

Euro Biopharma & Ethnopharmacology 2017



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Anti-inflammatory and antileukemia potential of *myrciaria Sp.* ethanol extract

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Most anti-inflammatory and anticancer drugs produced are derived from naturally occurring compounds or their derivatives. There is a constant search for new metabolites from natural origin, particularly from plants, which have potential for efficacious drugs. Myrtaceae, a plant family present in tropical areas, is one of the most studied on biological activities. *Myrciaria* genus belongs to this family and comprises several species; however, few studies have shown their therapeutic potential. The present study aimed to investigate the anti-inflammatory potential and cytotoxicity of the ethanol extract of a species of the *Myrciaria* genus on RAW 267.4 macrophage cells, human peripheral blood mononuclear cells (PBMCs) and Jurkat acute T-lymphocytic leukemia cells. First, RAW 267.4 and PBMCs were treated with increasing concentrations of the extract to assess cytotoxicity for 48 h and 96 h using Alamar blue and Trypan blue exclusion, respectively. In addition, lymphoproliferation was assayed on phytohemagglutinin (PHA)-stimulated PBMCs using MTT method. TNF- α levels were determined by ELISA after RAW 267.4 and PBMCs were pre-incubated with the extract and then challenged with LPS. Protein expression of inflammation-associated markers (NF- κ B, p38 α and p-p38) in LPS-activated RAW 264.7 cells was assessed by Western blot. In addition, the extract was screened for p38 MAPK inhibition using cell-free enzyme activity assay. Later, Jurkat cells were challenged for 24 h with the extract and cytotoxicity was determined by Trypan blue exclusion. After challenging RAW 264.7 and PBMCs with *Myrciaria* sp. extract, a slight decrease ($p < 0.05$) on RAW 264.7 viability was observed with the maximum concentration tested (200 μ g/mL), while PBMCs were not affected by the extract. However, PHA-stimulated PBMCs had a decreased proliferation when cultured with 200 μ g/mL extract. In addition, when both LPS-activated cells were pre-treated with the extract, there were dose-dependent decrease in TNF- α levels ($p < 0.001$), suggesting possible immunomodulatory and anti-inflammatory activities of the extract. Furthermore, Western blotting on RAW 264.7 cells showed that the extract was capable to inhibit LPS-induced NF- κ B activation and p38 phosphorylation. Besides, *Myrciaria* sp. extract presented a great p38 inhibitory activity. On Jurkat cells, the ethanol extract showed cytotoxicity after 24 h, indicating a selectivity. The IC₅₀ of the extract was 127.7 μ g/mL. The results suggest that *Myrciaria* sp. ethanol extract present great biological and is a potent inhibitor of p38 MAPK suggesting an action mechanism with selective activity that can be used in the development of anti-inflammatory and antileukemic drugs or phytomedicines.

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Molecule mechanism of shou tai decoction on URSA by regulating the function of dendritic cells

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Objective: To investigate the role of dendritic cells (DC) in unexplained recurrent spontaneous abortion (URSA) and to study the molecular mechanism of Traditional Chinese Medicine Shou Tai Decoction (STD) on URSA treatment by regulating the function of DC.

Methods: Thirty cases of normal pregnancy women and thirty cases of URSA patients were taken as control group and URSA group respectively. URSA patients were treated with Shou Tai Decoction. PBMC were taken from both control group and URSA group before and after STD administration. The proportion of CD11c+HLA-DR+, CD11c+CD80+ cells and CD11c+CD86+ cells in peripheral blood were measured by flow cytometry. Moreover, the mRNA expression of HLA-DR, CD80, CD86 and Indoleamine2,3-dioxygenase (IDO) in venous blood were detected by RT-PCR assay. The protein expression of IDO was detected by Western Blot. Furthermore, the cytokines, including IL-12p70 and IL-6, in the blood serum were measured by ELISA.

Results: Compared with normal pregnancy women, the proportion of CD11c+HLA-DR+, CD11c+CD80+, CD11c+CD86+ cells and the mRNA expression of HLA-DR, CD80, CD86 of URSA patients in peripheral blood were both increased significantly ($P<0.05$), while the mRNA and protein expression of IDO were decreased markedly ($P<0.05$). Additionally, the level of IL-12p70 and IL-6 in serum of URSA women were significantly increased ($P<0.01$). When compared with URSA patients before STD administration, the proportion of CD11c+HLA-DR+, CD11c+CD80+, CD11c+CD86+ cells and the mRNA expression of HLA-DR, CD80, CD86 decreased significantly after STD administration ($P<0.05$), while the mRNA and protein expression of IDO increased markedly after STD administration ($P<0.05$). Meanwhile, compared with before STD administration, serum protein level of IL-12p70 and IL-6 of URSA patients decreased significantly after STD treatment ($P<0.01$).

Conclusion: The changes of proportion and function of DC were involved in URSA. The regulatory effect of STD on DC proportion and function contribute to the treatment of URSA.

Biography

Xia Li has completed her PhD from Shandong University of Traditional Chinese Medicine and Postdoctoral studies from Shandong University. She is the PI of Laboratory for TCM Immunology and Epigenetics, Institute of Basic Medicine, Shandong Academy of Medical Sciences. She had published more than 10 papers in reputed journals and had been serving as an Editorial Board Member of repute.

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Antimicrobial and antineoplastic activity of fractions derived from geum *Urbanum L*

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Plants are a rich source of biologically active compounds with pleiotropic pharmacological effects. According to ethnopharmacological data the species from the genus *geum* are known for their anti-inflammatory and antioxidant activities. The herbaceous perennial plant species *Geum urbanum L.* had been used since ancient times in the folk medicine for gastro-intestinal diseases, disorders of the liver, biliary tract and uterus. However, the chemical composition and pharmacological properties of *G. urbanum* are still scantily studied. Aim of our study was to investigate the antimicrobial and cytotoxic activity of different fractions from *Geum urbanum L.*, including their bactericidal effect, total phenolic content, antioxidant and cytotoxic activity, as well as apoptosis induction in sensitive human normal and cancer cell lines. For this purpose roots and aerial parts were used to obtain methanol (MeOH) extracts, petroleum ether, ethyl acetate (EtOAc) and n-butanol (n-BuOH) fractions. Minimal inhibitory and bactericidal concentrations (MIC/MBC) were calculated by using broth microdilution method [ISO 20776-1:2006(E)]. The dehydrogenase activity of sensitive Gram-positive bacterial strains was measured based on tetrazolium salt reduction. Bacterial growth rate was determined by time-kill assay in dose-dependent manner. Bacterial cell surface structure was examined by SEM. The cytotoxicity was tested on normal transformed (HEK-294) and tumor (T-24, BC-3C, HeP-G2 and HuT-78) cell lines by MTT- (ISO 10993-5) and CFU assays. Apoptosis induction was observed by fluorescent microscopy and activation of caspase 3. Glutathione reduction was measured by colorimetric enzymatic assay. The statistical analysis of the data was performed with the GraphPad Prism Software. All fractions exhibited antibacterial activity against *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Bacillus cereus*. The EtOAc fraction of aerial parts was characterized by high content of polyphenols, antioxidant activity and lower cytotoxicity on normal transformed cells than on tumor cell lines. It induced apoptosis in the tumor cell line T-24 and inhibited the tumor cell proliferation as evidenced by the CFU-assay. In conclusion, the plant *Geum urbanum L.* possesses antimicrobial effect against Gram-positive bacteria, strong antioxidant properties, antineoplastic activity in tumor cell lines and favorable toxicological profile *in vitro*. Taken together, our results suggest that the EtOAc fraction from aerial parts might be beneficial for treatment of bladder carcinoma and antibacterial infections caused by sensitive Gram-positive pathogens and future *in vivo* investigations are needed to develop its medicinal potential.

Acknowledgements: The study was supported by DFNP-70/27.04.2016 of the Bulgarian Academy of Sciences and by Alumni Program "Equipment subsidies" of the Alexander von Humboldt Foundation.

