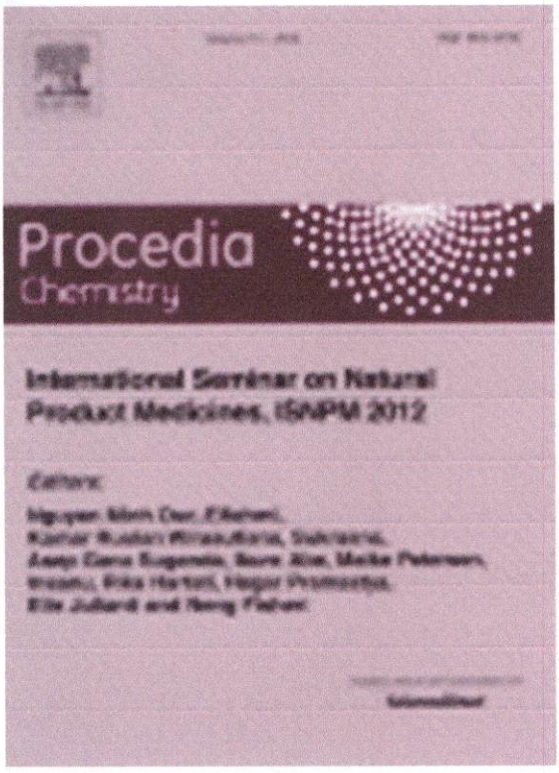
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Editorial: Current Issues on Future Researches and Applications of Natural Product Medicines ☆

Elfahmi [a, b]

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Abstract

The use of natural product medicine has emerged from traditional to modern therapy in order to increase the quality of health worldwide. To prove pharmacological effects of medicinal plants and to further develop the rational use of herbal medicines, scientific approaches are essential. The development of sciences and technologies have highly supported the research on natural product medicines in all aspects. Recent findings from research in natural product medicines were a major focus in the International Seminar on Natural Product Medicines 2012 which was held in Bandung and organized by School of Pharmacy, Bandung Institute of Technology (ITB) and Indonesian Society on Natural Product Researchers (PERHIPBA). Furthermore, there was an interesting sharing of experiences and knowledge on how to develop and practice some well-known traditional medicines such as Traditional Chinese Medicine (TCM), ayurveda and jamu

Keywords

ISNPM 2012; Natural Products Medicines; Bandung; Perhipba

* Peer-review under responsibility of the School of Pharmacy, Bandung Institute of Technology.

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Editorial *Open access*

Editorial: Current Issues on Future Researches and Applications of Natural Product Medicines

Elfahmi

Pages 1-2

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Abstract

Pharmaceutically active compounds risperidone has difficulty to be packaged into nanocarrier due to its positive charge. In this study, we encapsulated risperidone into chitosan-based nanocarrier via ionic interaction. The nanocarrier was consisted of chitosan:sodium TPP = 5.72:1 and prepared by ionic gelation method. Encapsulation efficiency was enhanced by modification of pH of chitosan, sodium tripolyphosphate (TPP), and the concentration as well as the surface charge of risperidone. The later was done by addition of SDS (Sodium Dodecyl Sulphate) minimum 0.005% (w/v). The particle size was in the range of 300-400 nm. Encapsulation efficiency of risperidone was reached up to 25.62% with addition of 0.050% SDS. Encapsulation of risperidone into chitosan-TPP nanocarrier was increased by modification of risperidone's charge with low concentration of SDS. Further amount of SDS higher than 0.1% increased the particle size to microparticles. Chitosan-based type nanocarrier was preferable for encapsulating anionic

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In vivo Antimalarial Activity of *Andrographis Paniculata* Tablets

Aty Widyawaruyanti, Muhammad Asrory, Wiwied Ekasari, Dwi Setiawan, ... Achmad Fuad Hafid

Pages 101-104

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Abstract

Abstract

The formulation of three phytopharmaceutical products of *Andrographispaniculata* fractions (AP fraction A and B) containing diterpene lactones as an active substance were developed and their antimalarial activities against *Plasmodium berghei*has been examined. *In vivo* antimalarial assay on *P. berghei* infected mice was carried out by oral administration,twice a dayfor four consecutive days of the AP fractions product, which were Tablet I: wet granulated formula of AP fraction A; Tablet II: wet granulated formula of AP fraction B; Tablet III: solid dispersion formula of AP fraction B.. The results revealed that three phytopharmaceutical products of *A.paniculata* were inhibited parasite's growth with inhibition range of 70.15% to 80.35%. There was no significant difference of antimalarial activities between Tablet II and III, meanwhile there was significant difference among Tablet I with Tablet II and Tablet III.It was concluded that antimalarial activity depending on raw material form of *A. paniculata* active substance

