Could inactivation of platelets be key to preventing thrombocytopenia in dengue virus infected patients?

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In recent years, dengue has been one of the most important mosquito-borne human viral diseases globally [1]. Although the disease is predominantly transmitted through the bite of *Aedes* mosquitoes carrying infectious dengue virus (DENV), recent evidence shows that other routes of introducing the virus to human beings are escalating and are likely to be underestimated [2]. Nevertheless, dynamic clinical presentations are the salient features in affected subjects, and numerous distinctive laboratory findings have been implicated in association with disease developments [3].

Wide clinical spectra of DENV infected people have been well documented, ranging from asymptomatic infection, mild dengue fever (DF) to potential life-threatening conditions, dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS), featured with plasma leakage likely from an increase in vascular permeability. In general, similar

to many febrile diseases induced by viruses, dengue is a time-dependent febrile illness. However, people often do not recall or notice when they were bitten by mosquitoes, but roughly recall the time of fever onset. Cumulative data reveals that the majority of DENV infected individuals seek professional help on the fourth day after the fever starts [4]. By which time, according to the reported human studies, the time lapse is about 10 days after the DENV being inoculated into human bodies by the bite of mosquitoes. Coincidentally, this time likely corresponds to the beginning of clinical investigations. Hence, there are many biomarkers being proposed as candidates for differential diagnosis of DF and DHF/DSS [5]. However, as of today, there are no consensus biomarkers available to differentiate DF from DHF. In addition, whether DHF is a consequence of the progression of DF remains to be further delineated

KEYWORDS

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and clarified. Despite there being many different hypotheses on the mechanisms of how DHF develops, the true etiological factors have not been identified.

Human platelets are small, anucleate, colorless blood cells, 2–4 μm in diameter containing granules, mitochondria and endoplasmic reticulum. Platelets are made in human bone marrow and travel through blood vessels to form clots to stop any bleeding that may happen if a blood vessel becomes damaged. Although the major role of platelets is maintaining hemostasis, they also play a significant role in many pathophysiologic processes, including thrombosis, hemorrhage, inflammation, pathogen host defense, tumor growth and metastasis [6]. In healthy human beings, the normal count of circulating platelets is approximately 150,000–450,000 platelets/ μl of blood and the turnover rate on average is 7–10 days. In general, when the platelet count drops to below 150,000 platelets/ μl in circulation, it is considered lower than normal and can be categorized as being a low platelet count, or so-called thrombocytopenia [6], and if a platelet count is below 50,000 platelets/ μl , which is a potential life threatening event, caused by internal bleeding, an emergency treatment may be required.

Hematological disorder is one of the most common findings in dengue patients [7], and yet its significant role, both biologically and clinically, in the pathogenesis of DHF/DSS remains largely unexplored. Thrombocytopenia is a salient laboratory finding in both DF and DHF patients [1]. Significantly, it has been shown that platelet counts correlate to the degree of severity of the disease development [8] and the nadir platelet count has been corresponded to the most critical period in dengue subjects [1]. As such, investigations on the mechanisms leading to low platelet counts in dengue patients has been the main focus for many years. As the platelet count in dengue patients are time sensitive events, the fever onset day primarily relies upon the recall of infected subjects; the dynamic change of platelet counts in infected individuals is the most challenging task to be tabulated. In addition, other underlying diseases or medicines taken by the affected individuals may contribute to the low platelet counts in patients. Nevertheless, two major mechanisms contributing to the thrombocytopenia have been proposed: impaired production and platelet destruction with or without dysfunction [9]. These result in several different scenarios, including direct infection of platelets by DENV, suppression of megakaryopoiesis in bone

marrow, reduction of megakaryocytes in bone marrow, inhibition of thrombopoiesis, shortening the life span, apoptosis, complement-mediated destruction and antibody-mediated phagocytosis by phagocytic cells [7,8,10–12].

There are at least three distinctive stages of the clinical course in patients with DHF/DSS: febrile, critical and convalescent. The progression of the platelet counts is the following; at normal range during the early febrile stage, at the lowest in the critical phase and rebounded to normal range during the convalescent period [7]. Importantly, the status of platelets is activated during the febrile stage, which has recently been demonstrated to be correlated with the severity of thrombocytopenia in dengue patients [8]. Moreover, the functionality of platelets during the febrile stage has been investigated and reveals that platelet dysfunction in the acute febrile stage are common [7]. Furthermore, parameters derived from platelets contributing to the vascular permeability have been reported recently [5]. This cumulative evidence indicates that activated platelets play a significant role in DHF/DSS. Researchers have, therefore, initiated the concept that therapeutic measures by inactivation of platelets might benefit dengue patients [8,13].

Biologically, inactivation of platelets may benefit the affected dengue patients by mitigating the progression to severe dengue. However, it has been demonstrated that platelet count, occasionally, does not correlate with the bleeding tendency in dengue patients [7], suggesting alternative factors may contribute to the DHF/DSS. Recent evidence suggests that platelets are part of the human immune system since the contents released in their active form play a role in modulating of the proper immune response in human hosts [14]. Interestingly, although abnormal immune response in dengue-infected subjects has been known for many decades, the causative mechanisms remain at large. This raises a scenario in which there are at least two additional and distinctive emerging functions of the platelets in dengue patients: mediating proper immune response in the host and assisting on the integrity of endothelial cells lining the blood vessel walls [15].

Scientifically, recent evidence demonstrates that platelets and their progenitor cells are likely to be infected by DENV in patients [8,16–19], suggesting that the timing of inactivated platelets occurs very early. Moreover, multifactorial factors engaging in the hemostasis of blood components and integrity, as well as functionality of endothelial cells, in dengue patients have been

well documented [5]. As such, therapeutic measures to inactivate platelets in dengue patients, to some extent, may have some benefits [20]. As a whole, in order to find a precise countermeasure to mitigate or improve the progression of clinical conditions, the identification of etiologic mechanisms leading to low platelet counts, in affected dengue subjects, should be further delineated.

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