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Hawthorn berry extract lowers kynurenic acid and anthranilic acid formation

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Background: *Hawthorn* berry “haws” are used to make wine, jelly and flavor brandy and this plant has been used as a remedy for heart problems and also to treat Alzheimer’s disease. Since elevated kynurenine metabolism has been documented in patients with cardiovascular problems and also in several brain pathologies including dementia we searched the biochemical properties of *Hawthorn* extract with respect to kynurenic acid (KYNA) and anthranilic acid (ANA) formation. KYNA is an endogenous metabolite of tryptophan degradation and is an antagonist of the glutamate ionotropic EAA and of the nicotine cholinergic receptors. KYNA and ANA, both influence the mitochondria respiratory parameters. We questioned whether *Hawthorn* drink has an ability to influence KYNA and ANA formation in the rat tissues, in an *in vitro* study as we have observed with other anti-dementia drugs.

Methods: The activities of the KYNA synthesising enzyme kynurenine aminotransferase II (KAT II) and ANA synthesising enzyme kynureninase in rat liver homogenates were analysed in the presence of different amount of *Hawthorn* drink (N= 5) and in respectively controls (N=5). Formed KYNA and ANA were measured using a HPLC and enzymatic method in the presence of 100 µM L-kynurenine, 70 µM pyridoxal 5'-phosphate and 150 mM Tris-acetate buffer, pH 7.4. The blanks were obtained by using tissue which has been heat inactivated for 30 min in a boiling water bath. As a comparison drug to block KAT II activity we used D-cycloserine in the assay, too (Eur Neuropsychopharmacol. 2014 24(4): 639-44).

Results: *Hawthorn* drink dose-dependently and significantly reduced KAT II activity of rat liver homogenate. Furthermore, *Hawthorn* drink exerted a dose-dependent inhibition of rat kynureninase activities, too. The inhibitory effect of *Hawthorn* drink was more pronounced for KAT II than for kynureninase under assay conditions. Under used assay conditions *Hawthorn* extract and D-cycloserine exerted similar dose-dependent inhibitory effect on KYNA synthesis.

Discussion: This study for the first time demonstrates the ability of *Hawthorn* berry to lower KYNA and ANA formation in rat liver homogenate. Components of *Hawthorn* berry extract are able to affect pyridoxal-5-phosphate complex causing a lowering of KYNA and ANA formation. It is to assume that the inhibitory effect can be seen in other tissue homogenates, as well. We propose *Hawthorn* extract as a drink susceptible of therapeutic exploitation in disorders associated with enhanced KYNA and ANA synthesis in the periphery and in the CNS particularly in diseases with cardiovascular problem and/or with memory impairment and dementia. We believe that frequent use of this berry extract can prevent the development of pathological condition with mostly significant advantage of use such as a lack of side effects. This study suggests anti-dementia action for *Hawthorn* berry due to lowering of KYNA formation.

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Treatment of deep vein thrombosis via regulating balance of Th1/Th2 subsets with chinese medicine compound

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Objective: To discuss the changing rule of Th1/Th2 subsets in deep vein thrombosis (DVT) and the mechanism of Chinese medicine compound Xiao Shuan Tong Mai Granules (XSTMG) in regulating the balance of Th1/Th2 subsets.

Methods: Sixty patients with acute DVT were divided into 2 groups; XSTMG and western medicine group (ITCWMG) and western medicine group (WMG), thirty healthy persons were selected to be in normal group. Flow cytometry and ELISA were performed to observe the frequency change of Th1 and Th2 cells in peripheral blood and the level of TNF- α , IFN- γ , IL-4, IL-10 in plasma.

Results: After using Xiao Shuan Tong Mai Granules, the TNF- α and IFN- γ levels in WMG were higher than those in ITCWMG; the levels of IL-4 and IL-10 in ITCWMG were higher than those in WMG ($P < 0.05$); the improvement of circumference was better in ITCWMG than that in WMG after treatment.

Conclusion: Xiao Shuan Tong Mai Granules can inhibit TNF- α and IFN- γ transcription, enhance IL-4 and IL-10 expression, and increase the proportion of Th2 subsets. It also can predominantly induce T cells response bias to Th2 subsets, reduce the inflammatory reaction, protect vascular endothelial cells. Thrombus recanalization can be accelerated and the limb swelling will be reduced.

Biography

Bin Wang completed his PhD degree from Shandong University of Traditional Chinese Medicine in 2008. He is now working in the Peripheral Vascular Surgery Department, Affiliated Hospital of Shandong University of Traditional Chinese Medicine. He had published more than 10 papers in reputed journals in the recent five years.

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Controlled protein release from defatted pine pollen

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Pine pollen with its two air sacs and a sporoplasmic central cavity is naturally capable of molecular loading owing to the void sac space. Pollen-defatting has been shown to remove lipid content and thus increase the pore size and volume favoring enhanced loading. Here natural pine pollen was defatted using diethyl ether and was physically and chemically characterized comparatively. Vacuum loading was employed and increase in bovine serum albumin (BSA) loading efficiency was observed upon defatting. Rapid release was observed with powdered formulation while tableting helped slow it down. Controlled release was achieved by using a binder (Xanthan Gum) for tableting and coating the tablets using sodium alginate separately, where minimal release was observed in SGF (simulated gastric fluid). As proof of concept, natural pine pollen was also shown to encapsulate other hydrophilic and hydrophobic molecules through both passive and vacuum loading separately and dually, opening exciting prospects for microencapsulation using natural particles.

Biography

Arun Kumar Prabhakar has completed his Master's (Biomedical Eng.) from IIT, Bombay (India) and Bachelor's (Biotechnology) from Anna University, Chennai (India). His area of interest is drug delivery and he has worked with both nanoparticles (polymerosomes, graphene quantum dots) and microparticles (pine pollen) for the same. He is currently working with pine pollen for protein delivery for his PhD thesis under Cho Nam Joon (Assoc. Prof. MSE dept., NTU). He has published a paper for his work on pine pollen capsules (A K Prabhakar, et al., Chemical processing strategies to obtain sporopollenin exine capsules from multicompartmental pine pollen. J. Ind. Eng. Chem. (2017)).

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Green synthesis of capped gold nanoparticles as drug delivery

Anderson J Gomes, Claire N Lunardi, Mirella P F Barros, and Marina L Rodrigues
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Metal nanoparticles is an area of research all over the globe due to their enchanting applications in various fields such as sensor technology, catalysis, optics, drug delivery, and biomedical. Gold nanoparticle (AuNP) and its colloidal dispersions are promising candidates for future scientific, industrial, and domestic applications. AuNP have been used as nano-biomaterials for molecular imaging and drug delivery in recent years. Because AuNP can be modified in different ways, and be used due to its association with receptors coupled with several forms of therapeutics. Microwave assisted synthesis using plant extracts as both reducing and capping is a rapid and facile green synthesis of metal nanoparticles. It has several attractive features such as short reaction time, lower energy consumption and better product yield. We used the MW approach as a simple, green and cost-effective method for the rapid and facile synthesis of plant latex AuNP as a potential drug delivery for cancer. Spectroscopic, size and zeta was measured.

Biography

Anderson J Gomes has completed his Undergraduate in Chemistry from Universidade Federal of Uberlandia (1995), Master's at Chemistry from Universidade de São Paulo (1998) and PhD at Chemistry from Universidade de São Paulo (2003). Has experience in Chemistry, focusing on Photochemistry and Nanotechnology. He has published more than 25 papers in reputed journals.

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Graphene oxide and photosensitizer interaction in drug delivery

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Graphene Oxide (GO) is an oxidized form of graphene, containing functional groups exposed all over its surface expanding the capacity to link drugs and other functional molecules through noncovalent/covalent interaction. Therefore, GO can be considered an appropriate platform, since it can easily physically absorb cationic charged dye compounds via strong p-p and electrostatic interaction on its surface resulting on the formation of GO-dye composite. GO is basically composed of carbon than it can be considered relatively biocompatible, being a suitable candidate for biological applications as well. We associated GO with a photosensitizer for photodynamic therapy. UV-vis spectroscopy and fluorescence emission measurement were used to characterize this system.

Biography

Claire N Lunardi has completed her Undergraduate in Chemistry from Universidade de São Paulo (1996), Master's at Chemistry from Universidade de São Paulo (1999) and PhD at Chemistry from Universidade de São Paulo (2004). She has experience in Chemistry, focusing on Photochemistry and Nanotechnology. She has published more than 45 papers in reputed journals.

