



The 8th International Society for the Development of Natural Products

OCTOBER 10-11, 2014



Organizer : Fudan University

Co-organizer : National Natural Science Foundation of China

Shanghai Pharmaceutical Association

Shanghai Traditional Chinese Medicine Trade Association

Shanghai Pharmacological Society

Shanghai Fudan-Zhangjiang Platform for Drug Discovery

The 8th ISDNP

Dear Friends and Colleagues,

On behalf of the organizing committee, we are pleased to meet you here in Shanghai for the 8th International Society for the Development of Natural Products (ISDNP) during October 10-12, 2014.

The objective of the conference is to provide a forum for international scholars, researchers, professionals involved in the areas of natural products. This time, the organizing committee has chosen “New Trends in the Development of Natural Products” as the main theme for the conference, with a focus on the latest developments and trends, as well as future prospective in the field of natural product discovery, development and application. The conference includes plenary lectures, oral and poster presentations, and various social programs for hundreds participants from all over the world.

The focus of this conference includes all aspects of natural products, including detection of natural product chemistry, pharmacology of natural product drug, new methods for natural product analysis, novel aspects of natural product exposure, etc.

For more detailed information of the conference and submission of abstract, please follow the link: <http://www.isdnp.fudan.edu.cn>.

We thank you for your participation at this great international conference. And we are very pleased to build up such a platform for scientific research communication as well as international friendship.

Sincerely,



Dr. Zhu Yi Zhun, Conference Chair
Professor, College of Pharmacy, Fudan University
826 Zhangheng Road, Shanghai, China

The 8th International Society for the Development of Natural Products

Agenda

2014 Oct.10 th 8:30 am—12:15 noon			
8:30—8:40	Yizhun ZHU Trevor YEE Rongxing WANG	Chairman of 8 th ISDNP Welcome speech: Founder of ISDNP President, Shanghai Pharmaceutical Association	
Chair	Prof. Yi-zhun ZHU, Prof. Wan-sheng CHEN		
8:40—9:05	Ren-xiang TAN	Nanjing University, Jiangsu, China	Mining bioactive molecules from symbionts
9:05—9:30	Trevor YEE	University of the West Indies, Mona, Jamaica	Hypoglycaemic and Hypotensive constituents from <i>Eucalyptus camaldulensis</i> .
9:30—9:55	Ge LIN	The Chinese University of Hong Kong, China	Herbal hepatotoxicity, development of specific biomarker for its diagnosis
9:55—10:20	De-an GUO	Shanghai Institute of Materia Medica, Chinese Academy of Sciences, Shanghai, China	Strategies to elaborate overall TCM quality monographs
10:20—10:35	Tea Break		
Chair	Prof. Ren-xiang TAN, Prof. Ling-yi KONG		
10:35—11:00	Wan-sheng CHEN	Second Military Medical University, Shanghai, China	Quality-design in Traditional chinese medicine: a case study about <i>Isatis indigotica</i> fort
11:00—11:25	Yan-bing ZHANG	Zhengzhou University, Zhengzhou, China	Isolation and purification of Chalcone, the synthesis of its derivatives and evaluation of their cytotoxicity
11:25—11:50	Wen-cai YE	Jinan University, Guangzhou, China	Securinega Alkaloids from the Plants of Flueggea spp
11:50—12:15	Qiang YU	Shanghai Institute of Materia Medica, Chinese Academy of Sciences, Shanghai, China	TCM compounds that regulate the JAK/STAT signaling pathway
12:15—13:30	Lunch		
2014 Oct.10 th 13:30pm—17:00pm			
Chair	Prof. Trevor YEE, Prof. Xiao-he XIAO		
13:30—13:55	Wei-dong ZHANG	Second Military Medical University, Shanghai, China	Chemical biology study in Traditional chinese medicine: target profiling of bioactive natural products
13:55—14:20	Ling-yi KONG	China Pharmaceutical University, Nanjing, China	Research of complex and bioactive terpenoids from medicinal plants of meliaceae family
14:20—14:45	Yang QIU	GSK R&D Shanghai, Shanghai, China	Development of innovative TCM

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14:45—15:10	Hong-min LIU	Zhengzhou University, Zhengzhou, China	The anti-tumor activity and molecular mechanisms of OP46, a novel ent-kaurene diterpenoid compound from <i>Isodonrubescens</i>
15:10—15:25	Tea Break		
Chair	Prof. Wei-dong ZHANG, Dr. Qiu YANG		
15:25—15:50	Yi-zhun ZHU	School of Pharmacy, Fudan University, Shanghai, China	Discovery of novel compounds for heart and brain: 2 preclinical trials
15:50—16:15	Xiao-he XIAO	China Military Institute of Chinese Medicine, Beijing, China	Standardization of Chinese herbal medicine: from lab to clinic
16:15—16:40	Hai-bo ZHU	Chinese Academy of Medical Sciences & Peking Union Medical College, Shanghai, China	H007, A cordycepin derivative improves cholesterol metabolism in macrophages
16:40—17:00	Lin LU	AmGene Technology Co. Ltd.	New Technology for molecular mechanisms
Conference dinner (by invitation only)			
2014 Oct. 11th 8:00am—12:05pm			
Chair	Prof. Xun SUN, Prof. Thejani Rupika DELGODA-CLARKE		
8:00—8:25	Yong-hong LIU	South China Sea Institute of Oceanology, Chinese Academy of Sciences, Guangzhou, China	Bioactive metabolites from the marine symbiotic microorganisms
8:25—8:50	Guo-qiang LI	Ocean University of China	Study on chemical and bioactive diversities from the Coral-associated organisms in South China Sea
8:50—9:15	Hou-wen LIN	Shanghai Jiao Tong University School of Medicine, China	Discovery of Structurally Intriguing and Biologically Active Secondary Metabolites from <i>Xisha</i> Marine Sponges and Associated Microbes
9:15—9:40	Yue-wei GUO	Shanghai Institute of Materia Medica, Chinese Academy of Sciences	Chemoecology guided discovery of drug leads from South China Sea marine invertebrates
9:40—9:55	Tea Break		
Chair	Prof. Dao-feng CHEN, Prof. Ge LIN		
9:55—10:20	Xun SUN	College of Pharmacy, Fudan University, Shanghai, China	Design, synthesis and biological evaluation of resveratrol dimmers and their derivatives
10:20—10:45	Rupika DELGODA	University of the West Indies, Mona, Jamaica	Meeting the gaps for developing natural products in the Caribbean: a Jamaican model
10:45—11:10	Ai-jun HOU	College of Pharmacy, Fudan University, Shanghai, China	The Moraceae Family: an Important Source of Drug Discovery and

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			Development
11:10—11:35	Lin-lin LI	Xinjiang Medical University, China	The extracts of <i>Coreopsis tinctoria</i> Nutt.on experimental diabetes Rats and its mechanism
11:35—12:05	Jun YAN	Tecan (Shanghai) Trading Co. Ltd.	New Technology for drug screening
12:05—13:30	Lunch		
2014 Oct. 11 th 13:30pm—17:30pm			
Chair	Prof. Yong-hong LIU, Prof. Ai-jun HOU		
13:30—13:55	Dao-feng CHEN	College of Pharmacy, Fudan University, Shanghai, China	Isolation and Characterization of Anti-complement Agents from Traditional Chinese Medicines
13:55—14:20	Xiao-yan SHEN	College of Pharmacy, Fudan University, Shanghai, China	Therapeutic effect of Cryptotanshinone on collagen-induced arthritis in DBA/1 mice via reciprocal regulation of Treg/Th17 balance by impairing STAT3 acetylation
14:20—14:45	Zheng-tao WANG	Shanghai University of Traditional Chinese Medicine	Investigation into the pharmacophore of iridoid glycosides from Chinese medicinal herbs
14:45—15:10	Hui WANG	Institute for Nutritional Sciences, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences	Japonicone A suppresses growth of Burkitt's lymphoma cells through its effect on NF-κB
15:10—15:25	Tea Break		
15:25—17:00	Poster section		
17:00—17:10	Best Poster Award and closing ceremony		
17:30—20:00	Farewell dinner (by invitation only)		

Speaker Section

Mining bioactive molecules from symbionts



Prof. Renxiang TAN

*Institute of Functional Biomolecules, State Key Laboratory of
Pharmaceutical Biotechnology, Nanjing University, Nanjing, P. R. China.*

Plants and animals could not live (well) without numerous benefits from a community of symbionts which are microorganisms residing inside their hosts. Symbionts have been evidenced to help hosts to survive in stressed conditions. A growing pile of data has indicated that the ‘host-helping’ effects of symbionts are at least in part ascribable to their production of functional compounds. As generally accepted, symbionts are specialized but poorly investigated microorganisms, and some of them may be a rich source of functional molecules with novel architectures and/or promising biological functions. This talk will mention a selection of the new findings about the topic and future trends of the field.

Hypoglycaemic and Hypotensive constituents from *Eucalyptus camaldulensis*.



Prof. Trevor H. YEE

Natural Products Institute, the University of the West Indies, Mona, Kingston, Jamaica.

Since the 1920's, eleven species of *Eucalyptus* have been introduced in Jamaica by the National Water Commission and Forestry Department, as a part of their forest and watershed management programs, and as a source for charcoal. A number of these species have become established in our forests and have been the sources of several ethno-medical claims.

One of these species is *Eucalyptus camaldulensis*, for which the leaves and stems are used in some areas of rural Jamaica for the treatment of high blood sugar levels. An extract of the leaves and stems confirmed its hypoglycaemic activity and that the activity was contained in the non-polar fractions. Further extractions led to the isolation of three compounds, which were responsible for the activity, two of these forming a synergistic interaction with each other. Still further investigations in the activities of these compounds showed that all three were also hypotensive.

The Identifications of the three compounds were done by spectroscopy and direct comparisons with the authentic compounds, and these confirmed their identity as a hydrocarbon and two fatty acids, the latter being widely consumed in foods. The oils which contain the fatty acids as the major components were also active against hypoglycaemia and administration of the fatty acids was found to be active against both disease states when done either intravenously and orally.

Comparisons with Metformin and Captopril as positive controls, finally confirmed the efficacies of the compounds.

