## Formulation and characterization of novel functional beverages with antioxidant and anti-acetylcholinesterase activities

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## ABSTRACT

**Background:** Nowadays, there is increased consumer demand for high-antioxidant foods. Drinking high-antioxidant beverages may help to protect against aging, Alzheimer's disease, and other chronic diseases. Grapes and some plants including *Phyllanthus emblica*, *Terminalia chebula*, *Kaempferia parviflora*, *Centella asiatica*, *Nelumbo nucifera*, *Rauvolfia serpentina*, *Ginkgo biloba*, *Crocus sativus*, *Clitoria ternatea* and others are well-known to possess antioxidant, neuroprotective and other health-promoting activities. Thus, it is possible to use these plants for the development of new functional beverages.

Methods: Ten formulations of beverages were produced. The 5 non-alcoholic beverages contained dried medicinal plants, fresh grapes and others and are as follows: beverage B1: 10.2% K. parviflora rhizomes, 5.1% brown sugar and 84.7% water; beverage B2: 0.45% Ardisia polycephala leaves, 0.45% C. asiatica leaves, 0.36% Clitoria ternatea flowers, 0.45% Crocus sativus pollens, 0.45% Ginkgo biloba leaves, 0.45% Melodorum fruticosum flowers, 0.90% N. nucifera petals, 0.45% Nymphaea lotus petals, 5.43% crystalline sugar and 90.58% water; beverage B3: 0.62% A. polycephala fruits, 0.35% C. ternatea flowers, 0.44% G. biloba leaves, 2.64% K. parviflora rhizomes, 1.76% P. emblica fruits, 0.88% T. chebula fruits, 5.28% brown sugar and 88.03% water; beverage B4: 0.51% Acorus calamus stems, 0.68% C. ternatea flowers, 4.23% K. parviflora rhizomes, 0.85% N. nucifera petals, 0.85% N. lotus petals, 0.85% *M. fruticosum* flowers, 0.34% *R. serpentina* roots, 0.34% *U. gambir*, 1.69% Zingiber officinale rhizomes, 5.08% brown sugar and 84.60% water; beverage B5: 53.09% fresh grapes, 2.65% brown sugar and 44.25% water. After heating, filtering, and cooling, these beverages were put in sterile bottles. One part of each beverage was stored at 4°C for 23 weeks before analyzing, but the other two parts were used to prepare the alcoholic beverage of each formulation. Grapes were mixed with the beverages B1, B2, B3, B4 and B5 in the ratio of 60:40 to prepare alcoholic beverages W1, W2, W3, W4 and W5, respectively. Two different fermentation conditions

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(fermentation with or without pieces of sliced medicinal plant residue, PMPR) were compared. After fermenting, racking and aging, all alcoholic beverages, as well as all non-alcoholic beverages, were analyzed for some phytochemical properties.

**Results:** Grape fermented with PMPR had higher anti-acetylcholinesterase and antioxidant activities, and total phenolics, flavonoids and tannins, compared to the others. Among all non-alcoholic beverages, the beverage B3 contained the highest anti-acetylcholinesterase (22.78% inhibition at 1:10,000 dilution) and antioxidant activities (reducing capacity, 4.22 mmol Fe(II)/100 mL), total phenolics, flavonoids, and tannins (494.44 mg gallic acid equivalents) (GAE)), 383.22 mg catechin equivalents (CE) and 338.29 mg tannic acid equivalents ((TAE)/100 mL, respectively). Among all alcoholic beverages, the beverage W3 (fermented with PMPR) exhibited the highest antioxidant activity (DPPH radical inhibition, 95.99 mg trolox equivalents and reducing capacity, 3.57 mmol Fe(II)) /100 mL), total phenolics, flavonoids and tannins (239.71 mg GAE, 372.67 mg CE and 157.67 mg TAE/100 mL, respectively). The beverage W2 (fermented with PMPR) had the highest anti-acetylcholinesterase activity (21.35% inhibition at 1:10,000 dilution).

**Conclusion:** The beverages B3, W2 and W3 contained valuable sources of natural antioxidants and acetylcholinesterase inhibitors, and may provide health benefits when consumed.

Keywords: medicinal plant, wine, grape, anti-Alzheimer's disease, antioxidant