

Implications of Dengue Outbreaks for Blood Supply, Australia

Helen M. Faddy, Clive R. Seed, Jesse J. Fryk, Catherine A. Hyland, Scott A. Ritchie, Carmel T. Taylor, Kathryn L. Van Der Merwe, Robert L.P. Flower,¹ and William J.H. McBride¹

Dengue outbreaks have increased in size and frequency in Australia, and transfusion-transmitted dengue poses a risk to transfusion safety. Using whole blood samples collected during the large 2008–2009 dengue epidemic, we estimated the risk for a dengue-infectious blood donation as ≈ 1 in 7,146 (range 2,218–50,021).

Dengue causes >50 million infections per year worldwide; however, the true incidence is expected to be higher given that asymptomatic infection is possible (1). Dengue virus types 1–4 (DENV-1–4) are emerging or re-emerging in many regions of the world (1,2), including Australia (3). One of the largest epidemics in at least 50 years occurred in Queensland, Australia, during 2008–2009, with separate outbreaks in Cairns (and surrounding regions; DENV-2, DENV-3; 2008–2009), Innisfail (DENV-4; 2009), and Townsville (DENV-1, DENV-3; 2009), totaling >1,000 confirmed clinical cases (3).

Infection with DENVs poses a risk for transfusion safety, and 5 cases of transfusion-transmitted dengue have been reported (4,5). In addition, DENV RNA has been detected in asymptomatic blood donors from areas to which dengue is endemic (6–8). Given the absence of an approved blood screening test for dengue in Australia, managing transfusion-transmission risk focuses on identifying donors at risk for exposure and temporarily excluding them from donating fresh blood components (erythrocytes, platelets, and clinical plasma) (referred to here as dengue management strategy) (9). Plasma collection for fractionation can continue because the process of manufacturing

concentrates inactivates the virus (10). This approach assists with meeting an expanding demand for intravenous Ig but may result in fresh component losses and be associated with considerable cost.

Risk to the blood supply correlates with asymptomatic donor viremia; understanding the rate of dengue subclinical infection in countries to which it is not endemic and local northern Queensland seroprevalence is necessary for assessing this risk. We examined dengue seroprevalence rates in Australian donors during this epidemic; used these data to estimate the subclinical infection rate, population prevalence, and associated transfusion-transmission risk; and estimated the economic effect of this epidemic to the Australian Red Cross Blood Service (Blood Service).

The Study

Whole blood samples collected during the 2008–2009 dengue epidemic were tested for DENV IgM by ELISA (Dengue IgM Capture ELISA; Panbio, Brisbane, QLD, Australia). All reactive samples were tested with a second ELISA (Anti-Dengue IgM ELISA; Standard Diagnostics Inc., Giheung-gu, South Korea) and by the Public Health Virology Laboratory at Queensland Health Forensic and Scientific Services (QHFSS) (11). Serologic evidence of recent exposure (presence of DENV IgM) was observed in 12 (0.22%) donors (Table 1). Of the 8 DENV IgM-positive samples that were examined for type specificity, 7 (88%) were DENV-3 specific, which was the dominant type during the epidemic (3).

We used these IgM seroprevalence rates to estimate the rate of subclinical dengue infection (online Technical Appendix, wwwnc.cdc.gov/EID/article/19/5/12-1664-Techapp1.pdf). We estimated 168–921 subclinical cases (clinical:subclinical ratio 1.0:0.59; range 0.18–1.0) in Cairns, the city where the epidemic was centered. Our estimate was toward the lower end of that observed in dengue-endemic areas (12,13) but higher than that estimated during a DENV-2 outbreak in Charters Towers (14), which probably reflects the different methods used in the respective studies.

