ceeding
1st International Conference
aceutics & Pharmaceutical Sciences
ICPPS 2014

ANTIHEPATITIS C VIRUS ACTIVITY SCREENING ON Harpullia arborea EXTRACTS AND ISOLATED COMPOUND

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INTRODUCTION

Hepatitis C is a major healthcare problem worldwide. Available therapy for hepatitis C treatment is very expensive and probably not be accessible for all patients. Regarding to this reason, the development of safe and inexpensive antiviral drugs is required. Natural products as a source of new drugs are potential to study. Some antiHCV substances from plants were obtained (Wahyuni, 2013; Adianti, 2014; Apryani, 2014).

Harpullia arborea (tulip wood tree) is a member of Sapindaceae family commonly known as lanas pacat in Indonesia (Basuni, 1997). Traditionally, watery exudates from barks and fruits is used as leech repellant, oil extracted from seeds is a source of antirheumatics (Singh, 2011). H.arborea seeds extract also shown antibacterial activities against various strains of bacteria. H.arborea seeds contain glycosides, steroids, saponins and resins (Gowri, 2009). A norhopane triterpenoid also isolated from the leaves of H.arborea (Poovapatthanachart, 2008).

This study was conducted to determine anti-HCV activity of H.arborea extracts and isolated compound.

MATERIALS AND METHOD

Plant material

Harpullia arborea was obtained from Alas Purwo National Park at Banyuwangi, East Java. Sample was authenticated by the authority of Purwodadi Botanical Garden, Pasuruan, East Java.

Extraction dan fractionation

H.arborea was extracted by ultrasonic assisted extraction method using 80% ethanol as a solvent. Liquid fractionation was conducted using dichloromethane, ethyl acetate and buthanol respectively.

AntiHCV activity test

Extract was examined for antiHCV activity against JFH1a and J6/JFH1 in a cell culture system using Huh7 cells at a multiplicity of infection (MOI) of 0.1.

RESULTS DAN DISCUSSION

Anti-Hepatitis C Virus (anti-HCV) activity screening of H.arborea leaves and stem extract revealed that leaves extract exhibited anti-HCV with IC50 value of 17.5 µg/ml and 12.4 µg/ml against HCV JFH1a and J6/JFH1 respectively, meanwhile stem extract was found to be not active against both HCV type.

Fractionation of leaves extract resulted in 4 fractions which were dichloromethane, ethyl acetate, buthanol and aqueous fraction. Anti-HCV activity screening at a concentration of 30 µg/ml revealed that buthanol fraction inhibited HCV JFH1a growth by 54% in which other fractions only inhibited by 15-30%. Buthanol fraction contains yellow spot on TLC profile as a major compound. Further separation of buthanol fraction using sephadex LH-20 and

methanol 90% as a solvent was obtained 7 fractions (B1-B7). Fraction B5 contain yellow precipitate and by recrystallization process obtained a yellow crystal as a glycosylated flavonoid compound which identified as Kaempferitrin (3,7-di-α-L-rhamnopyranosyl kaempferol). Structure determination of compound was done by nuclear magnetic resonance spectroscopy and data were compared with references (Ouyang Ming-An, 2003; De Sauza Menezes, 2007). Kaempferitrin was further tested against JFH1a. Anti-HCV activity test shown that kaempferitrin was not exhibited anti-HCV. It is possible to explain that anti-HCV activity of extract and buthanol fraction was produced by other compounds in the extract and buthanol fraction instead of kaempferitrin or the activity was created by synergism effect of many compounds. Previous studies were reported some activity of kaempferitrin. Like many flavonols, it has antimicrobial, antioxidant and antiinflamatory activities. It is also mimics insulin in stimulating glucose uptake in diabetic rats, but inhibits insulin-stimulated glucose uptake in 3T3-L1 cells (Jorge, 2004; Prasad, 2009). But no report about antiviral activity of kaempferitrin was found. Further study need to be done to investigate the anti-HCV compounds of H.arborea.

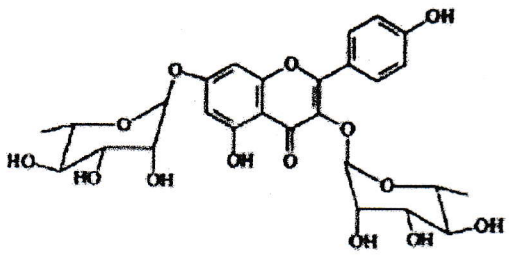

Figure 1. Kaempferitrin
(3,7-di-α-L-rhamnopyranosyl kaempferol)

CONCLUSION

In this sudy, we concluded that H.arborea leaves extract and buthanol fraction were exhibited anti-HCV activity against JFH1a virus, while the isolated compound, [illegible] was not.

