



PROCEEDING

The 1st University of Muhammadiyah Purwokerto Pharmacy Internasional Conference (UMP-PIC)

5-6 June 2015

"Colaboration Approach to Improve Research
and Management of Chronic Diseases"

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– PHARMACY INTERNATIONAL CONFERENCE (UMP-PIC)
PROCEEDING**

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UNIVERSITY OF MUHAMMADIYAH PURWOKERTO
Purwokerto, Central Java, INDONESIA**

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**Greetings from the Dean Faculty of Pharmacy
University of Muhammadiyah Purwokerto**

It is a great honour for Faculty of Pharmacy, University of Muhammadiyah Purwokerto to welcome all of invited speakers and researchers at The First University of Muhammadiyah Purwokerto – Pharmacy International Conference (UMP-PIC). Faculty of Pharmacy, UMP would like to held this confence periodically every two year. For the first time, we have a topic, “Collaborative Approach to Improve Research and Management of Cronic Diseases”. The aim of this conference is to promote the utilization and research development of cronic diseases toward patients management including prevention, therapy and health promotion.



Chronic diseases, such as heart disease, stroke, cancer, chronic respiratory diseases and diabetes, are by far the leading cause of mortality in the world, representing 68% of all deaths in 2012. Out of the 38 million people who died from chronic disease, more than 40% of them (16 million) were premature deaths under age 70 years. Almost three quarters of all Chronical diseases deaths (28 million), and the majority of premature deaths (82%), occur in low- and middle-income countries. This invisible epidemic is an under-appreciated cause of poverty and hinders the economic development of many countries that is why then called non communicable diseases (NCD) (WHO, 2014).

I hope that all scientists and researchers participating in this event will present and discuss current international strategies for prevention and control of chronic noncommunicable diseases (NCD), to make participants familiar with the Finnish experiences from the North Karelia Project and to train in planning, implementation and evaluation of NCD prevention interventions: 'from theory to practice'. I am sure that this event will give advantages for all of us.

Lastly, I would like to express my deep gratitude to all participants for their contributions to the success of this event. I hope all of you will have a fruitful meeting and a pleasant stay in Purwokerto.

Dr. Nunuk Aries Nurulita, M.Si., Apt.
Dean of Faculty of Pharmacy
University of Muhammadiyah Purwokerto

Greetings from the Chairperson of the Organizing Committee
The 1st University of Muhammadiyah Purwokerto – Pharmacy International Conference
(UMP-PIC)

Dear Colleagues,

The Organizing and Scientific Committee of the 1st UMP-PIC (University of Muhammadiyah Purwokerto-Pharmacy International Conference) is very pleased to announce its 1st Biannual Conference which will take place from June 5-6, 2015 at the Horison Hotel, Purwokerto, Central Java, Indonesia. The theme of this meeting is “Collaborative approach to improve research and management of chronic diseases”. It will focus on the emerging role of pharmacists in chronic care. We have been able to invite experts in the field of science and clinical pharmacy to share about their experience in managing chronic diseases. Conference sessions will include plenary lectures, oral and poster presentations.



The 1st UMP-PIC invites pharmacists and pharmaceutical scientists from all over the world to delve into research and healthcare management in chronic diseases. All the organizing and scientific committee are greatly looking forward to welcoming the participants of the 1st UMP-PIC in Purwokerto, Central Java, Indonesia to experience the wonderful Indonesian hospitality. Come to the 1st UMP-PIC and gather at the most important meeting for our global network of pharmacists. We look forward to seeing you here!

Githa Fungie Galistiani, M.Sc., Apt.
Chairperson

Antimicrobial Activity of Extracts and Fractions of Purwoceng (*Pimpinella alpina* Molk)

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ABSTRACT

Purwoceng (*Pimpinella alpina* Molk) is a traditional aphrodisiac plant. This plant is originated from Indonesia and grow at Dieng Plateu, Central Java. The antimicrobial activity of petroleum ether extract, ethyl acetate extract, ethanol extract and fractions of ethyl acetate extract were assayed in vitro by agar disc diffusion method against *Staphylococcus aureus*, *Escherichia coli*, MG42 bacterial isolate, *Candida albicans* and *Malassezia furfur*. Ciprofloxacin was used as positive control in antibacterial assay and Dimethylsulfoxide (DMSO) was used as negative control. Ethyl acetat extract and etanol extract of Purwoceng have antibacterial activity against *Staphylococcus aureus*, *Escherichia coli*, and MG42 bacterial isolate. None of the extracts and fractions of Purwoceng assayed was active against *C. albicans*. All of the extracts were active against *M. furfur*. The lowest concentration of ethyl acetate extract inhibiting *M. furfur* was 1.25%.

