REVIEW ARTICLE

Dengue viral infections in Pakistan and other Asian countries: a comprehensive review

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Abstract

Infections due to Dengue virus are widespread throughout the world. Disease starts with mild flu like sickness to a severe intricate condition which results in the death of the patient. Dengue illness has high morbidity and mortality in Pakistan as well as in other Asian countries. The Review article is a discourse analysis that explores the facts about the history, emergence and impact of dengue in Pakistan and other Asian countries. Data was collected from internet sources, mainly using Science Direct and PubMed. The final literature was reviewed and summarised. About 150 articles were identified and 47 articles were shortlisted for final review. Aedesaegypti was found to be a major vector for the transmission and spread of dengue illness. Treatment comprises supportive therapy as no specific treatment was available. During the last couple of years, the incidence of dengue fever was extraordinary in metropolitan cities of Pakistan.

Keywords: Dengue, Clinical features, Pathogenesis, Diagnosis, Treatment.

Introduction

Dengue virus (DV) causes one of the most prevalent infections in the world which are transmitted by mosquitoes. These infections may be asymptomatic or may produce dengue fever (DF), undifferentiated fever and dengue haemorrhagic fever or dengue shock syndrome (DHF/DSS). DV contains single-stranded ribonucleic acid (RNA) and belongs to family flaviviridae. Four antigenically related but distinct viruses, DEN 1, DEN 2, DEN 3, and DEN 4, are considered as aetiological agents of DF.[1] DF is an acute febrile condition. Infected Aedes mosquito (Aedesaegypti) is the most important vector for the transmission of virus to humans.[2] This virus is affecting human population at a large scale, leading to high morbidity and mortality and becoming a serious issue over the last decade. Some of the patients suffering from DHF may display characteristics of circulatory disorder and refractory shock which is known as DSS.[3] Overpopulation in areas which have poor infrastructure also contributes to the transmission of disease and pathogen dispersal.[4] About three billion people in more than 100 countries of tropical and subtropical regions are prone to DV infection.[5] Lymphoma, leukopenia, lymphocytosis, high haematocrit and thrombocytopenia are common clinical characteristics of patients suffering from DV infection.[6] Secondary infections associated with different serotypes which are responsible for primary infections are considered as factors linked with epidemiology of DHF/DSS.[7] Recently, a novel one-step multiplex real-time reverse transcription polymerase chain reaction (RT-PCR) has been developed for the early diagnosis of dengue. This assay can detect an anti-dengue immunoglobulin M (IgM) positive response during the first five days of illness and almost 14 days after infection.[8] Biosensors, quartz crystal microbalance (QCM), surface plasmon resonance (SPR), electrochemical impedance spectroscopy (EIS), micro-and nanofabrication, microarray technology and microfluidic platforms are some significant emerging tools for the diagnosis of dengue.[9] Strategies to control dengue include integrated mosquito control, disease surveillance and emergency control.[10] Purpose of this review was to overlap the different clinical aspects of dengue infections. By following several preventive measures, the dengue outbreaks can be lessened to a great extent. Many diagnostic tests are in practice for the dengue infection, but there is a need to introduce more specific and low-cost diagnostic tools for the early diagnosis of dengue illness. Delay in the diagnosis and failure to identify the complications may result into worse outcomes.

Epidemiology

Dengue occurs mainly in the tropical regions of Africa, Oceania, Asia and Americas. Distribution of dengue is limited majorly by the mosquitoes (Aedesaegypti). Before 1970, only nine countries were listed for dengue cases. Since then, the number is continuously increasing.[11] Dengue is now endemic in more than 140 countries of Asia, Africa, Mediterranean and Americas where about four billion people are in danger. It is suggested that approximately 390 infections with about 2,000 deaths occur each year due to DV. In areas where there is more

