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## ABSTRACT BOOK

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aqueous extract (SAE) in complete Freund's adjuvant (CFA) induced arthritis rat model. So, Male Wistar rats weighed 120-150 g were used. Animals were injected with 100 µl of CFA (100 mg/ml) into the right tibiotarsal joint. After complete establishment of arthritis on day 14, animals were placed in 6 groups (five animals in each); groups II, III, IV and V AIA rats were injected intraperitoneally with 100, 200, 400 and 800 mg/kg of SAE, respectively. Groups I and VI were injected with equal amounts of normal saline and dexamethasone (mg/kg body weight) as a standard reference, respectively. Saffron, normal saline and dexamethasone injections were repeated four more times every other day (days 2, 4, 6 and 8). Measurement of the paws footpad and tibiotarsal joints diameters on the injected and not-injected paws were performed every three days. An arthritis index was assigned to each animal based on footpad paw diameter, ankle joint diameter and hyperalgesia. Results: Our results show that SAE at 400 mg/ml has the best effect on reducing the footpad and tibiotarsal joint diameters, arthritis indexes and limitations of movement in CFA induced arthritis rats compared to non treated group. However this reduction is not as significant as dexamethasone treated group. Conclusion: It seems that aqueous extract of saffron stigma can alleviate the inflammatory condition of CFA induced arthritis. We showed the effect of saffron aqueous extract on improving disease condition in adjuvant-induced arthritis in rats.

Keywords: Saffron, Arthritis, Wistar Rat

#### 741. *Achillea millefolium* Hydro-Alcoholic Extract Downregulates IL-1 $\beta$ Gene Expression in Streptozotocin-Induced Diabetic Rats

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Background: Type I diabetes mellitus is an autoimmune disease that causes selective destruction of insulin producing  $\beta$ -cells. Interleukin-1 (IL-1 $\beta$ ) has been implicated as an effector molecule of  $\beta$ -cell destruction in autoimmune diabetes. IL-1 $\beta$  inhibits insulin secretion from pancreatic  $\beta$ -cells by stimulating the expression of inducible nitric oxide synthase (iNOS) that generates the free radical nitric oxide. Protective effect of *Achillea millefolium* extract which is traditionally used as a hypoglycemic agent may be the result of antioxidant action of its flavonoid content. The effect of *Achillea millefolium* extract on IL-1 $\beta$  gene expression in STZ-induced diabetic rats was investigated. Materials and Methods: The rats were divided into normal, diabetes mellitus (DM), and *Achillea millefolium*-treated groups (each group, n=7). Diabetes was induced by intraperitoneal injection of streptozotocin into wistar rats. Streptozotocin-induced diabetic rats were treated with IP injection of *Achillea millefolium* extract (100mg/kg) for 14 days. The expression level of IL-1 $\beta$  was examined in rat pancreas using real-time RT-PCR. Results and conclusion: In diabetic rats, elevated fasting blood glucose concentration was significantly ( $p<0.05$ ) reduced following administration of *Achillea millefolium* extract. The mRNA expression level of IL-1 $\beta$  gene was significantly ( $p<0.05$ ) increased in diabetic rats. Treatment with extract caused a significant ( $p<0.05$ ) reduction in IL-1 $\beta$  gene expression. In conclusion, *Achillea millefolium* extract showed a beneficial effect on  $\beta$ -cells at least in part due to its antioxidative effect through which it down regulates the IL-1 $\beta$  gene expression.

Keywords: *Achillea millefolium*, IL-1 $\beta$ , Streptozotocin-induced Diabetic Rats

#### 742. *Achillea millefolium* Inflorescence Aqueous Extract Attenuates Cyclophosphamide-induced Oxidative Apoptosis in Rat Testis: Immunohistochemical Evidence

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Background: Cyclophosphamide (CP) is a commonly used agent in cancer chemotherapy and immunosuppression. However, CP-induced apoptosis in spermatogenic cells may result in oligospermia and azoospermia, which limits CP clinical application. This study was conducted to assess the possible protective efficacy of *Achillea millefolium*, a medicinal plant with anti-oxidant property, on CP-induced apoptotic effects in rat testes. Materials and Methods: Male Wistar rats were categorized into four groups. Two groups of rats were administered CP at a dose of 5 mg in 5 ml saline/kg per day for 28 days by oral gavages. One of these groups received *Achillea millefolium* Inflorescences aqueous extract at a dose of 1.2 g/kg body weight/day orally four hours after CP administration. A vehicle treated control group and an *Achillea* control group were also included. The immunohistochemistry was used to determine the expression of caspase-3 in testicular tissue samples. Results: After 28 days, rats treated with CP alone displayed increase of cleaved caspase-3 abundance, while *Achillea* aqueous extract coadministration could effectively prevent nearly this abnormality. Conclusion: These findings provide evidence that *Achillea* would offset the apoptotic impact imposed by CP, and may attenuate the testicular toxicity of CP in clinical practice.

