Dengue is a mosquito borne viral infection which is endemic in over 100 countries putting almost half of the world’s population at risk. Dengue virus is single positive stranded RNA belonging to family Flaviviridae and has four serotypes (DEN1-4). The disease is transmitted to humans by the mosquito vector Aedes aegypti. Infection due to dengue virus varies as this may be sub-clinical infection or clinical which includes dengue fever, dengue hemorrhagic fever and dengue shock syndrome. In recent decades, dengue fever has become the second most prevalent mosquito-borne infection after malaria. In Pakistan, outbreaks of dengue were reported since 1994 to date and caused high morbidity and mortality especially during the last epidemic of 2011 in Lahore.

Diagnosis of dengue infection starts with the clinical symptoms followed by laboratory confirmation. During early phase of illness, dengue patients often present with sudden onset of high grade fever along with nausea, aches and pains. However, these symptoms are common in all febrile illnesses and are not specific to dengue. The typical sign and symptoms include maculopapular rash, retro-orbital pain, petechiae or bleeding from GI tract, nose or gums and these trigger a differential diagnosis for dengue fever. Diagnostic algorithm and logistic regression models have also been developed to differentiate dengue from other febrile illnesses however, their usefulness remains to be tested in different populations with different circulating strains.

A number of laboratory tests are available for the diagnosis of dengue. However, the suitability of these tests depends upon the time when the patient presents to the health facility. Similarly the sensitivity and specificity of these tests also vary. These tests include detection of viral RNA, antigen, antibody or combined antigen/antibody detection from patient’s blood or other body fluids like saliva etc.

Dengue viral RNA can be detected by using Reverse transcriptase PCR (RT-PCR) and Real time PCR using blood, serum or plasma samples. It is a very sensitive (48-100%) and specific (100%) method for confirmation of dengue infection but it give results in 24-48 hours and require expertise and expensive laboratory equipment. A technique was developed which use single reaction mixture at constant temperature (nucleic acid sequence based amplification [NASBA]) having a sensitivity of 98.5% and 100% specificity. Real time PCR can also detect dengue virus RNA in urine and saliva of patients but the level of RNA is very low in these samples when compared to serum samples. The RT-PCR is useful in early 5-6 days of illness and it is rarely positive after 6 days of illness.

Another test used for the early detection of primary dengue infection is NS1 (Non-structural protein). This protein is highly conserved glycoprotein and remains detectable for up to 18 days (peak at 6-10 days) after the onset of symptoms. It has a sensitivity of 82% and a specificity of 98.9%. It is easy to perform and less expensive than RNA. However, this test is less sensitive in cases having secondary dengue infection (67-77%) as compared to primary infection (94-98%).

The most widely used test in the diagnosis of dengue infection is the detection of anti-dengue antibodies (IgM and IgG). These tests have two versions: ELISA and strip (rapid test). The ELISA based detection kits have higher sensitivity ranging between 61-99% and a specificity between 79-97% as compared to rapid test which have a sensitivity 20-97% and a specificity of 76-90%. Anti-dengue IgM antibodies can be detected within 3-5 days of the onset of illness and they continue to increase for up to 15 days. They may persist for approximately 179 days in case of primary dengue infection and 139 days in secondary dengue infection.

Detection of IgM antibodies indicates probable dengue infection where its levels are high in primary infection and low in secondary dengue infection. The dengue IgG antibodies are detected after 10 days of onset of illness however, there can be seen as early as 4 days in those having a second attack also called a secondary infection. Both these tests are least expensive, easy to perform and can differentiate between primary and secondary dengue infection.

Combined tests are also available in which all the three tests (NS1, dengue IgM, Dengue IgG) are combined into single reaction for ease of use. They have a sensitivity of 89-93% and a specificity of 75-100%.

Corresponding Author:
Ibrar Rafique
Pakistan Medical Research Council (PMRC) Head Office, Islamabad.
Email: ibrarpmc@gmail.com
In future, integrated micro fluidic system can be used for the detection of dengue IgM and IgG using a chip which will give results in 30 minutes i.e.1/8th of the time required for traditional method. It utilizes virus bound magnetic bead complexes for rapid serological analysis of antibodies 26-27.

Monitoring of progressions of disease from mild to severe forms by regular observation of signs (abdominal pain, mucosal bleeding and lethargy) and symptoms (severe plasma leakage, severe bleeding, increased AST/ALT levels, impaired consciousness and thrombocytopenia <100,000/mm$^3$) can also help in taking early measures to save life from complications like dengue hemorrhagic fever and dengue shock syndrome 28-30.

References