Herbal hepatotoxicity, development of specific biomarker for its diagnosis



Prof. Ge LIN

School of Biomedical Sciences, Faculty of Medicine, The Chinese University of Hong Kong, Hong Kong SAR

Drug-induced liver injury (DILI) is one of global leading causes of acute liver failure. Although the true prevalence and incidence are unknown, herbal and dietary supplement induced hepatotoxicity has been reported to be 10-75% of DILI cases from different countries. Clinically, the diagnosis of DILI induced by herb and dietary supplement is challenging and very difficult due to the lack of characteristic clinical features and specific tests. Therefore, development of specific diagnosis and advances in the understanding of the risk involved are needed to improve herbal medicine safety and public healthcare.

Among different herbs and dietary supplements induced hepatotoxicity, pyrrolizidine alkaloid-induced liver injury (PA-ILI) has been known for a long time. However, to date, the specific and confirmative diagnostic method and targeted treatment are unavailable for PA-ILI. PAs are widely distributed in plants, and the intake of PA-containing medicinal herbs as well as PA-contaminated foodstuffs causes numerous cases of liver injury with a high mortality worldwide. In this presentation, using PA-ILI as an example, our translational research from the basic science to delineate the toxic mechanism to the clinical application to develop a mechanism-based biomarker for PA-ILI diagnosis will be presented. Moreover, the risk assessment of PA-ILI due to the intake of PA-containing medicinal herbs will also be addressed. [Supported by Research Grant Council of Hong Kong (GRF Grants no. CUHK469712, and CUHK471013) and CUHK (Direct Grants no. 2041744 and 4054047)]

Strategies to elaborate overall TCM quality monographs



Prof. De-an GUO

The National Engineering Laboratory for TCM Standardization Technology, Shanghai Research Center for TCM Modernization, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, Shanghai, P.R. China

Traditional Chinese medicine (TCM) is an extremely complex system with hundreds of or even thousands of chemical components. Hence, it is a great challenge to establish the quality control standards for such a complex TCM system. The first challenge would be to clarify the chemical composition of the herbs or herbal formulas. Secondly, the active or effective components should be clear in order to establish the chemical markers for assay. Thirdly, translation from the basic research results to the feasible quality standards should be performed by setting up a research model. As to the above-mentioned challenges, we have made a great endeavor, in the last decade, to perform chemical, metabolic and biological analyses for TCM complex system to aim at elaborating scientific and feasible quality standards of TCM herbal medicines and their preparations. A series of chemical analytical methods including fingerprinting have been developed for the comprehensive analysis of multiple component TCM system including multiple marker quantification of single herbs or herbal combinations. Metabolic analysis for TCM herbal medicines also plays an important role for clarifying the active components of TCM complex systems. Metabolic fingerprint profiling method was developed for a number of typical Chinese herbs including Licorice and Chinese Salvia. Advances in high-throughput ‘omics’ technologies to measure changes of genes, proteins, and other biomolecular components in complex biological systems have dramatically revolutionized research of traditional Chinese medicine. Proteomics techniques such as 2-DE and nano-LC-MS/MS were used in our lab to study the mechanisms of TCM including *Salvia miltiorrhiza*, *Ganoderma lucidum*, etc. On the basis of the above mentioned methods and techniques developed in the lab, comprehensive quality standard model has been established for TCM by taking these thoroughly investigated TCM herbs as exemplified cases. Finally, the comprehensive quality monographs of several above-mentioned herbs were elaborated and recorded in Chinese Pharmacopoeia and United States Pharmacopoeia.

Quality-design in Traditional chinese medicine: a case study about *Isatis indigotica* fort



Prof. Wan-sheng CHEN

Department of Pharmacy, Changzheng Hospital, Second Military Medical University

Quality is the basis for the efficacy of Traditional Chinese Medicine (TCM), affecting herbs, formulations, and even the practice of TCM itself. In our laboratory, we used *Isatis indigotica*, a prevalent Chinese medicinal herb, as a model to illustrate strategies and methods for TCM Quality-design study. **First of all**, tetraploid *I. indigotica* ($2n=28$) with better yield, higher antiviral activity and enhanced resistance was obtained from its natural diploid progenitor ($2n=14$) after selection for five years. Chemical investigation demonstrated the lignans including lariciresinol and larch lignan glycosides represented important efficacious substances for the antiviral effect, which accumulated more in tetraploid *I. indigotica* than diploid progenitor. A further comprehensive survey of global gene expression performed by using an *Arabidopsis thaliana* whole genome Affymetrix gene chip revealed the variation of gene expression between autotetraploid and diploid *I. indigotica*, providing a pool of candidate genes for improving *I. indigotica* quality through transgenic manipulation. **Next**, in order to facilitate the process of isolating these quality-related genes (especially gene family) such as that involved in biosynthesis of effective metabolites, *I. indigotica* transcriptome sequencing was performed. By using this gene database, a large number of quality-related genes were isolated and intensively investigated, including stress resistance-involved transcription factor family *liWRKYs*, signal transduction-involved gene *liCPK1* and *liCPK2*, plant development-involved gene *liSDD1*, lignans biosynthetic pathway regulatory factor family *libHLHs*, and a series of lariciresinol biosynthetic pathway genes such as *liPAL* (DQ115905), *liC4H* (GU014562), *li4CL* (GU937875), *liCCR* (GQ872418), *liCAD* (GU937874), *liC3H* (JF826963), *liCCoAOMT* (DQ115904), and *liPLR* (JF264893), as well as *liDifs* family. **Lastly**, gene-based metabolic engineering study was performed to improve quality. For example, overexpression of PLR greatly enhanced lariciresinol production in *I. indigotica* hairy root cultures with the content of 135 $\mu\text{g/g}$ DW, which was appropriately 6.8-fold more than that in the wild-type counterpart, transgenic *I. indigotica* expressing *Bt CryIAc* and *Pinellia ternata agglutinin (pta)* significantly enhanced plant tolerance to moths and aphids. Our study not only prompted the possibility to obtain *I. indigotica* with higher quality, but also provided a reference for the study of TCM Quality-Design.

Meeting the gaps for developing natural products in the Caribbean: a Jamaican model



Prof. Rupika DELGODA

Natural Products Institute, University of the West Indies, Mona, Jamaica

At the heart of bioprospecting and development of ethnopharmacological claims, lies the capacity for conducting biological activity screens, which is often the missing link, in many developing countries. High to medium throughput screening capabilities allow for large libraries of compounds and extracts to be screened for biological activity against specific targets. Like many parts of the developing world, the Caribbean has been lagging behind, and thus its full potential is yet to be unveiled. Blessed with a relatively high 25% endemism among the vascular plants, a rich marine fauna and flora, and a culture of ethnomedical practices, Jamaica holds much promise. Working in collaboration with chemists, the Natural Products Institute, at the University of the West Indies, in Jamaica, has expended much effort to develop capacity of bioactivity screening. This presentation highlights our efforts and current capacity that reveals value in cancer prevention and treatment of Jamaican biodiversity, that have led to a preliminary patent in chemoprevention; testing of herbal medicine safety to avoid drug-herb interactions; and thus the involvement and contribution to the nation's efforts to build a nutraceutical industry.

Securinega Alkaloids from the Plants of Flueggea spp.



Prof. Wen-Cai Ye

College of Pharmacy, Jinan University, Guangzhou, 510632, P. R. China

The plants of genus *Flueggea*, belonging to the family Euphorbiaceae, are reported to be a rich source of a class of structurally unique indolizidine alkaloids known as *Securinega* alkaloids. Previous phytochemistry investigations on several species of this genus had led to the isolation of a number of *Securinega* alkaloids, some of which had been reported to show significant biological activities on the central nervous system and cytotoxicity.

In recent years, our group carried out systematically chemical and biological investigations on three selected species of this genus, *Flueggea virosa*, *F. suffruticosa*, and *F. leucopyra*. From above three plants, over 100 alkaloids had been isolated including a series of novel alkaloids possessed more than 10 unusual carbon skeletons. Among them, 5 novel alkaloids with unprecedented skeletons had been selected and reported as “Hot off the press” by the journal *Natural Products Report*. The biological activities of the isolated alkaloids had been tested. Some of the tested alkaloids exhibited potency activity in central nervous system, including modulation activity on the GABA receptor and regulating the morphology of Neuro-2a cells. Based on the structures of active compounds, we semi-synthesized a series of novel dimeric *Securinega* alkaloid analogues. The bioassay results showed that several bis(n)-securinine derivatives exhibited potential activities on the central nervous system. In addition, the structure-activity relationships of these compounds were also systematically evaluated.

TCM compounds that regulate the JAK/STAT signaling pathway



Prof. Qiang YU

*Shanghai Institute of Materia Medica, Chinese Academy of Sciences,
Shanghai, P.R. China*

Many Traditional Chinese Medicines (TCM) are used for treating inflammation and inflammation-related diseases. The chemical constituents and their molecular mechanisms of the TCMs however have not been fully understood. We have systematically analyzed 3000 TCM herb extracts using various inflammation-related cell and signaling pathway-based assays and found that many TCM extracts and compounds affected the JAK/STAT signaling pathways. Detailed studies have identified new protein targets and revealed new mechanisms of the TCM compounds in regulating the JAK/STAT pathways. Our model systems and approaches have provided an experimental basis to evaluate and to understand the TCMs.

Chemical biology study in Traditional Chinese medicine: target profiling of bioactive natural products



Prof. Wei-dong ZHANG

Modern Research Center for Traditional Chinese Medicine, School of Pharmacy, Second Military Medical University, Shanghai, P. R. China

Traditional Chinese Medicine (TCM), whose therapeutic efficacy has long been proved by thousands of years' clinical usage, is an important part of public healthcare and medicine research in China and Asian countries. Natural products derived from TCM are appreciated for their structural and functional diversity, which allow them to be ideal tools for the chemical biology study. However, the target identification is still a big challenge for natural products with novel structure and significant bioactivity but no specific targets. These years, techniques of genomics and proteomics, as well as tools of chemobioinformatics were employed for the target profiling of natural products in our lab. Some successful examples, especially the identification and validation of Bcl-2, 5-LO, DPP-IV and falcipain 2, for anti-tumor, anti-inflammatory, anti-diabetic and anti-microbic natural products, will be introduced in this presentation.

Research of complex and bioactive terpenoids from medicinal plants of meliaceae family



Prof. Ling-yi KONG

State Key Laboratory of Natural Medicines, Department of Natural Medicinal Chemistry, China Pharmaceutical University, Nanjing, China

Objective: In China, many plants of Meliaceae, distributed mainly in Southern China, were well-known Chinese folk medicines using to treat dysentery and fever. Recently, plants from Meliaceae were the research focus of natural products because of their diverse structures and interesting biological properties. In order to discover novel and bioactive natural products from Chinese folk medicinal plants, we have investigated the chemical constituents and bioactivity of several medicinal plants from Meliaceae family mainly distributed in Xishuangbanna of China.

