 (1.3)	103 (1.9)	0.02
Rash	30 (0.3)	23 (0.6)	7 (0.1)	< 0.01
Body Pain	219 (2.4)	88 (2.4)	131 (2.4)	0.96
Headache	572 (6.3)	204 (5.6)	368 (6.8)	0.02
Fatigue	304 (3.4)	98 (2.7)	206 (3.8)	< 0.01

P values generated using a χ^2 test.

FIGURE 3. The presence of at least one symptom at the acute visit (left axis) and persistent rates at three follow-up time periods (right axis).

persistence were observed with an increasing time interval from symptom onset to follow-up for body pain ($p_{\text{trend}} < 0.05$), headache ($p_{\text{trend}} < 0.05$), and fatigue ($p_{\text{trend}} < 0.05$).

Association of demographic and DENV-related factors with any symptom persistence The association of gender and age with symptom persistence was estimated among DENV infected and febrile controls (Table 3). Among those with DENV infection, there was a significantly higher association of at least one symptom with 1) female gender (adjusted odds

ratio [aOR]: 1.44; 95% CI: 1.04, 2.00) and 2) age groups of 21–30 years (aOR: 3.77; 95% CI: 1.14, 12.4) and 41–50 years (aOR: 4.47; 95% CI: 1.31, 15.3) when compared with those 5–10 years of age. Among febrile controls, persistence of at least one symptom was significantly more associated with the age groups of 41–50 years (aOR: 1.92; 95% CI: 1.29, 2.87) and ≥ 51 years (aOR: 1.73; 95% CI: 1.14, 2.62) compared with those 5–10 years of age. The aOR for the association of persistence of at least one symptom with female gender and ages 11–20, 41–50, and ≥ 51 years were significantly higher among DENV-infected individuals compared with febrile controls (Table 3). Finally, there was significantly less symptom persistence with DENV-4 compared with the other DENV serotypes, but no difference in those experiencing their first compared with a secondary DENV infection.

DISCUSSION

In this study of 3,659 DENV-infected participants, we found the percentage of symptom persistence was 9.1%, 6.6%, and 4.3% for follow-up visits occurring at 10–20 days, 21–30 days, and 31–60 days, respectively, after the time from symptom onset. These values were significantly less than what was observed in febrile controls at all timeframes. Furthermore, among those with DENV infection, female gender and older age were more likely to be associated with persistent symptoms, whereas DENV-4 serotype was associated with less persistence.

Some of the symptoms in our DENV infected population—body pain and gastrointestinal symptoms—have also been prominent in other dengue studies that looked at symptom persistence beyond the acute presentation.^{9,13–15} Our most

FIGURE 4. The presence of specific symptom groups at the acute visit (left axis) and persistent rates at three follow-up periods (right axis). The six symptom groups and their corresponding symptoms were “respiratory” (cough, rhinorrhea, and/or expectoration), “gastrointestinal” (decreased appetite, abdominal pain, diarrhea, nausea, and/or vomiting), “headache” (headache), “fatigue” (malaise and/or asthenia), “body pain” (myalgia, bone pain, and/or joint pain), and “rash” (truncal rash, extremity rash, and/or facial rash).

TABLE 3
Association of demographic and DENV-related factors with persistence of at least one symptom

