学位論文の要旨(論文の内容の要旨) Summary of the Dissertation (Summary of Dissertation Contents)

論 文 題 目 Dissertation title

Biological Activities and Environmental Interactions of Momilactones A and B, and Phytochemicals in Rice

広島大学大学院国際協力研究科 Graduate School for International Development and Cooperation, Hiroshima University 博士課程後期 開発科学 専攻 **Doctoral Program** Division of Development Science 学生番号 D160630 Student ID No. TRUONG NGOC MINH 氏 名 Name Seal

Momilactones A and B (MA and MB, respectively) are phytoalexins and plant growth inhibitors available in rice husk. The isolation and purification of these two compounds are complicated, laborious, and ineffective. Thus, the known biological properties of MA and MB have been limited only to allelopathy, antioxidant, antifungal, and antimicrobial activities. The present study was conducted to establish a protocol to simplify and optimize quantities of MA and MB by using combinations of ethyl acetate (EtOAc), distilled water, methanol (MeOH), temperature, and pressure for extractions. The chemical components, antioxidant, and antimicrobial activities in rice husk were thus examined. By using a temperature at 100 °C combined with EtOAc and MeOH 100%, MA and MB were enriched to much greater quantities than in non-treated rice husk. Treatment with either ethyl acetate (100 °C, 1 h) or distilled water (100 °C, 2 h) combined with MeOH 100% provided maximum yields of MA (51.54–58.76 µg/g dry weight (DW)) and MB (102.23–104.43 µg/g DW). Because the melting points of MA and MB were 234-236 °C and 240 °C, respectively, the treatment at 100 °C may effectively increase the quantities of MA and MB. The enrichment increased the amounts of MA and MB purified by column chromatography by 5 and 15 times, respectively. The use of either only distilled water or MeOH \leq 50% at any temperature could not successfully isolate both MA and MB. Although the treatments that afforded maximum yields of MA and MB were not proportional to total contents of phenolics, flavonoids, or antioxidant and antimicrobial activities, the extractions established by this research were useful to utilize rice husk as a source of antioxidants and antimicrobial agents. The optimization of MA and MB yields aids simpler and more productive purification of these two compounds and contributes to the search for further biological activities of MA and MB.

Although investigations on phytochemicals in rice plant parts and root exudates have been extensively conducted, the chemical profile of essential oil (EO) and its potent biological activities are not well understood. In this study, chemical compositions of rice leaf EO and in vitro biological activities were investigated. From 1.5 kg fresh rice leaves, an amount of 20 mg EO was obtained by distillation and analyzed by gas chromatography-mass spectrometry (GC-MS), electrospray ionization (ESI), and atmospheric pressure chemical ionization (APCI). This revealed the presence of twelve volatile constituents, of which methyl ricinoleate (27.86%) was the principal compound, followed by palmitic acid (17.34%), and linolenic acid (11.16%), while 2-pentadecanone was the least (2.13%). Two phytoalexin momilactones A and B were identified for the first time in EO by using ultra-performance liquid chromatography coupled with electrospray mass spectrometry (UPLC/ESI-MS). The amounts of each were low (9.80 and 4.93 ng/g fresh weight, respectively for MA and MB). The assays of DPPH (IC₅₀ = 73.1 μ g/mL), ABTS (IC₅₀ = 198.3 μ g/mL), FRAP (IC₅₀ = 700.8 μ g/mL) and β -carotene oxidation (LPI = 79%) revealed that EO possesses excellent antioxidant activity. The xanthine oxidase assay indicated that the antihyperuricemia potential was at a moderate level ($IC_{50} = 526 \ \mu g/mL/mL$) compared with the standard allopurinol. The EO exerted potent inhibition on the growth of Raphanus sativus, Lactuca sativa, and two noxious weeds Echinochloa crus-galli and Bidens pilosa, but in contrast, the growth of rice seedlings was promoted. Among the examined plants, growth of E. crus-galli root was the most inhibited, suggesting that constituents found in EO may potential be used for the control of the problematic paddy weed E. crus-galli. It was found that the EO of rice leaves contains rich phytochemicals, which are potent as antioxidants

and in gout treatment, as well as weed management. The findings of this study highlight the potential value of rice leaves to provide extra benefits for rice farmers in developing countries.

The compounds secreted by plant roots have been documented to serve important roles as chemical attractants and repellants in the rhizosphere, the narrow zone of soil or wet soil immediately surrounding the root system. In this study, we investigated the allelochemical interaction of rice (Oryza sativa) and weed (Monochoria vaginalis and Eleocharis) plants in paddies and examined their growth inhibitory property from wet soil extracts. The results of field study showed that rice and weed exerted strong mutual inhibition, as their growth parameters were reduced by 2.47-42.22%. However, the slight inhibitory effects between tested weeds was observed to be 1.59-15.55%. Although the TPC and TFC of aqueous extracts from studied plants decreased (40% and 5%) significantly when they were interfered by their neighboring species, that of EtOAc extracts increased (43% and 9%). The exudation of two well-known phytoalexin compounds from rice momilactones A and B was measured to be 13% and 2% in field 1 (herbicide application), 15% and 5% in field 2 (non-herbicide application) using the chromatographic analyses HPLC and UPLC. Interestingly, the more abundant field contained a small amount of momilactones (75.6 ng/g fresh weigh (FW)). In the various wet soil extracts from studied fields, the growth of L. sativa was the most suppressed, whilst that of Oryza sativa was the least influenced. The abandoned field showed slight inhibition on test plant species. Findings in this study suggest that the chemicals exudate from rice and paddy weeds-including phenolic acids, flavonoids, and momilactones-inhibited successive plants and can be stored in wet soil for a long time.

備考 論文の要旨はA4判用紙を使用し、4,000字以内とする。ただし、英文の場合は1,500語以内と する。

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