

Aktivitas anti agregasi trombosit ekstrak kering phyllanthus niruri pada orang sehat = Anti platelet aggregation activity of dry extract phyllanthus niruri on healthy human subjects

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Abstrak

Penelitian ini dilakukan dengan tujuan untuk mengetahui apakah ekstrak kering P. niruri mempunyai efek menghambat agregasi trombosit secara in vitro dan in vivo pada orang sehat. Parameter penilaian adalah adanya perubahan nilai agregasi trombosit (% maksimal) dan besar hambatan (%). Sebelum penelitian dilakukan uji validasi yang meliputi uji ketelitian (within-run), penentuan bahan pelarut, uji validasi metoda in vitro, penetapan kadar P. niruri, penentuan waktu inkubasi, dan variasi pemeriksaan agregasi trombosit hari ke hari (intra individu). Pemeriksaan agregasi trombosit dilakukan dengan menggunakan aggregator Adenosin difosfat (ADP) dengan kadar akhir Id Kmolll dan alat Platelet Aggregation Chromogenic Kinetic System-4 (PACKS-4). Prinsip pemeriksaan menggunakan alat tersebut yaitu mengukur persentase perubahan intensitas transmisi cahaya yang dapat melewati plasma (PRP) setelah terjadinya agregasi trombosit. Pada penelitian in vitro, dilakukan inkubasi platelets-rich plasma (PRP) dengan larutan ekstrak kering P. niruri dalam 3 kadar selama 5 menit. Air suling ditentukan sebagai pelarut dan kadar ekstrak keying P. niruri ditetapkan 1.5, 3, dan 6 mg/mi. Pada studi in vivo digunakan desain penelitian paralel menyilang, tersamar ganda, acak dengan pembanding plasebo. Pada minggu pertama diberikan ekstrak kering P. niruri dengan dosis 300 mg atau piasebo, diminum sekali sehari selama 7 hari berturut-turut. Setelah periode bebas obat 14 hari, diberikan perlakuan sebaliknya dari perlakuan pertama. Pengukuran agregasi trambosit dilakukan pada awal dan akhir masa minum obat, kemudian ditentukan perubahannya. Penelitian ini mengikutsertakan 5 sukarelawan sehat untuk penelitian in vitro dan 16 orang sukarefawan sehat untuk in vivo. Subyek penelitian tidak minum obat selama 2 minggu terakhir. Analisis statistik penelitian in vitro dilakukan dengan menggunakan ANOVA satu arah, dan untuk in vivo digunakan paired Nest. Perbedaan dianggap bermakna bila diperoleh nilai $p < 0.05$.

Dari hasil uji ketelitian didapatkan nilai koefisien variasi 1,35 %. Dari penelitian in vitro didapatkan besar hambatan (%) agregasi trombosit oleh P. niruri pada kadar larutan P. niruri 0, 1.5, 3, dan 6 mg/ml berturut-turut adalah 0, 0, 3 dan 14% ($p = 0.33$). Dari penelitian in vivo didapatkan rerata nilai agregasi maksimal sebelum pemberian P. niruri 89.9% dan sesudah pemberian P. niruri adalah 86.9%, dengan nilai hambatan oleh P. niruri sebesar 3%. Rerata (Li- SD) perbedaan nilai agregasi maksimal sebelum dan sesudah pemberian plasebo serta sebelum dan sesudah pemberian P. niruri masing-masing adalah 4.6 ($\pm 17.3\%$) dan 2.9 ($\pm 7.5\%$) ($p = 0.194$). Selama penelitian in vivo dicatat adanya keluhan : pusing 4 orang, mengantuk 1 orang dan sering buang air kecil 1 orang pada subyek yang mendapat ekstrak kering P. niruri, serta 1 orang pusing dan 1 orang mengantuk pada subyek yang mendapat piasebo. Dari penelitian ini disimpulkan bahwa pada penelitian in vivo, larutan ekstrak kering P. niruri dalam air sampai dengan kadar 6 mg/ml tidak mempunyai efek menghambat terhadap agregasi trombosit PRP orang sehat, dan pada penelitian in vivo ekstrak kering P. niruri 300 mg, sekali sehari, yang diberikan selama 7 hari berturut-turut juga tidak mempunyai efek hambatan terhadap agregasi trombosit orang sehat.

<hr><i>This research was conducted to find out whether dry extract of P. Niruri was effective to inhibit platelet aggregation, in vitro and in vivo, on healthy human subjects. Parameter of examination was a change in value (% maximal) of platelet aggregation and extends of inhibition (%). For preliminary study, several validation tests were conducted which included precision test (within-run), determining the solvent material, validation test of in vitro method, determining the concentration of P. niruri, determining the incubation time and daily variation of examination of the platelet aggregation (intra-individual). Examination of the platelet aggregation was conducted by using Adenosin Diphosphate (ADP) as aggregating agent with final concentration 10 pmol/l and equipment Platelet Aggregation Chromogenic Kinetic System-4 (PACKS-4). The principle of the test is to measure the percentage of change of the intensity of the light that is able to pass through the platelets-rich plasma (PRP) after the occurrence of the platelet aggregation. On in vitro research PRP was conducted using the solution of dry extract P. niruri in 3 different concentrations for 5 minutes. Distilled water was chosen as the solvent for P. niruri extract and the concentrations prepared were 1.5, 3, and 6 mg/ml. On in vivo study, a randomized, crossover, parallel, double-blind, and placebo-controlled design was applied. in first week, 300 mg of P. niruri extract or placebo was given daily to the subjects for 7 days. After 14 days of wash out period, the procedure was repeated by giving alternative agent to the subjects. Measurement of platelet aggregation was done at the beginning and at the end of the each treatment period. This research involved 5 and 16 normal human subjects, for in vitro and in vivo studies, respectively and not on any drugs therapy for the last two weeks. Statistical test for in vitro study was ANOVA one way, and paired t-test was used for in vivo study. A difference was considered significant if p value < 0.05.

The coefficient of variation for the recovery test was 1.35%. In in vitro study, inhibition of platelet aggregation (%) by P. niruri of 0, 1.5, 3, and 6 mg/ml were 0, 0, 3 and 14%, respectively ($p = 0.33$). From in vivo study, mean maximum aggregation value before and after giving P. niruri were 89.9% and 86.9%, respectively, therefore the inhibition by P. niruri was 3%. Mean (SD) change of maximum aggregation value before and after giving placebo and before and after giving P. niruri were $4.6(\pm 17.3\%)$ and $2.9(\pm 7.5\%)$ ($p = 0.194$). During in vivo study, several adverse events in subjects given dry extract P. niruri were recorded: 4 persons had headache, 1 had drowsiness and 1 had frequent urination, while in subjects given placebo: 2 persons had headache and 1 had drowsiness. From this in vitro study, it is concluded that up to the concentration of 6 mg/ml, dry extract P. niruri solvent in water does not inhibit platelet aggregation activity and in in vivo study, dry extract P. niruri 300 mg, once a day for 7 days continuously, also does not affect platelet aggregation activity in healthy human subjects.</i>