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Diabetic conditions and drug vehicles alter drug metabolism

Kvell K, Varga E, Almási A, Pandur E, Horváth Gy, Bencsik T, Póor M, Pohóczky K, Pál Sz, Horváth B, Garai K and Perjési P
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Type II diabetes mellitus is a chronic disease, characterized by hyperglycemia and impaired metabolism. In diabetes, several metabolic pathways are altered, thus the biotransformation and pharmacokinetic of drugs can be influenced due to changes in enzymes and drug transporters. Streptozotocin (STZ) is a widely used glucosamine-nitrosourea compound to induce type II diabetes in murine models. Studies show that in STZ induced chronic uncontrolled diabetes different biotransformation enzymes are altered. Carotenoids are a class of natural antioxidants and according to epidemiological evidences, might have a protective role in chronic diseases. They are able to protect the body from long term consequences of diabetes, like neuronal and eye abnormalities or infectious diseases. Carotenoids have low solubility in water, therefore cyclic oligosaccharides, cyclodextrins can be used to improve the aqueous solubility of carotenoids. In pharmaceutical applications, cyclodextrins are able to enhance drug permeability through gastrointestinal tissues, accordingly these cyclic compounds can be also used for increase the bioavailability of carotenoids.

Biography

Kvell K, MD PhD has primary expertise in Immunology and Biotechnology. Currently he is working at the Department of Pharmaceutical Biotechnology, Faculty of Pharmacy, at the University of Pecs, Hungary. He is currently involved in interdisciplinary research utilizing nanoparticles. This field encouraged the collaborative research team to develop novel drug delivery strategies.

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Isolation and synthesis for secapin from bee venom (*Apis mellifera L.*)

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Bee venom is highly rich in peptides and enzymes constituents, some of which are well-characterized functional agents such as melittin, apamin, and phospholipase A2 with potent bioactivities. Secapin in particular has attracted growing interest due to its role in the insect innate immune system as a hemolytic agent and due to its known applications in neurological, cardiovascular and immunological research areas. Secapin was first identified in early 1970s, but due its scarcity, detailed phytochemical investigations of secapin have been limited. Thus, it had also been challenging to introduce secapin to a wider range of medical and pharmaceutical applications. In the current work, we isolated secapin from bee venom (*Apis mellifera*), employing RP-HPLC and mass spectrometry. We successfully synthesized secapin for the first time using microwave-assisted Fmoc solid-phase peptide synthesis, followed by oxidative folding. The secondary structure was elucidated by a combination of techniques including MS, LC-MS, MS2, and 2D-NMR to characterize the pure native peptide.

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Novel chemosensitizing effects for crocin and flavocoxid in a mouse eac-tumor model: cellular and molecular triggers

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Purpose: We evaluated the sole and doxorubicin (doxo)-combined chemotherapeutic and survival-effects of the phytomedicines; flavocoxid (flvcox) and crocin, using a mouse-Ehrlich-Ascites-Carcinoma-solid-tumor-model (EAC).

Methods: We analyzed tumor-burden, animal-survival, redox status, and levels of mediators for tumorigenesis/inflammation, host-immunity (serum-TNF- α and -IL-10) and tumor-apoptosis (Caspase-3-expression).

Results: EAC-bearing-mice had significantly-raised serum-TNF- α and tumor-lipid-peroxide (MDA) levels, but reduced serum-IL-10 levels and total-serum antioxidant-capacity (TAC), thereby inducing animal-fatalities after 3-weeks. Crocin administration significantly-shrank tumor-mass, -reduced tumor-MDA and serum-TNF- α levels; but -raised serum-IL-10, -TAC and tumor-caspase-3-levels; ultimately augmenting animal-survival. Furthermore, crocin appreciably optimized all responses to doxo to markedly extend animal-survival. Flvcox had similar but less-prominent effects than crocin.

Conclusions: Results reveal that: 1)-Doxo elicits superb cytotoxicity but lesser cytokine-, redox- and animal rescuing-profiles; 2)-Crocin and flvcox achieve significant-sole and -combined chemotherapeutic and animal-survival effects by modifying cytokine levels, optimizing redox-potential and promoting tumor apoptosis.

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Evaluation of *Lavandula angustifolia* oil in cosmetology and aromatherapy

FELICIA ANDREI

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Lavender oil is extracted mostly from the flowers of the lavender plant, a fragrant in nature, and have been used for making perfumes for centuries. The oil is very useful in aromatherapy and many aromatic preparations and combinations are made using lavender oil. Traditionally, lavender essential oil has also been used in dermocosmetics. The aim of this study is to show on human subjects the potential for diminishing the melanin of the skin, at the level of unreliable melanic spots (macules of diverse causes) by the mexametry method. The ointment in which we have introduced the lavender oil contains an increased percentage of lipophilic components and water. This is why we also included in this formulation: stabilizers, antioxidants, antimicrobial preservatives, emulgators, surfactants. Prior to the start of the volunteer study, a lavender flower extract was obtained that was compared to two other commercial samples and chemically characterized and analyzed by chromatography in the University of Agricultural Sciences of Banat, Timisoara, Romania. The results obtained are obvious and demonstrate the depigmenting effects of lavender oil but we encourage the long-term and associative study.

Key Words: Lavender, melanin, depigmentation, mexametry.

Biography

Felicia Andrei has the PhD degree in Medicine (University of Medicine and Pharmacy Timisoara) and is a pharmacist specialised in Clinical Pharmacy (University of Medicine and Pharmacy Bucharest). She has completed also two Masters degrees: one in Pharmacy -Formulation and evaluation of the dermatocosmetic product and the other in Polytechnic Computer Automation - Information Systems in Health Care. Now teaching in the Faculty of Pharmacy in Timisoara as an Assist. Prof. at the discipline of Dermatopharmacy and Cosmetology. She is a member of the College of Pharmacists in Romania and of the European Federation for Pharmaceutical Sciences.

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Evaluation of medicine distribution, regulatory privatization, social welfare services and its alternatives

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The strategy of price liberalization and privatization had been implemented in Sudan over the last decade, and has had a positive result on government deficit. The investment law approved recently has good statements and rules on the above strategy to pharmacy regulations. Under the pressure of the new privatization policy, the government introduced radical changes in the pharmacy regulations. To improve the effectiveness of the public pharmacy, resources should be switched towards areas of need, reducing inequalities and promoting better health conditions. Medicines are financed either through cost sharing or full private. The role of the private services is significant. A review of reform of financing medicines in Sudan is given in this article. Also, it highlights the current drug supply system in the public sector, which is currently responsibility of the Central Medical Supplies Public Corporation (CMS). In Sudan, the researchers did not identify any rigorous evaluations or quantitative studies about the impact of drug regulations on the quality of medicines and how to protect public health against counterfeit or low-quality medicines, although it is practically possible. However, the regulations must be continually evaluated to ensure the public health is protected against by marketing high quality medicines rather than commercial interests, and the drug companies are held accountable for their conducts.

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Structure- and ligand-guided screening of LptA inhibitors: An initial study for developing novel Neisserial antibacterials

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Pathogenic Gram-negative bacteria such as *Neisseria meningitidis* and *Neisseria gonorrhoeae* have developed resistance against antibiotics due to their ability in creating an envelope on the outer layer of lipooligosaccharides (LOS). The cationic phosphoethanolamine (PEA) decoration of LOS' lipid A is regulated by lipid A-PEA transferase A (LptA) which may serve as a prominent target for developing new antibiotics. The discovery of Neisserial LptA has provided a structural aspect to its catalytic mechanisms and ligand recognition that are crucial for inhibitor development. A combination of structure- and ligand-based approach has been employed to explore novel potent LptA inhibitors among millions of commercially-available compounds and approved drugs. A total of 4000 hit molecules obtained from LIDAEUS structure-based screening and PubMed ligand similarity search were further examined through semi-flexible docking simulation performed in MOE and Schrödinger's Glide. Best hits were therefore carefully selected based on their docking score, drug likeness, and pharmacological properties. Free energy of binding calculation and ligand interaction analysis suggest that the selected 20 hit compounds have a stronger binding affinity than LptA natural substrate and possess a more effective interaction with catalytically-essential residues. Further molecular dynamics (MD) simulation of these 20 compounds also confirms that they all maintained stable complex conformation showing low total RMSD, capability to maintain interactions with active site, and acceptable Ramachandran plot. This study provides an insight to drug repurposing which may serve as an initial step to develop novel potent LptA inhibitors to combat the virulence of multi-drug resistant *Neisseria*.