Selected samples were tested for DENV IgG by ELISA (Dengue IgG Indirect ELISA; Panbio). All reactive samples were tested at QHFSS. Serologic evidence of previous exposure (presence of DENV IgG) was influenced overall by donor location ($p < 0.05$) and age ($p < 0.05$). The proportion of the northern Queensland donor population with DENV IgG was 9.43% (95% CI 7.98%–10.89%), and this proportion increased with age (Table 2), which indicates cumulative previous exposure. The proportion of Melbourne (control area with no dengue activity) donors with DENV IgG was 6.78% (95% CI 4.48%–9.09%); however,

Author affiliations: Australian Red Cross Blood Service, Kelvin Grove, Queensland, Australia (H.M. Faddy, J.J. Fryk, C.A. Hyland, K.L. Van Der Merwe, R.L.P. Flower); Australian Red Cross Blood Service, Osborne Park, Western Australia, Australia (C.R. Seed); James Cook University, Cairns, Queensland, Australia (W.J.H. McBride, S.A. Ritchie); Queensland Health, Cairns (S.A. Ritchie); and Queensland Health Forensic and Scientific Services, Coopers Plains, Queensland, Australia (C.T. Taylor)

DOI: <http://dx.doi.org/10.3201/eid1905.121664>

¹Joint senior authors.

Table 1. Serologic evidence of recent dengue exposure by dengue virus IgM in blood donors, northern Queensland, Australia, 2008–2009 epidemic

Location	Donors		Donations	
	No. samples	Reactive samples, no. (% [95% CI])	No. samples	Reactive samples, no. (% [95% CI])
North Queensland	5,453	12 (0.22 [0.10–0.34])	10,026	13 (0.13 [0.06–0.20])
Cairns	2,416	8 (0.33 [0.10–0.56])	5,051	9 (0.18 [0.06–0.29])
Townsville	3,037	4 (0.13 [0.00–0.26])	4,975	4 (0.08 [0.00–0.16])

no change was observed with age (Table 2), demonstrating no cumulative exposure. Previous exposure in Melbourne was surprisingly high; these persons may have been exposed during travel to dengue-endemic areas (subsequent follow-up demonstrated 94% reported travel to countries to which dengue may be endemic).

The proportion of donors with DENV IgG did not change from the beginning to the end of the outbreaks in Cairns and Townsville, nor in northern Queensland as a whole (Table 2), which suggests that the epidemic was not of a scale to result in a change in population seroprevalence. This study was powered to detect a change in incidence of at least 10%; small changes might have been missed, which would be difficult to detect through such studies.

We used our seroprevalence data along with donation frequencies to estimate the risk of collecting a dengue-infectious donation, based on published models (9,15) (online Technical Appendix). Using this model, the risk of collecting a dengue-infectious donation in Cairns during the

epidemic was ≈ 1 in 7,146 (range 2,218–50,021) (Figure). These estimates are similar to those obtained by using a published probabilistic model (9) revised to incorporate the outbreak specific subclinical infection rate reported herein, which predicts the risk to be ≈ 1 in 9,303 (range 3,092–32,344) donations in Cairns. Because both methods derive estimates within comparable ranges, it would appear valid to use the revised probabilistic model as a predictive risk estimator during future outbreaks.

The dengue management strategy used during the epidemic cost the Blood Service ≈ 1 –3.8 million Australian dollars (2009 terms). This estimate is publically available and was based on: the number of donations affected by the dengue management strategy, collection targets for 2009, costs associated with whole blood collections, additional costs to meet national targets, transportation costs to meet demand in affected regions, and additional waste costs. An offset for any plasma obtained through a whole blood donation (used for fractionation) was included in selected estimates.

Conclusions

Subclinical dengue infection rates vary by population, specific outbreak, and area examined (12,14). We demonstrate that the clinical to subclinical infection rate during the 2008–2009 dengue epidemic in northern Queensland, where dengue occurs seasonally, was toward the lower end of that observed in dengue-endemic countries (12,13). This observation, together with our data suggesting that the incidence of dengue in the northern Queensland population did not change from the beginning to the end of the epidemic, suggests the control and clinical management of dengue during this epidemic was comprehensive.