Methods: Various methods and technologies, such as ODS, Sephadex LH-20, MCI, P-HPLC, Circular-P-HPLC, UPLC-Q/TOF/MS, were used in our systemic separation and purification research. 1D and 2D NMR, CD exciton chirality, X-ray, HRESIMS, and so on, such structural elucidation means and techniques, were applied to elucidate their planer structure, relative and absolute configuration. Meanwhile, the anti-inflammatory, insecticide, antifungal, HSP90 inhibitory and cytotoxicity activities of the isolated limonoids were evaluated.

Results: From 2007 to 2014, 15 species of 5 genera from Meliaceae have been researched which led to the isolation of 447 terpenoids (limonoids) including 257 new compounds and 5 types of novel carbon skeletons. Many methods were applied to identify their structures, especially the application of single-crystal X-ray diffraction and CD exciton chirality method to determine their absolute configuration. The novel isolates were evaluated for a series of biological activities, such as anti-inflammatory, insecticide, antifungal, cytotoxicity, and HSP90 inhibitory, more than 100 isolates showed notable bioactivities.

Conclusion: In our research program, many novel and complex terpenoids and limonoids were isolated and identified, which have enriched the types of natural organic compounds. Through bioactivities screening according to their traditional usage, the material foundation of these traditional medicines were elucidated basically. The findings of novel limonoids with significant bioactivity have potential academic value and scientific significance for the research and development of new drugs.

Development of innovative TCM



Dr. Yang QIU

Director, Head of Molecular Discovery Research, GSK R&D Shanghai, P. R. China

Traditional Chinese Medicine (TCM) has been widely practiced for thousands of years in China. It is based on ancient Chinese philosophy and views diseases as a disharmony between Yin and Yang and the five elements of the universe (metal, wood, water, fire and earth). Recent years, TCM has evolved to modern formulations and plays even more important role in health care system in China. However, current TCM products are mainly derived from ancient recipes and sold based on traditional belief and subjective experience. There is a general lack of objective and modern scientific evidence for their efficacies. In addition, product qualities are not well controlled and variable. The advancement of modern technology and the experience of western drug development provide the unique opportunity to develop innovative TCM products with demonstrated clinical efficacy and safety as well as quality assurance. The current trends in understanding potential underline mechanism of TCM using system biology, developing mixture with quality consistency, and demonstration of clinical efficacy will be discussed.

The anti-tumor activity and molecular mechanisms of OP46, a novel ent-kaurene diterpenoid compound from *Isodonrubescens*



Prof. Hong-min LIU

School of Pharmaceutical Sciences, New Drug Research & Development Center, Zhengzhou University, Zhengzhou, Henan, P. R. China

Objective: This current study is designed to determine the anti-tumor effects of OP46, a new ent-kaurene diterpenoid isolated from *Isodonrubescens* on human cancer cells and the underlying mechanisms.

Methods: Anti-tumor effect of OP46 was evaluated *in vitro* and *in vivo* with MTT method and Xenograft models. Cell morphology, apoptosis, autophagy and cell cycle distribution were observed or analyzed by fluorescence microscopy or flow cytometry.

Results: Compared with controls, OP46 caused strong anti-proliferative in human cancer cell lines HT-29, SGC-7901, MGC-803, EC109 cells *in vitro* and *in vivo*. OP46 also resulted in significant apoptotic, autophagy and G2/M phase cell cycle arrest in human esophageal carcinoma cell line EC9706. These cytotoxic effects of OP46 on EC9706 were induced by apoptosis via p53 mediating mitochondrial signaling pathway.

Conclusion: We found that OP46 remarkably inhibits proliferation in esophageal cancer cells. Our study also demonstrates that the OP46 induces apoptosis by p53 upregulating Bax, altering the mitochondrial membrane potential leading to the release of cytochrome c and consequently activating caspases-9 and -3. In addition, OP46 down-regulates the expressions of p70S6K, suggesting that OP46 may have a dual efficacy (p53 mediating mitochondria and mTOR pathway) in induction of apoptosis in EC cells.

Discovery of two novel compounds from Chinese herbs for heart and brain: 2 preclinical trials



Prof. Yi-zhun Zhu

Department of Pharmacology, School of Pharmacy, and Institute of Biomedical Sciences, Fudan University, Shanghai 201203, China, Department of Pharmacology, National University of Singapore, Singapore

E-mail: zhuyz@fudan.edu.cn or phczhuyz@nus.edu.sg

Over the last decades, the interest in compounds of natural origin was somewhat increased and the success of this approach has of course greatly improved disease management and life expectancy. The *Herba Leonuri*, a popular Chinese herb, has been reported by our previous study to improve coronary flow and microcirculation via a wide variety of pharmacological activities. Recently, we found that active ingredient--Leonurine (4-guanidino-n-butyl syringate, named also as SCM-198), the unique compound found only in *Herba Leonuri*, also confers vasodilator, antioxidative, and cardioprotective effects both *in vitro* and *in vivo*. More recently, our data extended these findings and demonstrated an athero-protective effect in hypercholesterolemic rabbits, which was associated with a reduction in smooth muscle cell migration, lower macrophages infiltration and reduced expression of several adhesion molecules in the atherogenic process. Our most recent studies also demonstrated that SCM-198 has neuroprotective effects on stroked rats through its angiogenic effect. We just completed its preclinical studies and developed both oral and *i.v.* formulations for clinical trials.

Another of our novel compound was extracted originally from garlic but structurally modified to become a novel sulfur-containing amino acid to release H₂S. H₂S is the third physiologically relevant gaseous signaling molecule with a diverse physiological profile. Therefore, modulation of endogenous H₂S production may be of therapeutic benefit in cardiovascular and CNS diseases. Interestingly, our group found that S-propargyl-cysteine (SPRC, named as ZYZ-802), exerted neuroprotective and cardiovascular-protective effects in *in vivo* and *in vitro* studies through modulation of endogenous cystathionine γ -lyase (CSE)/H₂S system. We identified that ZYZ-802 or nano-preparation of ZYZ-802 was shown to preserve endogenous CSE/H₂S levels and upregulated various anti-oxidative enzymes after myocardial infarction. In addition, ZYZ-802 also prevent against cytokine-mediated endothelial dysfunction. More recently, we also demonstrated that ZYZ-802 could induce angiogenesis involved in a STAT3-dependent mechanism, which may be a potential therapeutic strategy for ischemic disease through angiogenesis promotion. Based on the above studies, ZYZ-802 has been already by the end of the pre-clinical studies.

Taken together, above data suggest that both SCM-198 and ZYZ-802, could be used as a neuroprotective and cardiovascular-protective agent in the near future.

Standardization of Chinese herbal medicine: from lab to clinic



Prof. Xiao-he XIAO

China Military Institute of Chinese Medicine , Beijing, P. R. China

The issue about the quality control for Chinese medicines has been one of the main bottlenecks for Chinese medicines (CM) modernization. For a long time, finding and determining the indicative constituents has been the basic way for the establishment of CM quality standard, by which there are many inherent defects such as inaccuracy and little relationship between quality and effects. To get a radical and significant breakthrough in the investigation on the quality control of CM, the proposition “integrative quality of CM” and it’s preliminary performance route has been put forward in this paper. And its main aim is to provide some creative and practical ideas as well as methods for the establishment of CM quality control standard, and to meet with the requirements that the quality control standards for CM should be effectiveness- related, quantitative and accurate, controllable and assessable.

H007, A cordycepin derivative improves cholesterol metabolism in macrophages



Prof. Hai-bo ZHU

Institute of Materia Medica, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing, P.R.China.

Here we demonstrate that the role of H007 resulted in a decrease in macrophage cholesterol uptake. This functional change was caused by down-regulation of the mRNA and protein expression of LOX-1 in response to the synthetic AMPK activator H007 or AICAR. Mechanistically, AMPK activation increases Protein phosphatase 2A (PP2A) activity leading to Ser536 dephosphorylation of NF- κ B and down-regulation of LOX-1. Preventing activation of AMPK reduced the ability of H007 to suppress expression of pNF- κ B and LOX-1. Accordingly, AMPK activation reduced macrophage oxLDL uptake, an effect that was also reversed by inhibition of AMPK. Furthermore, the lesion size of atherosclerosis was smaller in H007 and AICAR-treated ApoE^{-/-} mice, and the expression of LOX-1 in aortas was modulated similar to that observed in macrophages.

Furthermore, we observed that IMM-H007 promoted cholesterol efflux via ABCA1-mediated pathway in the study of different cholesterol efflux pathways' contributions. In vivo RCT assay showed that IMM-H007 enhanced HDL-dependent cholesterol efflux function by promoting RCT to the plasma, liver, and feces. Finally, apoE^{-/-} mice fed with western diet containing 1.25% cholesterol and indicated doses of IMM-H007 treatment showed reduction of plaque size and lipid content, in addition, the number of macrophages decreased and collagen content increased in aortic root cryosection, suggesting IMM-H007 increase markers of plaque stability.

Conclusions: Our current findings suggest a novel mechanism of cholesterol metabolism regulation by H007 attenuating macrophage oxLDL uptake and enhancing cholesterol efflux function, eventually preventing atherosclerosis generation.

Bioactive metabolites from the marine symbiotic microorganisms



Prof. Yong-hong LIU

Research Center for Marine Microbes, South China Sea Institute of Oceanology, Chinese Academy of Sciences, Guangzhou, P.R.China.

Objective: Marine microorganisms are plentiful source of bioactive lead compounds. They are subjects of intensive screening program for new drug leads. Our research interest is a discovery of antitumor compounds from special eco-environment microorganisms isolated from biogenic and abiotic sources.

Methods:

1. Collection samples from biogenic (invertebrates, mangrove, sea weeds, etc) and abiotic (deep sea sediment and soils) sources.
2. Isolation of fungi and bacteria (especially actinomycetes) from the special eco-environment.
3. Mass culture of the symbiotic microorganisms.
4. Chemical analysis of the active components of symbiotic microorganisms
5. Simple in-house bioassays including brine shrimp assay, antimicrobial assay, antioxidant assay.

Results: 100 new and 100 known compounds were isolated from marine symbiotic microorganisms. The structures were established on the basis of NMR and MS analyses. The stereochemistry was defined by combined use of NMR, ECD, X-ray, and chemical derivation.

