THERAPEUTICS

NPARR 5(1), 2014-070 Evaluation of antiangiogenic and antiproliferative potential of the organic extract of green algae Chlorella pyrenoidosa

Algae isolates obtained from fresh and marine resources could be one of the richest sources of novel bioactive secondary metabolites expected to have pharmaceutical significance for new drug development. This study was conducted to evaluate the antiangiogenic and antiproliferative activity of Chlorella pyrenoidosa in experimental models of angiogenesis and by MTT assay. Lyophilized extract of C. pyrenoidosa was extracted using dichloromethane/methanol (2:1), concentrated and vacuum evaporated to obtain the dried extract. The crude extract was evaluated in the vascular endothelial growth factor (VEGF)-induced angiogenesis in in ovo chick chorioallantoic membrane assay (CAM) at various concentrations (n = 8) using thalidomide and normal saline as positive and untreated control groups, respectively. The crude extract was also subjected to the antiangiogenic activity in the silver nitrate/potassium nitrate cautery model of corneal neovascularization (CN) in rats where topical bevacizumab was used as a positive control. The vasculature was photographed and blood vessel density was quantified using Aphelion imaging software. The extract was also evaluated for its anti proliferative activity by microculture tetrazolium test (MTT) assay using HeLa cancer cell line (ATCC). VEGF increased the blood vessel density by 220% as compared to normal and thalidomide treatment decreased it to 67.2% in in ovo assay. In the in-vivo CN model, the mean neovascular density in the control group, the C. pyrenoidosa extract and bevacizumab group were found to be 100%, 59.02%, and 32.20%, respectively. The Chlorella pyrenoidosa extract negatively affected the viability of HeLa cells. An IC₅₀ value of the extract was 570 µg/ml respectively. A significant antiangiogenic activity was observed against VEGF-induced neovascularization and antiproliferative activity by MTT assay. In this study, it could be attributed that the activity may be due to the presence of secondary metabolites in the C. pyrenoidosa extract [Mahender Kyadari, Tasneem Fatma, Rajvardhan Azad and Thirumurthy Velpandian (Department of Ocular Pharmacology and Pharmacy, Dr. Rajendra Prasad Centre for Ophthalmic Sciences, All India Institute of Medical Sciences, New Delhi India), Indian J Pharmacol, 2013 45(6),569-574].

NPARR 5(1), 2014-071 The polyphenol-rich baobab fruit (Adansonia digitata L.) reduces starch digestion and glycemic response in humans

The baobab fruit (Adansonia digitata L.) is found throughout regions of Africa and is becoming increasingly recognized for its high nutrient and polyphenol content. Polyphenols have been beneficial for their effects on reducing the glycemic response (GR) and for improving various other metabolic parameters. Based on previous research, it was hypothesized that the baobab fruit extract would reduce starch digestion in vitro and would show potential for reducing the GR and for increasing satiety and diet-induced thermogenesis in humans. Six extracts of baobab from 6 different locations in Africa were measured for their antioxidant and polyphenol content using the ferric ion–reducing antioxidant power and the Folin-Ciocalteu methods, respectively. Baobab extract was baked into white bread at different doses to determine the optimal dose for reducing starch breakdown and sugar release from white bread after an in vitro digestion procedure. In vivo, baobab extract was consumed in solution at both a low-dose (18.5 g) and a high-dose (37 g) aqueous drink in 250 mL of water along with white bread, and resulting GR, satiety, and postprandial energy expenditure were measured. All extracts in this study were shown to be good sources of...
polyphenols. Baobab fruit extract added to white bread at 1.88 % significantly (P < .05) reduced rapidly digestible starch from white bread samples. In vivo, the baobab fruit extract at both low and high doses significantly (P < .05) reduced GR, although there was no significant effect on satiety or on energy expenditure [Shelly A. Coe, Miriam Clegg, Mar Armengol and Lisa Ryan* (Functional Food Centre, Oxford Brookes University, Gipsy Lane, Oxford, OX3 0BP, UK) Nutrition Research, 2013, 33(11), 888–896].

**NPARR 5(1), 2014-072 Antioxidant and anti-inflammatory activities of six flavonoids separated from licorice**

Licorice, the roots and rhizomes of several *Glycyrrhiza* species (Leguminosae), is an important natural sweetening agent and a widely used herbal medicine. In this work, six flavonoids, 5-(1,1-dimethylallyl)-3,4,4′-trihydroxy-2-methoxychalcone (1), licochalcone B (2), licochalcone A (3), echinatin (4), glycycoumarin (5) and glyurallin B (6), were isolated from the extracts of licorice (*Glycyrrhiza inflata* and *Glycyrrhiza uralensis*). Their structures were elucidated using various spectroscopic methods. To our knowledge, compound 1 was isolated from natural plants for the first time. All the isolates were tested by antioxidant and anti-inflammatory assays. Compounds 2, 4 and 5 showed strong scavenging activity toward the ABTS**+ radical, and compounds 1, 2, 3, 5 and 6 exhibited potent inhibition of lipid peroxidation in rat liver microsomes compared with the reference controls. Compounds 1–4 dose-dependently inhibited LPS induced reactive oxygen species (ROS) production in RAW 264.7 cells. Furthermore, compounds 1–5 were demonstrated to inhibit the production of nitric oxide (NO), interleukin-6 (IL-6) and prostaglandin E2 (PGE2) in LPS-induced macrophage cells. Moreover, the contents of the six compounds, in different *Glycyrrhiza* species, were quantified by HPLC–MS.