We also estimated that the risk of collecting a dengue-infectious blood donation in Cairns during the epidemic was ≈ 1 in 7,146 (range 2,218–50,021). Given these risks, the increasing need for plasma in Australia, and the absence of a screening test for blood donations in Australia, the continuation of the dengue management strategy during future outbreaks is warranted. However, this strategy may have added strain on the inventory available to meet clinical demand for fresh blood components and was associated with a cost to the Blood Service of >1 million Australian dollars. Although this strategy is a necessary precaution to maintain safety, alternative approaches may exist, such as implementation of a suitable screening test (were one available) or pathogen reduction technology (a process designed to inactivate

Table 2. Prevalence of dengue virus IgG in the blood donor population, northern Queensland, Australia

Variable	No. samples	Reactive, no. (% [95% CI])
Northern Queensland	1,548	146 (9.43 [7.98–10.89])
Age group, y		
<40	567	18 (3.17 [1.73–4.62])
40–60	742	70 (9.43 [7.33–11.54])
>60	239	58 (24.27 [18.83–29.70])
Outbreak		
Start	788	72 (9.14 [7.13–11.15])
End	760	74 (9.74 [7.63–11.84])
Cairns	738	53 (7.18 [5.32–9.04])
Age group, y		
<40	271	5 (1.85 [0.24–3.45])
40–60	360	26 (7.22 [4.55–9.90])
>60	107	22 (20.56 [12.90–28.22])
Outbreak		
Start	384	31 (8.07 [5.35–10.80])
End	354	22 (6.21 [3.70–8.73])
Townsville	810	93 (11.48 [9.29–13.68])
Age group, y		
<40	296	13 (4.39 [2.06–6.73])
40–60	382	44 (11.52 [8.32–14.72])
>60	132	36 (27.27 [19.68–34.87])
Outbreak		
Start	404	41 (10.15 [7.20–13.09])
End	406	52 (12.81 [9.5–16.066])
Melbourne	457	31 (6.78 [4.48–9.09])
Age group, y		
<40	151	11 (7.28 [3.14–11.43])
40–60	215	16 (7.44 [3.93–10.95])
>60	91	4 (4.40 [0.18–8.61])

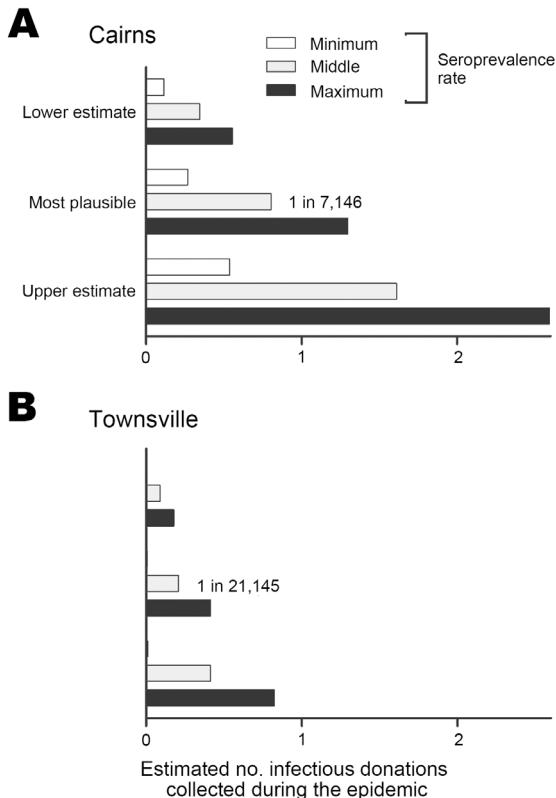


Figure. Risk of collecting a dengue-infectious blood donation, northern Queensland, Australia, 2008–2009 epidemic. Estimated risk calculated for Cairns (A) and Townsville (B).

pathogens in blood products), which may offer a similar level of safety but be more cost effective. With dengue becoming increasingly common in Australia (3) and the world (1), these alternative approaches may be needed in the future.