Conclusion: These compounds were screening for anticancer, antibacterial activities, some of them showed significant activities.

Study on chemical and bioactive diversities from the Coral-associated organisms in South China Sea



Prof. Guo-Qiang LI

School of Medicine and Pharmacy, Ocean University of China.

The Coral-associated organisms, especially the sponges, soft corals and gorgonians, have being an important origin of novel marine natural products. Recently, we systematically investigated one gorgonian rare species *Muriceides collaris* shaping in fan with unique azure branches collected off Weizhou islands, two Xisha (Paracel) Islands sponges *Aaptos suberitoides*¹ and *Plakortis simplex*², resulting in the isolation of series of new indene and azulene alkaloids, together with novel aaptamine dimmer alkaloids. The new structures isolated were identified on the basis of combined spectroscopic methods such as MS, NMR, and quantum chemical calculation. Evaluation of some isolates displayed potent bioactivity including cytotoxic, antifouling, and antiviral. Also an interesting phenomenon was observed in both the two species that, most of the new compounds were isolated as enantiomers, which apparently suggested a new problem worth of further investigation. Aforementioned study showed the coral-associated organisms in South China Sea to have plentiful chemical and bioactive diversities.

Discovery of Structurally Intriguing and Biologically Active Secondary Metabolites from *Xisha* Marine Sponges and Associated Microbes

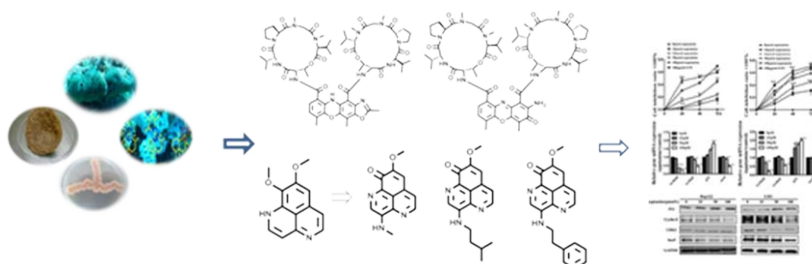


Prof. Hou-wen LIN

Key Laboratory for Marine Drugs, Department of Pharmacy, Renji Hospital,
School of Medicine, Shanghai Jiaotong University, Shanghai, P. R. China

Marine sponges rely on chemical defence as a major adaptation strategy to thrive in the extreme environmental condition of the sea, which may account for the diverse array of chemically intriguing, biologically active secondary metabolites that they produce.

By using spectrascopical, chemical and computational approaches, recently, our group has discovered a series of bioactive compounds with unusual structural features from various *Xisha* sponges, including sesquiterpene aminoquinones, aaptamine-like alkaloids, cyclopeptides, endoperoxides and actinomycin-like compounds with variety of potent activities. For example, dysifragilone C, an unusual sesquiterpene aminoquinone with a rearranged avarone skeleton, was isolated from the South China Sea sponge *Dysidea fragilis*, which inhibited the production of nitric oxide (NO) stimulated by lipopolysaccharide (LPS) in mouse RAW 264.7 macrophages with an IC_{50} value of $6.61 \pm 0.54 \mu M$. Sixteen new aaptamine type alkaloids, isolated from the sponge *Aaptos aaptos*, showed potent cytotoxicity against six human tumour cell lines with IC_{50} values ranging from 0.03 to $8.48 \mu M$. Furthermore, two new analogues of actinomycin D, isolated from the sponge *phyllospongia foliasce* associated bacteria *Streptomyce sp. PFS10*, showed the nM activities selectively against multiple cancer cell lines, which led us to begin the optimizing process of fermentation to probe the basis of its biosynthetic pathway.



Isolation and Characterization of Anti-complement Agents from Traditional Chinese Medicines*



Prof. Daofeng CHEN

(dfchen@shmu.edu.cn)

Department of Pharmacognosy, School of Pharmacy, Fudan University

826 Zhangheng Road, Shanghai 201203, China

The inappropriate activation of complement system may cause some severe diseases such as acute respiratory distress syndrome (ARDS) and systemic lupus erythematosus (SLE). In order to search for the natural anti-complement agents from the traditional Chinese medicines as the therapeutic medicines for prevention and treatment of the complement-associated diseases, this effort was initiated. Bioactivity-guided fractionation and isolation was performed to obtain the anti-complement agents from the traditional Chinese medicines. The structure, anti-complement activity and target in complement activation cascade of bioactive constituents were investigated and characterized. Animal study was performed as well to evaluate beneficial medical properties of the obtained anti-complement agents. A number of anti-complement constituents were isolated and characterized from several Chinese herbs. Animal study demonstrated that the polysaccharides obtained from *Bupleuri Radix* and *Houttuyniae Herba* had anti-complementary and immuno-modulating activities, and showed beneficial effects on the acute lung injury (ALI) induced by lipopolysaccharide or influenza virus A in rats/mice. Beneficial effect of the *Bupleurum* polysaccharides was also found on the SLE in mice. Our study indicated that the polysaccharides from the traditional Chinese medicines are valuable anti-complement agents, which might be useful in prevention and treatment of the complement-associated diseases. In addition, some small molecules were isolated as the anti-complement agents from *Arnebia euchroma* and *Paeonia suffruticosa*, their targets in complement activation cascade were identified and found to be diverse.

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Chemoecology guided discovery of drug leads from South China Sea marine invertebrates



Prof. Yue-Wei GUO

Shanghai Institute of Material Medica, Chinese Academy of Sciences

Natural products have played a significant role in the drug discovery process throughout the last hundreds years. What is the best strategy to investigate the biological potentialities of secondary metabolites? It is generally accepted that there are two ways to explore the pharmaceutical potentialities of natural products. The first way is so called bioassay guided isolation of bioactive natural products; the second one is so called random screening methodology. In fact, every procedure could be only partially satisfactory. Apart the above mentioned two solutions, an alternative way could be a good choice through studying the compounds that really play a biological role in the organism where they are present. This could be the starting point to discover other biological potentialities. Of course, to perform studies like these one needs a careful selection of promising biological systems and, also, the close collaboration among chemists, biologists and pharmacologists.

Trying to follow this bio-chemical approach some years ago we started to investigate marine nudibranchs that are extremely interesting from an ecological point of view. In fact, these mollusks are completely devoid of the mechanical protection of the shell. But, in spite of this apparent vulnerability, they are rarely victims of predators. This is due to a series of defensive strategies that include the use of chemicals that either derive from their food habits or are biosynthesized “de novo” by themselves.

In this lecture we will report the recent chemical studies on opisthobranch molluscs collected from South China Sea. All work has been performed in close collaboration with marine biologists who have correctly submitted the biological problems to the chemical analysis, and with pharmacologists who have carried out bioassay based on the clue provided through chemoecology studies.

Investigation into the pharmacophore of iridoid glycosides from Chinese medicinal herbs



Prof. Zheng-tao WANG

*Institute of Chinese Materia Medica, Shanghai University of TCM, 1200
Cailun Road, Zhangjiang Hi-Tech Park, Shanghai 201210, PR CHINA*

The present research aimed to investigate the *in vivo* pharmacophore of iridoid glycosides isolated from herbal medicines with cholagogic effect such as *Gentianae Radix et Rhizoma* and *Swertiae Herba*. Chemical and metabolic method have been applied to comparative study on the metabolic pathways of typical iridoid glycosides. The results was shown that seco-iridoid glycosides were readily subject to hydrolysis under biological conditions, and the aglycones were susceptible to phase I, phase II and multiple metabolism, while the iridoid glycosides was predominately transformed into aglycone forms as their principal circulating metabolites after oral administration. Furthermore, the major metabolites were isolated from bio-samples via chemical separation process, and several potential active products were prepared using enzyme and intestinal bacterial. In addition, hepatic stellate cell and H₂O₂-induced cytotoxicity model were used to study on the cytoprotective effect iridoid glycosides and their metabolites. The results revealed that, the metabolites could obviously increased cell survival rate, processing stronger hepatoprotective effect than prodrugs. According to the findings in this research, comprehensive and accurate understanding of iridoid glycosides ultimate *in vivo* behavior has been revealed and the aglycone moieties were proposed as the *in vivo* pharmacophore structures of iridoid glycosides, which demonstrated the medicinal material base of iridoid glycosides *in vivo* and also provided a new idea in study on the material foundation of therapeutic effectiveness of traditional Chinese medicine.

Design, Synthesis and Biological Evaluation of Natural Resveratrol Dimers and their Derivatives



Prof. Xun SUN

Department of Natural Products Chemistry, School of Pharmacy, Fudan University, 826 Zhangheng Road, Shanghai 201203, China

Resveratrol is a natural polyphenol stilbene from natural products, and reported to have a diverse range of biological and pharmacological properties, including anti-inflammatory, anti-platelet and antioxidant activities. A series of Resveratrol oligomers were isolated from *Caragana sinica* (Buc'hoz) Rehd., in which pallidol, a natural resveratrol dimer, possesses weak estrogen-like as well as anti-estrogenic effects. Therefore, it is important to develop efficient methodologies to synthesize these compounds for further biological and clinical investigation.

Concise total syntheses of isopaucifloral F, quadrangularin A and pallidol, starting from commercially available 3,5-dimethoxybenzoic acid, have been achieved through a sequential reaction processing involving Nazarov cyclization, Ramberg-Backlund olefination, and Friedel-Crafts alkylation.

Furthermore, a novel resveratrol dimer-based compounds database was built, including four structural types: indenone-, indene-, octahdropentalene- and sulfur-indene type, and the structure-activity relationship of these compounds were studied, in which indene- and octahdropentalene-based compounds exhibited good in vitro activities against paraquat-induced apoptosis in SH-SY5Y cells, while methyl substituted indenone-type compounds tended to exhibit good in vitro inhibitory activities on LPS-induced NO production in RAW 264.7 cells. Especially, isopaucifloral F was found to improve the symptoms of osteoporosis of ovariectomized female rats in a dose-dependent and time-dependent manner in vivo.