ACKNOWLEDGEMENT

This study was supported by Scien[illegible] nology Research Partnership for Sus[illegible] Development (SATREPS) from Japan [illegible] and Technology Agency (JST), Japan [illegible] tional Cooperation Agency (JICA) and [illegible] sitas Airlangga Indonesia.

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CERTIFICATE

This is to acknowledge that

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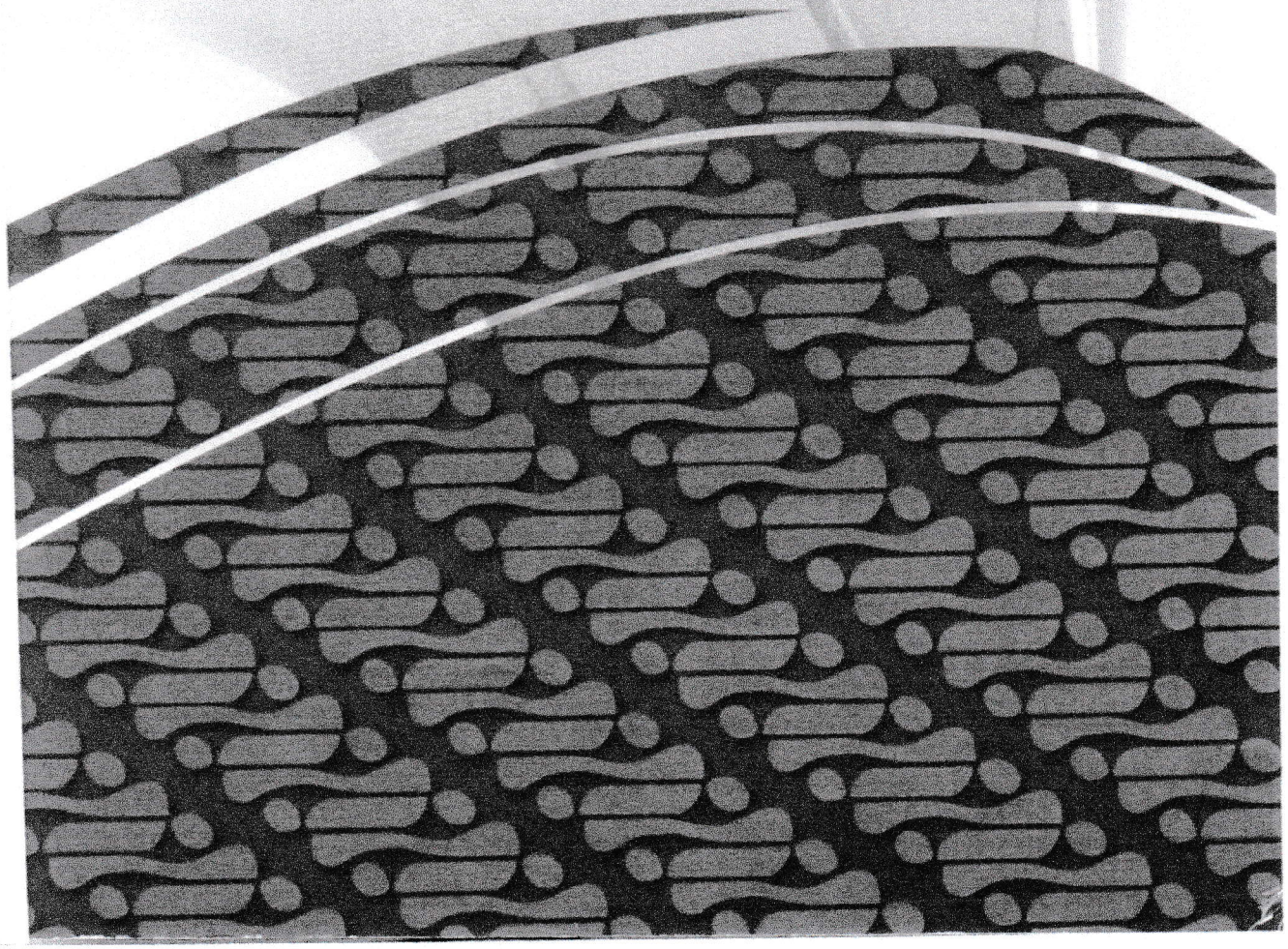

