inhalational phytoncide oil on cyclophosphamide(CPA)-induced immunosuppression in mice.

Materials and Methods: A total of 30 male ICR mice (6weeks old) were divided into 3 groups (n=10/group): CPA group was treated with CPA 150 and 100 mg/kg on experimental day 0 and 3, normal control group was substituted with saline on the same days, intraperitoneally, and CPA plus phytoncide group was administered phytoncide oil(*Pinus sylvestris*) in an inhalation apparatus for 20 min daily. On day 12, blood hematological parameter including total white blood cell (WBC) numbers, neutrophil and lymphocyte ratio, and immunologic organ weights such as spleen and thymus were measured. The levels of splenic cytokines (IL-2, IL-4, IL-10, IL-12, TNF-α, and IFN-γ)were determined by enzyme-linked immunosorbent assay. CCK-8 assay, immunohistochemistry for CD3+ and CD4+and histopathological analysis of spleen were performed.

Results: Inhalation of phytoncides ameliorated CPA-induced immunosuppressive changes such as the decreases of WBC numbers and lymphocyte ratio and the increase of relative spleen weight in mice. The levels of splenic cytokines including IL-2, IL-10, IL-12, IFN-γ and TNF-α decreased by CPA treatment were significantly recovered by inhalation of phytoncides. In addition, the splenocyte proliferation and numbers of specific splenocytes expressing CD3+ and CD4+ were also increased and further the atrophic changes of spleen accompanied by CPA treatment was inhibited by inhalation of phytoncides.

Conclusions: These results suggest that phytoncides are restorable the CPA-induced immunosuppression in mice through modulations of the immunocytes recruitment, cytokine production, and mitogenic effect that may have a possible therapeutic role for preventing immunosuppressive diseases.

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Theinhibitory Mechanism of *Rumex acetosaextract* in Collagen-induced Platelet Aggregation

Da Hye Jung, Man Hee Rhee*

Department of Veterinary Physiology, College of Veterinary Medicine, Kyungpook National University

Introduction: The *Rumex* species for many centuries have been used in medicinal treatment. *Rumex acetosa (R. acetosa)*,often called sorrel, is a perennial herb belonging to the family polygonaceae. Sorrels are used in food technology and for phytotherapeutic use in Korea. Previous studies reported their anti-oxidant and anti-herpes simplex viral activities. Platelet aggregation is an essential part of the haemostatic process following vascular insult. However, research on the anti-platelet activity of *R. acetosa* is scarce.

Materials and Methods: Platelet aggregation in washed rat platelets was measured using aggregometer under collagen stimulation. Intracellular calcium levels were measured using Fura 2AM. Furthermore, ATP release, and western blot were studied using washed rat platelets.

Results: Our results showed that *R. acetosa* markedly inhibited collagen-induced platelet aggregation and collagen-induced ATP release in a dose-dependent manner. It also suppressed[Ca^{2+}]i mobilization in collagen-stimulated platelets. Moreover, immunoblotting revealed that *R. acetosa* significantly suppressed extracellular signal-regulated protein kinase 1/2 (ERK1/2), c-Jun N-terminal Kinases (JNK), Akt and phosphoinositide 3-kinase (PI3K) phosphorylation.

Conclusions: These results indicate the inhibitory effect of *R. acetosa* on platelet aggregation and granule secretion that is mediated by suppression of calcium mobilization. Finally, ourresults support that *R. acetosa* could be potent candidate as a therapeutic agent against platelet-related cardiovascular diseases.

References

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Comparison of Cellular Senescence of Fetal and Adult Dog Fibroblasts

Minjung Kim, Yoo Bin Choi, Hyun Ju Oh, Geon A Kim, Young Kwang Jo, Erif Maha Nugraha Setyawan, Seok Hee Lee, Byeong Chun Lee*

Department of Theriogenology and Biotechnology, College of Veterinary Medicine, Seoul National University, 1 Gwanak-ro, Gwanak-gu, Seoul 151-742, Republic of Korea

Introduction: Production of transgenic cloned dogs using somatic cell nuclear transfer can be a valuable tool to investigate human diseases because of similarities in their size, longevity and physiology between dog and human. Although only fetal fibroblasts were used to transgenic cloned dogs (1, 2), adult fibroblasts derived from a dog having superior ability can also be used to preserve and increase the dog's talent. Therefore, the aim of present study was to compare cellular senescence of fetal and adult dog fibroblasts for determining their further usage to produce a transgenic cell line.

Materials and Methods: Six fetal fibroblasts were isolated from the fetuses of a beagle bitch obtained at 28 days after