Isolation and purification of Chalcone, the synthesis of its derivatives and evaluation of their cytotoxicity

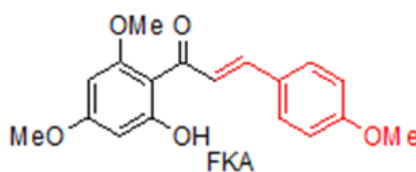


Prof. Yan-bing ZHANG

School of Pharmaceutical Sciences, Zhengzhou University, Zhengzhou, Henan, P.R.China

Previously, we found that FKA, a compound of chalcone, consists in KAVA plant spread over South America and has an outstanding anti-cancer activity on urinary bladder carcinoma. The similar effects of FKA were also reported in esophageal squamous carcinoma, gastric carcinoma, breast carcinoma, prostatic carcinoma.

In the current study, we designed and synthesized several classes of chalcone derivatives, including alkoxy, halogen, coumarins, heterocycle, thioester, triazole, double chalcone, taking FKA as a lead compound. We obtained more than 200 derivatives in total, all of them were characterized by means of IR, ¹H-NMR, ¹³C-NMR and MS spectral data. More than 80% of them had not been reported by literatures. Interestingly, we found that most of them showed a good anti-cancer activity in vitro. Based on the results, structure-function relationship were also summarized. Using MTT assay, flow cytometry, cell staining, western blotting, the mechanism of the compounds were also investigated. Our results indicate that this kind of compound induces cell death through activating oxidative stress and mitochondria-related apoptosis passway.



The Moraceae Family: an Important Source of Drug Discovery and Development



Prof. Ai-Jun HOU
School of Pharmacy, Fudan University

Moraceae is a large family, including some important groups such as *Morus*, *Artocarpus*, *Ficus*, and *Cudrania*. The moraceous plants are of important economic and medicinal values and are rich in prenylated phenolics with structural and biological diversity, which have attracted a lot of attentions as an important source of drug discovery.

Recently, a series of new compounds were isolated from some Moraceous plants by our group, including some compounds with new skeletons, novel 2-arylbenzofuran dimers, bioflavonoids, and Diels-Alder adducts. It is more important that some compounds promoted adipogenesis and induced up-regulation of the expression of glucose transporter 4 (GLUT4) and that some compounds showed inhibitory effects on pancreatic lipase (PL) and protein tyrosine phosphatase 1B (PTP1B). This will be of significance for exploring new lead compounds against diabetes and obesity. Furthermore, the antitumor constituents of *Cudrania tricuspidata* were explored, an antitumor traditional Chinese medicine. Mechanism of the active compound was investigated. Based on the investigations on *C. tricuspidata*, a new antitumor drug candidate is undergoing preclinical studies. In addition, the total syntheses of some bioactive isoprenylated flavonoids from the Moraceae family were also completed.

The extracts of *Coreopsis tinctoria* Nutt.on experimental diabetes Rats and its mechanism



Prof. Lin-lin LI
Xinjiang Medical University, P.R.China

Objective: To assay the quantitative determination of Trigonelline and diosgenin in different extract of Fenugreek by HPLC. And antioxidant activity on DPPH and hydroxyl freebase ($\cdot\text{OH}$) were studied. To observe therapeutic effects of *Trigonella foenum graecum*.L extract of alcohol (TFGe) on alcoholic fatty liver (AFL) of rats, trying to clarify the mechanism of its therapeutic effects.

Method: Using the CHP method to determinate Trigonelline ; diosgenin was analyzed by HPLC on Hypersil ODS-1 columns(4.6x250mm, 5 μm) with the mobile phase of methanol acetonitrile-water (86:14,v/v),the flow rate 1.0mL/min,detection wavelength 203nm,column temperature 35°C. Vitro antioxidant effects were studied by UV method. When pathological inspection showed that the model of alcoholic fatty liver of rats was established, intragastrically fed rats with TFGe for 4 weeks and silymarin to be the positive control. Serum lipid, total bilirubin (TBili), albumin(ALB) was detected : activity of Aspartate aminotransferase (AST), alanine aminotransferase (ATL), glutamyltranspeptidase (GGT) was detected; hepatic contents of TC, TG were tested : Serum level of tumor necrosis factor(TNF- α), adiponectin and AMPK was examined by enzyme-linked immunospecific assay (ELISA)method.

Results: The liner range of diosgenin was 43.2 $\mu\text{g/ml}$ ~ 864 $\mu\text{g/ml}$ ($r=0.9995$); average recovery ($n=9$) was 100.3%. Results showed that: Fenugreek extract have Scavenging effects on DPPH and hydroxyl radicals ($\cdot\text{OH}$). With TFGe treatment, contents of serum TG, TC, LDL-C of AFL rats were significantly decreased ; activity of AST, ATL was depressed ; hepatic contents of TC, TG were significantly reduced : the level of Serum TNF- α decreased while level of adiponectin increased highly. Steatosis of hepatic tissue obviously improved. But Serum level of TG increased as compared with that of the normal group.the level of Serum TNF- α decreased while level of adiponectin , SOD, AMPK increased highly.

Conclusion: Fenugreek extract have effects on Scavenging DPPH and hydroxyl radicals.TFGs have therapeutic effects on alcoholic fatty liver of rats.

Therapeutic effect of Cryptotanshinone on collagen-induced arthritis in DBA/1 mice via reciprocal regulation of Treg/Th17 balance by impairing STAT3 acetylation



Prof. Xiao-yan SHEN

Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, China

From an immunologic standpoint, the balance between Treg and Th17 cells is the major determinant in the pathogenesis of RA. Among the inflammatory cytokines implicated in RA, IL-6 and thus its mediated STAT3 signaling decide the differentiation of Treg and Th17 cells. To investigate the STAT3 inhibition of *Cryptotanshinone* (CTS), an active component isolated from the root of *Salvia miltiorrhiza* Bunge, on Treg function and the underlying mechanisms in RA, collagen-induced arthritis in DBA/1 mice and cell models including Raw264.7 and rat FLS were used. Our results revealed CTS treatment could prevent on joint inflammation and bone erosion. The level of collagen-specific IgG2 α was significantly reduced in CTS-treated mice. The percentage of Treg in CD4⁺ T cells was increased in a dose dependent manner by CTS treatment. CIA mice without treatment showed significant loss of Treg suppression (TGF- β and IL-10) on Th17 (IL-6 and IL-17 α). However, CTS treatment reversed the Th17/ Treg imbalance in a dose dependent manner. Further studies found that CTS significantly inhibited the overactivation of STAT3 signaling in CIA mice, and reduced the acetylation of STAT3 and the expression of p300. Ace-p65, the subtract of p300, also showed a decrease after CTS treatment. Those results indicate the anti-rheumatic effect of CTS might be achieved by inhibiting STAT3 acetylation via impairing P300. It provides the possibility to develop CTS as a potential therapeutic agent for RA patients.

Japonicone A suppresses growth of Burkitt's lymphoma cells through its effect on NF- κ B



Prof. Hui WANG

Key Laboratory of Food Safety Research, Institute for Nutritional Sciences, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200031, China

The tradition herbal medicines, composed with multiple biologically active compounds, are widely claimed to control human cancers with minimal adverse effects. To uncover the molecular mechanism of these agents is extraordinarily interesting and urgently needed for modern pharmacotherapy. Nuclear factor- κ B (NF- κ B), a transcriptional regulator of diverse genes, involved in cell survival, proliferation and apoptosis, has been implicated in various malignancies, inflammatory diseases and immunity disorders. *Inula japonica* Thunb is a traditional medicinal herb used for treatment of bronchitis, digestive disorders, diabetes, and inflammation. Here, we report that japonicone A (JA), a natural compound from *Inula japonica* Thunb, possesses antitumor activity against Burkitt's lymphoma via targeting the NF- κ B signaling cascade. A screen of various cell types revealed that JA killed cancer cells but had low cytotoxicity to normal cells. Burkitt's lymphoma cells were particularly sensitive. JA inhibited the growth and proliferation of Raji, BJAB, and NAMALWA lymphoma cells and resulted in G2/M phase arrest and apoptosis. Further, exposure of cells to JA caused inactivation of the TNF α -TAK1-IKK-NF- κ B signaling axis and inhibition of TNF α -stimulated NF- κ B activity and nuclear translocation, followed by down-regulation of NF- κ B target genes involved in cell apoptosis (Bcl-2, Bcl-XL, XIAP, TRAF2) and in the cell cycle and growth (Cyclin D, c-Myc). Moreover, for two mouse lymphoma models, JA inhibited local growth and dissemination of cancer cells to multiple organs. These results highlight the potential of Japonicone A as a chemotherapeutic agent and warrant its development as a therapy for lymphomas.

New sources of essential oils and aroma chemicals from the western ghats of India



Prof. Palpu Pushpangadan
*Amity Institute for Herbal and Biotech Products Development,
Peroorkada, Trivandrum -695 005*

The Western Ghats are a mountain range that runs almost parallel to the western coast of Peninsular India. It is a world heritage site and is one of the eight hottest hotspots of biological diversity in the world. The range starts from the south of river Tapti in Gujarat and extends to a distance of 1,600 kms through the Indian states of Maharashtra, Goa, Karnataka, Tamil Nadu and Kerala. The Western Ghats is the natural habitat of a rich biodiversity with over 5,000 flowering plant species, 139 mammalian species, 508 bird species, 179 amphibian species and 288 fresh water fish species. The damp forested slopes of the Western Ghats are the original location of the renowned spice black pepper (*Piper nigrum*) of commerce and history. A high level of endemism has been observed in the flora of Western Ghats with over 1,400 endemic species. Among the spices found in the Western Ghats are different species of pepper, cinnamon, cardamom, ginger, nutmeg, turmeric, bay leaf, star anise etc. In addition to these spices, the Western Ghats are natural sources of a large number of aromatic plants which are never investigated for their essential oil and aroma constituents. In this presentation, the essential oil constituents obtained from the fruits, rhizomes, leaves and flowers of a large number of hitherto uninvestigated aromatic plants belonging to the families Umbelliferae, Zingiberaceae, Rutaceae, Lauraceae, Myrtaceae, Aristolochiaceae etc, collected from the rain forest of Western Ghats in the state of Kerala, India will be discussed.