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frequency of infection, the disease load favours to deviate from adults to children. There are several reasons for which Aedesaegypti is a powerful vector for the transmission of dengue; it prefers to feed on human blood, it is more vulnerable to DV, it can live and multiply in the close vicinity to human, its bite is insignificant, it feeds during daytime and it bites many people to get a sole blood meal.[12] Travellers play a significant role in the transmission, and, hence, in global epidemiology of dengue infections. Infected travellers pass on the virus in areas having more population of mosquitoes which then play a critical part for the transmission of infections (World Health Organisation (WHO).[13] In Southeast Asia, the trend of dengue epidemiology was greatly changed during World War II. Ecological changes and disruption due to war efforts broadened vector geographical distribution, making many areas permissive for epidemic transmission. After World War II, many people migrated to Southeast Asia searching for work, resulting in increased population in urban areas of the region. These areas were deficient in proper sewerage system, housing and water supply. Such conditions were ideal for Aedesaegypti breeding and making dengue an epidemic.[14] DHF spread in Sri Lanka, Pakistan, India, the Maldives and China. Now DF/DHF is dominant cause for morbidity of children in Asia.[15] In Pakistan, first documentation of dengue infection was made in 1982 in Punjab, reported in 12 patients out of a sample of 174.[16] DF was recorded in 1994, 1995 and in 1997. Two deaths were reported in Karachi due to dengue infection in 1995. After a period of 10 years, dengue re-emerged in Pakistan and about 395 cases were noted, all of them from Karachi. In 2006, the disease expanded to the north of Pakistan and 5,800 cases were announced from all over Pakistan. About 3,280 cases with 30 deaths were illustrated from across the country. It was followed by an epidemic in 2007 which caused significant morbidity and mortality.[17] In 2009, the number of people suffering from dengue halved compared to the preceding year.[18] Pakistan faced a major outbreak of dengue in 2010. More than 21,204 cases were reported from all over the country, mainly from Punjab. As a result, 350 out of 16,000 affected people died in Punjab. Out of these, 14,000 cases with 300 deaths were only from Lahore.[19] Hassan et al.[20] found that about 41,354 patients were screened for dengue infection. Result revealed 3.1% IgM positive and 0.3% immunoglobulin G (IgG) positive. Borderline positives for IgM were 1.7% and for IgG were 0.26%.

Clinical features of DF and DHF

It is believed that most clinical infections are asymptomatic; however, the disease starts from mild flu-like sickness to severe intricate condition, characterised by leakage from vessels, haemorrhage and shock, which results in the death of the patient.[21] DF is described as febrile illness. Fever remains for 5-7 days and about 50% patients face the problems associated with skin. During first 24-48 hours flushed faces are commonly observed. During the period of defervesence, maculopapular or petechial rashes are detected in patients.[22] Clinical symptoms are described as headache, fever, skin rashes, leukocytopenia and pain in bones and muscles. Because of intransigent pain DF is also known as breakbone fever. Skin rashes arise around a day after the absence of fever. Pal et al.[23] found that there is an important correlation between dengue viral load (VL) and the level of interferon-gamma (IFN-g). They concluded that there is powerful correlation of nausea, fever, pain, aches, rash and pain, constant vomiting and leukopenia. Such patients who have high values of VL displayed constant vomiting, abdominal pain, leukopenia and fluid accumulation. Premaratnaet al.[24] recorded 12 patients suffering from DF mimicking acute appendicitis. All 12 patients were suffering from right iliac fossa pain with serious tenderness and eight of them from leukopenia. Severity of dengue infection may lead to a condition known as DHF.[25] Most serious form of DHF can affect the life of patient, mainly by increasing the vascular permeability and shock. Subcutaneous bleeding, petechiae and readily bruised skin are the most common symptoms of DHF.[15] This catastrophic condition is mostly characterised by haemorrhages and shock.[26] It is suggested that virus antigens and response of host immune system can reduce the association of endothelial cells which is more common in DHF.[27] Boosted vascular permeability and leakage of plasma are the fundamentals to discriminate DHF/DSS from DF. Symptoms of DSS are the same as that of DHF supplemented with circulatory collapse, hypotension and shock.[28] There can also be moderate liver dysfunction. The occurrence of DHF and DSS is about 5%; however incidence may increase in previously infected individuals with other serotypes of dengue.[29] Recently, Lardo et al[30] presented a study related to the multi-serotype Dengue viral infection in Indonesia in patients with severe clinical signs i.e. haemorrhage, severe plasma leakage and organ damage concerning the liver, lung and kidney. In this study, Infection with dengue virus serotype 2 and 3 was detected and was concluded that viremia due to dengue virus 3 was higher as compared to dengue virus 2.

