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学 位 論 文 要 旨
Dissertation Abstract

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学位論文題目： Screening of bioactive phytochemicals from mangrove fruits
Title of Dissertation （マングローブ果実からの生理活性植物成分の選別）

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The interest in functional foods has increased gradually. In addition to their nutritive value, the functional foods provide health benefit or have a role in disease prevention since they contain a wide range of chemical constituent which represents functional or bioactive compounds. The mangrove forest is one of the most recognized sources of various natural products such as timber, fuel, food, and medicine and also as a source of bioactive compounds. The bioactive compounds derived from mangroves are used not only for the medicinal purpose by indigenous people for centuries but also for functional food consumption because of their bioactivities. In Indonesia, mangrove fruit from species *Rhizophora stylosa* has been processed as food products, which is known as coffee and tea mangrove. Local folks who daily take both products recognize physiological activities, such as rejuvenation and fitness improvement. It means that some bioactive compounds may present in these products and exhibit such effects; however, no scientific evidence has been elucidated and the objectives of this study were to investigate the bioactive compounds of mangrove food products (coffee and tea mangrove) and their medicinal potentials: antioxidant activity, α-glucosidase inhibitory activity, and anti-bacterial activity.

Chapter 2: The bioactive compounds and antioxidant activity were screened from coffee and tea mangrove. Furthermore, three raw materials of coffee mangrove were also investigated to clarify their potencies. The crude extracts of five samples were subjected to antioxidant assay using DPPH. The results show that the extract of tea mangrove has the strongest activity; then, it was successfully fractionated using different polarity of solvents and yielded acetone and methanol fractions that had high antioxidant activity. The acetone fraction was purified and gave fractions A1, A2, A3, A4, A5, and A6, but only A2 and A3 indicated antioxidant activity and, therefore, they were subjected to further purification. From fraction A2, four compounds were obtained (AS1–AS4) based on the peaks which amounted to only 14.1 mg. Most of A2 that could not be separated well seemed to be oligomer or polymer-like as methanol fraction; A3 was rather single compound. The fractions were identified as caffeine (A3) and from A2: AS1 (N,N-dimethyl-L-alanine), AS2 (quercetin-3-O-galactopyranoside), AS3 (dodecanoic acid), and AS4 that had the ^{1}H-NMR

spectrometric result similar to that of AS2, while the methanol fraction did not exhibit clear peaks on the chromatogram by HPLC. Therefore, the precipitation method was conducted to purify this fraction, and the precipitate was analyzed by NMR spectra. The results from ^{1}H and ^{13}C NMR indicate that this fraction is a typical polymer of condensed tannins, containing procyanidin and prodelphinidin units with the average DP (degree of polymerization) is estimated to be around 4 and 5. The evaluation of the antioxidant activity of 6 isolated compounds showed, condensed tannin, which mainly constitutes the acetone and methanol fractions of tea mangrove extract, has high antioxidant activity (IC_{50} 2.69 μg/mL) compared with the other compounds.

Chapter 3: The inhibitory activity of coffee and tea mangrove on α-glucosidase and anti-bacterial activity against *Streptococcus mutans* have been evaluated. The potential of coffee and tea mangrove were tested to inhibit the activity of α-glucosidase enzyme from yeast; and *p*-NPG as a substrate. Mangrove powder, ginger, Javanese long pepper, green tea, black tea and regular coffee extracts were also tested with the same manner; acarbose and quercetin were used as positive controls. Meanwhile, the anti-bacterial activities of the samples were evaluated by disk diffusion method in comparison with 0.2% aqueous solution of chlorhexidine used as positive control. It found that mangrove food products (coffee and tea including mangrove powder) showed mixed competitive inhibition type and strongly suppressed α-glucosidase activity compared to the other samples and the positive controls. At the lower concentration of 0.05 µg/mL, mangrove extracts indicated the % inhibition for α-glucosidase activities were ranged from 25.51 to 34.68, and at the highest concentration of 0.15 µg/mL, the extracts inhibited α-glucosidase ranging from 74.17% to 83.36%; while their IC_{50} values were ranged from 0.07 to 0.08 µg/mL. These products also showed anti-bacterial activities in disk diffusion method through measuring inhibition zone. The anti-bacterial activities of products were increased with the increasing of concentration of the extracts. At lower concentration (1 mg/mL) the diameter of inhibition zones were ranged from 2.33 to 8.67 mm; and at highest concentration (25 mg/mL) the diameter of inhibition zones were ranged from 9.17 to 13.33 mm. As mentioned above (chapter 2), the mangrove food products, in particular, the acetone and methanol fractions of tea mangrove were found to be mostly composed of the condensed tannins. Tea and coffee mangrove, and mangrove powder were all originated from *R. stylosa* fruit, and the potent activity of those products to inhibit α-glucosidase and as an anti-bacterial agent might be of the presence of condensed tannins.

In conclusion, coffee and tea mangrove can be the potential of antioxidant, anti α-glucosidase and anti-bacterial agent due to the presence of condensed tannins as the active main compound.