Perfection of Traditional medicine through the identification of metabolomics by metabolomic engineering



Prof. Varughese GEOGRE

Amity Institute for Herbal and Biotech Products Development, Peroorkada, Trivandrum

The traditional medicines used to be an individual based treatment regime wherein the traditional physicians used handpicked plant materials to prepare drugs/ formulations to treat their patients. The prescription and preparations of the drugs or remedies were also used to be person specific and based on the constitutional nature of the patient called 'Prakriti' as per the 'Tridosha' concept of Ayurveda/ Siddha. But the transformation of traditional medicine from such an individualized system to a commercial manufacturing system has resulted in great deterioration in the whole procedure and process of traditional medicine. Indeed, quality of the drugs became the casualty in this transformation. But with the advancement of biotechnology, a combination of genomic, proteomic and bioinformatics tools, it is now possible to replace the traditional plant collectors / plant breeders by genetic engineers, who would identify or design plants with the increased yields of metabolites. In spite of the advances in synthetic, computational and combinatorial chemistry and high throughput analysis, plants still continue to be the major sources of drugs, drug intermediates and lead molecules in drug development. However due to the lack of agro technology, only a few medicinal plants have been brought under commercial cultivation, while majority of the plants used by the pharmaceutical industry are extracted from their habitat. This has resulted in shortage of raw materials or loss of genetic materials and in a few cases the endangerment of the species itself. Metabolomic studies rely on the analysis of the multitude of small molecules (metabolites) present in the biological system. Most commonly, metabolomics is heavily supported by Mass Spectrometry (MS) and Nuclear Magnetic Resonance (NMR) as parallel technologies that provide an overview of the metabolome and high power compound elucidation. The combination of liquid chromatography (LC) - MS and NMR is a powerful methodology for identifying metabolites. Better chemical characterization of the metabolome will undoubtedly enlarge knowledge on the specific aspects of the product.

Poster Section

P01

Novel LSD1 inactivator inhibit gastric cancer cell growth, invasion and migration

Yi-Chao Zheng¹, Jin-Lian Ma¹, Zhi-Ru Wang¹, Jin-Feng Li¹, Li-Juan Zhao¹, Li-Jie Zhao¹, Qian-Na Zhan¹, Wen Zhao¹, Hong-Min Liu^{*,1}

1. New Drug Research & Development Center, School of Pharmaceutical Sciences, Zhengzhou University, 100 Kexue Avenue, Zhengzhou, Henan, P.R. China

The post-translational modifications of histone, e.g. methylation and demethylation, regulate chromatin structure as well as gene activation and repression. Lysine-specific demethylase 1 (LSD1), the first identified histone demethylase since 2004, has been reported to be up-regulated in numbers of cancers, including gastric cancer. Targeting LSD1 by specific inhibitors has shown to be a potential strategy to prevent cancer. Here, we generated and screened a series of novel 1, 2, 3-triazole-dithiocarbamate hybrids, which can specifically inhibit LSD1 activity with IC₅₀ less than 3 μM. Interestingly, these LSD1 inhibitors exhibit FAD competitive, with selective and potent cytotoxicity against LSD1 overexpressed human gastric cancer cell line MGC-803 and HGC-27, while they do not show any toxic effects on several normal cell lines. While cells were exposed to MGC-803, EMT mediated cell migration and invasion can be inhibited. In vivo study revealed that the 1, 2, 3-triazole-dithiocarbamate based LSD1 inhibitor performed strong anti-cancer effect. Our findings indicate that these LSD1 inhibitors may be potential candidates to target LSD1 overexpressed gastric cancer cells with the ability to induce cell apoptosis, arrest cell cycle at G2/M as well as inhibit cell migration and invasion.

P02

Synthesis and biological evaluation of novel Leonurine derivatives and Leonurine-Aspirin hybrids as cardioprotective agents

Huan Gao^{1,2}, Xianfeng Gu², Yihun Zhu²

1. Department of Medicinal Chemistry, School of Pharmacy, Jilin University, Changchun, P.R. China

2. Department of Pharmacology, School of Pharmacy and Institute of Biomedical Sciences, Fudan University, Shanghai, P.R. China

Objective: Leonurine (3,5-dimethoxy-4-hydroxy-benzoic acid 4-guanidino-butyl ester), an effective ingredient from Chinese motherwort, is reported to have cardioprotective effect due to its anti-oxidation and anti-apoptosis properties. In the early structure-activity relationship (SAR) study of Leonurine, we have proved that the length of aliphatic chain and the replacement of methoxyl group have no direct effect on its activity. But the necessity of the guanidyl group remained to be studied. Considering Aspirin's ability to protect ischemic myocardial cells, we planned to synthesize a series of Leonurine-Aspirin hybrids and do some further work on Leonurine derivatives to fulfill the SAR study, and then to study the mechanisms of their anti-oxidation and anti-apoptosis properties. We intended to obtain some more effective and less toxic compounds.

Methods: Part one: Choosing Syringate as the starting material, we got target hybrids by carbonylation, esterification, amidation and deacetylation. Part two: Related Leonurine derivatives were acquired by modifying the guanidyl group and the phenolic hydroxyl group. Part three: Using the model of oxidative stressed H9c2 cells, we detected cell survival rate by MTT and evaluated the antioxidant activity by SOD、CAT、MDA and LDH tests.

Results: Three synthetic approaches were established. A serial of Leonurine derivatives and Leonurine-Aspirin hybrids were synthesized, and all the structures were identified by MS, ¹H-NMR and ¹³C-NMR.

The modification on Leonurine showed that removing the guanidyl group will greatly diminished the activity of myocardial protection while replacing it with nitrate or ureido group keep it sustained.

Among all the derivatives, LA-2, LA-3 and L-u showed promising data in cellular tests. MTT test showed that the cell survival rate elevated significantly after pretreatment with 0.1μmol/L of LA-2, LA-3 and L-u(P<0.05), which were consistent with SOD and CAT results. The study also indicated potent cardioprotective effects of LA-2 and LA-3 against hypoxia-induced H9C2 cell damage at lower molar concentration (10-fold less than Leonurine required and 100-fold less than aspirin required).

Conclusions: We completed the SAR study of Leonurine and proved that guanidyl group was the necessary part for activity. Pharmacological evaluation showed that anti-oxidative stress and anti-apoptosis of these compounds contributed to the cardioprotective effect. All these results demonstrated that LA-2 and LA-3 is worth to be further investigated as the potential anti-myocardial ischemia drugs.

P03

A novel hydrogen sulfide-nitric oxide-releasing hybrid (ZYZ-803) in stimulation of angiogenesis: new insights into the biology of hydrogen sulfide and nitric oxide

Qingxun Hu¹, Dan Wu¹, Xianfeng Gu¹, Yi Zhun Zhu^{1*}

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R.China

Aims: Angiogenesis is a complex process of extracellular matrix remodeling and changing, in which endothelial cells (ECs) play an important role. In endothelial cells, hydrogen sulfide (H₂S) and nitric oxide (NO) are synthesized respectively by cystathionine γ-lyase (CSE) and the endothelial isoform of NO synthase (eNOS). Growing evidence has suggested that H₂S and NO are important regulators of cell angiogenesis. However, it is hardly conclusive that whether H₂S and NO react together better or not. Herein, we synthesize a novel hydrogen sulfide-nitric oxide-releasing hybrid (ZYZ-803) with H₂S donor and NO donor. This hybrid is used as a tool to unravel the mystery and investigate the cardiovascular biology of these gases.

Methods: The novel hybrid was developed by H₂S donor coupled with NO donor. Cell viability and p53 expression were determined to assess cellular toxicity. The releases of H₂S and NO from ZYZ-803 were measured via the spectrometer assay. Angiogenesis was assessed using *in vitro* parameters (*i.e.* endothelial cell proliferation, transwell migration assay and formation of tube-like structure) and *in vivo* by matrigel plug assay and aortic ring assay. Cyclic guanosine 5'-monophosphate (cGMP) content in cell was measured by enzyme-linked immunosorbent assay. Western blot was used to determine the expression levels of proteins.

Results: Treatment of ECs with ZYZ-803 did not cause significant cytotoxicity or apoptosis. ZYZ-803 released H₂S and NO slowly after administration *in vivo*. ZYZ-803 concentration-dependently accelerated cell growth, migration and tube-like structure formation *ex vivo* and *in vivo*. At the same concentration, the hybrid was more potent than either one alone. These effects of ZYZ-803 on cell proliferation and migration were prevented by PI3K inhibitor LY294002, ERK inhibitor UO126 and p38 inhibitor SB203580. Incubation of ECs with ZYZ-803 resulted in the increase of phosphorylation of PI3K, Akt, ERK and p38, and these effects could be blocked by respective inhibitors. ZYZ-803 increased the content of cGMP and activated protein kinase G (PKG) as well as its downstream effectors.

Conclusion: The present study proves that H₂S and NO react together much potent than either one alone in cell proliferation. As a slow-releasing H₂S and NO hybrid, ZYZ-803 is a useful tool to study the interactions between H₂S and NO in many biological effects. Moreover ZYZ-803 could be recognized as an important potential drug in the therapy of cardiovascular disease.

P04

Effects of Betaine hydrochloride on cerebral Ischemia reperfusion Injury in rats

Lin Li¹, Xiao-Ming Xin¹, Yun-Sheng Gao¹

Department of Pharmacology, School of Pharmacy, Taishan Medical University, Taian, Shandong, P.R.China

Objective: Betaine hydrochloride(BTH) is the active ingredient in traditional Chinese medicine, such as Chinese wolfberry and Achyranthes etc. It has been proven to protect the heart and blood vessels, besides that it also can improve Oxidative Stress and Hyperhomocysteinemia etc. The aim of this study was to evaluate the neuroprotective effects of the Betaine Hydrochloride (BTH) against blood brain barrier disruption and neurological injury in the cerebral ischemia/reperfusion in rats.

Methods: Focal cerebral I/R injury in rats was induced by left middle cerebral artery occlusion(MCAO) according to previously described methods with minor modifications for 1h followed by reperfusion for 48h. Adult male Sprague-Dawley (SD) rats weighing 250 to 300 g were randomly assigned to five groups: Sham group; I/R group; BTH-treated groups at doses of 10mg/kg and 20mg/kg; Edaravone(ED) -treated group. The cerebral blood flow (CBF) was measured by laser Doppler flowmetry. Rats were evaluated for neurological deficits just before sacrifice. Brains were harvested for infarct size estimation and 2,3,5-triphenyltetrazolium chloride (TTC) staining after 48h reperfusion. Various biochemical indexes in brain tissue homogenate were assayed by colorimetry, including malondialdehyde (MDA), superoxide dismutase (SOD) and the level of homocysteine (HCY) and activity of MMP-9 by ELISA; To evaluated the morphological changes in brain tissues among the experimental groups, hematoxylin and eosin (HE) staining was performed.