Keyword: *Achillea millefolium*, Cyclophosphamide, Apoptosis, Testis

#### 743. Preparation of Turmeric Ointment Based on Traditional Iranian Medicine References and Investigation of its Efficacy on Rheumatoid Arthritis in Rat

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Background: Rheumatoid arthritis is inflammation of one or more joints which causes joint pain, swelling, stiffness and limited movement. Corticosteroids and NSAIDs are usually used for treatment but they have some limitations including lack of efficacy and high incidence of side effects. In traditional medicine, *Curcuma longa* (turmeric) is used as a powerful anti-inflammatory agent in different inflammatory disorders. In traditional Iranian medicine, combination of turmeric and egg yolk is used for inflammations due to trauma and strains. The major component of egg yolk is lecithin. In the present study, in order to evaluate the efficacy of turmeric and egg yolk combination in treatment of inflammation, the effect of turmeric ointment with or without lecithin on Rheumatoid arthritis has been studied in rat. Materials and Methods: Powder of turmeric was extracted by using methanol 80% and maceration method. Two types of ointments were prepared by using vaseline, eucerine, paraffin and bees wax. The first type contained turmeric extract alone and second one was prepared by turmeric extract and lecithin. Inflammation was induced by Freund's adjuvant subcutaneous injection in right tibiotarsal joint in rats. The ointments were applied on the joints for 20 days. Arthritis index and joint diameter were recorded before and after of 20 day's treatment and compared with control group. Serum TNF- $\alpha$  was measured at the last day of treatment. Results: The results showed the variations in the results of joint diameter and arthritis index which cannot be interpreted. While, the results of TNF- $\alpha$  measurement established that all products had anti-inflammatory activity with no significant differences between turmeric alone and turmeric with lecithin groups ( $P>0.05$ ). Moreover, no differences were observed between 2.5% and 5% turmeric formulations. Conclusion: It is concluded that turmeric ointment has reasonable anti-inflammatory activity in rheumatoid arthritis model in rat. Since, lecithin had no effect on anti-inflammatory activity of turmeric and 2.5% and 5% turmeric groups showed the same activity, therefore, the product containing 2.5% turmeric extract is suggested as the best formulation among the tested products.

Keywords: Topical dosage form, *Curcuma longa*, Rheumatoid arthritis, Traditional medicine

### Poster Presentation

#### 744. Suppressive Effect of Peppermint Extract on IL-13 Production

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Background: Medicinal plants have been broadly used in treatment of various diseases. Mints are a group of plants belonging to Labiatae family have anti-bacterial, anti-tumoral and anti-inflammatory effects. Peppermint is a mint species extensively used in therapy of several disorders such as common cold and bronchitis. The anti-bronchospasmodic and anti-allergic effects of peppermint have also been shown. Elevation of

interleukin-13 (IL-13) (a Th2 cytokine profile) level is a well known indicator of allergy. In the present study the effect of aqueous extract of peppermint on IL-13 production in human peripheral blood mononuclear cells (hPBMCs) has been assessed in vitro. Materials and Methods: The hPBMCs were isolated from the venous blood of healthy volunteers by ficoll-hypaque-gradient centrifugation. Then the PBMCs were cultured in complete RPMI medium. The cells at logarithmic growth phase, were incubated with different concentrations of aqueous extract of peppermint leaves (0.01-10 mg/ml) in triplicate for 24 hours. Afterward the cell culture supernats were collected and the IL-13 concentration was measured using a standard ELISA kit. Results: Peppermint aqueous extract significantly decreased the IL-13 production in hPBMCs dose-dependently. Conclusions: The results of this study indicate that peppermint aqueous extract down-regulates the production of IL-13 in hPBMCs. Regarding the important role of IL-13 in atopic allergy, the anti-allergic activity of peppermint and also its inhibitory effect on bronchospasm (a symptom of respiratory allergy), may be partly due to its inhibitory effects on IL-13 production.

