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RESEARCH ARTICLE

A REVIEW OF LABORATORY DIAGNOSIS OF DENGUE FEVER

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ABSTRACT

Dengue, a mosquito-transmitted viral disease that produces variable symptoms, ranging from asymptomatic infection to life-threatening disease, is present in about 110 tropical and subtropical countries. As dengue is increasing in incidence, improved diagnosis, early detection of severe cases, and efficient medical management are of primary importance in all areas where dengue is endemic. Traditionally, dengue has been diagnosed by virus isolation or serological methods, but with recent advances in molecular techniques and in rapid detection technology, a range of novel diagnostic tests will soon be commercially available that will improve case management and aid disease control efforts.

INTRODUCTION

Dengue fever, also known as break bone fever, is an infectious tropical disease caused by the dengue virus. Symptoms include fever, headache, muscle and joint pains, and a characteristic skin rash that is similar to measles. In a small proportion of cases the disease develops into the life-threatening dengue hemorrhagic fever, resulting in bleeding, low levels of blood platelets and blood plasma leakage, or into dengue shock syndrome, where dangerously low blood pressure occurs. Dengue is transmitted by several species of mosquito within the genus *Aedes*, principally *A. aegypti*. The virus has four different types; infection with one type usually gives lifelong immunity to that type, but only short-term immunity to the others. Subsequent infection with a different type increases the risk of severe complications. As there is no commercially available vaccine, prevention is sought by reducing the habitat and the number of mosquitoes and limiting exposure to bites.

Treatment of acute dengue is supportive, using either oral or intravenous rehydration for mild or moderate disease, and intravenous fluids and blood transfusion for more severe cases. The incidence of dengue fever has increased dramatically since the 1960s, with around 50–100 million people infected yearly. Early descriptions of the condition date from 1779, and its viral cause and the transmission were elucidated in the early 20th century. Dengue has become a global problem since the Second World War and is endemic in more than 110 countries. Apart from eliminating the mosquitoes, work is ongoing on a vaccine, as well as medication targeted directly at the virus. The acquired immune response to infection with dengue virus consists of the production of IgM and IgG antibodies primarily directed against the virus envelope proteins. The immune response varies depending on whether the individual has a primary or a secondary infection (Vorndam V & Kuno G; 1997) In general, sero diagnosis of dengue is dependent on the stage of the infection.

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A primary infection with dengue is characterized by a slow and low-titre antibody response. IgM antibody is the first immunoglobulin isotype to appear. Anti-dengue IgG at low titre is detectable at the end of the first week of illness, increasing slowly thereafter. In contrast, during a secondary infection (a dengue infection in a host that had been previously infected by a dengue virus or other flavivirus) antibody titres rise extremely rapidly and antibody reacts broadly with many flaviviruses (Innis B). High levels of IgG are detectable even in the acute phase and they rise dramatically over the following 2 weeks. The kinetics of the IgM response are more variable. Since IgM levels are significantly lower in secondary dengue infections, some false-negative results in tests for anti-dengue IgM are observed during secondary infections. According to the Pan American Health Organization (PAHO) guidelines (PAHO; 1994), IgM antibody is detectable by day 5 of illness in 80% of all dengue cases, and by day 6–10 of illness in 93–99% of cases, and may then remain detectable for more than 90 days. IgM antibody-capture enzyme-linked immunosorbent assay (MAC-ELISA) has become an important tool in the routine diagnosis of dengue; this technique has a sensitivity and specificity of approximately 90% and 98%, respectively, but only when used 5 or more days after the onset of fever.

Laboratory Diagnosis

The diagnosis of dengue fever can be done by virus isolation in cell cultures, nucleic acid detection by PCR, viral antigen detection (such as for NS1) or specific antibodies (serology) (Guzman MG *et al.*; 2010 & WHO; 2009). Virus isolation and nucleic acid detection are more accurate than antigen detection, but these tests are not widely available due to their greater cost (WHO; 2009). Detection of NS1 during the febrile phase of a primary infection may be greater than 90% however is only 60-80% in subsequent infections (Simmons CP *et al.*; 2012). All tests may be negative in the early stages of the disease (Ranjit S & Kissoon N; 2011 & Guzman MG *et al.*; 2010). PCR and viral antigen detection are more accurate in the first seven days (Simmons CP *et al.*, 2012). In 2012 a PCR test was introduced