Acknowledgments

We thank Donor Services staff involved in collecting additional samples; members of the Brisbane manufacturing team, especially H. Holwell, for assistance; staff at QHFSS, especially S. Wynwood, for performing confirmatory testing; W. Zayonce and P. Diaz for assisting with data acquisition; and A. Olds and J. Pink for critically reviewing the manuscript.

This study was conducted under approval from the Blood Service Human Research Ethics Committee. We acknowledge Australian governments that fully fund the Australian Red Cross Blood Service to provide blood products and services to the Australian community.

Dr Faddy is a research fellow in the Research and Development Division of the Australian Red Cross Blood Service. Her current research interests and activities focus on providing an evidence base to enable evaluation of current emerging infectious risks to the safety of the Australian blood supply.

References

- Guzman MG, Halstead SB, Artsob H, Buchy P, Farrar J, Gubler DJ, et al. Dengue: a continuing global threat. *Nat Rev Microbiol.* 2010;8(Suppl):S7–16. <http://dx.doi.org/10.1038/nrmicro2460>
- Radke EG, Gregory CJ, Kintziger KW, Sauber-Schatz EK, Hunsperger EA, Gallagher GR, et al. Dengue outbreak in Key West, Florida, USA, 2009. *Emerg Infect Dis.* 2012;18:135–7. <http://dx.doi.org/10.3201/eid1801.110130>
- Fitzsimmons GJ, Wright P, Johansen CA, Whelan PI. Arboviral diseases and malaria in Australia, 2008–09: annual report of the National Arbovirus and Malaria Advisory Committee. *Commun Dis Intell Q Rep.* 2010;34:225–40.
- Lanteri MC, Busch MP. Dengue in the context of “safe blood” and global epidemiology: to screen or not to screen? *Transfusion.* 2012;52:1634–9. <http://dx.doi.org/10.1111/j.1537-2995.2012.03747.x>
- Stramer SL, Linnen JM, Carrick JM, Foster GA, Krysztos DE, Zou S, et al. Dengue viremia in blood donors identified by RNA and detection of dengue transfusion transmission during the 2007 dengue outbreak in Puerto Rico. *Transfusion.* 2012;52:1657–66. <http://dx.doi.org/10.1111/j.1537-2995.2012.03566.x>
- Linnen JM, Vinelli E, Sabino EC, Tobler LH, Hyland C, Lee TH, et al. Dengue viremia in blood donors from Honduras, Brazil, and Australia. *Transfusion.* 2008;48:1355–62. <http://dx.doi.org/10.1111/j.1537-2995.2008.01772.x>
- Petersen LR, Tomashek KM, Biggerstaff BJ. Estimated prevalence of dengue viremia in Puerto Rican blood donations, 1995 through 2010. *Transfusion.* 2012;52:1647–51. <http://dx.doi.org/10.1111/j.1537-2995.2011.03529.x>
- Dias LL, Amarilla AA, Poloni TR, Covas DT, Aquino VH, Figueiredo LT. Detection of dengue virus in sera of Brazilian blood donors. *Transfusion.* 2012;52:1667–71. <http://dx.doi.org/10.1111/j.1537-2995.2012.03729.x>
- Seed CR, Kiely P, Hyland CA, Keller AJ. The risk of dengue transmission by blood during a 2004 outbreak in Cairns, Australia. *Transfusion.* 2009;49:1482–7. <http://dx.doi.org/10.1111/j.1537-2995.2009.02159.x>
- Xie YW, Chan PK, Szeto CK, Kwok SY, Chu IM, Chu SS, et al. Clearance of dengue virus in the plasma-derived therapeutic proteins. *Transfusion.* 2008;48:1342–7. <http://dx.doi.org/10.1111/j.1537-2995.2008.01647.x>
- Taylor C, Simmons R, Smith I. Development of immunoglobulin M capture enzyme-linked immunosorbent assay to differentiate human flavivirus infections occurring in Australia. *Clin Diagn Lab Immunol.* 2005;12:371–4.
- Rodriguez-Figueroa L, Rigau-Perez JG, Suarez EL, Reiter P. Risk factors for dengue infection during an outbreak in Yanes, Puerto Rico in 1991. *Am J Trop Med Hyg.* 1995;52:496–502.
- Burke DS, Nisalak A, Johnson DE, Scott RM. A prospective study of dengue infections in Bangkok. *Am J Trop Med Hyg.* 1988;38:172–80.
- McBride WJ, Mullner H, LaBrooy JT, Wronski I. The 1993 dengue 2 epidemic in Charters Towers, north Queensland: clinical features and public health impact. *Epidemiol Infect.* 1998;121:151–6. <http://dx.doi.org/10.1017/S0950268898001058>
- Weinstein P, Weinstein SR, Rowe RJ. Transfusions: how many cases of Ross River virus infection do we cause? *Med J Aust.* 1995;163:276.