Keywords: Peppermint, IL-13, hPBMCs

#### 745. Recombinant Production of an Anti-HIV Lectin, Griffithsin, with Two Leader Peptide in Soybean

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Background: Plant bioreactor, also called molecular farming, has enormous potential to produce recombinant proteins infinitely. Plant bioreactor could be a safe, economic and convenient production system, and can be widely applied in industries and agricultures, especially in the life science and pharmaceutical industry. The application of transgenic plant in the production of vaccines, antibodies and pharmaceutical proteins has become a hot point in the plant genetic engineering in recent years. Griffithsin (GRFT) is a anti-HIV lectin which isolated from red alga, *Griffithsia sp.* The potent anti-viral activity of GRFT against both laboratory and primary isolates of HIV at picomolar concentrations makes this protein an attractive candidate microbicide to prevent sexual transmission of HIV. Materials and Methods: Codon optimization of GRFT based on soybean's genome was done. Three vectors were designed, one used KDEL sequence after for protein targeting in ER, second extensin sequence before GRFT sequence for protein targeting in apoplasmic area and third without any leader peptide. For both RNA extraction and cDNA synthesis Biozol method was used. The level of transcription and protein expression was obtained by real time PCR and ELISA method respectively. Results: The results of real time PCR confirmed that recombinant GRFT was expressed 11.819, 7.807 and 67.72 times more than what is expressed in control plant respectively in KDEL construction, extensin construction and in no leader peptide construction. Also, ELISA results showed respectively 32 and 29 times in extensin and KDEL leader peptide more protein retention than when no leader peptide was used. Conclusion: Our results suggested that although the level of expression between KDEL and extensin construction is statistically significant but protein retention in both ER and apoplasmic area is same. On the basis of the results of this work and similar studies we could propose ER as a safe place for this protein retention.

Keywords: Recombinant, Griffithsin, Soybean

#### 746. Inhibition of Pro-inflammatory Cytokines Production by Ethyl Acetate Extract of *Scrophularia striata*

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Background: The aim of this study was to investigate the effect of ethyl acetate (EA) extract of *Scrophularia striata* (*S. striata*) on the production of pro-inflammatory cytokines. Materials and Methods: Mouse peritoneal macrophages cultured with/ without different concentrations (1, 10, 50, 100 and 200 µg/ml) of EA extract of *S. striata* and 2 µg/ml lipopolysaccharide (LPS) for 24 h. In order to recognize chemical components of the extract, Thin Layer Chromatography (TLC) was used. Production of pro-inflammatory cytokines including Interleukin 1β (IL-1β) and Tumor necrosis factor - α (TNF-α) were examined using ELISA. Results: Phytochemical assay showed main components; including phenyl propanoids, phenolic compounds and flavonoids were presented in the extract. EA extract in concentrations of (10- 200 µg/ml) significantly inhibited pro-inflammatory cytokines (IL-1β and TNF-α) production by LPS stimulated peritoneal macrophages. Conclusion: The effect of EA extract of *S. striata* on pro-inflammatory cytokines may lead to improving inflammatory diseases, and possibly acts as anti-inflammatory agent via immunomodulatory effect.

Keywords: Pro-inflammatory, Ethyl acetate extract, *Scrophularia striata*

#### 747. The Effect of Garlic Consumption on Blood Total Antioxidant Status and Some Biochemical/Hematological Parameters in Rat

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Background: The effect of garlic supplementation on blood serum total antioxidant status (TAS), nitrate and routine biochemical/hematological parameters were investigated in rats. Materials and Methods: A total of 30 rats were randomly divided into two groups. Each of fifteen rats of garlic group was fed 600mg/kg garlic solution in water and controls received distilled water by gavage. After garlic consumption for one month, serum total antioxidant, nitrate and some routine biochemical/hematological tests including serum lipids parameters, blood sugar, complete blood count (CBC), Hemoglobin and so on were performed. Results: the mean levels of TAS (1.18±0.11 mmol/L) and nitrate (1.44±0.27 µmol/L) in the blood serum of rats which treated with garlic significantly (p<0.01), (p<0.05), increased in compare with the controls (0.77±0.10 mmol/L), (0.78±0.06 µmol/L) respectively. But there were no meaningful differences with regard to the routine biochemical/hematological parameters. Conclusion: this investigation confirms that garlic has antioxidant property and may have no effect on lipids profile and total blood cell counts.

Keywords: Garlic, Blood Serum Total Antioxidant Status, Rat

#### 748. In vitro Cytotoxic Effect of *Cuscuta chinensis* whole extract on Human Acute Lymphoblastic Leukemia (ALL) Cell Line

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