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Development of Immunonutrient from Pumpkin (*Cucurbita Moschata* Duchense Ex. Lamk.) Seed

Maria Immaculata Iwo, Muhamad Insanu, Caryn Anne Santhana Dass

Pages 105-111

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Abstract

Abstract

Pumpkin seeds has long been used as a source of nutrition. Based on its content, it is possible that part of this plant used as immunonutrient. The purpose of this research is to study the immunomodulatory activity of pumpkin seeds. It effects on the non-specific immune response was determined through carbon clearance test, organ indexes (liver, spleen and thymic gland) and mice peritoneal exudate activity (PEA). The effects on specific immune response were

determined through total antibody titre, delayed type of hypersensitivity (DTH) reaction and number of lymphocytes. On non specific immune response test, pumpkin seed at a low dose (PLD-3.8 g/kg bw) and at a high dose (PHD-7.6 g/kg bw) showed immunostimulanting effect with phagocytic index of 1.219 and 1.347, respectively. PHD increased PEA activity ($p<0.01$) in lysing microbes. On specific immune response test, PLD and PHD showed

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Analysis of Secondary Metabolite Production in Somatic Embryo of Pasak Bumi (*Eurycoma Longifolia* Jack.)

Iriawati, Andira Rahmawati, Rizkita R. Esyanti

Pages 112-118

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Abstract

Abstract

Pasak bumi (*Eurycoma longifolia* Jack.) has been known as a plants that can produce secondary metabolites for medicinal purposes such as: aphrosidiac, antimalaria, dysentri, antitumor, etc. Poor seed germination of pasak bumi will affect the avaibility of plant material for drug extraction. Over exploitation of this plant will also reduce plant population in its natural habitat. In vitro culture, i.e. through somatic embryogenesis, therefore, can be used as one of an alternative method for plant regeneration as well as for in vitro metabolite production. Based on this reason, the research has been done with an objective to analyze the presence of secondary metabolite in somatic embryo of pasak bumi. Seed-derived callus was used as an explant. This callus was maintained to proliferate in MS (Murashige&Skoog, 1962) medium supplemented with 2.25 mg/L 2,4-D and 2.0 mg/L kinetin. A half gram of callus from proliferation medium was transferred into the MS liquid medium containing 1.0 or 2.25 mg/L 2,4-D and 2.0 mg/L BAP or 2.0 mg/L kinetin

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Influence of β-cyclodextrin on Cefixime Stability in Liquid Suspension Dosage Form

Jessie Sofia Pamudji, Rachmat Mauludin, Nurhabibah

Pages 119-127

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Abstract

Abstract

Cefixime is a third generation of cefalosporin antibiotic which is easily to hydrolize in water and usually prepared in the form of tablet and dry suspension dosage form. Formation of active ingredient in the form of inclusion complex was one of the methods which can be employed to increase its stability to hydrolytic degradation.The purpose of this study was to evaluate the stability of cefixime-β-cyclodextrin inclusion complex in liquid suspension dosage form. The Inclusion complex of cefixime with β-cyclodextrin were prepared by utilizing of kneading and freeze drying method with weight ratio of 1:1, 1:2 and 1:3. The Inclusion complex formation were confirmed by infrared spectroscopy (FTIR), powder X-ray difractometer (PXRD) and *Scanning Electron Microscopy* (SEM). The formulation of cefixime liquid suspensions were prepared in the citrate buffer pH of 3.5 for pure cefixime, physical mixture of cefixime with β-cyclodextrin and inclusion complex from each method. Further studies werestability tested for each formulae of

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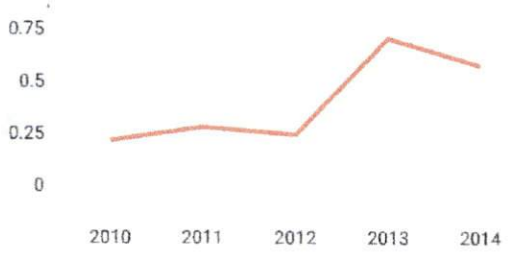

