

concentration of OPCs and antiradical activity in tests "in vitro". More than 40 samples of food and medicinal plants were investigated. Total polyphenolics were determined employing Folin-Ciocalteu photometric method, sum of OPC — by Bate-Smith acid butanol assay, free radical scavenging activity (FRSA) — by the DPPH test.

The study was carried out to search the antioxidant ingredients for diet enrichment. No direct correlation between the antioxidant in vitro tests and the content of polyphenolic compounds and proanthocyanidins has been found. On the basis of the above test results the feasibility of using products based on cinnamon, red beans, cocoa and blueberry for diet enrichment has been shown.

IN VITRO CYTOTOXIC AND ANTIOXIDANT ACTIVITIES OF SOME PLANT EXTRACTS ON DIFFERENT HUMAN CANCER CELL LINES

© **El-Hallouty Salwa M.¹, Khalifa Noha S.², Barakat H. M.², Salim Dina²**

¹Pharmacognosy Department, National Research Centre, Giza, Cairo, Egypt

²Botany Department, Ain Shams University, Cairo, Egypt

This study involves a bioassay screening of 22 methanol extracts from 19 plants that are commonly used in Egypt for many purposes based on their ethnic values. The aim of this study is to evaluate the cytotoxic and antioxidant activities of 22 methanol extracts. The methanol plant extracts were tested *in vitro* against four human cancer cell lines (by using MTT method) to determine their cytotoxic effect. The cell viability was examined after 24 h exposure to 100 µg/ml of the extract in the medium. Negative (dimethyl sulfoxide) and positive (*Annonacherimolia* methanol extract) controls were simultaneously used. Moreover, the antioxidant effect was determined using DPPH assay. Extracts showing cytotoxic and antioxidant activities were further subjected to determine their (lethal concentration) LC₅₀ and LC₉₀ values. Confirmed *in vitro* cytotoxic activity was found in four plant extracts. In HepG-2, *Cympopogonproximus*, *Perseaamericana* fruits and *Vignaunguiculata* seeds showed LC₅₀=57.4, 13.3 and 56.4, respectively. In

A-549, *Perseaamericana* fruits and *Tabernamontana-divaricata* leaves showed LC₅₀=35.4 and 70.7, respectively. In HT-29, *Cympopogonproximus*, *Perseaamericana* fruits and *Tabernamontana-divaricata* leaves showed LC₅₀=58.6, 22 and 67.5, respectively. In MCF-7, *Perseaamericana* fruits showed LC₅₀=54.5. *In vitro* antioxidant activity was confirmed in three plant extracts *Ceratoniasiliqua* leaves, *Perseaamericana* leaves, *Abrusprecatorius* seeds showed LC₅₀=10.6, 46.5 and 25.7, respectively. 5 Extracts out of the 22 studied methanol extracts exhibited potent antioxidant properties when tested at the concentration of 100 ppm against DPPH. The methanol extract of *Persea americana* leaves, *Abrus precatorius* seeds, *Ceratoniasiliqua* leaves had LC₅₀=46.5, 25.7, 10.6 µg/ml, respectively Table 4. The methanol extracts of *Ocimum basilicum* and *Hilanthus annuus* had lost their activity below 100 ppm. The plants under study may represent promising natural sources for cytotoxic and antioxidant drug discovery.

PHYTOCHEMICAL SCREENING AND ANTIOXIDANT CAPACITIES OF SOME COMPOSITAE PLANTS

© **Ergene B.¹, Bahadır Acıkara Ö.¹, Bakar F.², Saltan G., Nebioğlu S.²**

¹Ankara University Faculty of Pharmacy, Department of Pharmacognosy, Ankara, Turkey

²Ankara University Faculty of Pharmacy, Department of Biochemistry, Ankara, Turkey