	DENV infected			Febrile controls			P§
	Total (N = 3,659)	n (%)	aOR* (95% CI)	Total (N = 5,408)	n (%)	aOR† (95% CI)	
Gender							
Male	1,740 (47.5)	112 (6.4)	1.0	2,620 (48.5)	278 (10.6)	1.0	
Female	1,919 (52.5)	169 (8.8)	1.44 (1.04, 2.00)	2,788 (51.5)	288 (10.3)	0.97 (0.81, 1.16)	0.03
Age							
5–10	183 (5.0)	6 (3.3)	1.0	456 (8.4)	41 (8.9)	1.0	
11–20	1,076 (29.4)	56 (5.2)	1.78 (0.53, 6.00)	1,553 (28.7)	125 (8.1)	0.94 (0.64, 1.36)	0.91
21–30	1,046 (28.6)	85 (8.1)	3.77 (1.14, 12.4)	1,442 (26.7)	149 (10.3)	1.28 (0.89, 1.85)	0.02
31–40	640 (17.5)	57 (8.9)	2.97 (0.87, 10.1)	896 (16.6)	95 (10.6)	1.26 (0.86, 1.86)	0.08
41–50	440 (12.0)	50 (11.4)	4.47 (1.31, 15.3)	584 (10.8)	88 (15.1)	1.92 (1.29, 2.87)	< 0.01
≥ 51	274 (7.5)	27 (9.9)	3.48 (0.96, 12.6)	477 (8.8)	68 (14.3)	1.73 (1.14, 2.62)	< 0.01
			P _{trend} < 0.01			P _{trend} < 0.01	
DENV serotype¶							
1	219 (10.7)	29 (13.2)	1.65 (0.99, 2.73)				
2	34 (1.6)	4 (11.8)	1.95 (0.65, 5.81)				
3	1,048 (51.1)	94 (8.9)	1.08 (0.72, 1.64)				
4	751 (36.6)	42 (5.6)	0.61 (0.39, 0.93)				
Prior DENV infection status							
Primary infection	1,449 (39.6)	121 (8.4)	1.0				
Secondary infection	2,165 (59.2)	157 (7.3)	0.89 (0.62, 1.27)				
Indeterminate	45 (1.2)	3 (6.7)	0.55 (0.13, 2.44)				

* Mutually adjusted for age, gender, city, year of diagnosis, time between visits, time from symptom onset to follow-up visit, dengue virus (DENV) serotype, and prior DENV infection status.

† Mutually adjusted for age, gender, city, year of diagnosis, time between visits, and time from symptom onset to follow-up visit.

§ P value estimates the difference in the odds ratio for the association of factors with persistence of at least one symptom between DENV infected and febrile controls.

¶ Restricted to those with DENV serotype information obtained from culture or PCR (N = 2,052).

aOR = adjusted odds ratio; CI = confidence interval.

common symptom, headache, was found in 23% of DENV-infected patients from Cuba after 3 weeks, more than five times higher than what we noted (Figure 4).⁹ Fatigue, our second most common symptom, also affected a much larger proportion in other studies compared with ours (32–73% versus < 4%).^{9,12,14,15} One possible explanation for our lower rates of persistence of headache and fatigue is that our study occurred during a time in Peru when nearly all dengue cases were dengue fever and not the more severe DHF.²⁰

Rash was the only manifestation with a higher rate of persistence among DENV-infected subjects compared with febrile controls. Cutaneous manifestations have been well-described in dengue disease.^{3,16,21} Typical descriptions include a macular or morbilliform rash, which may be pruritic, often appearing at the time of defervescence, and lasting from 2 to 5 days. Given that 76.7% of study subjects still had fever at their acute visit, a significant portion may not have developed cutaneous symptoms until immediately after their initial presentation. As a result, the rate of persistence of rash may be underestimated. Indeed, the prevalence of cutaneous symptoms at the initial acute visit was significantly lower among those reporting fever as compared with those who did not (18.7% versus 26.7%; $P < 0.01$). Furthermore, the prevalence of rash among individuals who had a follow-up visit 31–60 days after their initial visit was significantly lower (0.3%) compared with those who presented 10–20 days for their follow-up visit (0.9%), thereby reinforcing previous observations that rash is a transient symptom developing a short time period after defervescence. Interestingly, the prevalence of rash at the follow-up visit remained elevated among DENV-infected individuals compared with controls irrespective of the time interval since the acute visit (data not shown). Therefore, although the frequency of persistence of rash may be underestimated in this study given the nature of the sampling, the overall pattern of higher rates of persistence of this symptom among DENV infected individuals remains consistent.

Women with DENV infection were more likely to have symptom persistence across follow-up compared with DENV-infected men in our study. One study looking at acute symptoms noted that DENV-infected women had more body pain, joint pain, and rash than men.²² Looking at time periods beyond the acute visit, a study from Singapore evaluating fatigue after 2 months and a study from Cuba examining multiple symptoms after 2 years both noted higher levels of persistent symptoms in females.^{10,13} Females have also been noted to have a worse prognosis or slower recovery after West Nile virus (WNV), CHIKV, and tick-borne encephalitis virus (TBEV) infections.^{23–25}