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Increasing the VP1-VP2/GFP complex yield with single amino acid substitutions of the VP2 interaction site for biopharmaceuticals drug delivery

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Virus-Like Particles (VLPs) are complex proteins resembling viruses, but deprived of virus DNA and therefore non-infectious. For that reason, they have an excellent potential to be great drug delivery systems. Polyomavirus VLPs are constructed of VP1-VP2 proteins complexes. VP2 is often truncated to the middle of VP1 interacting sequence in solution. During my internship at the University of Queensland in Australia I tried to increase the yield of the VP1-VP2/GFP complex by making single amino acid substitutions in the VP2 truncation site and therefore avoid the truncation. I planned single aa substitutions in the VP2 truncation site and performed site-directed mutagenesis of the template plasmid and expressed the proteins in the autoinduction media after bacterial transformation. A modified ELISA test, named FLISA - a fluorescence-linked immunosorbent assay, verified the integrity of the complex by revealing GFP fluorescence in complexes bound by an anticomplex antibody. The results confirmed the presence of a large amount of complex in 5 out of 20 prepared mutations; 3 of them were located in the 294 locus, suggesting that the original glycine is not necessary for the complex integrity and can potentially be replaced also with different candidate amino acids. Overall the results need to be investigated further, but the work can be pronounced hopeful, as the GFP protein in the complex can be substituted for a cytotoxic or therapeutic protein or compound and used with the VLP method to target particular cells in the body, giving a powerful and promising tool for biopharmaceutical treatment.

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Evaluation of medicine distribution, regulatory privatisation, social welfare services and its alternatives

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The strategy of price liberalisation and privatisation had been implemented in Sudan over the last decade, and has had a positive result on government deficit. The investment law approved recently has good statements and rules on the above strategy in particular to pharmacy regulations. Under the pressure of the new privatisation policy, the government introduced radical changes in the pharmacy regulations. To improve the effectiveness of the public pharmacy, resources should be switched towards areas of need, reducing inequalities and promoting better health conditions. Medicines are financed either through cost sharing or full private and the role of the private services is significant. A review of reform of financing medicines in Sudan is given in this article. Also, it highlights the current drug supply system in the public sector, which is currently responsibility of the Central Medical Supplies Public Corporation (CMS). In Sudan, the researchers did not identify any rigorous evaluations or quantitative studies about the impact of drug regulations on the quality of medicines and how to protect public health against counterfeit or low-quality medicines, although it is practically possible. However, the regulations must be continually evaluated to ensure the public health is protected against by marketing high quality medicines rather than commercial interests, and the drug companies are held accountable for their conducts.

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November 09-11, 2017 Vienna, Austria

Immunotherapy of glioblastoma spheroids tumor cultured in fibrin gel by atorvastatin

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Glioblastoma multiform (GBM) is the most aggressive glial neoplasm. Absolutely, the survival, growth, and invasion of GBM cells are promoted by various inflammatory cytokines. Statins, such as atorvastatin, are known to exert anti-inflammatory effects. Chronic inflammation is a pathological feature of cancer. Growth of solid tumors results in most cases in a hypoxic microenvironment and the release of various cytokines and growth factors, which together increase inflammation, angiogenesis in tumor stroma, and triggering signaling cascades that activate NFκB and STAT3 that produces predominantly by a specific subset of T helper cells (Th cells), namely Th17 cells. Interleukin-17 (IL-17) has emerged as a central player in the mammalian immune system. IL-17RA is expressed in most tissues examined to activate many of the same signaling cascades as innate cytokines such as TNFα and IL-1β. Furthermore, emerging knowledge regarding IL-17A/IL-17RA signaling in numerous tissues suggests an important role in health and disease beyond the immune system. This increasing evidence suggests that IL-17A and Th17 play a main role in autoimmune inflammation. A VEGF independent pathway was also found via NF-κB, which leads to suppression of the immune response targeting cancer cell. In this study, we investigated the anti-inflammatory and anti-angiogenesis activity of atorvastatin on engineered three-dimensional (3D) human tumor models using glioma spheroids and Human Umbilical Vein Endothelial cells (HUVECs) in fibrin gel as tumor models in different concentrations of atorvastatin (1, 5, 10 μM). After 48 hours exposing with different concentrations of atorvastatin, cell migration of HUVECs were investigated. After 24 and 48 hours exposing with atorvastatin VEGF, CD31, IL-17R genes expression by real time PCR were assayed. In the current study, results have demonstrated a potential impact of IL-17R in glioma growth and progression. The results showed that atorvastatin has potent anti-inflammatory and anti-angiogenic effect against glioma spheroids by downregulates IL-17RA and VEGF expression especially at 10 μM concentration. The most likely mechanisms are the inhibition of inflammation by IL-17RA interaction with NFκB signaling pathway. Finally, these results suggest that this biomimetic model with fibrin may provide a vastly applicable 3D culture system to study the effect of anti-cancer drugs such as atorvastatin on tumor malignancy *in vitro* and *in vivo* and atorvastatin could be used as agent for glioblastoma treatment.

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November 09-11, 2017 Vienna, Austria

Overview of avicenna (ibn sina) opinion on stomach swelling

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The stomach is the most dilated portion of the GI tract (1). In Iranian traditional medicine (ITM) stomach is very important organ and its disorders can affect other organs (2-5). Avicenna (Ibn Sina), a Muslim scientist of the tenth and eleventh centuries has an important role in the history of medicine in Iran and the world (6). Canon, his famous book that was the greatest work by Avicenna, was translated into Latin in the 12th century and used as a major reference in medical education from the 12th until the 17th centuries (7). In addition, this book is one the most important textbooks in ITM. The foundation of ITM system was based on the balancing humors in the human body (8). In ITM, Mizaj (temperament) plays a key role in preventive, therapeutic, and lifestyle recommendations (9). Normally there are four humors in the human body: Phlegm or Balgham, Blood or Dam, Yellow bile or Safra, and Black bile or Sauda (10). Their imbalances in the body causes some disorders such as stomach swelling (2, 8). Persistent or repeated stomach pain that does not resolve with suitable treatments is the main symptom of stomach swelling (8). According to Avicenna, swelling is a complex disease (8). Swelling is thickness and bulging occurring in organ by material accumulation that expand it and fills the cavity. This material is usually infused by another organ or the organ of swelling itself (2, 8). Stomach swelling is divided into different types according to the materials: "warm" (Haar Safravi or Damavi), "cold Phlegm" (Barede Balghami), and "Solbe Ghaliz" (Hard and thick swelling) (8). The warm swelling is made by not only blood or yellow bile, but also any substance that is warm in essence or is heated by infection (2, 8). Prolonged and progressive pain, severe inflammation, strong burned, thirst and associated fever, and radiating pain are the symptoms of warm swelling. It may even lead to a mixing of mind, melancholy, and meningitis (8). "Cold phlegm" swelling is made by phlegm and predisposing factors such as moisture, indigestion, exercise reduction, and other moisture-generating causes. The symptoms and signs of "cold Phlegm" swelling are persistent or repeated pain even during sleep, diluted and increased mouth water, leaden face color, little thirst, poor digestion, poor appetite, hate and suffer moist food, tendency to dry food, without fever, inflammation, and obsession (8). Sometimes, hard and thick swelling in the stomach is formed primarily by "Sauda" or from transmission and conversion of warm swellings. Phlegm swelling seldom becomes the hard stomach swelling. There are strength to the touch, a lot of dryness, loss of body weight, and other symptoms associated with swelling in the stomach (2, 8). Treatment in warm swelling includes reducing the swelling matter by venesection or prescribed laxatives, reducing the swelling by rubbing astringent and cold medications over swollen stomach, and avoiding vomiting and potent laxatives. Topical use of Radea (deterrent) drugs, Quince oil, and grated cucurbit, avoiding food, and use of easily digestible food at first and then laxative food and drugs such as beer, cucurbit, and Khiareshanbar (Cassia fistula) is recommended. Safravi swelling remedy is consisted of topical cold drugs such as Sandal (Santalum album), Kafoor (Cinnamomum camphora), alcohol-free beer, and Enabolsalab (Solanum nigrum L.). Treatment in "cold Phlegm" swelling include oral use of astringent solvent drugs such as extract of Celery and Fennel, almond oil, topical use of Baboone (Matricaria chamomilla) and shebet (Anethum graveolens), and avoiding vomiting. Finally, potent solvent drugs is recommended in hard swelling astringent and use of camel bone marrow and cow calf brain in topical drugs is obligatory (2, 8). Based on this study, diagnosis and treatments of stomach swelling can be helpful in patients with persistent or repeated episodes stomachache.

