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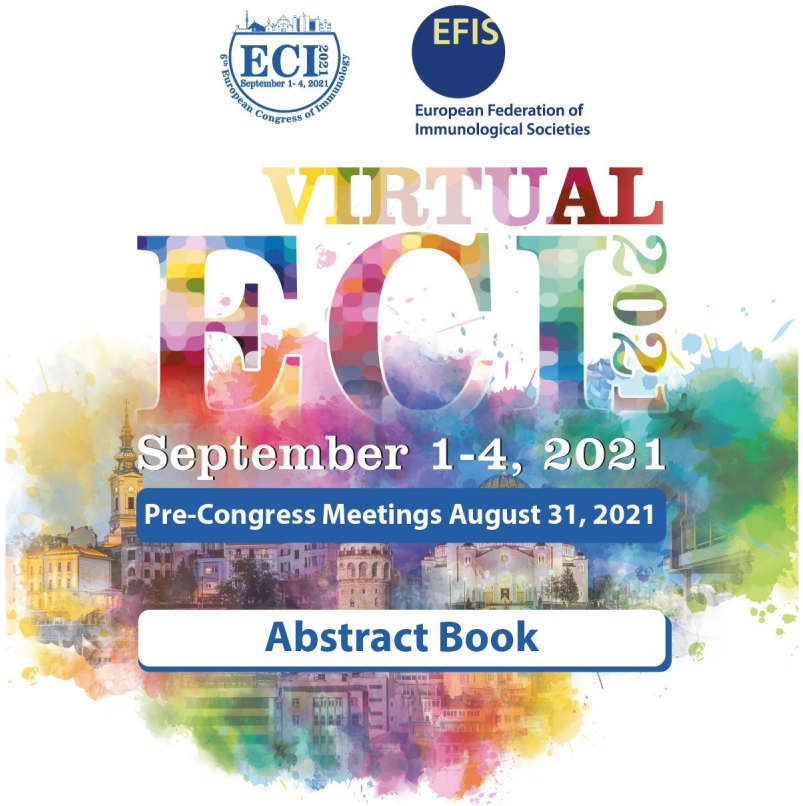

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6th European Congress of Immunology

1-4 September 2021

Virtual meeting

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POSTER PRESENTATIONS

P-0625

(Bio)Chemical characterization of moss *Hypnum cupressiforme* extracts as potential immunomodulators

Tanja M Lunić[1], Mariana M Oalđe[2], Marija R Mandić[1], Aneta D Sabovljević[2], Marko S Sabovljević[2], Uroš M Gašić[3], Sonja N Duletić Laušević[2], Bojan Đ Božić[1], Biljana Đ Božić Nedeljković[1]

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In this study, we aimed to examine the chemical composition and biochemical activities (antioxidant, antidiabetic, and antineurodegenerative) of seasonal aspects (spring, summer, autumn) of moss *Hypnum cupressiforme* extracts. The extracts were prepared using Soxhlet extractor. Chemical characterization of extracts was performed via spectrophotometric assays and LC-MS. The antioxidant activity of the extracts was determined by DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate), total reduction power, and β-carotene bleaching assays. Extracts were analyzed for their inhibitory activity on α-glucosidase, α-amylase, acetylcholinesterase, and tyrosinase. The chemical analysis of the moss extracts revealed the presence of flavonoids, phenolic acids, and triterpenoids. The highest concentration of these compounds was found in the moss from summer season, and was significantly higher in comparison to the spring and autumn aspects. Major compounds identified by LC-MS in *H. cupressiforme* extracts were kaempferol, *p*-hydroxybenzoic, protocatechuic, *p*-coumaric, gallic, and caffeic acid. According to biochemical assays investigated extracts exhibited significant antineurodegenerative potential, evaluated by the capacity to inhibit acetylcholinesterase and tyrosinase. Among the examined aspects, summer and autumn were found particularly efficient, especially at the lowest tested concentrations. Regarding β-carotene bleaching test, the summer aspect showed the best activity at the highest concentration, while the spring aspect was found more efficient at the lowest investigated concentration. The obtained data suggest that the most favorable season in terms of chemical composition and biochemical characteristics of moss *H. cupressiforme* is summer. This moss is a promising source of biologically active compounds that may be useful in the treatment of various pathological conditions.