Results: BTH afforded neuroprotection as evident through significant reduction in cerebral blood flow and infarct volume, improvement in neurological deficit. The treatment of BTH lowered MDA content, up-regulated SOD, down-regulated HCY and MMP-9 levels in brain tissue homogenate. Furthermore, compared with I/R group, BTH(10mg/kg or 20mg/kg) could ameliorate the structure of cerebral cortex and CA region characterized by largely survived neurons and dense neuropil.

Conclusion: The results suggest that BTH could improve neurological injury, attenuate blood brain barrier disruption and have the ameliorative effect on cerebral I/R injury in rats.

P05

Synthesis of novel chalcone derivatives containing thiazole and sulfydryl heterocyclic

Lei Liu¹, Sai Yang Zhang¹, Xiao-Long Shi¹, Yi Hou¹, Jiang-Hao Wang¹, Ting-yu Li¹, Hong-min Liu^{1*}, Yan-Bing Zhang^{1*}

School of Pharmaceutical Science, Zhengzhou University, Zhengzhou, P. R. China

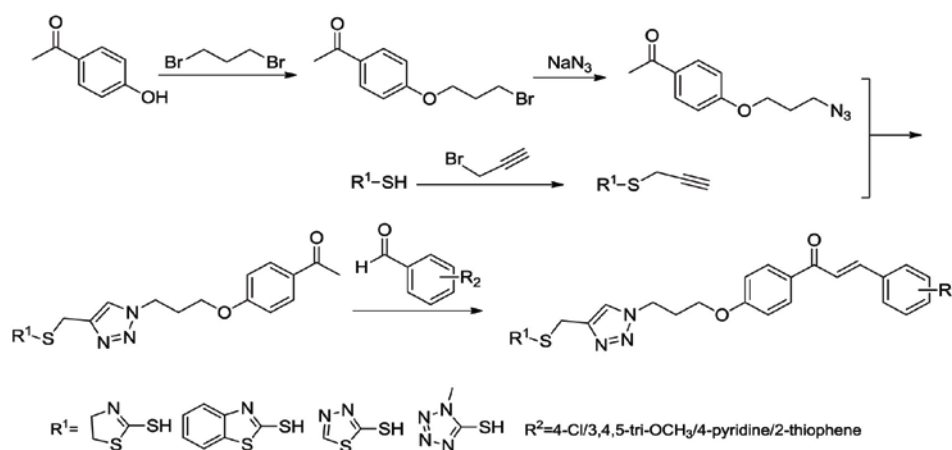
Chalcones, which are abundant in plants as a class of flavonoids, are confirmed to possess a diverse array of pharmacological activities and considered as a promising template for drug design, and especially as antitumor drugs.

Objective: In order to obtain novel chalcone derivatives with high biological activity, a series of new type of chalcone derivatives with thiazole and sulfydryl heterocyclic were synthesized.

Methods: As material, 4'-Hydroxyacetophenone was used to synthesize etherified group and then connected with azide. Containing triazole ring and thiol heterocyclic acetophenone intermediates were synthesized. Then acetophenone intermediates and different substituted benzaldehyde were used to obtain target compounds through Claisen-Schmidt reaction.

Results: 16 new compounds have been synthesized and research on the bioactivities of all the compounds is in progress.

Conclusion: All the compounds were characterized by means of IR, ¹H-NMR and ¹³C-NMR spectral data.



P06

Evaluating the *In Vitro* Inhibition of Human UGT1A1-Mediated Bilirubin Glucuronidation by Danshen (*Salvia Miltiorrhiza*) injection and Compound Danshen injection in Predicting Herb-Induced Jaundice and Hyperbilirubinemia

Guo Ma¹, Ying Zhang¹, Jiayuan Lin¹, Ping Yang¹, Xiaoming Xin², Jing Liu¹, Shanshan Tong¹, Peng Zhang¹, Weimin Cai^{2*} and Ming Hu^{3§}

1. Department of Clinical Pharmacy, School of Pharmacy, Fudan University, Shanghai, P.R. China

2. School of pharmaceutical sciences, Taishan Medical University, Taian, Shandong, P.R. China
3. Department of Pharmacological and Pharmaceutical Sciences, College of Pharmacy, University of Houston, Houston, Texas, United States

Objective: Main purpose of this paper was to disclose the cause and mechanism of Danshen (*Radix Salvia Miltiorrhiza*) injection (DSI) and Compound Danshen injection (CDI)-induced jaundice or hyperbilirubinemia based on evaluating the *in vitro* inhibition of the two herbal injections and their constituents on human UGT1A1-mediated bilirubin glucuronidation.

Methods: In the present study, *in vitro* inhibitory effects of DSI, CDI and their constituents [i.e., salvianolic acid A (SAA), salvianolic acid B (SAB), danshensu (DSS), protocatechuic aldehyde (PA) and rosmarinic acid (RA)] on human UGT1A1-mediated bilirubin glucuronidation was evaluated, respectively. Experiment was performed by incubating bilirubin with expressed human UGT1A1 in the presence of different concentrations of DSI, CDI or their constituents at 37 °C. Apparent kinetic parameters [e.g., reaction velocity (*V*), Michaelis–Menten constant (*K_m*), maximum rate of metabolism (*V_{max}*), concentration at which inhibitor achieves 50% inhibition (*IC*₅₀), and the Lineweaver–Burk plots were used to evaluate the apparent kinetic mechanisms of inhibition of bilirubin glucuronidation.

Results: The results indicated whether it was in the presence of inhibitors or not, average formation rates of three bilirubin glucuronides [i.e., bilirubin monoglucuronides (BMGs, including BMG1 and BMG2 isomers) and diglucuronide (BDG)] demonstrated a significant difference ($p < 0.05$), and the rank order was $\text{BMG2} > \text{BMG1} > \text{BDG}$ [$p < 0.05$, TBG (total bilirubin glucuronides) = $\text{BMG1} + \text{BMG2} + \text{BDG}$]. All the inhibition kinetics of BMG1, BMG2 and BDG by DSI, CDI, SAA and SAB obeyed the mixed-type inhibition and demonstrated substrate concentration dependence in the tested concentrations of substrate and inhibitors. Average *IC*₅₀ values of DSI, CDI, SAA and SAB on glucuronidation of bilirubin with respect to different substrate concentrations (i.e., 0.2–5 μM bilirubin) demonstrated a significant difference ($p < 0.05$) and found to be in the range $0.18 \pm 0.02 \mu\text{M} - 0.32 \pm 0.01 \mu\text{M}$, $0.14 \pm 0.04 \mu\text{M} - 0.34 \pm 0.03 \mu\text{M}$, $0.88 \pm 0.03 \mu\text{M} - 1.75 \pm 0.09 \mu\text{M}$ and $5.87 \pm 0.42 \mu\text{M} - 17.39 \pm 1.29 \mu\text{M}$ for TBG, respectively. DSS, PA and RA exhibited very poor inhibition on bilirubin glucuronidation, and their average *IC*₅₀ values were $340.20 \pm 6.13 \mu\text{M}$, $738.01 \pm 20.75 \mu\text{M}$, $149.53 \pm 2.54 \mu\text{M}$ for TBG at 0.75 μM bilirubin, respectively.

Conclusion: In conclusion, DSI, CDI and their phenolic acid constituents SAA and SAB were the inhibitors of UGT1A1, and displayed potent inhibition on human UGT1A1-mediated bilirubin glucuronidation via a mixed-type inhibitory mechanism. Inhibition of bilirubin glucuronidation by phenolic acids (e.g., SAA and SAB) was one of important pathogeny and pathogenesis of herbs (e.g., DSI and CDI)-induced jaundice or hyperbilirubinemia.

P07

Hydrogen sulfide mitigate infarct of myocardial infarction via governing migration and infiltration of macrophages

Lei Miao¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy and Institute of Biomedical Sciences, Fudan University, Shanghai, P.R. China

Background: Myocardial infarction (MI), associated with the inflammatory response, has long been postulated to be key determinant of rapid necrosis of cardiac myocytes. Activated macrophages infiltrating into the necrotic myocardium, as a part of the robust inflammatory response, is prerequisite for healing process and scar formation during the post-infarction remodeling. RAW264.7 express cystathionine gamma-lyase (CSE) and produce significant amount of H₂S. Although Hydrogen sulfide (H₂S) have been recognized to have potent cyto-protective effects and play an important role in the regulation of cardiovascular functions, less is known about the contribution of the roles and mechanisms of NaHS on macrophage during infarct repair. Thus, the present study was to evaluate the roles of hydrogen sulfide on macrophage migration and polarization using C57 Wild-type and CSE knockout (KO) mice.

Methods: WT mice and CSE KO mice were subjected to myocardial ischemia. NaHS (2 mg/kg, 4 mg/kg) were administered, respectively. Cardiac function were assessed by echocardiography imaging, macrophage recruitment and activation was evaluated by immunohistochemistry and qRT-PCR in WT mice, as well as CSE KO mice at 3, 5, 8 days post-MI. Additionally, macrophage phenotype was analyzed by flow cytometry and the mRNA level of related cytokines in CD11b positive macrophages. Moreover, the migration mechanism of RAW264.7 and peritoneal macrophages was also determined.

Results: The cardiac function were improved in both WT mice and CSE KO mice, pretreated with different concentrations of NaHS (Exogenous sources of H₂S). Meanwhile, the cardiac tissues that treated with NaHS exerted an increased immunohistochemical staining for macrophage marker (Galectin-3). Enhanced M2 macrophage polarization was observed in the infarcted heart treatment with NaHS following MI, along with increased production of M2 signature cytokines, such as interleukin-10 and CD163. In vitro, NaHS accelerated the migration of macrophage cell line RAW264.7. While, the inhibitors (refer to Pyk2, FAK, Src, and Rac1), not only significantly restored the migratory ability in response to the NaHS, but also blocked the activation of phospho-Src, -Pyk2, -FAK397, and -FAK925. Furthermore, the internalization of integrin β 1 on the macrophage surface was induced by NaHS. Whereas, integrin β 1 silencing inhibited the macrophage migration and the Src signaling activation that responsible for NaHS.

Conclusion: These results suggest that H₂S may have the potential as an anti-infarct of MI by governing M2 polarization and migration of macrophages, and the migration regulation was via the integrin β 1-Src-FAK/Pyk2-Rac1 pathway.