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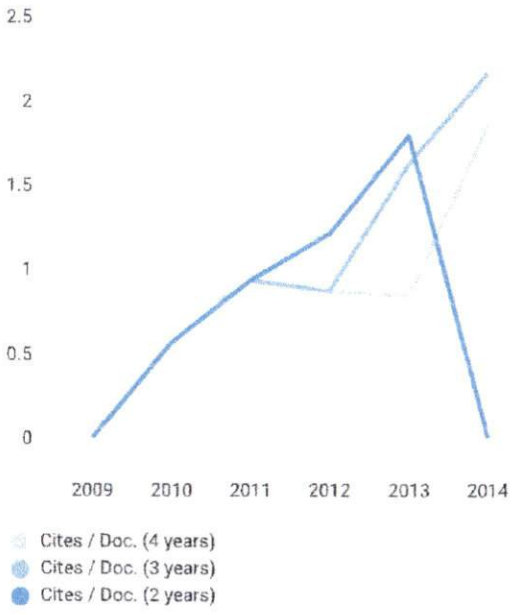

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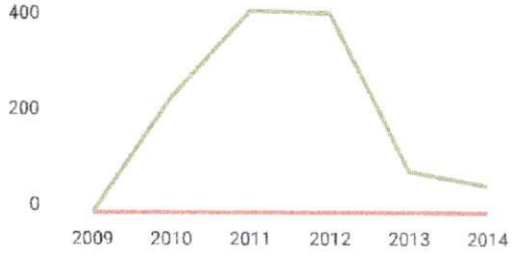

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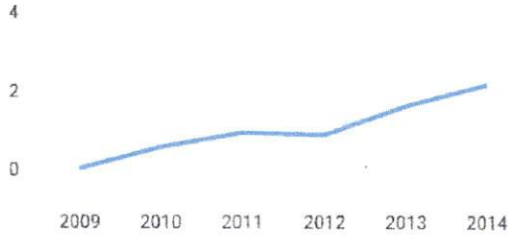

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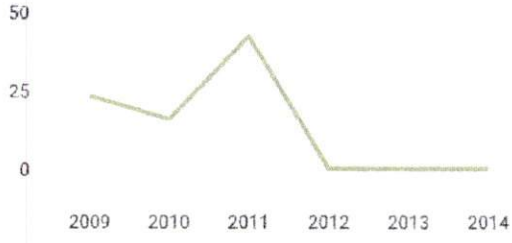

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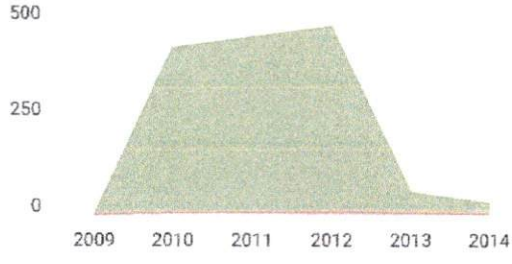

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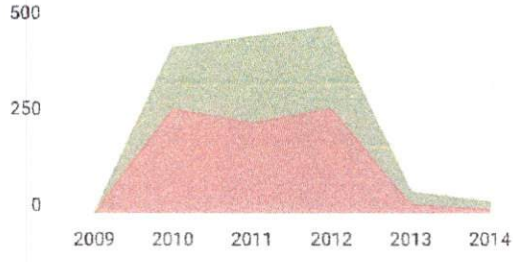

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In vivo Antimalarial Activity of *Andrographis paniculata* Tablets

Aty Widyawaruyanti[a,b]*, Muhammad Asrory[a,b], Wiwied Ekasari[a], Dwi Setiawan[a], Achmad Radjaram[a], Lidya Tumewu[b], Achmad Fuad Hafid[a,b]

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[b]*Institute of Tropical Disease, Universitas Airlangga, Surabaya 60115, Indonesia*

Abstract

The formulation of three phytopharmaceutical products of *Andrographispaniculata* fractions (AP fraction A and B) containing diterpene lactones as an active substance were developed and their antimalarial activities against *Plasmodium berghei*has been examined. *In vivo* antimalarial assay on *P. berghei* infected mice was carried out by oral administration,twice a dayfor four consecutive days of the AP fractions product, which were Tablet I : wet granulated formula of AP fraction A; Tablet II : wet granulated formula of AP fraction B; Tablet III : solid dispersion formula of AP fraction B.. The results revealed that three phytopharmaceutical products of *A.paniculata* were inhibited parasite's growth with inhibition range of 70.15% to 80.35%. There was no significant difference of antimalarial activities between Tablet II and III, meanwhile there was significant difference among Tablet I with Tablet II and Tablet III.It was concluded that antimalarial activity depending on raw material form of *A. paniculata* active substance.