that can run on equipment used to diagnose influenza; this is likely to improve access to PCR-based diagnosis (CDC; 2012). These laboratory tests are only of diagnostic value during the acute phase of the illness with the exception of serology. Tests for dengue virus-specific antibodies, types IgG and IgM, can be useful in confirming a diagnosis in the later stages of the infection. Both IgG and IgM are produced after 5–7 days. The highest levels (titres) of IgM are detected following a primary infection, but IgM is also produced in reinfection. IgM becomes undetectable 30–90 days after a primary infection, but earlier following re-infections. IgG, by contrast, remains detectable for over 60 years and, in the absence of symptoms, is a useful indicator of past infection. After a primary infection IgG reaches peak levels in the blood after 14–21 days. In subsequent re-infections, levels peak earlier and the titres are usually higher. Both IgG and IgM provide protective immunity to the infecting serotype of the virus (Chen LH and Wilson EM; 2010; Guzman MG *et al*.; 2010 and Gubler DJ; 2010). The laboratory test for IgG and IgM antibodies can cross-react with other flaviviruses and may result in a false positive after recent infections or vaccinations with yellow fever virus or Japanese encephalitis (Simmons CP *et al*.; 2012). The detection of IgG alone is not considered diagnostic unless blood samples are collected 14 days apart and a greater than fourfold increase in levels of specific IgG is detected. In a person with symptoms, the detection of IgM is considered diagnostic (Gubler DJ; 2010).

NS1 assays

The NS1 gene product is a glycoprotein produced by all flaviviruses and is essential for viral replication and viability. During viral replication, NS1 is localized to cellular organelles. The protein is secreted by mammalian cells, but not by insect cells. The secreted form of the protein is a hexamer composed of dimer subunits. Glycosylation of this protein is believed to be important for secretion. NS1 antigen appears as early as day 1 after the onset of fever and declines to undetectable levels after day 5–6. NS1 is also a complement-fixing antigen and it produces a very strong humoral response. Because this protein is secreted, many studies have been dedicated to the utility of NS1 as a tool for the diagnosis of infection with dengue virus. These studies focus on various aspects of diagnosis, including antigen-capture enzyme-linked immunosorbent assay (ELISA), and NS1-specific IgM and IgG responses. Commercial kits for the detection of NS1 antigen in serum samples are available. These assays do not differentiate between the serotypes. As NS1 antigen appears early in infection and before the appearance of antibody, such assays are useful for early case detection and for outbreak investigations. Evaluations of these assays should be performed to assess their utility and cost–effectiveness.

Conclusions

To improve case management, surveillance, outbreak investigations and to ensure the success of dengue vaccine trials, quality diagnostic tools are essential. However, current diagnostic tools available for dengue are not practical for point-of-care use or during the febrile phase of the disease. Many tools are commercially available but their performance and operational characteristics have not been widely evaluated. More novel diagnostic techniques need to be developed for patient management. The goal of a new diagnostic tool would combine antigen (e.g. NS1 antigen) and IgM/IgG detection in a single test and ideally prognostic markers of disease severity would be paired with etiologic diagnosis. The recommended new tools, reference material collection and specimen banks discussed within this document address these needs.

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REFERENCES

"New CDC test for dengue approved". Centers for Disease Control and Prevention, 2012.

Chen LH, Wilson ME. 2010. "Dengue and chikungunya infections in travelers". Curr. Opin. Infect. Dis. 23 (5): 438–444.

Gubler DJ. 2010. "Dengue viruses". In Mahy BWJ, Van Regenmortel MHV. Desk Encyclopedia of Human and Medical Virology. Boston: Academic Press, 372–82.

Guzman MG, Halstead SB, Artsob H. 2010. "Dengue: a continuing global threat". Nat. Rev. Microbiol. 8 (12 Suppl): S7–S16.

Innis B. An enzyme-linked immunosorbent assay to characterize dengue infections where dengue and Japanese encephalitis co-circulate. American Journal of Tropical Medicine and Hygiene, 40: 418–427.

PAHO. 1994. Dengue and dengue hemorrhagic fever in the Americas: guidelines for prevention and control (Scientific Publication No. 548). Washington DC, Pan American Health Organization.

Ranjit S, Kissoon N. 2011. "Dengue hemorrhagic fever and shock syndromes". Pediatr. Crit. Care Med. 12 (1): 90–100.

Simmons CP, Farrar JJ, Nguyen vV, Wills B. 2012. "Dengue". N Engl J Med 366 (15): 1423–1432.

Vorndam V, Kuno G. 1997. Laboratory diagnosis of dengue virus infections. In: Gubler DJ, Kuno G, (eds), Dengue and dengue hemorrhagic fever. New York, CAB International, pp. 313–333.

WHO, 2009. Dengue Guidelines for Diagnosis, Treatment, Prevention and Control. Geneva: World Health Organization, 90-95.

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