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Up-regulation of cd4+ t-lymphocytes by isomeric mixture of quercetin-3-o-rutinoside and quercetin-3-o-robinobioside isolated from the leave extract of millettia aboensis

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Context: *Millettia aboensis* (Hook. F.) Baker (Fabaceae) is popular in ethno-medicine due to its acclaimed efficacy in a number of disease conditions.

Objective: To evaluate the immunomodulatory effect of the leaf extract of. Materials and method: Humoral and cellular immune responses of albino mice to tetanus toxoid and cyclophosphamide, respectively, were used to monitor immunomodulatory activities of the ethanol leaf extract and fractions of *M. aboensis* at 200, 300 and 400 mg/kg. Most active fraction was subjected to chromatographic purification and isolation of active compounds. Stimulation of specific T-lymphocytes was used to evaluate immune enhancing activity of the isolated compounds.

Results: The extract and fractions evoked increase in both humoral and cellular immunity. At 400 mg/kg, normalized mean secondary IgG₁ and IgG_{2a} antibodies response of the butanol fraction were 9.0 and 7.7 respectively compared to 13.2 and 16.5 produced by Noni capsule[®]. Findings from the nature of cytokine up-regulation by butanol fraction following secondary challenge with tetanus toxoid revealed that IL-12, IL-17A, IFN- γ and IL-4 were expressed by 48.14, 41.37, 38.22, and 31.03%, respectively. Structural elucidation of the active compounds revealed presence of isomeric mixtures of quercetin-3-O-rutinoside and quercetin-3-O-robinobioside. This compound mixture exhibited *in vitro* up-regulation of specific CD₄⁺ lymphocytes that were largely interferon gamma (IFN γ) releasing. Up to 43.7% stimulatory effect of IFN γ was produced at 6.25 μ g/mL compared to the baseline effect in DMSO control group.

Discussion and conclusion: *M. aboensis* expressed strong immune-enhancing properties, which may explain its ethnopharmacological use in disease management.

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Design and activity of intelligent cellulose-based dressings for growth factor protection in chronic wounds

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Intelligent dressing design has been a goal of chronic wound dressing development for more than a quarter century. Since the report of the first dressing that modulated water vapor transmission rates numerous approaches to semi occlusive dressing designs have enabled improvement in chronic wound healing rates. Among the approaches to improved wound healing, the delivery of therapeutic growth factors like platelet derive growth factor has played an important role. However, maintaining stability and controlled release of growth factor activity has met with mixed success. This in part is due to the high proteolytic activity in chronic wounds which degrades growth factors and extracellular matrix proteins. Accordingly, we report here an approach to an intelligent dressing design based on protease point of care sensors combined with protease sequestrants and/or inhibitors in semi occlusive dressing motifs. Protease biosensors detected levels of elastase at those found in chronic wound fluid (0.025 U/mL). A sensor-based dressing is applied to monitor protease activity while release of the protease inhibitor oleic acid (18:1) is triggered and taken up by albumin binding to the dressing and subsequent inhibition of elastase. Transfer from albumin to inhibition of elastase was observed when four per cent albumin solutions were used and it was most effective in binding cellulose bound-18:1. However, 2% albumin was sufficient to transfer quantities of 18:1 necessary to achieve a significant elastase-lowering effect. Formulations with 128 mg 18:1/g cellulose gauze had equivalent elastase lowering with 1 - 4% albumin. 18:1 bound to cotton wound dressings may have promise in the selective lowering of cationic serine protease activity useful in topical application for chronic inflammatory pathogenesis.

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November 09-11, 2017 Vienna, Austria

The late phase of drug development: A bioanalytical dealing with the health authorities

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Background: Bioanalytics are necessary and crucial during the drug's development, especially in the most advanced phase when approaching the health authorities' requests. At this phase, bioanalytics are not only involved in the preparation of regulatory dossiers, but also in a series of additional investigations to meet the demands from health authority (HA) itself. The present report summarizes some of the investigations related to the biological activity evaluation, upon approval request from HAs. As case report, the successful example of the just approved Bavencio (Avelumab) has been chosen. The biological activity is monitored, according to the regulatory dossiers, in terms of cell binding activity and ADCC (Antibody Dependent Cell Cytotoxicity) of Avelumab (Anti PDL-1 therapeutic antibody) as result of physicochemical modifications. Anti-PDL-1 is a fully human IgG1 monoclonal antibody direct against the PDL-1, a receptor highly expressed in a variety of human cancer cells. The Anti-PDL-1 binds PD-L1 blocking the interaction between PD-L1 and the PD-1 expressed on activated immune cells, thereby releasing cells from immunosuppression and strongly enhancing anti-tumor immunity. Beside this major MoA, Anti-PDL-1 is also able to mediate an ADCC.

Analytical Approach/Methods: Upon HA requirement, for a complete biological characterization of the product, physicochemical modifications have been evaluated starting from a highly modified form of the Anti PDL-1 in term of aggregation (HMW) fragmentation (LMW) and fucosylation level to identify a correlation between different percentage of each modified form and its biological activity. The biological effect of each modifications on both Fab and Fc portions of the molecule have been verified applying both ADCC assay and cell binding assays.

Results: Each physicochemical modification have highlighted a biological impact. Regarding the fucosylation level, a clear correlation between the ADCC activity and the level of fucose in Anti PDL1 has been shown, confirming that the removal of fucose residues greatly enhances therapeutic MAb ADCC activity. Significant biological results have been proven also by the effect of aggregation (HMW) and fragmentation (LMW) on Anti PDL-1. Upon each HA requirement, it has been demonstrated the complete knowledge of the molecule from the physicochemical and biological point of view. The critical quality attributes are monitored by a dedicated analytical panel and the possible modifications are under control, ensuring to satisfy the expectations of HAs, the market and the patients.

Conclusions: These studies contributed the submission to the FDA of a biological license application for Anti PDL-1 (Bavencio) as the first metastatic carcinoma of Merkel cells (mMCC). Subsequently, a second US FDA approval for urothelial or advanced metastatic carcinoma (UC), commonly diagnosed as a metastatic bladder tumor has been obtained.

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The preventive effect of date palm (*Phoenix dactylifera*) seed and fruit hydro-alcoholic extracts on carrageenan-induced inflammation in male rat's hind paw

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Background & Objective: The side effects of NSAIDS drugs, have caused increasing interest of scientists in herbal medicines as alternative treatment. In this study, the effect of anti-inflammatory of seed and fruit of date palm hydro-alcoholic extracts, due to having antioxidants, was studied.

Materials & Methods: In this study, the extracts of date palm seed and fruit were prepared by maceration method in 70% alcohol. Eighty male rats Wistar, divided into 10 groups of eight in each, 4 groups received different doses (100, 200, 400 and 600 mg/kg) of seed extract and 4 other groups different doses (100, 200, 400 and 600 mg/kg) of fruits extract of the palm, and the positive control aspirin (300 mg/kg) and the negative control group saline (5 ml/kg) via injection intraperitoneally. Half an hour later all animals received 100 µl of 1% carrageenan into the rat's hind paw subcutaneous. The changes in rat's paw edema was measured by plethysmometer every hour for five hours.

Results: The effect of the doses of date palm seed extract on edema were less than aspirin ($P < 0.05$). But there was no significant difference between the group that received 400 and 600 mg/kg date palm fruit extract when compared with aspirin group. The dose 400 mg/kg of fruit extract showed the most anti-inflammatory effect and it was assigned as the best dose.