Peer-review under responsibility of the School of Pharmacy, Bandung Institute of Technology

Keywords:Andrographis paniculata Ness;tablets;*in vivo* antimalarial activity;*Plasmodium berghei;* inhibition of parasite's growth

1. Introduction

Malaria was one of the oldest recorded diseases in the world. Each year from 300 to 500 million new cases were diagnosed and approximately 1.5 million people diedbecause the disease; majority were children[1]. The re-emerging of malaria in many parts of the world was due to the rapid increasing of resistance to most of the available antimalarial drugs, as well as resistance of vectors to insecticides[2,3]. Drug resistant for strains of *Plasmodium. falciparum* has been found in many endemic areas of the world and many of conventional anti-malarial drugs have

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been associated with treatment failure. Furthermore, the difficulty of creating efficient vaccines and also adverse side-effects of the existing antimalarial drugs highlight the urgent need for novel and well-tolerated antimalarial drugs[2] for both prophylaxis and treatment of malaria.

Andrographis paniculata Nees which known as sambiloto was traditionally used for antimalaria in Indonesia. The previous research had shown that *A. paniculata* extract have antimalarial activity both in vitro and in vivo[4,5]. Methanol extract of *A. paniculata* exhibited antimalarial activity against *P. falciparum* with IC_{50} value of 7.2 µg/ml. Futhermore, the results of in vivo toxicity assay indicating no toxicity associated with the use of this extract in mice system[5]. Andrographolide was the main content of sambiloto which reported have antimalarial activity. Andrographolide has IC_{50} value of 9.1 µM[6]. Andrographolide was considered to be active compound and has good antimalarial activity based on its IC_{50} value which was less than 20 µM[7]. Our previous research also found that andrographolide shown antiplasmodial activity as blood schizontocidal activity and also gametocytocidal with IC_{50} value of 12.16 and 3.61 µg/ml, respectively[8].

Regarding to the potential antimalarial activity of *A. paniculata*, Widyawaruyanti et al[9] was developed phytopharmaceutical products by conducted formulation study of two fractions from ethanol extract of this plant which were AP fraction A and B. AP fraction A was obtained from fractionated ethanol extract using ethyl acetate, while further purification of AP fraction A resulted as AP fraction B. Although AP fractions have different physicochemical properties, both of them were contain andrographolide (diterpene lactone compound) as an active substance. The formulation study of AP fractions A and B was performed based on two methods which were wet granulation and solid dispersion, therefore three phytopharmaceutical products were obtained which were Tablet I: wet granulated formula of AP fraction A; Tablet II: wet granulated formula of AP fraction B; and Tablet III: solid dispersion formula of AP fraction B.

The aim of this study was to observe in vivo antimalarial activities of Tablet I, II and III based on The Peter's test (The 4-days suppressive test) by oral administration of products at a dose of 12.55 mg andrographolide/kg mice body weight, twice a day for four consecutive days..

2. Experiments

2.1. Material

A. paniculata dry herbs powder was obtained from pharmaceutical company PT. Kimia Farma Tbk, Bandung, Indonesia. *A. paniculata* was extracted by maceration method using ethanol 96% as a solvent. Ethanol extract of *A. paniculata*was further fractionated by liquid-liquid fractionation method using ethyl acetate and water to obtain AP fraction A. Dried AP fraction A was further purified so that AP fraction B was obtained. AP fraction A was dark green and sticky, meanwhile AP fraction B was greenish amorphous powder.

Tablet I contained of 75 mg of AP fraction A per 400 mg tabletformulated by wet granulation method. Tablet II contained of 15 mg of AP fraction B per 300 mg tabletformulated by wet granulation method. Tablet III contained of 60 mg of AP fraction B per 150 mg tablet formulated by solid dispersion method.

The chloroquine sensitive *Plasmodium berghei* ANKA strain was obtained from Institute of Biomolecular Eijkmann, Jakarta and maintained on mice at Malaria Laboratorium, Faculty of Pharmacy, Universitas Airlangga. The inoculum of *P. berghei* parasitized erythrocytes was prepared by determining the percentage parasitemia of mice donor and diluting the blood with alceivers solutions in proportion indicated by the determination. The mice were infected intraperitonially with 200 µL of 5% parasite blood stock from frozen deposits. Once the percent of parasitemiaon this donor mice reached 20%, the mice blood taken intracardially and diluted with PBS or alceivers solutions (1:3) up to 5% parasitemia. The test mice has been infected by 200 µL of this diluted parasitized blood that contain 5% parasite.

Male albino Swiss mice Balb-C strain (20-30 g weight) were obtained from Animal Laboratory, Faculty of Pharmacy, Universitas Airlangga. They were maintained on standard animal pellets and water ad libitum. Permission and approval for animal studies were obtained from Faculty of Veterinary Medicine, Universitas Airlangga.