Yu Fu, Jun Chen*, Yan-Jing Li, Yun-Feng Zheng and Ping Li (State Key Laboratory of Natural Medicines, China Pharmaceutical University, Nanjing 210009, China) *Food Chemistry*, 2013, 141(2), 1063-1071.

**NPARR 5(1), 2014-073 Pomegranate seed oil prevents bone loss in a mice model of osteoporosis, through osteoblastic stimulation, osteoclastic inhibition and decreased inflammatory status**

In the current context of longer life expectancy, the prevalence of osteoporosis is increasingly important. This is why development of new strategies of prevention is highly suitable. Pomegranate seed oil (PSO) and its major component, punicic acid (a conjugated linolenic acid), have potent anti-inflammatory and antioxidative properties both in vitro and in vivo, two processes strongly involved in osteoporosis establishment. In this study, we demonstrated that PSO consumption (5% of the diet) improved significantly bone mineral density (240.24±11.85 vs. 203.04±34.19 mg/cm³) and prevented trabecular microarchitecture impairment in ovariectomized (OVX) mice C57BL/6J, compared to OVX control animals. Those findings are associated with transcriptional changes in bone tissue, suggesting involvement of both osteoclastogenesis inhibition and osteoblastogenesis improvement. In addition, thanks to an ex vivo experiment, we provided evidence that serum from mice fed PSO (5% by gavage) had the ability to significantly down-regulate the expression of specific osteoclast differentiation markers and RANK-RANKL downstream signaling targets in osteoclast-like cells (RAW264.7) (RANK: negative 0.49-fold vs. control conditions). Moreover, in osteoblast-like cells (MC3T3-E1), it elicited significant increase in alkaline phosphatase activity (+159% at day 7), matrix mineralization (+271% on day 21) and transcriptional levels of major osteoblast lineage markers involving the Wnt/β-catenin signaling pathways. Our data also reveal that PSO inhibited pro-inflammatory factors expression while
stimulating anti-inflammatory ones. These results demonstrate that PSO is highly relevant regarding osteoporosis. Indeed, it offers promising alternatives in the design of new strategies in nutritional management of age-related bone complications. [Mélanie Spilmont, Laurent Léotoing, Marie-Jeanne Davicco, Patrice Lebecque, Sylvie Mercier, Elisabeth Miot-Noirault, Paul Pilet, Laurent Rios, Yohann Wittrant and Véronique Coxam* (INRA, UMR 1019, UNH, CRNH Auvergne, F-63009 Clermont-Ferrand, France), The Journal of Nutritional Biochemistry, 2013, 24(11), 1840–1848]

NPARR 5(1), 2014-074 Screening of phytochemical, physico-chemical and bioactivity of different parts of Acmella oleracea Murr. (Asteraceae), a natural remedy for toothache

Acmella oleracea Murr. (Asteraceae) is a natural source of bioactive secondary metabolites which are responsible for an array of therapeutic properties. Therapeutic effects are mainly due to the secondary metabolites and antioxidant activity present in different parts of the plant. Purpose of this work was to compare the physico-chemical, phytochemical, total phenol content (TPC), total antioxidant capacity (TAC) and cytotoxicity of different parts of A. oleracea. Physico-chemical and phytochemical parameters were performed according to the methods described in WHO guidelines. The TAC and TPC were determined using Ferric Reducing Antioxidant Power assay (FRAP) and modified Folin–Ciocalteu colorimetric method respectively. Presence of a prominent, bright light green color spot (Rf – 0.78) in TLC fingerprints was characteristic for flower extracts. The highest values for all the physico-chemical parameters, TAC and TPC were found in leaves while higher cytotoxicity was exhibited from flower extracts. Order of cytotoxicity was flower > leaf > stem. Presence of higher cytotoxicity in flower and leaf extracts scientifically validates the extensive use of flower and leaf in traditional systems of medicine in Sri Lanka. Information generated are vital important for the quality control and standardization of A. oleracea in order to validate/upgrade the Sri Lankan pharmacopeia. [G.R.P.I. Abeysiri, R.M. Dharmadasa*, D.C. Abeysinghe and K. Samarasinghe(Industrial Technology Institute, BaudhalokaMawatha, Colombo, Sri Lanka) Industrial Crops and Products, 2013, 50, 852–856]