## 学位論文要旨 Dissertation Abstract

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学位論文題目: Evaluation and utilization of *Melaleuca leucadendron* Linn. leaf oils Title of Dissertation (メラルーカ・ロイカデンドロン葉精油の評価と利用)

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Essential oils from *Melaleuca sp.* are endowed with many physiological and pharmacological properties. *M. leucadendron* oil is one of the most important non-timber forest products in Indonesia. Leaves and stems of this species produce essential oil, which have useful medicinal properties. This study elucidated the chemical compositions, qualities, antioxidant, anti-hyaluronidase, antifungal effects, physiological effect on human, and insecticidal activities of *M. leucadendron* leaf oils from Indonesia in order to evaluate their potency and improve their utilization.

Samples of *M. leucadendron* leaves were collected from plantations of *M. leucadendron* in Gundih-Central Java, Indonesia. Leaves were distillated by water-steam distillation. Identification of oil components was conducted by GC-MS analysis, and qualities of oils were analyzed based on Indonesia National Standard (*SNI*) number 06-3954-2006 for standard quality of *Melaleuca* oils. Antioxidant activities were investigated *in vitro* by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) assay, anti-hyaluronidase activities using hyaluronidase enzyme, antifungal activities by agar diffusion method against nine fungal species, insecticidal activities by fumigant assay against whitefly, and effect of *M. leucadendron* oil odor on human physiological responses was analyzed before and after sniffing the essential oil. For the physiological tests of this essential oil, the systolic (max.), diastolic (min.) blood pressures, pulse rate, stress index, and intensities of brain wave were investigated.

Twenty-six components were identified in *M. leucadendron* oils, which 1,8-cineole (49.22-55.04 %),  $\alpha$ -terpineol (8.79-10.70 %), *d*-limonene (5.58-6.39 %), and  $\beta$ -caryophyllene (5.03-7.64 %) were the main compounds in these oils. The organoleptic profiles and physicochemical properties of *M. leucadendron* oils satisfied the quality prescribed by the Indonesian National Standard (*SNI*) number 06-3954-2006; only a few values of specific gravity were below the standard.

The antioxidant assay and anti-hyaluronidase assay showed that M. leucadendron leaf

oils possessed mild antioxidant activity with  $IC_{50}$  between 7.21 and 9.23 mg/ml and anti-hyaluronidase activity with  $IC_{50}$  between 1.94 and 3.03 mg/ml. Antioxidant activities of these oils are probably due to the existence of eugenol. This study also indicates that *M. leucadendron* leaf oils possess moderate anti-hyaluronidase activity caused from  $\beta$ -caryophyllene and slightly 1,8-cineole. Although anti-hyaluronidase activity of *M. leucadendron* leaf oil is not so large, it seems to increase in proportion to the content of  $\beta$ -caryophyllene.

The antifungal assay showed the activities of these essential oils against Fomitopsis palustris (IC<sub>50</sub>: 0.12-3.16 mg/ml), Trametes versicolor (IC<sub>50</sub>: 0.01-0.06 mg/ml), Fusarium oxysporum (IC<sub>50</sub>: 0.02-0.05 mg/ml), Thanatephorus cucumeris (IC<sub>50</sub>: 0.52-3.96 mg/ml), Rhizopus oryzae (IC<sub>50</sub>:1.35-6.43 mg/ml), Aspergillus niger (IC<sub>50</sub>: 10.24-10.97 mg/ml), Cladosporium cladosporioides (IC<sub>50</sub>: 0.03-0.49 mg/ml), Chaetomium globosum (IC<sub>50</sub>: 0.06-0.15 mg/ml), and Penicilium citrinum (IC<sub>50</sub>: 5.84-8.70 mg/ml). The antifungal activity shows that M. leucadendron oils are effective against F. palustris, T. versicolor, F. oxysporum, T. cucumeris, C. cladosporioides and C. globosum, but they are low effective against R. oryzae, A. niger and P. citrinum. The effectiveness of these essential oils is probably due to the existence of  $\alpha$ -terpineol.

Physiological responses showed the systolic and diastolic blood pressures after sniffing *M. leucadendron* oil were reduced. Stress index was evaluated by amylase activity of saliva found that stress index decreased after sniffing this oil sample. *M. leucadendron* oil also can increased  $\alpha$  brain wave or increased relaxation value. This study shows that essential oils have certain therapeutic effects on user or human when its fragrance is inhaled and *M. leucadendron* oil possesses sedative effect and it can be used as aromatherapy (anti-depressants).

*M. leucadendron* oil also has potency to be used as pesticide against greenhouse whitefly *Trialeurodes vaporariorum*. LC<sub>50</sub> of treated whitefly was evaluated after 0.5h (30 min.) exposure time indicated that *M. leucadendron* oil (15.28  $\mu$ l/l) and its main compounds exhibited fumigant toxicity against *T. vaporariorum*. The effectiveness of *M. leucadendron* oil as insect control fumigant is likely influenced by main compounds of this oils such as 1,8-cineole and  $\alpha$ -terpineol.

In conclusion, *M. leucadendron* oil from Indonesia possesses multi-effective properties, such as antioxidant, anti-hyaluronidase, antifungal, insecticidal, and sedative effects. Each compound in *M. leucadendron* oil has different properties and biological activities. The effective components for each property are different such as eugenol for antioxidant,  $\beta$ -caryophyllene for anti-hyaluronidase,  $\alpha$ -terpineol for antifungal, and 1,8-cineole and  $\alpha$ -terpineol for insecticidal.