Key words: Antimicrobial activity, Purwoceng (*Pimpinella alpina* Molk).

INTRODUCTION

Purwoceng (*Pimpinella alpina* Molk) is one of traditional medicinal plants possessed an androgenic effect and used as aphrodisiac. Purwoceng is endemic plant of Indonesia (Usmiati, 2010). This plant is originally from Indonesia that grows at Dieng Plateu, Central Java. Purwoceng have been reported contain steroid, flavonoids, glycosides, saponins, tannins, and phenolic (Ma'mun et al., 2011). Based on the secondary metabolites of Purwoceng, a research to determine antimicrobial activity of Purwoceng is needed to be done.

Many studies reported the androgenic effect of Purwoceng. Treatment with Purwoceng extract could enhance vitality, indicated by increasing testosterone and luteinizing hormone (LH) (Nasihun, 2009). Chloroform extract of this plant had the highest aphrodisiac activity than other extract. The root of Purwoceng contain of stigmastero (Suzery, 2005)l. Unfortunately, this plant doesn't have estrogenic activity (Susanti and Dhiani, 2012).

The previous study showed that ethyl acetat extract and its fractions containing flavonoid and phenolic compounds, scondary metabolites commonly possessing antimicrobial activity. In this research, antimicrobial activity of ethyl acetate extract and its fractions were assessed using agar diffusion method against *Staphylococcus aureus*, *Escherichia coli*, MG42 bacterial isolate, *Candida albicans* and *Malassezia furfur*.

EXPERIMENTAL

2.1 Plant material

Purwoceng was collected from Dieng, Wonosobo, Central Java, Indonesia.

2.2 Chemical and Reagents

Petroleum ether, ethyl acetate, ethanol, n-hexane, methanol, silica gel 60 (0,2-0,5 mm) (Merck), nutrient agar (NA), nutrient broth (NB), Saboroud Dextrose Agar (SDA), Saboroud Dextrose Broth (SDB), ciprofloxacin, dimethyl sulfoxide.

2.3 Microorganism strains

Staphylococcus aureus, *Escherichia coli*, *Candida albicans* used in this study were obtained from Microbiology Laboratory, University of Muhammadiyah Purwokerto. MG42 bacterial isolate was an isolated bacteria from soil that was resistance to amoxicillin, cotrimoxazole, and oxytetracyclin.

2.4 Extraction and Fractionation

Powder plant material was macerated in petroleum ether for 3 days. The residue then macerated in ethyl acetate for 3 days. The last, residue was macerated in ethanol. Each filtrated was evaporated using rotary evaporator and yielded 3 extract that is petroleum axtract, ethy acetate extract and ethanol extract. Based on antibacterial activity screening, ethyl acetat extract was the most active among three extract so ethyl acetate extract was fractionated by column chromatography and resulted 25 fraction. Fractions that had same TLC profile were collected. Fraction number 11-25 were collected in one fraction, so there were 10 fraction tested against microorganism.

2.5 Antimicrobial activity

The antimicrobial activity was conducted with agar diffusion technique. Ten microliter (10 µL) of extract or fraction solution in various concentration was applied on plate. To compare the activity with standard antibiotic, ciprofloxacin (2.5 µg/disc) was used. Disc containing 10 µL DMSO was used as a negative control. The discs were then incubated at 37°C for 24 hours for bacteri and 25°C for 48 hours for fungi, after which the zones of inhibition of desired growth could be easily measured. The zone of inhibition was considered as an indicator for the antibacterial activity. At the end of the incubation period the antibacterial activity was evaluated by measuring the inhibition zones in mm.

RESULTS AND DISCUSSION

The antimicrobial activities of different extract were tested by agar diffusion method were shown in table 1. All of the three extract showed weak antibacterial activity (inhibition zones ≤ 12 mm) and had lower zone inhibition than positive control ciprofloxacin. Ciprofloxacin had strong with inhibition zone ≥ 18 mm. Among of three extract, ethyl acetate extract had strongest antibacterial activity against all microorganism tested. Ciprofloxacin was used as positive control in antibacterial activity assay due to the sensitivity of MG42 bacterial isolate. MG42 bacterial isolate was resistant to oxytetracyclin, chloramphenicol, gentamicin dan amoxicillin.

The result showed that Gram negative bacterium *E. coli* was more sensitive than Gram positive bacterium *S. aureus* to ethyl acetate extract. But in contrast to the activity of ethyl extract, *S. aureus* was more sensitive than *E. coli* to ethanol extract. Al-Bayati (2008) reported that essential oil and methanol extract of *Pimpinella anisum*, a plant with same genus, inhibited several bacterial growth but their effectiveness varied. Gram positive bacterium *S. aureus* and *Bacillus cereus* were more sensitive to the essential oils than the Gram negative bacterium except *Proteus vulgaris*. The presence of a hydrophilic outer membrane in Gram negative bacteria blocks the penetration of hydrophobic essential oils into target cell membrane.