Address for correspondence: Helen M. Faddy, Research and Development, Australian Red Cross Blood Service, PO Box 145, Kelvin Grove, QLD 4059, Australia; email: hfaddy@redcrossblood.org.au

Implications of Dengue Outbreaks for Blood Supply, Australia

Technical Appendix

Statistical Analysis

Sample numbers for dengue virus (DENV) IgG testing were determined by using a power analysis (1) to enable detection of a 10% difference in prevalence between the beginning and end of the Cairns and Townsville outbreaks with at least 85% power. The proportion of donations/donors with the presence of antibody was determined and a 95% CI calculated. Logistic regression modeling was used to assess the relationship of donor sex, age, blood group (ABO and Rh antigens), and location with DENV IgG status. The antibody response (reactive or nonreactive) was the dependent variable, with sex, location, blood group, and age group as factors. The goodness-of-fit of the model was assessed by using the Deviance and Pearson χ^2 tests. The Statistical Package for the Social Sciences (SPSS; IBM Australia Ltd., St. Leonards, NSW, Australia) was used for analyses. Significance was determined with a p value ≤ 0.05 .

Subclinical Infection Estimate

Given the length of DENV IgM persistence (6 months for primary dengue infection) (2) and the timing of the last reported locally acquired dengue case in North Queensland before this epidemic (early 2008) (3), it is likely that all donors donating during the 2008–2009 dengue epidemic and who were positive for DENV IgM were exposed during this epidemic. To be eligible to donate blood in Australia, donors must satisfy a number of criteria, including being well at the time of the donation. It is therefore highly unlikely that a donor would have been eligible to donate if he or she had a symptomatic dengue infection. Furthermore, donors are requested to inform the Blood Service if they experience any illness within 7 days of donation so their donation can be quarantined or recalled, ruling out donors with symptomatic infection who may have donated during the presymptomatic viremic period. Assuming 100% compliance with Blood Service policy, that any donor with DENV IgM was exposed during the 2008–2009 epidemic and that the blood donor population had a

similar level of dengue exposure as the general population, we utilized IgM seroprevalence rates, along with estimates of the Cairns (164,354) and Townsville (181,740) populations in 2009 (4), to estimate the rate of subclinical dengue infection. Although seroprevalence data without detailed clinical information will not produce a definitive answer here, it will provide an estimate of the scale of the problem, independent of current notification systems. These estimates of subclinical infection do not include ill “non-reporters” who did not seek medical attention and who were unable to donate, so although our estimate is realistic, it is likely to underestimate the true number of infections during the epidemic. Moreover, our estimate assumes that all dengue cases during the epidemic were actually notified; the subclinical rate could of course be lower depending on the number of cases that were not notified as a ratio of those that were.

