Universitas Indonesia Library >> UI - Disertasi Membership

Potensi, mekanisme kerja dan toksisitas ekstrak etanol daun cassia alata linn terhadap virus dengue serotipe-2 in vitro, in vivo dan in silico = Potential, mechanism of actions and toxicity of ethanol extract of cassia alatas leaves againts dengue virus serotype-2 in vitro, in vivo and in silico

Marissa Angelina, author

Deskripsi Lengkap: https://lib.ui.ac.id/detail?id=20493437&lokasi=lokal

---

Abstrak

Latar Belakang: Infeksi virus dengue (DENV) masih endemis di Indonesia dan di banyak negara tropis. Hingga saat ini belum ada antivirus terhadap DENV. Penelitian ini bertujuan untuk mendapatkan antivirus dari tanaman Cassia alata Linn (CA) terhadap DENV-2 secara in vitro, in vivo, dan in silico.
Metode: Penelitian ini dilakukan dilakukan di Laboratorium LIPI dan Departemen Mikrobiologi FKUI, 2017-2019. Penelitian in vitro menggunakan DENV serotipe 2 strain New Guinea C (NGC) dan sel Huh 7it-1. DENV diberi perlakuan ekstrak CA dan fraksi dan senyawa murni hasil isolasi dengan bebagai dan konsentrasi untuk menentukan nilai IC₅₀ dan CC₅₀. Penentuan nilai IC₅₀ dan CC₅₀melalui uji fokus dan MTT secara berurutan. Selanjutnya dilakukan percobaan untuk menentukan mekanisme penghambatan pada tahapan reseptor, pre, post dan pre/post infeksi dari ektrak CA dan fraksinya. Uji efikasi ekstrak CA in vivo dilakukan pada model mencit Balb/c dengan melakukan pengukuran titer virus dengue, jumlah trombosit, leukosit, IL-6 dan IL-10 yang dilanjutkan dengan uji toksisitas akut ekstrak CA. Dilakukan juga uji in silico untuk mengetahui interaksi antara senyawa dengan protein DENVmenggunakan software Autodock 1.5.6.
Hasil: Uji in vitro menunjukkan nilai IC₅₀ ekstrak CA, fraksi heksan, etil asetat, butanol, dan air berturut-turut adalah 0,026; 0,004; 0,0013; 4,6; dan 2,5 mg/ml dengan nilai CC₅₀berturut-turut adalah 208,9; 47,46; 57,2; 753,8; 311,33 mg/ml. Hasil uji mekanisme pada dosis 10 mg/ml, ekstrak CA, fraksi heksan dan etil asetat menunjukkan hambatan pada tahapan reseptor, pre, post dan pre/post infeksi yang lebih baik dibandingkan fraksi butanol dan etil asetat. Ekstrak CA dapat menghambat keempat mekanisme di atas dengan nilai >95%. Fraksi heksan dan etil asetat menghambat 100% pada post dan pre/post infeksi. Hasil uji in vivo dengan pemberian ekstrak 1 hari setelah infeksi menunjukkan bahwa ekstrak CA dosis 0,2; 0,4; 1 g/kg bb menurunkan titer virus DENV-2 dan menaikkan hitung trombosit secara bermakna dibandingkan dengan kelompok DENV-2 tanpa ekstrak . Ekstrak CA tidak memberikan efek terhadap jumlah leukosit dan kadar sitokin IL-6 dan IL-10. LD₅₀ semu ekstrak CA > 15 g/kg bb. Aloe-emodin diisolasi dari ekstrak CA dengan metode kolom kromaografi. IC₅₀ senyawa kaempferol, emodin dan aloe-emodin terhadap DENV-2 berturut-turut adalah 22,24; 42,47; 7,51 mg/ml, dan CC₅₀ terhadap Huh7-it 1 berturut-turut 68,28; 74,19; 68,28 mg/ml. Uji in silico ketiga senyawa menunjukkan bahwa mekanisme penghambatan ekstrak CA yang paling stabil adalah terhadap protein NS5 (IL9K4) dimana diperoleh tingkat energi bebas (DG) terendah.
Kesimpulan: Ekstrak CA menghambat DENV-2 secara in vitro dan in

vivo. Selain menurunkan titer virus dengue, ekstrak CA juga meningkatkan hitung trombosit, dengan mekanisme penghambatan in vitro >95% pada tahap pre, post, pre/post dan reseptor. Mekanisme penghambatan fraksi heksan dan EA terbaik pada post dan pre/post infeksi sebesar 100%. Ikatan paling stabil senyawa yang terdapat didalam ekstrak CA adalah ikatan dengan protein NS5.

<hr />

Background: Dengue virus infection (DENV) is still endemic in Indonesia and in many tropical countries. Until now there is no anti viral available against dengue virus. This study aimed to investigate the antiviral effects of Cassia alata Linn (CA) leaves on DENV in vitro, in vivo, and in silico.
Methods: This research was carried out at Laboratories of LIPI, Department of Microbiology FMUI, 2017-2019. In vitro tests of CA extract,fractions and isolated compound were carried out to determine the IC₅₀, CC₅₀ and the inhibition mechanism at receptor, pre, post and pre/post infection stages. In vivo efficacy of CA extract was tested in mice Balb/c model. Dengue virus titers, platelet, leukocytes and IL-6 and IL-10 in bloods were measured. Acute oral toxicity test was carried out to determine the LD₅₀ of CA extract. Isolation of compounds was carried out from CA extract. In silico test was carried out to know interaction test compound with DENV protein using Autodock 1.5.6 software.
Results: The results of the in vitro test showed that the IC₅₀ of CA extract, hexane, ethyl acetate, butanol, and water fraction against DENV-2 were 0.026; 0,004; 0.0013; 4.6; and 2.5 mg/ml and the CC₅₀ to Huh 7 it-1 were 208.9; 47.46; 57.2; 753,8; 311.33 mg/ ml, respectively. The results of the mechanism study showed that at a dose of 10 mg/ml, CA extract, the hexane and ethyl acetate fractions inhibited DENV-2 at the receptor stage, pre, post and pre/post infection which were better than the butanol and ethyl acetate fractions. CA extract inhibited the four mechanisms above by more than 95%. Hexane and ethyl acetate fractions inhibited DENV-2 100% at post and pre-post infection stages. In vivo test showed that the administration of CA extract at doses of 0.2; 0.4; 1 g/kg bw 1 day after DENV-2 infection significantly reduced virus titers and increased platelet counts compared to DENV-2 infected group only. CA extract did not affect the number of leukocytes and cytokines of IL-6 and IL-10 back to normal which had been altered in the DENV-2 group. LD₅₀ of CA extract was more than 15 g/kg bw. Aloe-emodin was isolated from CA extract used column chromatography. The IC₅₀ of kaempferol, emodin and aloe emodin to Huh7-it 1, respectively were 22.24; 42,47; 7.51 mg ml, and the CC₅₀respectively were 68.28; 74,19; 68.28 mg/ml. In silico study of the three compounds showed that the most stable inhibition mechanism of CA extract was on protein NS5 (IL9K4) which had the lowest free energy (DG) level.
Conclusion: CA extracts have high inhibitory activity against DENV-2 in vitro and in vivo. In addition to reducing dengue virus titers, CA extract also increased platelet count, with in vitro inhibition mechanism >95% at the pre, post, pre/post and receptor stages. Hexane and ethyl acetate fractions inhibit DENV-2 100% at post and pre/post infections. The most stable bond of the compounds contained in CA extract is the bond with NS5 protein.