

***In Vitro* Assessment of Bioactive Properties and Dose-Dependent Antioxidant Activities of Commercial Grape Cultivars in Taiwan**

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Abstract

Grapes are excellent sources of bioactive compounds, which have been suggested to be responsible for lowering the risk of chronic diseases. Fresh and freeze-dried extracts of Kyoho and Jubilee, commercial grape varieties available in Taiwan and attractive for their quality berries, were investigated for their total phenolics and total flavonoids contents and related dose-dependent antioxidants properties using various *in vitro* assays. The efficiency of the extraction yield ranged from 7.10 % to 25.53 % (w/w), depending on solvent used. Fresh samples of Kyoho and Jubilee exhibited total polyphenolic contents (351.56 ± 23.08 and 328.67 ± 16.54 $\mu\text{g GAE/mL}$, respectively), whereas Kyoho freeze-dried methanol:water extracts contains the good levels of total flavonoids (4767.82 ± 22.20 $\mu\text{g QE/mL}$). Kyoho and Jubilee freeze-dried extracts exhibited the highest total flavonoid contents. There was a weak correlation between total phenolic and flavonoid assays ($r = -0.05$, $R^2 = 0.02$, $p > 0.05$). Kyoho fresh and freeze-dried samples showed the DPPH (11.51 – 77.82 %), superoxide scavenging activity (33.61 – 81.95 %), and total antioxidant inhibition (92.01 – 99.28 %), respectively. Total flavonoids were statistically correlated with EC_{50} DPPH scavenging radicals ($r = 0.91$, $p < 0.01$), EC_{50} nitric oxide ($r = 0.25$, $p > 0.05$), and EC_{50} lipid peroxidation radicals ($r = 0.38$, $p > 0.05$). These results suggested that the two commercial grape cultivars in Taiwan could be used as a good source of natural antioxidants. Thus, consumption of grapes as a source antioxidant might lower the risk of chronic diseases. Moreover, future studies will investigate and develop phenolic acid profile for the cultivars in Taiwan.

Keywords: antioxidants, EC_{50} radical scavenging activity, grape cultivars, total phenolics

***IN VITRO* ASSESSMENT OF BIOACTIVE PROPERTIES AND DOSE-DEPENDENT ANTIOXIDANT ACTIVITIES OF COMMERCIAL GRAPE CULTIVARS IN TAIWAN**

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MAIN CONCLUSION

In the current study, polyphenolic and antioxidant activities of two commercial grape cultivars (Kyoho and Jubilee) in Taiwan were evaluated. The results from the various antioxidant activities revealed that the two commercial grape cultivars had significant antioxidant activities.

INTRODUCTION

There is strong evidence that the plant-derived compounds and natural sources (what our ancestors ate) plays a vital role in health maintenance, reduction of chronic diseases, and extending life [1]. Therefore, the aims of this study were to determine the composition of total phenolics and total flavonoids content and *in vitro* analysis of the dose-dependent antioxidants properties of fresh and freeze-dried commercial Kyoho and Jubilee extracts, respectively. Two different solvent systems (methanol: water (4:1, v/v), and water) were used to extract polyphenolic compounds from fresh and freeze-dried samples of commercial Kyoho and Jubilee, respectively.

MATERIALS AND METHODS

Sample preparation was done by two methods i.e. i) fresh and ii) freeze-dried sample. Prior to freeze drying, the pieces were frozen at $-80\text{ }^{\circ}\text{C}$ for 12 h and dried using a freeze dryer (Kansas, USA) at the condenser temperature of $-50\text{ }^{\circ}\text{C}$. For the extraction of bioactive compounds using different solvents, Annegowda, Bhat [2] standard procedure was used for extraction. The determination of phenolic and flavonoid compounds were performed according to Moniruzzaman, Yung An [3]. Antioxidant assays, such as DPPH assay [4], superoxide [5], nitric oxide radical scavenging assay (NOSA) [6], hydrogen peroxide (H_2O_2) scavenging assay by Sroka and Cisowski [7], and total antioxidant activity [8] were conducted in order to determine antioxidant properties of extracts. The effective concentration (EC_{50}) was reported as the amount of antioxidant required to decrease the initial concentration of scavenging radicals by 50 %.

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RESULTS AND DISCUSSION

Fresh samples of Kyoho and Jubilee presented the highest total polyphenolic content (0.35 ± 0.02 and 0.32 ± 0.01 mg GAE/g, respectively), compared to those freeze-dried samples of Kyoho and Jubilee, respectively. Kyoho freeze-dried methanol:water extracts contains the highest levels of total flavonoids (4.76 ± 0.02 mg QE/g), which was approximately 1.80 times lower than Kyoho fresh methanol:water extracts (2.60 ± 0.01 mg QE/g). The radical scavenging activity of Kyoho fresh and freeze-dried methanol: water extracts showed 11.51 % and 12.98 %, respectively at 50 μ L/mL. The results indicated that the grape extracts significantly ($p < 0.05$) decrease in the superoxide radical generation. Nitric oxide radical scavenging assay (NOSA) indicated that the fresh and freeze-dried Kyoho and Jubilee extracts exhibited the more than 60 % inhibition with compared to reference gallic acid (15.67 %) at a minimum concentration (50 μ L/mL). The scavenging activity of grape extracts for hydrogen peroxide (H_2O_2) scavenging assay ranged from 8.84 to 80.53 % at an arithmetic series of concentration (50 – 100 μ L/mL). The EC_{50} values for grapes extracts were in the following order: hydrogen peroxide radical scavenging activity > superoxide radical > DPPH > nitric oxide > total antioxidant radical scavenging activity. Total phenolics was positively correlated with EC_{50} DPPH scavenging radicals ($r = 0.32$, $p > 0.05$) and EC_{50} hydrogen peroxide radical scavenging ability ($r = 0.02$). The positive and negative correlations among EC_{50} radical scavengers might be due to the presence of different chemical compounds and reaction among antioxidants.

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