Conclusion: It is likely that with further studies on different model of animals and on human model the palm fruit extract could be used for pain treatment.

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Nano multi arms biodegradable polymers for drug delivery

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Novel triblock copolymers of poly(L-lactide)-poly(ethylene glycol)-sebacate-poly(ethylene glycol)-poly(L-lactide) were synthesized by Ring-Opening Polymerization of different ratio of L-lactide using Diazabicyclo-[5.4.0]-undec-7-ene as catalyst. These copolymers were fully characterized. They were used to prepare IPN's which had a Nano fibre characteristic as examined by Scanning Electron Microscopy. The swelling ratio, hydrolytic degradation and Insulin release studies were performed at three different pH. It was found that the swelling ratio and the time needed to degrade fibre IPN's increased with increasing PEG chain length. *In vitro* insulin release showed that the longer the PLA chain length the slower the release rate. Insulin release could be potentially controlled by PEG molecular weight, PLA content, pH, presence of Pluronic F-127 in the IPN's, crosslinking agent ratio, and the number of coated layers.

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Silymarin: As a multi-potential, novel and effective compound on diabetes and chemotherapy

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Silymarin (SMN) is a polyphenolic mixture of flavonolignans extracted from seeds of the milk thistle (*Silybum marianum*). It was found attribute to its capability in the reduction of NO and MDA levels and myeloperoxidase activity, suggesting it's not only antioxidant but also anti-nitrosative capacities. Traditionally, SMN has been used as a natural remedy for digestive problems and in particular for diseases of the liver and the biliary tract, for menstrual disorders and varicose veins. There are increasing data indicating beneficial effects of SMN on various disorders and diseases in different tissues. We in our investigations during the last ten years found that SMN exerts remarkable protective and regulatory effects on drug and xenobiotic biotransforming enzymes in experimentally-induced diabetic animals. At the same series of investigation we explored that although both SMN and melatonin treatment was able to normalize the antioxidant status, while only SMN administration could restore the β cells of Langerhans islets in diabetic rats. The anti-inflammatory property of SMN on mono-iodoacetate- induced osteoarthritis and antinociceptive effects on acetic acid-induced reaction were also clarified. In another study the SMN protective and preventive effects on doxorubicin-induced carbonyl stress, DNA damage, and its capability in the alteration of c-myc gene expression were demonstrated. SMN beneficial effects on mycophenolate mofetil-induced duodenal disorders

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November 09-11, 2017 Vienna, Austria

Relationships between serum adipokine levels (adiponectin, leptin) in diabetic and non-diabetic osteoporosis patients

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The purpose of this study was to investigate the relationship between serum adipokine levels (adiponectin, leptin) in diabetic and non-diabetic osteoporosis patients. We studied 72 osteoporosis patients (36 diabetic and 36 non-diabetic with body mass index [BMI] 28.1 ± 5.1 and 27.1 ± 6.8 , respectively). BMD was studied by dual-energy X-ray absorptiometry from the lumbar spine (L1–L4) and femoral neck and fasting blood samples were taken for biochemical measurement of fasting blood glucose, Glycosylated hemoglobin, leptin, adiponectin. Fasting levels of plasma adiponectin had a significant positive correlation with BMD of the lumbar spine and a no significant positive correlation with BMD of the femoral neck in the diabetic osteoporosis group ($r=0.9$, $P=0.02$ / $r=0.18$, $P=0.31$, respectively), but a no significant negative correlation with BMD of the femoral neck and lumbar spine in the non-diabetic osteoporosis group ($r=-0.02$, $P=0.95$ / $r=-0.01$, $P=0.95$, respectively). Leptin did not have a significant correlation with BMD in either the diabetic and non-diabetic osteoporosis groups ($P>0.05$). The correlation between adiponectin and leptin are not inconclusive.

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Antioxidant activity and hepatoprotective effects of *Centaurea incana* on CCl₄-induced liver toxicity in rats

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Aim: The aim of the present study was to investigate the potential antioxidant and hepatoprotective effects of *Centaurea incana* on the free radical damage of liver caused by carbon tetrachloride in rats.

Methods: For the study of preventive effect of methanolic extract of *Centaurea incana* on CCl₄-induced hepatotoxicity, our study was carried out on rats. The animals were randomly divided in to 4 different groups comprising 7 animals each. Group I served as controls and received an injection of vehicle (olive oil) alone; Acute liver injury in rats was induced by a single intraperitoneal injection with CCl₄ dissolved in an equal volume of olive oil at a dose of 3 ml/kg body weight, group II, which is well documented to induce hepatotoxicity. Group III was administered methanolic extract of *Centaurea incana* at a dose of 500 mg/kg alone. Group IV was administered methanolic extract of *Centaurea incana* at a dose of 500 mg/kg and was injected by CCl₄ i.p., at a dose of 3 ml/kg body weight. After 4 weeks of treatment, all of the animals were sacrificed 24 h after administration of CCl₄, and blood was collected, serum was separated and stored at -20°C.

Results: The single intraperitoneal injection with CCl₄ caused severe hepatotoxicity in rats, as evidenced by the significant elevation of serum AST and ALT activities after the administration of CCl₄. The concentration of MDA, an end product of lipid peroxidation, in the rats treated with CCl₄ was increased 2.7-fold when compared with the vehicle control rats. However, pre-treatment with *Centaurea incana* significantly prevented the elevation of serum AST and ALT activities induced by CCl₄ treatment. Consistent with the serum AST and ALT activities, pre-treatment with *Centaurea incana* for 4 weeks to the rats resulted in a significant decrease in the concentration of hepatic MDA when compared with the CCl₄ group.

Conclusion: Our investigation provided convincing data that *Centaurea incana* decrease the lipid per-oxidation and liver enzymes, and increase the anti-oxidant defense system activity in the CCl₄-treated rats. The mechanisms underlying hepatoprotection of the methanolic extract of *Centaurea incana* may be related to both its radical scavenging properties and indicate effects as a regulator of antioxidative systems.

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***In vitro* antiplasmodial and antifungal activity of *morinda morindoides* (baker) milne-redh (rubiaceae), an ivoirian traditional medicine plant**

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Introduction: In Côte d'Ivoire and elsewhere in Africa, medicinal plants occupy a place of choice in the treatment of various diseases. The survival and intensification of this practice today despite the prodigious development of modern medicine are related to several factors, among which may be mentioned economic constraints and sociocultural factors. It is study that we are interested in antimalarial and antifungal activity of *Morinda morindoides*, a plant used against fever and diarrhea in Côte d'Ivoire.

Method: *M. morindoides* leaves were collected, air dried and made into a fine powder. Aqueous extracts (Aqe), ethanol (Eeth), ethyl acetate (EAc) and acetate-water (EAc- H₂O) were performed. Each extract was tested on *Plasmodium falciparum* and *Aspergillus fumigatus*.

Results: IC₅₀ values of different extracts are ranked in the following order: 6.1 (EAc) <17.8 (Eeth) <21.5 (Aqe) <46.5 (EAc-H₂O) for *P. falciparum* and 1.3 (EAc) <6.1 (Eeth) <12.47 (Aqe) < more than 300 (EAc-H₂O) for *A. fumigatus*, the ethyl acetate extract being the most active against both pathogens.

Conclusion: These results show that *M. morindoides* leaves display significant antiplasmodial and antifungal activity, which justifies its use in traditional medicine against malaria and mycoses.

Keywords : *Aspergillus fumigatus*, Malaria, *Plasmodium falciparum*

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Evaluation of antiangiogenic and antitumor properties of *Anogeissus leiocarpus*

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Background: *Anogeissus leiocarpus* is a traditional medicinal plant with strong antioxidant and hypoglycemic properties.

Hypothesis/Purpose: This study was aimed to investigate the antiangiogenic and cytotoxic effects of eight extracts from *Anogeissus leiocarpa* (leaves and bark).

Study Design: Eventually, the most active extract was subjected to a series of *in vitro* and *in vivo* studies to elucidate the mechanism of action.

