

Efikasi Fermentasi Buah Berenuk (*Crescentia cujete* L.) terhadap Neurodefisit, Gambaran Histopatologis dan Imuno-Ekspresi IL-6 pada Otak Tikus *Sprague Dawley* Model *Ischemic Stroke*

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ABSTRAK

Stroke adalah disfungsi neuronal akibat gangguan sirkulasi darah ke otak. Pengobatan stroke dengan antikoagulasi cenderung menimbulkan efek samping. Oleh karena itu dibutuhkan terapi alternatif, salah satunya buah berenuk (*Crescentia cujete* L.). Penelitian ini menggunakan 25 ekor tikus *Sprague Dawley* sebagai model pasca stroke. Tikus dibagi dalam 5 perlakuan, sebagai berikut: P1 = kelompok kontrol positif, P2 = kelompok induksi stroke (IS) tanpa terapi, P3 = kelompok IS + fermentasi buah berenuk (FBB) dosis 0,74 mg/kg BB, P4 = kelompok IS + FBB dosis 1,28 mg/kg BB, dan P5 = kelompok IS + 2,96 mg/kg BB. Buah berenuk difermentasi selama 30 hari dan kandungan *choline* dianalisis dengan *liquid chromatography mass spectrometry* (LC-MS/MS). Hewan diinduksi stroke selama 4 jam dengan ligasi arteri karotis dan diterapi selama 7 hari. Tikus dianalisis skor neurodefisitnya dan dieuthanasia di hari ke-7. Cerebrum diambil dan dianalisis area infark menggunakan *tetrazolium chloride* 1%, pewarnaan histopatologi rutin, dan imunohistokimia *interleukin-6* (IL-6). Data pewarnaan H&E, badan nissl's dan imunohistokimia dianalisis secara semikuantitatif dengan skoring 1-4. Pengambilan foto histopat dilakukan sebagai data pendukung. Foto histopat di analisa secara kuantitatif menggunakan bantuan aplikasi *imageJ*®. Analisa statistik dengan ANOVA dan dilanjutkan dengan *post hoc test*. Taraf kepercayaan yang digunakan sebesar 95%. Hasil penelitian memperlihatkan bahwa buah berenuk mampu memperbaiki skor neurodefisit pada tikus model *ischemic stroke* ($p \leq 0.05$). Fermentasi buah berenuk mampu menurunkan persentase area infark secara makroskopis ($p \leq 0.05$). Pemeriksaan histopatologi dan imunohistokimia memperlihatkan bahwa fermentasi buah berenuk dosis 2,96 mg/kg BB menurunkan skor nekrosis, mikrogliosis, edema neuronal, hemoragi sekunder dan edema perivaskular ($p \leq 0.05$), memperbaiki badan Nissl's ($p \leq 0.05$) dan peningkatan imunoekspresi *IL-6* pada neuron ($p \leq 0.05$) yang lebih baik jika dibandingkan kelompok P2, P3, dan P4. Dosis fermentasi buah berenuk dengan efektifitas terbaik ditunjukkan pada kelompok dosis 2,96 mg/kg BB. Penelitian ini dapat disimpulkan bahwa buah berenuk memiliki potensi sebagai agen terapi alternatif *ischemic stroke* pada tikus *Sprague Dawley* model *ischemic stroke*.

Kata kunci : buah berenuk, *choline*, IL-6, model *ischemic stroke*, neurodefisit, otak.

Efficacy of Berenuk Fruit Fermentation (*Crescentia cujete* L.) on the Neurodeficits, Histopathology, and Immune-Expression of IL-6 on the Brains of Rat *Sprague Dawley* Model of *Ischemic Stroke*

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ABSTRACT

Stroke is neuronal dysfunction resulting from impaired blood circulation to the brain. Stroke treatment with anticoagulants tends to cause side effects. Therefore, alternative therapies are needed, one of which is berenuk fruit (*Crescentia cujete* L.). This study used 25 Sprague Dawley rats as a post-stroke model. Rats were divided into 5 treatments, as follows: P1 = positive control group, P2 = stroke induction (IS) group without therapy, P3 = IS + fermented berenuk fruit (FBB) group at a dose of 0.74 mg/kg BW, P4 = IS group + FBB dose of 1.28 mg/kg BW, and P5 = IS group + 2.96 mg/kg BW. Berenuk fruit was fermented for 30 days and the choline content was analyzed using liquid chromatography mass spectrometry (LC-MS/MS). Animals were induced to stroke for 4 hours with carotid artery ligation and treated for 7 days. Mice were analyzed for neurodeficit scores and euthanized on day 7. The cerebrum was removed and analyzed for the infarct area using 1% tetrazolium chloride, routine histopathological staining, and interleukin-6 (IL-6) immunohistochemistry. H&E staining data, Nissl's bodies and immunohistochemistry were analyzed semiquantitatively with scoring 1-4. Histopath photos were taken as supporting data. Histopath photos were analyzed quantitatively using the ImageJ® application. Statistical analysis with SPSS we use ANOVA test and continued with the post hoc test. The confidence level used was 95%. The results of the study showed that berenuk fruit was able to improve neurodeficit scores in mice with ischemic stroke models ($p \leq 0.05$). Berenuk fruit fermentation was able to reduce the percentage of infarct area macroscopically ($p \leq 0.05$). Histopathological and immunohistochemical examination showed that fermented berenuk fruit at a dose of 2.96 mg/kg BW reduced necrosis scores, microgliosis, neuronal edema, secondary hemorrhage and perivascular edema ($p \leq 0.05$), improved Nissl's bodies ($p \leq 0.05$) and increased IL-immunoexpression. 6 on neurons ($p \leq 0.05$) which was better when compared to groups P2, P3, and P4. The fermented berenuk fruit dose with the best effectiveness was shown in the 2.96 mg/kg BW dose group. This research can be concluded that berenuk fruit has efficacy as an alternative therapeutic agent for ischemic stroke in the Sprague Dawley rat model of ischemic stroke.

Keyword : Calabash fruit, choline, IL-6, model ischemic stroke, neurodeficit, brain.