Example Calculation (Cairns)

NOTE: All calculations were performed by using Microsoft Excel (Redmond, WA, USA). As such, rounding may have led to slight variation when compared with manual calculations.

Population = 164,354 (4)

DENV IgM seroprevalence rate (Cairns) = 0.33% (95% CI 0.10–0.56%) (Table 1)

Estimated number of subclinical cases (lower limit) = DENV IgM seroprevalence rate (lower 95% CI) × Population (or $0.10\% \times 164,354 = 168$)

Estimated number of subclinical cases (most probable) = DENV IgM seroprevalence rate × Population (or $0.33\% \times 164,354 = 544$)

Estimated number of subclinical cases (upper limit) = DENV IgM seroprevalence rate (upper 95% CI) × Population (or $0.56\% \times 164,354 = 921$)

Therefore, we estimate that there were between 168 and 921 subclinical cases in Cairns during the epidemic.

The number of clinical cases in Cairns during the epidemic was 917 (3). Using the subclinical estimates described above, we estimate a clinical to subclinical ratio of 1.0:0.59 ($(1/917) \times 544$) with a range of 1.0:0.18 ($(1/917) \times 168$) to 1.0:1.0 ($(1/917) \times 921$).

Risk Analyses

Two different models were used to estimate the transfusion-transmission risk during the 2008–2009 dengue epidemic. The first estimates the risk of collecting a dengue-infectious donation based on our seroprevalence data from blood donations (Table 1) along with the donor donation frequency for each city during the outbreak period (which included 3 months after the last confirmed case) and is based on published models (5,6). Theoretically there are 3 sources of obtaining a viremic donation: 1) asymptomatic cases; 2) clinical cases that may slip through the screening process; and 3) donors who may donate during the 1–2 day presymptomatic viremic period (7). As mentioned earlier, the Blood Service has policies in place to ensure the latter 2 scenarios are minimized, so this risk model is based on the first source only.

The probability of collecting a dengue-infectious donation was estimated for the duration of the Cairns and Townsville outbreaks separately. We estimate the probability of collecting an infectious donation ($P_{\text{infectious donation}}$) as

$$P_{\text{infectious donation}} = (\text{length of viremia in days for asymptomatic infection} / \text{length of IgM persistence in days [plus the IgM-negative viremic period]}) \times \text{IgM donation seroprevalence during the outbreak} \times \text{donor donation frequency during the outbreak.}$$

This probability was then used to predict the number of infectious blood donations ($N_{\text{infectious donations}}$) collected over the course of the outbreak by

$$N_{\text{infectious donations}} = P_{\text{infectious donation}} \times \text{number of donations collected during the outbreak.}$$

The duration of viremia was derived from a range of published studies, and assuming the duration of viremia in asymptomatic infection is similar to what is observed in clinical cases (6,8–10), we have estimated that it ranges from 3 to 14 days (most plausible estimate 7 days). The duration of IgM seropositivity was based on 6 months of persistence (2). Given that a period of viremia exists before the development of an IgM response, approximated as 5 days for the purposes of this analysis, it was necessary to adjust for this by adding this period of time to the duration of IgM seropositivity. The IgM seroprevalence rate obtained for donations collected during the outbreak for each region was modeled (middle estimate) along with the upper (maximum estimate) and lower (minimum estimate) 95% CI for each proportion. In addition, the number of donations, which had the potential to result in a blood component, collected during the epidemic was retrospectively obtained.

Example Calculation (Cairns, most plausible estimate [length of asymptomatic viremia], medium estimate for IgM seroprevalence)

NOTE: All calculations were performed by using Microsoft Excel. As such, rounding may have led to slight variation when compared with manual calculations.