Proceeding

The 1st International Conference on Pharmaceutics & Pharmaceutical Sciences

Published and Organized by
Faculty of Pharmacy
Universitas Airlangga
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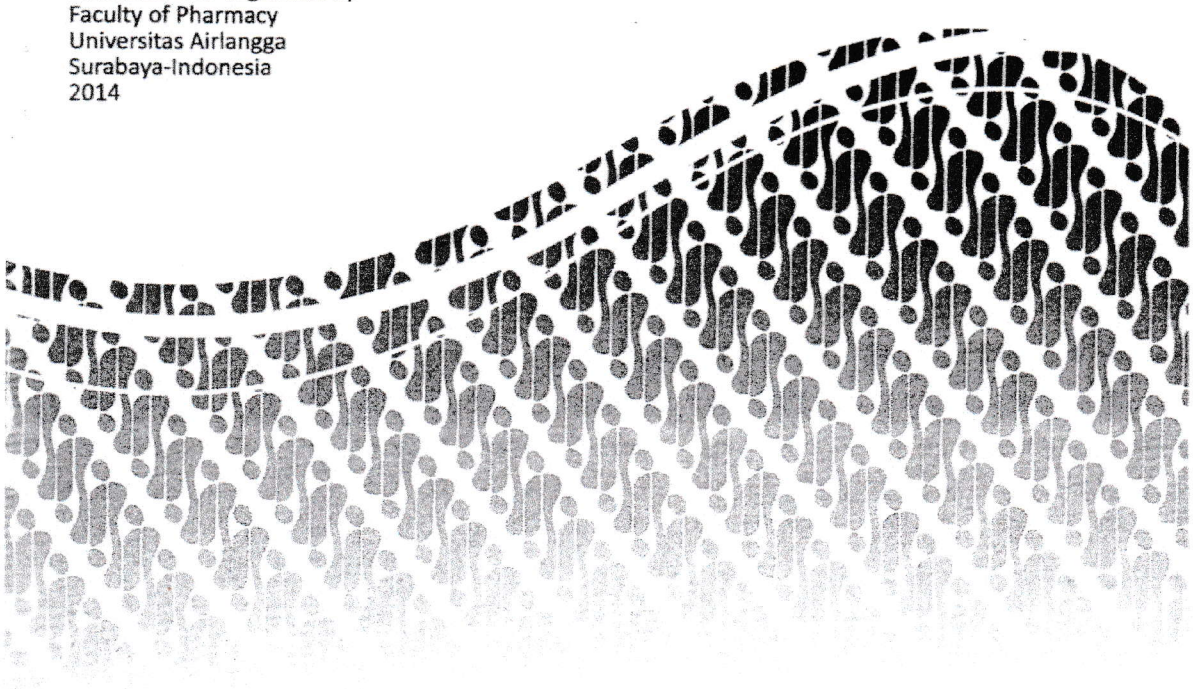

The 1st International Conference on Pharmaceutics & Pharmaceutical Sciences Proceedings

ISBN : 978-602-72333-0-0
(Letter of National ISBN Agency No. 4127/E.8/p/03.2015 Date 18 March 2015)

1st edition Proceeding
Published by:
Faculty of Pharmacy Universitas Airlangga
Surabaya, Indonesia

Address:
Kampus B Jl. Dharmawangsa Dalam
Surabaya 60286
Phone +62 31 5033710
Fax +62 31 5020514
Website: www.icpps2014.com or www.ff.unair.ac.id
Email: icppinfo@gmail.com

ISBN 978-602-72333-0-0

PREFACE From Chairman

It is our pleasure to present you the proceedings of The 1st International Conference on Pharmaceutics and Pharmaceutical Sciences (ICPPS) organized by The Faculty of Pharmacy Universitas Airlangga Surabaya Indonesia.

The proceeding was produced based on papers and posters presented at The 1st International Conference on Pharmaceutics and Pharmaceutical Sciences (ICPPS), held in Surabaya, Indonesia, 14-15 November 2014.

The proceeding clearly reflects broad interest, from the participants that coming from all around the world.

The papers presented were pharmaceutics and biopharmaceutics; requirements on how to evaluate molecules in discovery and their appropriateness for selection as potential candidate; their development in context of challenges and benefits, together with associated time and cost implications and also requirements to progress through pre-clinical and clinical.

In this an opportunity, I would like to express my appreciation to the editorial team of the proceeding who have been working hard to review manuscripts, and making the first edition of this proceeding be possible.

I would like also to thanks to all invited speakers and presenters who participated in The 1st International Conference on Pharmaceutics and Pharmaceutical Sciences (ICPPS) and your contribution to this proceeding.

Finally, I hope this proceeding will give contribution to the Pharmaceutics and Pharmaceutical Sciences research.

Chairman,

Dra. Esti Hendradi, MSI., Ph.D., Apt

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