Methods: In order to confirm the effect of the extract on motility of human endothelial cells, cell migration assay was conducted. In addition, VEGF suppressive effect of the extract was assessed in endothelial cells. Finally, the antitumor effect of the extract was evaluated using *in vivo* human tumor xenograft model.

Results: Results of the present study indicated that, hexane extract of the stem bark of *A. leiocarpus* was found as the most active extract on inhibition of sprouting of microvessels (89.56%). Additionally, ethanol extract of the leaves exerted high antiangiogenic (inhibition 82.12%) in rat aortic ring assay. Hexane extract of the stem bark displayed significant inhibitory effect on endothelial cells proliferation (76.87 %) while ethanol extract of the leaves was save on HUVEC cell lines (inhibition 5.80%). The two extracts inhibited HUVEC migration by 87.57 and 65.23% respectively. The extracts demonstrated significant inhibition of VEGF levels (45.32 and 30.52 % respectively) in treated endothelial cells. Finally the extracts exhibited potent anti-tumorigenic effect in athymic mice with $\Delta T/\Delta C = 8.43$ and 12.54% at doses 400 and 200mg/kg, respectively.

Conclusion: These results may provide novel guidelines towards improved strategies using *Anogeissus leiocarpus* extracts based on the suppression of angiogenesis to curb the growth of tumors. The plant can be used as promising candidate for anti-neoplastic drug development.

Key words: Medicinal plants, Antitumor *in vivo*, antiangiogenic *Anogeissus leiocarpus*

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November 09-11, 2017 Vienna, Austria

Selenium and its compounds as prospective drugs in oncology

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In last years, there have been some notable developments in several areas of inorganic pharmaceutical that have potentially far-reaching importance for future medicinal applications and research. One of the highly significant developments in the field of oncology and hematology is the application of selenium compounds. Selenium ions play an important role in biological systems, and without their catalytic presence in trace or ultratrace amounts many essential co-factors for many biochemical reactions would endogenous antioxidant enzymes. Selenium is an essential component of several endogenous antioxidant enzymes. Selenium affects the transcription of multiple defense and repair genes to protect against metal-induced pathologies. Selenium creates stable complexes with a wide variety of organic molecules that can provide required biological affinity and therapeutic activity veritable for targeting specific locations in the body. The aim of this review is a critical evaluation of selenium complexes used for therapy or diagnosis of various diseases in oncology.

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November 09-11, 2017 Vienna, Austria

Exploration of phytotherapies common among local communities of rawalakot, district poonch azad jammu and kashmir

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Ethnopharmacology act as bridge among medical, natural, and social sciences with most of its research focusing on chemical, biological, and pharmacological sciences. Medicinal plants are a basic source of health care in the Pearl Valley District Poonch of Azad Jammu and Kashmir, a mountainous region of Pakistan. Although some ethnobotanical researches have been carried out in the district, the work reported here is the first field study on medical ethnobotany in Rawalakot area. Information about the therapeutic properties of the medicinal plants was collected from 46 laypeople and 18 herbalists by using an open ended and semistructured questionnaire. The data about the use of plants was recorded into a synoptic table containing ethnobotanical inventory of plants, parts used, therapeutic indication and mode of application or administration. Different ethnobotanical indices were calculated in order to quantify the knowledge on the medicinal plants reported in the study. Our study recorded 136 species of medicinal plants belonging to 45 families. Asteraceae (14 species) was the dominant family in the area, followed by Lamiaceae (11 species), Fabaceae, and Rosaceae (5 species each). Herbaceous plants (55%) were the most used, with leaves (31%) as the most exploited plant part. Decoction (26 species), juice and powder (24 species each) were the most common methods of preparation. The highest use values (UVs) were reported for *Berberis lyceum* and *Ajuga bracteosa* (1.13 each), *Abies pindrow* (1.03), *Prunella vulgaris* and *Adiantum capillus-veneris* (1.00 each). Highest informant consensus (ICF) values were recorded for digestive system diseases (ICF = 0.90), muscular and skeletal system diseases (ICF = 0.89), and mouth/pharynx diseases and diabetes (ICF = 0.86 each). When we compared data of this study with those of other studies carried out in neighboring areas, we observed that the percentage of similarity in uses of plant species ranged from 13.33% to 34.62% with an average value of 22.53%. The present study revealed the importance to document and launch list of all the possible plants that are used in phytotherapies in the unexplored study area. The present study is useful in preservation of indigenous knowledge and could attract future researchers to investigate and explore phytochemicals responsible for medicinal properties of these plants.

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November 09-11, 2017 Vienna, Austria

Ethno-gynaecological knowledge and preliminary phytochemical screenings of medicinal plants used by women in Lagos State, Southwest, Nigeria

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The prevalence of gynaecological disorders worldwide has given rise to increased female infertility, morbidity and mortality. The aim of this study was to establish a regional profile of the indigenous knowledge on the treatment of various gynaecological disorders in Lagos State, Nigeria. Using oral and semi-structured interview methods, 100 local informants from five local government areas of Lagos State, Nigeria were interviewed. Preliminary phytochemical screening was carried out using standard procedures. Fifty (50) plant species belonging to 35 families were identified for the treatment of different gynaecological disorders in the study area. Family Fabaceae had the highest number of plant species (10 %) followed by Euphorbiaceae (8 %). Ethno botanical uses of 16 plant species for amenorrhea, 9 species for aphrodisiac, 7 species for vaginal infections and 6 plant species for sexually transmitted diseases were discovered in the study area. The most commonly used plant parts were the leaves (29.03 %) while decoction (48.08 %) was the most commonly used mode of preparations. The highest percentage of occurrence was observed in trees (52 %) followed by the herbs (20 %) while shrubs and creepers were 16 % and 12 % respectively. Phenols and flavonoids were present in all the plants while phlobatannins were present only in ten (10) plant species. The identification and documentation of medicinal plants used in folkloric medicine for the treatment of gynaecological disorders in Lagos State, Nigeria would be useful for the conservation of the plant species, also the plants can serve as precursors in the development of novel drugs for the treatment of various gynaecological disorders.

Keywords: Gynaecological disorders, Lagos State, Ethnobotanical survey, Medicinal plants, Phytochemicals.

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November 09-11, 2017 Vienna, Austria

Phytochemical screening and antioxidant activity of *cardiospermum corindum* l faux persil from botswana

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The aim of this study was to compare the phytochemical composition and antioxidant activity of roots and shoots of *Cardiospermum corindum* collected from two geographically distant regions of Botswana (Tswapong Hills and Kgale Hills). Qualitative phytochemical analysis revealed presence of alkaloids, reducing sugars, saponins, phytosterols, phenols, flavonoids and terpenoids. Analysis by thin layer chromatography, revealed that both shoots and roots of plant collected from the two respective regions showed no differences in phytochemical constituents. Total phenol and flavonoid contents were quantitatively estimated. Total phenolic content measured by Folin-Ciocalteu method varied from 164.4±2.2 to 364.2±3.1mg/L (GAE) Gallic Acid Equivalents. The order of total phenol contents were [364.2±3.1](Roots from Tswapong Hills)>[356.0±4.5] (Roots from Kgale Hills)>[169.1±2.6](Shoots from Tswapong Hills)>[164.4±2.2mg/lGAE](Shoots from Kgale Hills). The total flavonoid contents as measured by aluminium Chloride method varied from 56.7±1.1 to 124.1±1.5mg/L(QE) Quercetin Equivalents. The order of the total flavonoid contents were [124.1±1.5](Shoots from Kgale Hills)>[118.8±2.6](Shoots from Tswapong Hills)>[63.3±1.6](Roots from Tswapong Hills)>[56.7±1.1mg/l QE](Roots from Kgale Hills). The antioxidant activity as determined by the DPPH radical scavenging assay, revealed that, at all tested concentrations, root extracts exhibited greater (≥86%) scavenging potency than shoot extracts (≤83%). A direct correlation between total phenolic content and free radical scavenging activity was revealed. This work has validated the use of this plant as a health improving tool. However, structural identification of the bioactive constituents should be carried out.