2.2. Methods

2.2.1. Preparation of samples

Tablet I, Tablet II and Tablet IIIof which were equal to 12.55mg andrographolide/kg mice body weight were powdered and suspended in 0.5% CMCNa solution. Sample solutions were administered per oral (1 ml/mouse) twice a day.

2.2.2. In vivo antimalarial assay

Antimalarial activity of Tablet I, II and IIIwere evaluated using the Peter's test methods (The 4-days suppressive test). Each mouse was inoculated intraperitoneally on the first day (day 0) with 0.2 ml of infected blood containing *P. berghei* parasitized erythrocytes (5% parasitemia). The animals were divided into four groups of five mice each and administered orally with suspension of Tablet I, Tablet II and Tablet III containing andrographolide 12.55 mg/kg body weight twice a day and an equivalent volume of CMC-Na solutions (as negative control) at one day after inoculation, for four consecutive days (day 0 to day 3). On the fifth day (day 4), thin films were made from the tail blood of each mouse and the parasitemia level was determined by counting the number of parasitized erythrocytes out of 1000 erythrocytes in random fields of the microscope. Average percentage parasite's inhibition was calculated as following:

$$100\% - \left(\frac{Xe}{Xk} \, x \, 100\%\right)$$

Xe: % parasitemia growth of experimental group
Xk: % parasitemia growth of negative control

3. Results and Discussion

Tablet I, II and III produced inhibition effect at a dose employed in this study. The parasite's inhibition were 70.35%, 78.16% and 80.35% for sample equal to andrographolide 20.10 mg/kg/day dose of Tablet I, II and III respectively. The inhibition produced by the products were significant ($P< 0.05$) compared to control (Table 1).

Tabel 1.AntiplasmodialActiviry of Tablet I, II, and III during 4-days Test

Sample	Active substance	Andrographolide dose (mg/kg bw/day)	Average parasitemia (%)	Average inhibition (%)
Tablet I	AP fraction A	25.10	3.37±0.29	70.15
Tablet II	AP fraction B	25.10	2.85±0.48	78.16
Tablet III	AP fraction B	25.10	2.96±0.41	80.35
Negative control	-	-	10.85±1.54	-

Data were expressed as mean±SD for five animals per group F=53.789

*P<0.001 compared to control

The data was analyzed by one-way analysis of variance (one way ANOVA) at 95% confidence limit (α =0.05) to detect the significant differences ininhibition of parasite's growth of each test group. ANOVA and Post Hoc LSD (Least Significant Difference) test resulted the calculating F(53.789) and P <(0.001). It concluded that there were significant difference among Tablet I and Tablet II, Tablet I and Tablet III, but no significant difference between Tablet II and Tablet III. Tablet I and II displayed different antimalarial activity, they were formulated by the same

method, but different in active substance. Meanwhile, Tablet II and Tablet III displayed similar antimalarial activity, in which they were formulated by different method but contain the same active substance.

The antimalarial activities of Tablet I, II and III were mainly due to the active substances which contained in tablets instead of formulation method which conducted to produce tablets. Regarding to the fact that andrographolide was one of the active substances which proven active as an antimalarial and assumed to be responsible to the antimalarial activity of *A. paniculata*, then tablets were produced based on the content of andrographolide. Tablet I, II and III were containing the same andrographolide level as an effort to reach the same antimalarial activity level. Tablet I contain AP fraction A which was crude ethyl acetate fraction of *A. paniculata* extract with multi-components content and lower concentration of andrographolide, meanwhile tablet II and III contain AP fraction B which was diterpene lactone fraction obtained from purification of AP fraction A, more concentrate with higher andrographolide concentration. AP fraction B has shown better antimalarial activity than AP fraction A at the same level of andrographolide. Physicochemical properties of AP fraction B was greenish amorphous powder shown different form with AP fraction A which was dark green and sticky. There was a possibility that the sticky form of AP fraction A affected the dissolution profile of andrographolide as an active substance and produced the low bioavailability in the in vivo system. The results shown that the form of raw material of *A. paniculata* fractions were influenced the antimalarial activities of tablets.

4. Conclusion

A. paniculata formulated products containing AP fraction B as an active substance in Tablet II and III showed higher parasite growth's inhibition than product containing AP fraction A in Tablet I. The different formulation method was not influenced the antimalarial activity. The antimalarial activity productsdepending on the raw material form of *A. paniculata* fraction which contained in the products.

Acknowledgement

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