P2-19-4 Active constituents of the extract from Nandina domestica Thunberg responsible for inhibition of lipopolysaccharide-induced cyclooxygenase-2 expression in A549 cells
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We have previously found that the extract from Nandina domestica Thunberg (NDE: 0.1-10 μg/ml) inhibits lipopolysaccharide (LPS)-induced cyclooxygenase-2 (COX-2) expression and prostanandin E production in human pulmonary epithelial A549 cells. To identify active constituent(s) of NDE responsible for the anti-inflammatory effect, we fractionated NDE and assessed their effects in cultured A549 cells. NDE was introduced in a polyaromatic absorbent resin column and stepwise eluted to yield water fraction, 20% methanol fraction, 40% methanol fraction, 99.8% methanol fraction and 99.5% acetone fraction. However, none of the five fractions alone inhibited LPS-induced COX-2 expression. On the other hand, removal of water fraction from NDE abolished the inhibitory effect of NDE on LPS-induced COX-2 expression. These results suggest that constituent(s) present in water fraction is required but not sufficient for the anti-inflammatory activity of NDE, which may result from interactions among multiple constituents.

P2-19-5 Larix sibirica defend experimental fulminant hepatitis induced by ConA
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Fulminant hepatitis is induced by infection. Oxidative stress is also one of etiologies. Extract substances of Larix sibirica; Dihydroquercitin (DHQ) have some pharmacological action which include the inhibition and elimination of free radical, antioxidative properties, anti-inflammatory and so on. Genetically, Concanaavalin A (ConA) raises the incidence of hepatitis in the mice medicated with ConA. Therefore, we investigated the effect of DHQ on fulminant hepatitis using animal models having hepatitis induced by ConA. Male C57BL/6 mice were divided into 3 groups as follows: (1) normal; normal; free feed and no treatment(2)ConA: injection of ConA by iv.(3)ConA+DHQ: DHQ was administrated 3 times at 1,2 and 3 days before ConA. After the injection of ConA, DHQ was continuously administrated 1 time/day. Alamine amino transferase, aspartate amino transferase and inflammation in liver, spleen and serum increased in the ConA group compared to the normal group; moreover these levels significantly took effect in the ConA+DHQ group. Gene expressions of IL-1β, IL-17, IL-4, IFN and IL-10 detected by qRT-PCR also decreased in ConA+DHQ group compared to ConA group, DHQ of Larix sibirica significantly inhibit the onset of fulminant hepatitis by anti-oxidant and anti-inflammation mechanisms.

P2-19-6 Improvement of an Atopic Dermatitis Model Prepared by Repeated Application of Dermatophagoides farinae in Mice
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An atopic dermatitis model prepared by repeated application of Dermatophagoides farinae (DF) in mice has been established to assess the pharmacological effect of drugs. However, we were unable to use about 30% of the animal models treated with DF for evaluation of drugs, since they had low dermatitis scores. For this study, the dosing frequency of 4% sodium dodecyl sulfate (SDS) was increased to assess whether the ratio of animals with low dermatitis scores was decreased. Specifically, 4% SDS was administered to the dorsal skin of male NC mice (aged 8 weeks) 2 or 4 times a week, and then DF was given into the back and auricular (twice a week, 8 times in total). After dermatitis was induced, the animals were treated with betamethasone sodium phosphate (BP, 100 μg/animal) and tacrolimus ointment (TO, 100 μg/animal) once a day for 14 consecutive days. As a result, the ratio of animals in which we were able to evaluate efficacy of drugs was found to be increased by a change in dosing frequency from 2 to 4 times. Also, dermatitis symptoms and auricular thickness, which were caused by DF, were improved by BP and TO. Increased dosing frequency of 4% SDS did not affect efficacy evaluation of either drug.

P2-19-7 In vivo imaging of damage associated molecular patterns (DAMPs) DNA in mouse lungs with ARDS
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Many pathogenic non-viral and viral conditions lead to the development of the acute respiratory distress syndrome (ARDS), the most severe form of acute lung injury. ARDS is characterized in the early phase by neutrophil-mediated, excessive pulmonary inflammation. We previously reported that damage associated molecular patterns (DAMPs) derived from damaged cells activate TLR4-TRIF pathway, which is responsible for the pathogenesis of ARDS caused by multiple stimuli (Imai et al. Cell 2008). Also, recent accumulating evidence suggests that double-stranded DNA, independent of CpG motifs, possesses immunomodulatory effects when introduced into the cytosol or its homeostatic clearance is hampered. In the present study, using in vivo mouse lung imaging system with SYTOX Green, we observed massive accumulation of neutrophil extracellular traps-like DNA structures in the alveolar space in the lungs with non-viral (acid-induced) and viral (influenza virus-induced) ARDS. Also, the concentrations of free DNA were significantly higher in the BAL fluids obtained from lungs with non-viral and viral ARDS than controls in mice. These data suggest that DAMPs DNA were generated in the lungs with ARDS, which might be associated with the development of ARDS.