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November 09-11, 2017 Vienna, Austria

Medicaments and pregnancy

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Drugs administered during pregnancy there are in interest of professionals as they could influence embryogenesis. These can have temporary or permanent effects on the fetus development. Any drug with relation a dose can act during embryonic or fetal development to produce a permanent alteration of form or function. Study this problems in the territory of Slovakia our retrospective study was performed between May 2016 and November 2016. A method of questioning concerning pregnant women about their drug consumption. The examined group consisted of 300 women becoming from region of Bratislava. The data recovered from questionnaire were obtained up to 4 days after delivery. The data were statistically analysed. Based on the results of this study no statistically significant correlation was found between drug intake during pregnancy and increase in prevalence of the congenital malformations in the group. However the causal coincidence between drug intake and congenital disorder is rather difficult to establish. The number of drug consumers in the region of Bratislava reached 88%. That includes also a group of consumers of supplements. Women that consumed vitamins, minerals and Fe form 49% of all examined women. Pregnant women which used vitamins during the first trimester were 32% of all. The group of women that used Fe in the third trimester was 40% of all examined women. The most frequently used drugs except supplements were hormones in 11%, anticoagulants in 7% and antibiotics in 5%. Similar studies with pregnant women performed in the U.S.A. or in Western Europe have shown higher exposition levels of the prescription drugs. The choice of drugs used in different countries is also quite specific, which makes extrapolation between countries rather difficult. The drug registration process, prescription customs, drug availability etc. also needs to be taken in consideration.

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November 09-11, 2017 Vienna, Austria

An overexploitation approach for cell culture based modular bioprocessing

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In the battle of controlling healthcare costs, the high cost pharmaceuticals, especially biologics has become an important issue. Successful production planning requires consideration of cost factors such as manufacturing cost, capital investment to build new facilities or to retrofit existing ones, as well as the inventory costs. The cost attributed to the opportunity lost in selling the product or failure to meet market demand is also considered as the inventory cost. Any unplanned downtime could severely affect the customer service level, if facility utilization is too high. Underutilization of manufacturing facilities conversely suggests a misplaced investment in capacity. The overexploitation approach is designed to mitigate the above described facility constrains due to its power of combining the benefits of perfusion & fed batch processing mode with adequate elimination of facility constrains of fed-batch bioprocessing. The key factors considered are: 1. Adapting to modular single use technologies for facility design. 2. Smallest mall footprint for upstream processing saves initial investment, significantly. 3. Multiproduct facilities to be designed capable of offering production output in kilograms per month. 4. Completely avoiding the use of conventional Stainless steel bioreactors. 5. Culture revived from single low-passage cell bank can be expanded for several months without compromising the product quality. With continuous processing advantage while adapting an overexploitation approach, the cultured cells can be processed up to 60 passages, typically.

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November 09-11, 2017 Vienna, Austria

Preparation and evaluation of silicon-containing coatings on bioabsorbable Mg/27.5HA nanocomposite for bone tissue engineering

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A powder metallurgy technique to fabricate silicon and magnesium-oxide coated magnesium-based bionanocomposite with hydroxyapatite has been developed. The sample coated with double layer MgO/Si was compared with bare Mg/27.5HA nanocomposite and the sample coated by Si layer only. The surface microstructure and the cross section of bare and coated samples before and after corrosion were examined by field-emission scanning electron microscopy (FE-SEM). The corrosion performance of bare and coated samples was evaluated using potentiodynamic polarization, electrochemical impedance spectroscopy and immersion tests. A decrease from 4.28 to 0.65 mm/year was observed in corrosion rate of Mg/27.5HA nanocomposite with monolayer Si-coating. The results indicated that the corrosion resistance of Mg/27.5HA nanocomposite was significantly improved by MgO/Si double layer coating. Moreover, MgO/Si coating is promising to enhance biocompatibility of Mg/27.5HA nanocomposite for implant application. Antibacterial tests of the bare and coated samples revealed their antibacterial activity against *Staphylococcus aureus* and *Escherichia coli*.

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November 09-11, 2017 Vienna, Austria

Refolding control of highly disulfide bonded proteins by multi gradient on-column strategy

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Correct formation of disulfide bonds is crucial in the refolding of multi disulfide bonded inclusion body proteins. Covalent aggregation due to wrong cross linking of disulfide bonds leads to a low refolding yield. On-column refolding can be used to refold complex proteins under controlled conditions. This study reports the development of a multi gradient strategy for on-column refolding of high disulfide bonded proteins using size exclusion chromatography. Recombinant tissue plasminogen activator (r-PA, reteplase) with 9 disulfide bonds was used as the protein model. Applying a four-gradient strategy, including decreasing linear gradient of urea concentration and increasing linear gradients of pH, cysteine, and arginine concentration at the same time, resulted in 49.72% activity recovery and 91% mass recovery, which was 4 and 2.5-fold more than conventional on-column refolding with non-gradient and urea-arginine gradient strategies respectively. Successful application of multi gradient strategy in improving refolding yield of reteplase demonstrated controllability of the refolding process using size exclusion chromatography with a rationally designed gradient strategy which can be used in the refolding of other complex proteins too.

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November 09-11, 2017 Vienna, Austria

***Sarcopoterium spinosum*: An antidiabetic medicinal plant with a novel mechanism of action**

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Sarcopoterium spinosum (*S. spinosum*) is an abundant plant in Israel, used by Bedouin traditional medicine for the treatment of diabetes. In our previous studies the glucose lowering properties of this herb were validated *in vitro* and *in-vivo*. The goal of this study is to clarify the mechanisms of action mediating the effects of *S. spinosum* on glucose uptake. *S. spinosum* facilitates glucose uptake by a unique mechanism, different from that induced by either insulin or metformin; *S. spinosum* increased glucose uptake by 3T3-L1 adipocytes in a mechanism involving Glut4 translocation, independent of AMPK or PI3K activity. Akt activation is required to induce *S. spinosum*-dependent glucose uptake, however its mechanism of activation is still unclear; while neither ser473 nor thr308 were phosphorylated by *S. spinosum*, translocation of Akt from cytoplasm to membrane and nucleus was detected. In addition, substrates of Akt were phosphorylated by the extract. The hypothesis that *S. spinosum* utilizes a different set of proteins to induce glucose uptake was supported by results demonstrating that differentiating adipocytes respond differently to insulin and *S. spinosum*; while insulin gradually enhanced glucose uptake from the 11th day of differentiation, *S. spinosum* increased glucose uptake from the 8th day of differentiation. In addition, *S. spinosum* and insulin had additive effect on glucose uptake in fully differentiated adipocytes. Phosphoproteomics of serine/threonine residues phosphorylated by *S. spinosum* followed by bioinformatic analysis indicate for the activation of insulin-receptor pathway. We conclude that active ingredients in *S. spinosum* activate insulin signaling by a unique mechanism. Clarifying this mechanism of action may lead to the development of new agents for the treatment of diabetes.

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6th International Conference and Exhibition on PHARMACOLOGY AND ETHNOPHARMACOLOGY

November 09-11, 2017 Vienna, Austria

Small is big: Magic microfluidic droplets

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Droplets of nanoliter and subnanoliter are useful in a wide range of applications, particularly when their size is uniform and controllable. Examples include biochemistry, biomedical engineering, food industry, pharmaceuticals, and material sciences. One example of their many fundamental medical applications is the therapeutic delivery system for delivering site-specific therapy to targeted organs in the body and as the carriers for newer therapeutic options. The size, the size distribution, the generation rate and the effective manipulation of droplets at a scale of nano, pico, femto and even atto liters are critical in all these applications. We make an overview of microfluidic droplet generation of either passive or active means and report a glass capillary microfluidic system for synthesizing precisely controlled monodisperse multiple emulsions and their applications in engineering materials, nanofluids, microfibers, embolic particles and colloidosome systems. Our review of passive approaches focuses on the characteristics and mechanisms of breakup modes of droplet generation occurring in microfluidic cross-flow, co-flow, flow-focusing, and step emulsification configurations. The review of active approaches covers the state-of-the-art techniques employing either external forces from electrical, magnetic and centrifugal fields or methods of modifying intrinsic properties of flows or fluids such as velocity, viscosity, interfacial tension, channel wettability, and fluid density, with a focus on their implementations and actuation mechanisms. Also included is the contrast among different approaches of either passive or active nature.

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