Table 1. Antimicrobial activity of Purwoceng extracts

Microorganism	Diameter of Inhibition Zones (mm $\pm$ SD) ^a				
	Petroleum ether extract	Ethyl acetate extract	Ethanol extract	Ciprofloxacin	DMSO 10%
<i>S. aureus</i>	10.3 $\pm$ 0.96	9.97 $\pm$ 0.25	9.5 $\pm$ 0.6	28.06 $\pm$ 0.32	0.00 $\pm$ 0.00
<i>E. coli</i>	10.2 $\pm$ 0.46	11.0 $\pm$ 0.60	8.4 $\pm$ 0.6	28.56 $\pm$ 0.58	0.00 $\pm$ 0.00
MG42	9.53 $\pm$ 0.96	9.67 $\pm$ 1.76	7.11 $\pm$ 0.39	27.46 $\pm$ 0.50	0.00 $\pm$ 0.00
<i>C. albicans</i>	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	-	0.00 $\pm$ 0.00
<i>M. furfur</i>	10.04 $\pm$ 1.01	9.11 $\pm$ 1.82	7.76 $\pm$ 0.58	-	0.00 $\pm$ 0.00

a : results were means of three different plate

Concentration extract for antibacterial activity was 200 mg/ml

Concentration extract for antifungal activity was 100 mg/ml

Screening of chemical groups in the extract of Purwoceng revealed presence of alkaloid, tanin, flavonoid, triterpenoid, steroid, dan glikosida (Ma'mun et al., 2006). Based on the result of chemical composition, we conclude that antibacterial activity of the three extracts especially the ethyl acetate extract is apparently related to these compounds. Genistein, genistin, daidzein, daidzin and other sioflavon werw found in Apiaceae family especially in *Pimpinella* genus

(Abdulmanea, 2012). Essential oils from *P. anisetum* and *P. flabellifolia* (Tepe et al., 2006), *P. Affinis* (Mohammadreza, 2008) showed moderate antimicrobial activity against several microorganism. The vast majority of pharmacological activity studies of *Pimpinella* species were focused on the essential oil (Tepe, 2015).

None of the extract assayed was active against *C. albicans*. All of the extract were active against *M. furfur*. Ethyl acetate extract was re-assayed against *M. furfur* with different concentration and give result that the lowest concentration can inhibit *M. furfur* was 1.25%.

The antibacterial activity of fraction of Purwoceng extract shown in table 2. The data indicated that fractions had weak activity against all bacterial tested. Meanwhile, there were 4 fraction have no antibacterial activity against all tested bacterial. Fraction number 3 posses antibacterial activity with strongest inhibition among all fractions tested. But, its activity was lower than ciprofloxacin as positive control.

Table 2. Antibacterial activity of fractions of Purwoceng extract

Sample	Diameter of Inhibition Zones (mm $\pm$ SD) ^a		
	<i>S. aureus</i>	<i>E. coli</i>	MG42 bacterial isolate
Fraction 1	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00
Fraction 2	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00
Fraction 3	6,90 $\pm$ 0,85	6,77 $\pm$ 0,37	8,85 $\pm$ 0,21
Fraction 4	6,00 $\pm$ 0,00	6,63 $\pm$ 0,60	6,80 $\pm$ 1,06
Fraction 5	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00
Fraction 6	0,00 $\pm$ 0,00	6,30 $\pm$ 0,26	0,00 $\pm$ 0,00
Fraction 7	0,00 $\pm$ 0,00	6,10 $\pm$ 0,14	7,37 $\pm$ 0,55
Fraction 8	0,00 $\pm$ 0,00	6,20 $\pm$ 0,14	0,00 $\pm$ 0,00
Fraction 9	0,00 $\pm$ 0,00	6,05 $\pm$ 0,07	0,00 $\pm$ 0,00
Fraction 10	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00
Ciprofloxacin	29,43 $\pm$ 1,42	28,6 $\pm$ 2,04	29,20 $\pm$ 1,68
DMSO 10%	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00	0,00 $\pm$ 0,00

a : results were means of three different plate

CONCLUSION

It can be concluded from this study that petroleum ether, ethyl acetat extract and ethanol extract of Purwoceng have antibacterial activity against *S. aureus*, *E. coli*, and MG42 bacterial isolate. None of the extracts and fractions of Purwoceng assayed were active against *C. albicans*. All of the extracts were active against *M. furfur*. The lowest concentration of ethyl acetate extract that can inhibit *M. furfur* was 1.25%.

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