EP336

Anticancer activity of Orobanche crenata Forssk. leaves extracts against different human cancer cell lines

F. D'Angeli[1], C. Genovese[2,3], V. D'Argenio[1], M. Leo[1], A. Spila[1,4], P. Ferroni[1,4], F. Guadagni[1,4]

[1]*Dep. of Promotion of Human Sciences and Quality of Life, San Raffaele Roma Open University, 00166 Rome, Italy*

[2]*Dep. of Medicine and Surgery, "Kore" University of Enna, Contrada Santa Panasia, 94100, Enna, Italy*

[3]*Nacture S.r.l, Spin-off University of Catania, Via Santa Sofia 97, 95123 Catania, Italy.*

[4]*InterInstitutional Multidisciplinary Biobank (BioBIM), IRCCS San Raffaele, 00166 Rome, Italy*

Cancer is the second cause of death worldwide and it was estimated to become the leading in 2060. Cancer treatment is mainly based on chemotherapy, which is associated with a large variety of side effects. The use of natural adjuvants endowed with anticancer properties is considered a good strategy to reduce the chemotherapy dose, thus increasing its tolerability. In our previous work, we proved the anticancer activity of O. crenata leaves acetonic extract (OCLAE) against human breast cancer cell line MCF-7. The Human Foreskin Fibroblasts (HFF-1) cell line was used as the control cell line. The cytotoxic activity of the extract was compared to the standard drug Doxorubicin. MCF-7 cells were treated with increasing concentrations of OCLAE (75-1200mg/mL) for 24h. The cytotoxic effect of the extract was evaluated by MTT and LDH assays. The extract induced a significant reduction of MCF-7 cell viability and a significant increase in LDH release. These effects were correlated to the antioxidant properties of the extract. The obtained results led us to further explore the anticancer properties of O. crenata. In the present study, we evaluated the anticancer activity of O. crenata leaves aqueous extract (OCLAqE) against human colorectal cancer cell lines Caco-2 and HCT-116. Cisplatin, a potent chemotherapeutic agent, was used as the standard drug. The effect of both extract and Cisplatin was also tested on non-cancerous Human Dermal Fibroblast (HDF). Caco-2 and HCT-116 cell lines were exposed to increasing amounts of OCLAqE (10-160mg/mL) or Cisplatin (0.1 to 100 μM) for 24h, 48h, and 72h. The anticancer activity was evaluated by MTT assay. The potential synergistic effect between the two agents was assessed through MTT and Annexin V/ Propidium Iodide assays. The effect of the extract on ROS levels was revealed using 2,7-dichlorodihydrofluorescein diacetate. Finally, by UPLC-Ms/Ms the chemical profile of OCLAqE was obtained. The extract affected Caco-2 and HCT-116 cell viability at all time points. However, it dose-dependently reduced Caco-2 cell viability, starting from 40mg/mL. The treatment of HDF with the extract induced a significant reduction of cell viability only at the highest tested concentration (160mg/mL). Co-treatment of extract (80μg/ml) with a subtoxic concentration of cisplatin (1 μM) potentiated the drug effect. The extract was also able to modulate ROS production. Finally, the chemical analysis detected different polyphenolic compounds that could mediate the observed effects. These findings highlighted the anticancer properties of OCLAqE, thus suggesting its potential value as a promising therapeutic adjuvant.

EP337

Health and nutritional biomarkers in honeybees: opportunities and challenges under field conditions

G. Isani[1], C. Rudelli [1], E. Bellei[2], G. Andreani[1]

[1]*Dept. of Veterinary Medical Sciences, University of Bologna, Bologna, Italy*

[2]*Dept. of Surgery, Medicine, Dentistry and Morphological Sciences, Proteomic lab, University of Modena and Reggio Emilia, Modena, Italy*

The decline of honeybee (Apis mellifera) populations has negative consequences not only for agriculture and beekeeping, but also for ecosystems. In human and veterinary medicine, proteomics and metabolomics provide valuable biomarkers to assess the health and nutritional status of organisms. In honeybees, the application of these techniques is still in its infancy and remains underexplored from a clinical perspective. This study aims to investigate the most abundant proteins of honeybee hemolymph as potential biomarkers of health and nutritional status at the colony level. In addition, an untargeted metabolomics-based approach was applied to honeybee extracts.

Samples of hemolymph were collected from honeybees in different apiaries in the province of Bologna in different periods of the year, from April to November. Hemolymph proteins were separated and quantified by 1D SDS-PAGE. Honeybee cytosolic extracts were fractionated using size exclusion chromatography (SEC) and metabolites were analyzed in fractions using mass spectrometry (Orbitrap Exploris 480, Thermo Fisher).

The five most abundant hemolymph proteins, namely vitellogenin, apolipophorin I and II, transferrin, and hexamerin 70a, represent a panel of biomarkers related to key metabolic processes. These proteins are subject to interesting variations depending on many physiological and environmental factors, including the honeybees' age (nurse bees had the highest vitellogenin concentration compared to the other two sub-castes), the season (in November, a peak of vitellogenin and transferrin concentration was observed in winter bees), and the presence of parasites (in bees parasitized by Varroa, a decrease of vitellogenin, apolipophorin II, transferrin, and hexamerin 70a was detected). One hundred and ninety-eight different pathways and more than 2000 metabolites were identified. The most abundant metabolites belonged to the flavone pathway, followed by the lipoxygenase pathway. Many metabolites were of plant origin and may be related to the environmental availability of nectar and pollen, which in turn are essential for honeybee nutrition, suggesting a possible role as biomarkers of nutritional status.

This research was in part funded by Agritech National Research Center and received funding from the European Union Next-Generation EU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR)–MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4–D.D. 1032 17/06/2022, CN00000022).