RAPID IDENTIFICATION OF QUINONE METHIDE TRITERPENES USING LCMS/MS APPROACH

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1. Letter of Report Submission

26th April 2017

To,

Ass. Prof. Dr. Ahmad Taufek Abdul Rahman
Director of Research Management Centre (RMC)
Universiti Teknologi MARA
40450 Shah Alam

Subject: **End Report Submission of Project**
Project Title: "Rapid identification of quinone methide triterpenes using LCMS/MS approach" 600-RMI/FRGS 5/3 (49/2014)

Respected Sir,

I am pleased to informed you that we have successfully completed the above mentioned project. Please find the detailed report about the project as attached.

Thanks for your prompt assistance for this matter.

Sincerely,

Dr. Humera Naz
Senior Lecturer
Faculty of Pharmacy
University Teknologi MARA
Puncak Alam Campus

5.2 Enhanced Executive Summary

(Abstract of the research) – 1 page only

Lophopetalum javanicum is one of the specie in Celastraceae family. The family Celastraceae is rich in quinone methide triterpenes and many biological activities have been reported from members of this class, including antitumor, antioxidant and trypanocidal.

The stem bark of *Lophopetalum javanicum* was investigated for its chemical constituents. The plant was obtained from the herbarium unit, Universiti Malaya (Series No. : 4795). Around 1.5 Kg bark of *L. javanicum* were extracted by using *n*-hexane, chloroform, ethyl acetate, and methanol. Several chromatographic methods were used to isolate and purify the compounds from hexane and chloroform extracts such as VLC, glass column, thin layer chromatography and preparative thin layer chromatography. The phytochemical studies on *n*-hexane and chloroform extracts on *Lophopetalum javanicum* resulted in the isolation of three known triterpenes including lupeol, betulin, betulinic acid, and one known steroid namely β-sitosterol. Structural identification was achieved by using spectroscopic methods such as ultraviolet (UV), infrared (IR), mass spectroscopy (MS), 1D and 2D-NMR. Although Celestraceaous plants are rich in quinone-methide triterpenes, but none of this type of compounds were isolated from *Lophopetalum javanicum*.

5.3 Introduction

Medicinal plants have been used by mankind as a source of medicines since immemorial time. More than 35,000 plant species have been reported to be used in various human cultures around the world for medicinal purposes (Lewington & Network, 1993). Burkill (1966), in his extensive compilation of the economic products of the Malay Peninsula, recorded around 1,300 plants that have been used in the local traditional medicine.

The diverse flora of Malaysian forest offers a great abundance of chemical diversity of secondary metabolites and bioactive compounds such as alkaloids, flavonoids, steroids, xanthones and genetic variability that have great potential to serve the needs of modern medicine (Ahmad *et al.,* 2003). The discovery of calanolide A and their derivatives from *Calophyllum lanigerum* (bintangor tree) with anti-HIV activities shows the potential of the forest in the drug discovery program and the importance of a well-planned research. Other Malaysian plants such as *Eurycoma longifolia* (tongkat Ali), *Centella asitatica* (pegaga), *Labisia pumila* (kacip Fatimah),

Elephantopus scaber (tapak Sulaiman) and *Phyllanthus niruri* (dukung anak) also showed good potential to be developed into useful drugs (Ahmad et *al.,* 2003).

The plant extracts of the family Celastraceae have been used throughout South America and China as insect repellents and insecticides in traditional agriculture, and also as a remedy for stomach complaints, fever, rheumatoid arthritis and cancer (Spivey *et al*., 2002). Quinone methides are secondary metabolites of the Celastraceae family. It is considered as chemotoxonomic indicators due to its major presence in Celastraceae family. Pristimerin, a quinonemethide triterpenoid derived from Celastraceae, has been proven by research to suppress tumor promotion, metastasis and angiogenesis (Kim *et al*. 2013).

The bioactive compounds from *L. javanicum* is not thoroughly investigated. Therefore, this study is carried out in an attempt to characterise the phtochemical constituents and biological activities of this plant.

5.4 Brief Literature Review

Lophopetalum javanicum is a member of Celastraceae family. The family Celastraceae is indigenous to tropical and subtropical regions of the world including North Africa, South America, and many parts of East Asia, particularly China. These plants generally grow as small trees, shrubs, or lianas that have resinous stems and leaves (Alarcón *et al*, 2015). Plants belonging to the Celastraceae family produce a variety of characteristic secondary metabolites including bioactive celastroloids, pentacyclic triterpenes, sesquiterpene pyridine alkaloids, and dihydro-β-agarofuran (Nunez *et al*., 2016). The triterpenes of this family exhibited antimicrobial, cytotoxic, antiviral, anti-inflammatory, hepatoprotective, antifeedant and insecticidal activities (Zhou *et al*.,2017).

The genus *Lophopetalum* is consists of about 18 species of evergreen trees that can be found in India, Cambodia, Laos, Malaysia, Thailand and Vietnam. Phytochemical investigations of the genus *Lophopetalum* showed the presence of cardiac glycosides and triterpenoids. Bioactive compounds of *Lophopetalum wallichii* have cytotoxic and positive inotropic activity (Sturm *et al*., 1996).

L. javanicum (known as 'perupok' in Malaysia) belongs to the family Celastraceae. It is an indigenous plant in Indonesia, Malaysia, Papua New Guinea, Philippines, Thailand and Vietnam. The bark has been used as a constituent of dart poison. Phytochemical screening on the leaves of this plant showed the presence of