Length of viremia in days for asymptomatic infection = 7 days

Length of IgM persistence in days (plus the IgM negative viremic period) = 187 (6 months + 5 days)

IgM donation seroprevalence during the outbreak (Cairns) = 0.18%

Number of donations collected during the outbreak (Cairns) = 5,753

Number of donors giving successful donations during the outbreak (Cairns) = 2,770

Donor donation frequency during the outbreak = 2.0769

$P_{\text{infectious donation}} = (7/187) \times 0.18\% \times 2.0769 = 0.000139941$

$N_{\text{infectious donations}} = 0.000139941 \times 5,753 = 0.805077923$

$\text{Risk}_{\text{collecting an infectious unit}} = (1/0.805077923) \times 5,753 = 7,146$ (or 1 in every 7,146 donations)

A published probabilistic model was also used, which estimates the risk for dengue transmission by blood using the number of confirmed cases during the epidemic (also including 3 months after the last confirmed case) (6). This model was adapted to incorporate the subclinical infection rate (including the upper and lower 95% CI) estimated from the seroprevalence data for these outbreaks from this study, which addresses 1 key limitation of the original model. The specifics of this model are described elsewhere (6). In this study, we applied this model to the 2008–2009 dengue outbreaks in Cairns and Townsville.

References

1. Moore DS, McCabe GP. Introduction to the practice of statistics. 3rd ed. New York: WH Freeman and Company; 1999.
2. Prince HE, Matud JL. Estimation of dengue virus IgM persistence using regression analysis. Clin Vaccine Immunol. 2011;18:2183–5. [PubMed http://dx.doi.org/10.1128/CVI.05425-11](http://dx.doi.org/10.1128/CVI.05425-11)
3. Fitzsimmons GJ, Wright P, Johansen CA, Whelan PI. Arboviral diseases and malaria in Australia, 200809: annual report of the National Arbovirus and Malaria Advisory Committee. Commun Dis Intell. 2010;34:225–40.

4. Australian Bureau of Statistics. 3218.0 Regional population growth, Australia as of 30 March 2012 [cited 2012 May 14]. <http://www.abs.gov.au/AUSSTATS/abs@.nsf/DetailsPage/3218.02010-11>
5. Weinstein P, Weinstein SR, Rowe RJ. Transfusions: how many cases of Ross River virus infection do we cause? *Med J Aust*. 1995;163:276. [PubMed](#)
6. Seed CR, Kiely P, Hyland CA, Keller AJ. The risk of dengue transmission by blood during a 2004 outbreak in Cairns, Australia. *Transfusion*. 2009;49:1482–7. [PubMed](#)
<http://dx.doi.org/10.1111/j.1537-2995.2009.02159.x>
7. Guzmán MG, Kouri G. Dengue: an update. *Lancet Infect Dis*. 2002;2:33–42. [PubMed](#)
[http://dx.doi.org/10.1016/S1473-3099\(01\)00171-2](http://dx.doi.org/10.1016/S1473-3099(01)00171-2)
8. Vaughn DW, Green S, Kalayanarooj S, Innis BL, Nimmannitya S, Suntayakorn S, et al. Dengue viremia titer, antibody response pattern, and virus serotype correlate with disease severity. *J Infect Dis*. 2000;181:2–9. [PubMed](#) <http://dx.doi.org/10.1086/315215>
9. Gubler DJ. Dengue and dengue hemorrhagic fever. *Clin Microbiol Rev*. 1998;11:480–96. [PubMed](#)
10. Murgue B, Roche C, Chungue E, Deparis X. Prospective study of the duration and magnitude of viraemia in children hospitalised during the 1996-1997 dengue-2 outbreak in French Polynesia. *J Med Virol*. 2000;60:432–8. [PubMed](#) [http://dx.doi.org/10.1002/\(SICI\)1096-9071\(200004\)60:4<432::AID-JMV11>3.0.CO;2-7](http://dx.doi.org/10.1002/(SICI)1096-9071(200004)60:4<432::AID-JMV11>3.0.CO;2-7)