Consistent with our findings, others have shown that older age impacts many different outcomes of DENV infection, both in the acute presentation and subsequent outcomes. A study from Singapore noted that older subjects infected with DENV were hospitalized and had severe disease significantly more frequently than younger individuals.²⁶ Another study showed numerous acute clinical findings more often in Vietnamese adults than children, including arthralgia, myalgia, upper gastrointestinal bleeding, abdominal pain, thrombocytopenia, and elevated transaminases.²⁷ Finally, a study from Singapore that evaluated non-acute symptoms such as fatigue 2 months after first detection of DENV infection found a greater proportion in older subjects as compared with younger subjects.¹⁰ Similar age-related findings have also been noted with CHIKV and WNV infections.^{28,29}

Compared with the other DENV serotypes, we noted that individuals with DENV-4 infection were significantly less likely to have persistence of at least one symptom, independent of demographic factors, temporal factors, and prior DENV infection status. Although no longitudinal study has evaluated the association of symptom persistence with specific DENV serotype, many cross-sectional studies of acute DENV infection have assessed clinical features among the different DENV serotypes, including variations in acute

severity and specific manifestations^{16,30,31}; our findings provide more evidence of the distinct clinical differences among DENV serotypes.

Contrasts between primary and secondary DENV infection have been a topic of intense research and debate. One theory, antibody-dependent enhancement, contends that pre-existing, non-neutralizing antibodies from a previous DENV exposure may lead to more severe disease such as DHF and DSS upon subsequent infection with a heterologous DENV serotype. Alternative viewpoints incorporate a more complex interplay of factors including population dynamics and viral genetics that influence the severity of disease³²; the lack of an association of prior DENV infection status with symptom persistence in this study lends support and is consistent with this second hypothesis. A study from Singapore similarly did not find a link between the duration of DENV-associated symptoms and prior DENV exposure.¹⁵ Nevertheless, host immunological factors may be important, as others have shown that genotypic differences in the immune receptor FcγRII correlate with long-term persistence of symptoms after DENV infection.¹³

Although only a handful of prior reports describe and enumerate symptom persistence after the acute febrile period with dengue fever, this phenomenon has been described for other arboviral infections. For example, the alphaviruses CHIKV, MAYV, and Ross River virus may cause joint pain that lasts weeks to years.^{33–35} Symptom persistence with CHIKV infection has been the most documented, with rates of persistent arthralgia ranging between 14% and 87% 1 month after the acute phase.³⁶ Infection with non-DENV flaviviruses, such as WNV, TBEV, Japanese encephalitis virus (JEV), St. Louis encephalitis virus, and Murray Valley virus, may result in long-standing objective findings such as permanent neurologic damage following infection.^{23,37} Of those with symptomatic JEV infection, approximately two-thirds have encephalitis, leading to death in 20–30% and neuropsychiatric sequelae in 50–60% at hospital discharge.

A few caveats to our study exist. First, subjects were only assessed at one follow-up visit, and those who attended their follow-up visit earlier may have been significantly different with respect to reporting their symptoms or access to care compared with those who attended a follow-up visit at later time periods. However, individuals who attended follow-up at the earlier time period of 10–20 days from symptom onset did not differ with respect to demographic factors such as age, sex, and city and the proportion that were DENV infected compared with those who attended follow-up at the later time periods of 21–30 or 31–60 days. Second, attendance at the follow-up visit was voluntary (i.e., passive follow-up). As a result, the group of individuals who attended a follow-up visit may not be representative of the total group of individuals who initially presented with symptoms. Third, this study did not explore symptom severity or the effect of symptoms on subjects' quality of life. Fourth, this study did not collect information on medication usage or co-morbidities that may have influenced the likelihood of symptoms at the follow-up visit. Finally, because this study focused on subjects in the first two months after DENV infection, it limited our ability to discriminate persistent-but-resolving from permanent symptoms (whether predating the DENV infection or not).

Our study possessed many strengths not found in other studies, including a large sample size and a febrile control

group. The 7.7% frequency of symptom persistence in our DENV-infected subjects was less than what we observed in febrile controls and lower than what prior studies reported in infection with other arboviruses.^{23,33–36} However, assuming 96 million people suffer from acute, symptomatic DENV infection annually, this represents a potential disease burden of 7.4 million cases, a substantial number.¹ We hope that our results will better inform patients—and their family members, physicians, and employers—of what to expect in the weeks after DENV infection. Our future work will focus on longer follow-up times and quality of life, as we have done for other arboviruses.³⁵

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