Asteraceae is one of the families which include many members containing phenolic compounds (1). The beneficial effects of many of the phenolics in human health have been attributed to their reactive oxygen and nitrogen scavenging and antioxidant capacity. The consumption of vegetables, fruits and flavonoid-rich beverages has been reported to prevent against neurodegenerative diseases, cancer and aging (2). Phenolic compounds have antioxidant potential due to their tendency to act as reducing agents, hydrogen donors, sin-

glet oxygen quenchers and metal chelators, chelating agents which can bind metal ions, could be added to enhance the activity of natural preservatives in food stuffs (2, 3, 4). In this study, the antioxidant activity of three members of Asteraceae family; *Crepis foetida* subsp. *rhoeadifolia*, *Leontodon crispus* var. *asper* and *Pilosella hoppeana* subsp. *testimonialis* were evaluated using 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging assay and measuring malondialdehyde measuring levels. In DPPH free radical scavenging activity as-

say; the extract prepared from the roots of *P. hoppeana* subsp. *testimonialis* was found to be the most active one with IC_{50} values of 0,231 M. The MDA level of this sample was found as 3.91 nmol/ml. Phytochemical screening of the tested extracts was performed by RP-HPLC using some flavonoids and phenolic acids as standards.

References: (1) Wojdylo A., Oszmianski J., Czemerys R. Antioxidant and phenolic compounds in 32 selected herbs. *Food Chem.* 105: 940–949 (2007). (2) Erdogan-Orhan I., Sever-Yilmaz B., Altun L., Saltan G. Radical quenching activity, fer-

ric reducing antioxidant power and ferrous ion-chelating capacity of 16 Ballota species and their total phenol and flavonoid contents. *J. Med. Food* 13 (6): 1537–1543 (2010). (3) Pietta P.G. Flavonoids as antioxidants. *J. Nat. Prod.* 63: 1035–1042 (2000). (4) Stanojevic L., Stankovic M., Nikolic V., Nolic L., Ristic D., Canadanovic-Brunet J., Tumbas V. Antioxidant activity and total phenolic and flavonoid contents of *Hieracium pilosella* L. extracts. *Sensors*, 9: 5702–5714 (2009).

DEVELOPMENT AND VALIDATION OF TLC METHOD FOR THE IDENTIFICATION OF EUCALYPTUS LEAVES

© **Evdokimova O. V., Obukhova V. V., Kuzminova L. V.**

JSC "Krasnogorskleksredstva", Krasnogorsk, Moscow region, Russia

According to "Krasnogorskleksredstva" House monograph 42–8000–06 "Eucalyptus leaves" the determination of lipophilic compounds in this herbal medicinal product is carried out by TLC method. This method requires only one reference substance — Sudan III. But it is more reasonable to use two reference substances in TLC analysis that allows to evaluate the suitability of chromatographic system. The purpose of our study was to develop and validate a new identification method for Eucalyptus leaves with use of two reference substances. The study object was Eucalyptus leaves; TLC was chosen as an analytical method for identification of lipophilic compounds profile in the samples. In order to harmonize with leading Pharmacopoeias the mobile phase was changed from toluene — ethyl acetate (95:5) to toluene — ethyl acetate (90:10; according to European Pharmacopoeia 7.001/2008:1320) and the plates were changed from TLC-P-A (Sorbfil, Russia) to TLC Silica gel 60 (aluminium sheets, Merck, Germany). Solutions of Sudan Red G and Sudan III were used as reference solutions. Anisaldehyde solution was used for detection. The following zones were present in daylight in the chromatograms obtained with the reference solutions: the zone with red color (R_f approx. 0.4 that was accepted as $R_s = 1.0$, due to Sudan Red G) and the zone with blue or blue-violet color (R_s approx. 1.4, due to Sudan III). The

following zones of lipophilic compounds were present in daylight in the chromatograms obtained with the test solution: 3 zones with violet color (R_s (by Sudan Red G) approx. 0.2; 0.6; 2.0); zone with dark violet color on start line and others zones could be present. Method validation included evaluation of chromatographic system specificity and suitability. The method specificity was evaluated by coincidence of the different batches chromatographic profiles by the main zones and by compliance of abovementioned profiles with the test solution chromatogram description. The method validation was performed on 6 industrial batches. Chromatographic profiles of the different batches have coincided and been compliant with the test solution chromatogram description. The resolution between zones of Sudan Red G (R_s approx. 1.0) and Sudan III (R_s approx. 1.4) in the chromatogram obtained with the reference solutions has been chosen as a chromatographic system suitability index. The resolution value between the chosen zones must not be less than 1.5. The resolution values that had been calculated by the chromatograms obtained from different batches satisfied the method requirements. The suggested method can be used for identification of Eucalyptus leaves in routine analyses. This method has been included into the draft version of "Krasnogorskleksredstva" House monograph on Eucalyptus leaves.