Pathogenesis

Pathogenesis of dengue is still poorly understood. However, it is said that there are successive intracellular events which play a part in the viral pathogenesis. After

the entry of virus, there is response of unfolded proteins followed by lipid bodies and lipophagy, endoplasmic reticulum and autophagy.[31] Dengue viral infection of the dendritic cells (DCs) of the skin is main as well as interesting focus point of current research. Such dendritic cells are thought to be the place of dengue viral residence and its interaction with the key actors of human immune system when virus enters the body after the bite of the infected mosquito.[32] It is suggested that pathogenesis of dengue infections is strongly related to the immune response of the host as the virus shows its serious phenotypes at the time when it is being cleared by the host immune responses. In humans, the major target in DV infections are monocytes, dendritic cells and macrophages. Langerhans cells in the dermis and epidermis are the first target of DV after its injection by infected mosquitoes.[33] It is also suggested that cross-reactivity of DV non-structural protein1 (NS1) with endothelial and platelet results into the damage of these cells with inflammatory response. These conclusions indicate that anti-dengue viral NS1 is associated in DV infections.[34] Ability of a viral infection is increased by antibody-dependent improvement; this phenomenon can also repress antiviral responses which are mediated by IFN type-1. Abnormal activated T cells and excessive production of soluble factors result in increased vascular permeability. Complement system activated as a result of cytokine production or activated immune is also associated in the process of plasma leakage. C3a and C5a proteins are the complement fragments which are known to be involved in vascular permeability.[35] Abnormal activation of platelets, endothelial cells and coagulators in DV infections is due to the production of induced autoantibodies.[36] Plasma leakage usually in pulmonary and peritoneal chambers in DHF is due to the exaggeration in vascular permeability mediated by cytokines. Walls of the vessels are not damaged because there is no vasculitis observed in DHF.[37] Povo'aet al.[38] analysed the lesions of different organs infected with dengue. Oedema and haemorrhages were commonly observed in all organs under that study. Necrotic areas as well as macro and microsteatosis were observed in the liver. Of all the organs, lung was the most damaged. Cardiac lesions involved myocarditis with destructed fibres. Spleens showed atrophy of lymphoid follicles and loss of germinal centres. Presence of megakaryocytes in alveolar spaces correlates with increased respiratory distress. Rathiet al.[39] performed autopsies of six patients who had died of acute dengue infection, and concluded that most common causes of death were hypotension, bleeding diathesis, hepatic failure, acute respiratory distress and acute renal failure.