Keywords: Immune regulation and therapy, immunopharmacology, inflammatory disease

P-0626

A nonsense mutation in DIAPH1 gene presents with major T cell defects

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Diaphanous related formin 1 (DIAPH1) is a member of formin family proteins and is an important regulator of actin polymerization and microtubule stability. DIAPH1 loss of function mutations are associated seizures, cortical blindness, and microcephaly syndrome (SCBMS). Very recently DIAPH1 mutations have been linked to a combined immunodeficiency. In this study we characterized a patients' lymphocytes presented with SCBMS and symptoms of immunodeficiency. Next generation sequencing of samples from a 10-year-old female patient, who was diagnosed with SCBMS revealed a nonsense mutation in DIAPH1 gene P.R31*. The patient has been followed up by pediatric neurology and pediatric immunology due to epilepsy, autism, cortical blindness and lymphopenia and had frequent infections (pneumonia and bronchiolitis), frequent ear drainage and otitis history. Peripheral blood mononuclear cells were studied with respect to cell proliferation, NK cell cytotoxicity, IL-2-mediated STAT5 phosphorylation, and induced Treg cell generation *ex vivo* and examined on FACSAria III. DIAPH1 deficient T cells showed proliferation defects in response to both CD3/28 and PHA stimulation. NK cells had cytotoxicity defects against K562 cells. As expected, DIAPH1 deficient PBMCs had migration in a trans-well plate. In addition, generation of Treg cells from naïve T cells was impaired. This impairment had more to do with cell expansion rather than conversion of naïve T cells into Foxp3+ T cells. DIAPH1 deficient PBMCs had impaired IL-2-mediated STAT5 phosphorylation. Lastly, DIAPH1 Mutant PBMCs have reduced CD4/CD8 ratio. Our data reveal that DIAPH deficiency results in major T cell defects in patients.

Keywords: Molecular immunology, NK cells, adaptive immunity, immunodeficiency

P-0627

Immunomodulatory properties of extracts of moss *Hypnum cupressiforme* from various seasons

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In this study, we aimed to examine the immunomodulatory potential (anti-neuroinflammatory/neuroprotective and antitumor activities) of various seasonal aspects (spring, summer, autumn) of moss *Hypnum cupressiforme* extracts. Moss extracts have been obtained using solvents of different polarities and their mixtures applying Soxhlet extractor. Activities of the extracts were tested on cell lines MRC-5, BV2, SH-SY5Y, HCT-116, and MDA-MB-231, for potential anti-neuroinflammatory and antitumor activities. The impact of extracts on cell viability/metabolic activity was measured by MTT assay, while the influence on the production of ROS and NO was determined using NBT and Griess assays, respectively. The biocompatibility of extracts was confirmed using MRC-5 cells. Extracts of moss from spring season have shown significant antiproliferative activity against MDA-MB-231 cell line (inhibition rate ~50%), as well as anti-neuroinflammatory activity. Furthermore, the effects of seasonal variation (spring, summer, autumn) on the immunomodulatory potential of ethyl-acetate extract were examined. Summer aspect of moss has led to the greatest reduction of ROS production, while autumn aspect had the greatest impact on the reduction of NO production by LPS-activated BV2 cells. Additionally, the supernatant-transfer model proved that treatment with moss extracts from summer and autumn seasons of LPS-activated BV2 cells exhibit neuroprotective activity towards SH-SY5Y neurons. The obtained data suggest that extracts from moss *H. cupressiforme* possess significant immunomodulatory potential evaluated *in vitro*, which may be useful in conditions such as Alzheimer's disease, Parkinson's disease, and breast cancer. In terms of anti-neuroinflammatory/neuroprotective activity, summer and autumn were characterized as the most favorable seasons.

Keywords: Cancer immunology, immunotherapy, inflammatory disease