

뽕보리수 추출물의 항혈소판 응집반응과 항염증활성

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Anti-platelet aggregation and anti-inflammatory activity for extracts of *Elaeagnus multiflora*

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Objective

Elaeagnus multiflora is medicinal plant and the fruit, leaves, and root has been used on treatment for cough, diarrhea, itch and foul sores, and even cancer for long time in China. In the present study, we investigated anti-platelet aggregation, anti-inflammation and detected the chemical compounds in *E. multiflora* by GC/MS assay.

Materials and Methods

- Material extract : 40 g of dried *E. multiflora* fruit were extracted with 200 ml each of petroleum ether (PE), chloroform (CE), ethyl acetate (EE), butanol (BE) and water (WE) in a Soxhlet extractor for 24hrs. The solvents were concentrated to dryness with a vacuum rotary evaporator under controlled temperature (<50°C).
- Anti-platelet aggregation activity : Platelet aggregation was performed *in vitro* with an aggregometer. Experiments with various extracts were monitored by pre-incubating the washed platelet sample with each extract individually for 1 min.
- Nitric oxide (NO) production : Experiment cell was RAW264.7 cells (1×10⁶ cells/ml).
- GC-MS analysis : Extract compounds were analyzed using HP 6890 GC and HP 5974MS apparatus with an HP-5 MS column (length 30m; inner diameter 0.25mm; film thickness 0.25μm). 1 μl sample was injected in split mode 50 : 1. Carrier gas was helium (flow rate; 0.7 ml/min, injector and detector temperatures; 200 °C-300 °C).

Results and Discussion

- Only CE exerted weak effects, inhibited about 19.29% of platelet aggregation, at 250μg/ml in comparison with control. However, at higher concentration (500 μg/ml), CE was almost inactive on inhibiting platelet aggregation too (Fig. 1).
- All fractions of *E. multiflora* fruit used in the present study inhibited LPS-induced nitrite production in a dose-dependent manner, and especially, the CE showed the highest inhibitory effects among the fractions with almost 100% inhibition at 500 μg/ml.

- We detected the chemical compounds in CE and EE by GC-MS assay, and only small amounts of phenolic compounds, such as Phenol, 2,4-bis(1,1-dimethylethyl) in CE and EE (Table 1 and 2), were found.

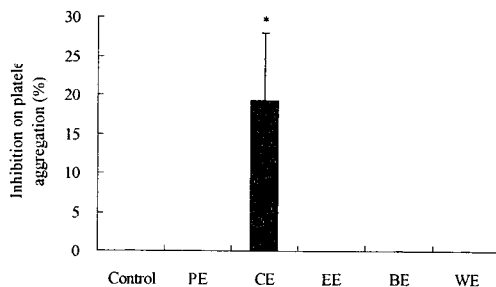


Fig. 1. Inhibitory effects of *E. multiflora* fruit on platelet aggregation at 250 µg/ml. *, p<0.05 vs. control. Petroleum ether extract (PE), chloroform extract (CE), ethyl acetate extract (EE), butanol extract (BE), water extract (WE).

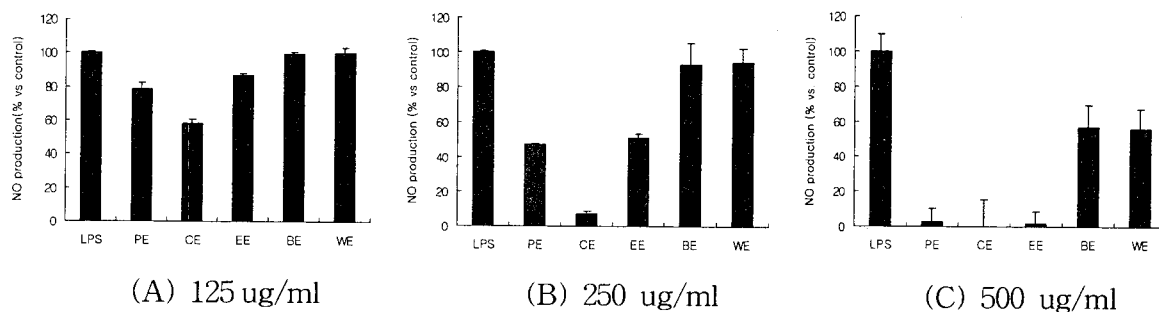


Fig. 2. Effects of *E. multiflora* fruit on Nitric Oxide production in murine RAW264.7 macrophages stimulated by LPS (1 µg/ml). Bars with different characters mean significantly different (p<0.05).

Table 1. Main compounds detected by GC-MS in chloroform extract of *E. multiflora*

Compound	Retention time(min)	% of Extract	Compound	Retention time(min)	% of Extract
Phenol, 2,4-bis(1,1-dimethylethyl)	13.676	6.84	Hexadecanoic acid, 2-hydroxy-1-(jdroxymethyl) ethyl ester	27.389	10.16
Dotriacontane	29.393	2.12	Octadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester	29.619	49.90
Eicosane	30.401	2.02	Tranylcypromine, pentafluorobenzoyl ester	31.17	1.95
Benzoflex	32.792	6.71	Cyclotrisiloxane, hexamethyl	36.1	3.49

Table 2. Main compounds detected by GC-MS in ethyl acetate extract of *E. multiflora*

Compound	Retention time (min)	% of Extract	Compound	Retention time (min)	% of Extract
2,3-Butanediol	2.30	2.10	2,3-Butanediol	2.36	3.79
Octane,4-methyl	2.63	2.00	2-Propanol	2.94	6.56
2-Hexanol	2.99	3.08	Octane,3,3-dimethyl-	5.15	1.39
Undecane,5-methyl	5.78	10.76	Dodecane,2,6,10-trimethyl-	5.88	2.57
Octane,6-ethyl-2-methyl-	6.63	5.72	Heptadecane	6.74	2.03
Pentadecane	10.35	2.30	Phenol,2,4-bis(1,1-dimethylethyl)-	15.21	5.99