Laboratory Diagnosis

DF represents acute illness with two or more of the following: myalgia, rash, arthralgia, retro-orbital pain and leukopenia. DHF is manifested by haemorrhagic presentations i.e. purpura, petechiae, bleeding from gastrointestinal tract (GIT) and mucosa, platelet count is less than 100000/mm^3. DSS includes all the manifestations of DHF with hypotension and lower pulse pressure (<20mmHg).[12] Tourniquet test is also widely used for the diagnosis of DHF and DSS. It depicts thrombocytopenia and capillary fragility and is recommended by the WHO.[40] Other laboratory methods for the diagnosis of dengue infections consist of isolation of virus, serological and molecular approaches. The virus can be isolated from the plasma, serum and leucocytes. However, from post-mortem cases it can be isolated from the spleen, liver, lung, thymus, lymph nodes, pleural fluid or cerebrospinal fluid. Different methods used for virus isolation include mosquito inoculation technique, mosquito cell line and vertebral cell culture. Serological tests include haemagglutination inhibition test, complement fixation test (CFT), enzyme-linked immunosorbent assay (ELISA), antigen capture ELISA and neutralisation test, while molecular test used for diagnosis is RT-PCR.[1] For the detection and isolation of virus, mosquito cell culture has been preferably used but due to low sensitivity it has been combined with RT-PCR for the swift diagnosis of dengue. Isolation of viruses can easily be executed with cultured mosquito cells i.e. Tra-284 (Toxorynchites amboinensis), conjugated linoleic acid 1 (CLA-1), AP-64 (clone of A. pseudoscutellaris cell line), C6/36 (A. albopictus) cell lines. Mammalian cell lines are also under practice i.e. LLCMK2 (monkey kidney), BHK21 (baby hamster kidney) and Vero (monkey kidney) cell lines. However, nowadays C6/36 cell line is also still in practice as a method of choice for routine isolation of DV.[41] This method is specific and feasible for the detection of serotypes but it has certain limitations such as need of good skill, it takes more than one week and is not able to differentiate between primary and secondary infections. Serological diagnosis has more advantages, i.e. it is effective for differential diagnosis, easy to operate and can discriminate between primary and secondary infection. Its drawback lies in the fact that serological tests may miss the dengue cases due to low or immeasurable level of IgM in secondary infections, requires two samples and there is delay in the confirmation of diagnosis.[13] Another test known as Dengue Duo Rapid Strip Test(DDRST) is also commercially available for the rapid diagnosis of dengue. It is an immune-chromatographic test which identifies IgM and IgG in capture format within 15 minutes.[42] RT-PCR has also proved a valuable tool for the serological

diagnosis of DF.[43]

Prevention and Treatment

There are two scenarios for the prevention of dengue infections. First concerns the control of mosquitoes and second includes avoiding the bite of mosquitoes.[11] Strategies involved in vector control include appropriate disposal of solid waste material, upgraded systems for the storage of water, implementation of suitable insecticides at larval dwelling sites, use of insecticide treated nets and spraying insecticides by focusing the mosquito breeding areas.[44] However, use of mosquito repellent, long dresses with full sleeves, mosquito coils and electric vapour mats and use of curtains can prevent mosquito bites.[45] Luz et al.[46] developed a model and found that control of dengue vector at larval stage lessens the dengue trouble up to two years while application of control strategies at adult stage minimised the burden for up to four years. Biological control is also significant in the control of Aedesaegypti. In this method, living organisms such as guppy fish or mesocyclops are used which feed on mosquito larvae. Some bacteria such as Bacillus thuringiensisisraelensis are also used for this purpose. A toxin is released from bacteria which kills the larvae after its ingestion.[13] There is no precise treatment of dengue infection.[13] Supportive therapy with the combination of fluid and analgesics-antipyretics is suggested.[47] Ahmed et al.[48] showed that aqueous extract of leaves of Carica papaya displays a strong effect against DF. There is still no effective vaccine available against dengue because of subsequent infections with enhanced severity caused by four serotypes.[49]

Discussion

Dengue is spreading globally, mainly in the developing and overpopulated countries. Dengue is now endemic in more than 125 countries. It has been estimated that about 40% of world population resides in an area presenting high risk for the transmission of DV. There is strong involvement of immunity in the production of disease resulting into high morbidity and mortality.[33] Initiatives which result into the decline of dengue outbursts to a great extent are needed. As there is no therapy and vaccine for the treatment of dengue, efforts should be made to introduce effective control strategies to check its global expansion. Good surveillance and reporting can support for better strategies in vector control. The production of effective vaccine and antiviral therapy is strongly needed for the global eradication of dengue epidemics.

Conclusion

Dengue is expanding geographically, especially in developing countries. In Pakistan, the incidence of DF has been extraordinary in metropolitan cities over the last couple of years.

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