EVALUATION OF THE MUTAGENIC AND ANTIMUTAGENIC ACTIVITY OF AMMODAUCUS LEUCOTRICHUS AND PERILLALDEHYDE BY FLOW CYTOMETRIC ANALYSIS

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Introduction: Numerous experimental and epidemiological evidences show how many naturally occurring substances modulate cellular and molecular targets critical in the development of numerous pathologies. However, their use require an exhaustive and complete characterization of their toxicological profile to demonstrate the safety of use, in particular in terms of mutagenicity. In the present study we evaluated on a human lymphoblastoid cell line (TK6) the cytotoxic and genotoxic activity of the ethanolic extract of Ammodaucus leucotrichus and of Perillaldehyde, the aromatic substance present in higher percentage.

Materials and methods: TK6cells were treated for 3h with Ammodaucus leucotrichus 0.1mg/mL, Perillaldehyde 0.05mM, two known clastogens Mitomycin-C and Cytosine Arabinoside or two known aneuploidogens Colchicina and Vinblastina, followed by 21h of recovery in fresh medium. Subsequently, the cytotoxicity, in terms of cell viability and induction of apoptosis was analyzed by Guava Via Count reagent and Guava Nexin reagent, respectively. The mutagenic activity was measured in terms of micronuclei frequency increasing in treated cultures respect to the controls by flow cytometric analysis using an automated protocol published by Lenzi et al. All analyzes were performed by flow cytometer Guava easyCyte 5HT.

Results: The results obtained show how the viability remains abundantly above 80% in all treated cultures and the induction of apoptosis was comparable in treated cultures and in the controls.
The MN test, did not reveal a mutagenic potential for Ammodaucus leucotrichus and Perillaldehyde.
On the basis of obtained results we decided to investigate a possible antimutagenic activity of the extract and molecule. For this purpose, TK6cells were treated with the four known mutagens in the absence or presence of Ammodaucus leucotricus and Perillaldehyde (co-treatment). The flow cytometric analysis allowed to observe a statistically significant decrease in the frequency of MN in presence of Ammodaucus leucotricus or Perillaldehyde compared to the cultures treated with clastogens alone. On the contrary, neither Ammodaucus leucotricus nor Perillaldehyde show antimutagenic activity against the two aneuploidogens.

Conclusions: Ammodaucus leucotrichus and Perillaldehyde at the concentrations tested did not prove either cytotoxic or mutagenic effects. Moreover, the extract and molecule were able to exert antimutagenic activity, showing a different behavior versus clastogens and aneuploidogens, and in particular towards cytosine Arabinoside.
The ability to counteract the mutagenicity activity has an important therapeutic value, in fact it represents a possible chemopreventive mechanism.