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POTENTIAL OF FLAVONOIDS AS ENTRY INHIBITORS FOR DENGUE VIRUS TYPE-2 USING MOLECULAR DYNAMICS SIMULATION AND MOLECULAR DOCKING

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ABSTRACT

Dengue infections are currently estimated to be 390 million cases annually. Yet, there is no vaccine or specific therapy available. Envelope glycoprotein E (E protein) of DENV mediates viral attachment and entry into the host cells. Several flavonoids have been shown to inhibit HIV-1 and hepatitis C virus entry during the virus–host membrane fusion. Herein, we report the findings from protein modelling, molecular dynamics simulation and docking of the E-M heterodimer model of DENV type-2 for Malaysia (DENV2-My) in an attempt to suggest the entry inhibitor for DENV. Two main interactions discovered during molecular dynamics simulation of both DENV2-My and its template, DENV2-Thai, in both different conditions; normal and low pH conditions. Hydrophobic contacts were mainly observed between the N-terminal loop of M protein and the soluble ectodomain E protein in three different pockets. In addition, hydrogen bond interactions were discovered between the stem-anchor region of the E and M proteins. Both interactions diminished at low pH due to the increasing of distance between the E and M proteins triggered by the low pH. This reduction leads to the assumption that both interactions control the conformational change that responsible in the dissociation between E and M during the virus maturation and fusion. Molecular docking method was employed to predict the binding of nine flavonoids (baicalin, baicalein, EGCG, fisetin, glabranine, hyperoside, ladanein, quercetin and flavone) to the soluble domain E protein of both DENV2. Eight flavonoids were found to dock into the same binding pocket located between the domain I and domain II of different subunits of E protein. Consistent docking results were observed not only for both DENV2 but also for the E protein structures of tick-borne encephalitis virus and Japanese encephalitis virus. Apart from docking, molecular dynamics simulations were performed to further evaluate the interaction profile of the docked E protein-flavonoid complexes. Ile4, Gly5, Asp98, Gly100, and Val151 residues of DENV2-My are dominantly forming hydrogen bonds with baicalein, quercetin, and EGCG during the simulations. In computational studies, our proposed flavonoids binding pocket could be potentially used as a drug discovery target and the selected flavonoids confirm promising results to work as potential entry inhibitors for dengue infection. Further experimentation on the proposed flavonoids can result in the development of strong inhibitors.

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TABLE OF CONTENTS

CHAPTER ONE
INTRODUCTION

1.1 RESEARCH BACKGROUND

The increasing incidence of dengue virus (DENV) infections in most tropical and subtropical areas of the world, leads to 500,000 hospitalization and 20,000 death annually estimated by World Health Organization (WHO) ((WHO), 2012). Malaysia has been experiencing remarkable dengue outbreaks and ranks in the top ten countries with the highest dengue infection and death in the world.

Dengue virus serotype-2 (DENV2) is the most dominant in Malaysian dengue cases. Research from Infectious Diseases Research Centre (IDRC) in Institute for Medical Research Malaysia's (IMR) have found that DENV2 is more effective in multiplying itself causing a higher viral load in some individuals body. It has the capability to evade the immune system stronger than the other dengue serotypes and might cause dengue hemorrhagic fever (DHF) or dengue shock syndrome (DSS) which could lead to fetal if it is not detected at an early stage (Arumugam, 7 March 2016).

Dengue is a mosquito-borne disease transmitted to humans by *Aedes aqgypti* and*Aedes albopictus.* Dengue infection varies from asymptomatic infections to life-threatening DHF or DSS (Gould & Solomon, 2008; Guzman, 2003; Kuhn, 2002). Recovery from infection with one dengue serotype can give long lasting immunity against reinfection with that particular serotype, but not against the other serotypes. Subsequent infection by a different dengue serotype contributed to the risk of DHF. This phenomenon is known as an antibody-dependent enhancement (ADE) (Guzman et al., 2010; Halstead, 2007).

Envelope glycoprotein (E protein) and membrane protein (M protein) are two structural proteins in DENV polypeptide besides capsid protein (C). The E protein plays a vital role in viral entry by mediating the viral attachment to the host cell receptors and involve in the fusion process between the viral and the host membrane (Acosta, Castilla, & Damonte, 2008; Lindenbach, 2007). It is initially complexes with the precursor membrane protein (prM) forming non-infectious immature particles. The