P08

Synthesis and antitumor activities of petasin derivatives

Xiaolong Shi¹, Saiyang Zhang¹, Yi Hou¹, Lei Liu¹, Janghao Wang¹, Tingyu Li¹, Hongmin Liu^{1*}, Yanbing Zhang^{1*}

1. School of Pharmaceutical Science, Zhengzhou University, Zhengzhou, P. R. China

Ligularia fischeri, one of Asteraceae Senecioneae Ligularia perennial herb, contains a larger amount of terpenes in the root of the plant, airyalkylphenol type sesquiterpenoids-petasin is one of the main natural products which is large abundant in the *Ligularia fischeri*. From the vitro pharmacological experiment result, we can see that petasin has a good anti-tumor activity for human neuroblastoma,

the antitumor activity IC_{50} can reach $0.8\mu\text{mol}\cdot\text{L}^{-1}$.

Objective: Due to the petasin's excellent antitumor activity, The preliminary study is the petasin derivatives' synthesis and the antitumor activity determination in vitro.

Methods: Petasin which was extracted from *Ligularia fischeri*, via the hydrolysis reaction to get isopetasinol. Using isopetasinol as the starting materials, we designed and synthesized a series of isopetasin derivatives.

Results: The petasin derivatives were measured their anti-tumor activity in vitro for SKN-SH, MGC803, HepG2 cell lines by MTT method, we discovered that the petasin derivatives showed more excellent anti-tumor activity in vitro.

Conclusion: The research focus on the Synthesis and antitumor activities of petasin. We will analyze their structure-activity relationships, learn the necessary structure of petasin and design better effective chemicals based on the analysis of the structure-activity relationships. With the complement of the present experiment, we will understand more about petasin and develop new medicine for anti-neuroblastom.

P09

Improving photodynamic therapeutic and anti-inflammatory effects of curcumin by solid lipid nanoparticles

Jiao Wang¹, Shan Jiang¹, Rongrong Zhu¹, Shilong Wang^{1*}

1. Research Center for Translational Medicine at East Hospital, School of Life Sciences and Technology, Tongji University, P.R. China

Objective: Curcumin (Cur), a natural component of the rhizome of turmeric, is known to exhibit multiple pharmacological actions. However, its poor solubility in water limits its clinical utilization. We have encapsulated Cur into solid lipid nanoparticles (SLNs) to conquer its shortages and improve its photodynamic therapeutic and anti-inflammatory effects.

Methods: The Cur-SLNs were prepared using emulsification and low-temperature solidification methods. Its dispersity in PBS (0.01 M, pH 7.4) was performed. In terms of its photodynamic therapeutic effects, the cytotoxic and apoptotic effects on A549 cells under the visible light of 400–700 nm were determined with MTT assay and annexin V-FITC/PI staining. Meanwhile, the production of reactive oxygen species (ROS) in treated cells was detected by the ROS Assay Kit. Besides, the cellular uptake was studied using confocal microscopy. To evaluate its anti-inflammatory effects, we founded a sepsis model through intraperitoneal administration of LPS (3 mg/kg) in cHS4I-hIL-1 β P-Luc transgenic mice firstly. Then the luciferase and inflammatory cytokines expression in different treating groups were measured using living imaging technology and ELISA assay.

Results: Cur-SLNs were well dispersed in aqueous medium. It presented an increased intracellular uptake over time in A549 cells. The Cur-SLNs, even treated at a very low concentration ($0.625\mu\text{g/ml}$) under the visible light of 400–700 nm, induced a significantly higher apoptosis ratio in cancer cells. Cur-SLNs can produce more ROS, which played as a strong oxidant to kill cancer cells. We found that Cur-SLNs were more active than free Cur in reducing the luciferase expression in the LPS-induced sepsis model. Cur-SLNs were more effective than free Cur at reducing the expression levels of several pro-inflammatory mediators in serum, including inflammatory cytokines (TNF- α ,

IL-6, and IL-1 β).

Conclusion: SLNs could be used as a potential carrier to improve the therapeutic effects of free Cur.

P10

Novel apoptotic protease activating factor-1 (apaf-1) inhibitor, WY-488, is potent against apoptosis induced by myocardial hypoxia

Ying Wang¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy and Institute of Biomedical Sciences, Fudan University, Shanghai, P.R. China

Cardiac apoptosis plays an important role in the pathology of myocardial ischemia. Apaf-1 is a central component of the intrinsic pathway of apoptosis. In present work, we synthesized, evaluated and indentified WY-488 as a novel kind of apaf-1 inhibitor, H9C2 rat ventricular cells were induced by hypoxia, as a model of ischemia to assess the protection effect of WY-488. Treatment with WY-488 significantly increased cell viability, accompanied by a consistent decline in lactate dehydrogenase (LDH) and creatine kinase leakage (CK). The apoptotic fraction of H9C2 measured through both annexin V-fluorescein and Confocal scanning microscope was increased by hypoxia but reduced by WY-488, which implies WY-488 may elicits its cardioprotective function by potent anti-apoptotic effect. Apaf-1, one of the most critical protein in cell apoptosis was indentified the potential target for WY-488 using pharmacophore mapping approach. Protein-ligand interactions are studied by docking WY-488 transported by apaf-1. Structural analyses revealed the close contact of WY-488 with active-site amino acids (Ser23, Glu40, Asp27) in caspase recruitment domain(CARD) which directly bind and activate procaspase-9 then induce apoptosis. The carboxyl group of WY-488 has a strong interaction with Arg44, making the contact between apaf-1 and the small molecular much more stable. Results of western blot indicated that WY-488 reduced the activation of procaspase-9 while having no impact on the expression of apaf-1 itself. These results suggest that WY-488 maybe a potential competitive inhibitor of apaf-1.

P11

Pharmacological researches of SPRC-TYR as the structure optimization subject of SPRC

Weijun Wu¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R. China

SPRC, the chemical name is S-propargyl-cysteine. As the one of the H₂S donors, SPRC can be catalyzed by CSE, CBS and 3-MST. The pharmacological researches have been already discussed thoroughly by our group, which are inspiring, not only in the cardiovascular field, but also in the area the brain. The effect of treating the HF and brain diseases such as AD and stroke have been proved efficient. However, the side effect of the weight loss for long-term use and the congenital defects of large dose and big polarity could not be ignored. Based on the discussions above, we project an analogue of SPRC named as SPRC-TYR combining with the existing research results with the hope of penetration of the blood brain barrier through the large amino acid transporter(LAT1) and

lowering the amount of effective dose. As the name implied, SPRC is linked with the natural amino acid tyrosine by ester bond. Owing to the tyrosine is the substrate of LAT1, we predict that SPRC-TYR could penetrate the BBB and release more small gas molecular H₂S. To prove the prediction true, we firstly tested the SPRC and SPRC-TYR on PC-12 cell line. On the same level, we find that SPRC-TYR can release much more H₂S. To mimic the same symptoms of stroke in vitro, oxygen glucose deprivation was executed on PC-12. The result shows that the SPRC-TYR group elevates the cell viability compared with SPRC group. The phenomenon may be explained by SPRC-TYR increasing the concentration of SOD in cells and reducing the level of ROS and MDA, elevating the ratio of Bcl-2/Bax to alleviate the damage of inflammatory and inhibiting the cell apoptosis process, which should be proved in the next stage. Meanwhile if the SPRC-TYR could breakthrough the BBB also need to be verified.

P12

Total synthesis of (±)-*Cephalotaxine* and related ester side chain

Hua Yang

School of Pharmaceutical Science, Zhengzhou University, Zhengzhou, P.R. China

Several alkaloids isolated from the genus *Cephalotaxus* have been shown to exhibit interesting antileukemic activities. Among them harringtonine, deoxyharringtonine, homodeoxyharringtonine, neoharringtonine, anhydroharringtonine, and homoharringtonine have been studied extensively. Homoharringtonine is the most potent in this family of antitumor alkaloids. Indeed, it is currently in phase II–III of clinical trials in the United States for the treatment of chronic myeloid leukaemia. The limited availability of these alkaloids has led to the development of a number of strategies for their synthesis.

Objective: Total synthesis of (±)-*Cephalotaxine* and its related ester side chain including harringtonine, homoharringtonine, bis-homoharringtonine, deoxyharringtonine and homoharringtonine

Methods: Using the Stevens rearrangement-acid lactonization sequence as a key transformation from readily available (3,4-dimethoxyphenyl)acetic acid, methyl proline, and allyl bromide to synthesis Cephalotaxine; Construction of chiral tertiary alcohol stereocenters via the [2,3]-Wittig rearrangement to synthesis the Side-Chain Acids of Homoharringtonine and Harringtonine.

Results: The highly efficient formal synthesis of (±)-Cephalotaxine using the Stevens rearrangement-acid lactonization sequence as a key transformation has been finished through eight stage operations in total 51% yields. In addition, we also realized the enantioselective synthesis of the side chain acid of homoharringtonine using the [2, 3] Wittig rearrangement as a key transformation.

Conclusion: The methodology developed for the construction of chiral tertiary alcohol structural unit in the side chain acid of homoharringtonine is suitable to other complex bioactive molecules. Its application to the synthesis of forstrocin, Erythronolide A and Integerrimine are underway and results will be reported in due course. Currently, one process study for large scale industrial production on the synthesis of homoharringtonine is underway and we wish related work will solve supply problem of homoharringtonine.

P13

The protective role of Cordyceps cicadae Shing in ConA induced liver injury

Shaowei Yang¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R. China

Objective: To investigate the effect of 4 fractions (fraction 1-4) of water extracts from Cordyceps cicadae Shing in ConA induced liver injury.

Methods: Totally 70 C57BL/6 mice were randomly divided into 7 groups, i.e., the blank control group, the model group, the bicyclol group (250mg/kg), the fraction 1 group (100mg/kg), the fraction 2 (100mg/kg) group, the fraction 3 group (100mg/kg) and the fraction 4 (100mg/kg) group, 10 mice in each group. The treatment groups and the bicyclol group were pre-treated via gavage daily for 7 days. The control group was given the equal volume of saline. One hour after the last gastrogavage, acute liver injury was induced by ConA(20mg/kg) via tail vein. The control group was injected with saline via tail vein. Mice were sacrificed 12h after modeling. Blood and liver were collected to detect lab indicators such as serum ALT, AST, TNF- α , IL-1 β T-AOC and tissue SOD, MDA. HE staining was performed to determine the pathological condition of the liver.

Results: Compared to the model group, ALT and AST levels were lower in all the treatment group ($P<0.05$); T-AOC was remarkably increased in the fraction 1 group, fraction 2 group ($P<0.01$), fraction 3 group and fraction 4 group ($P<0.05$); levels of TNF- α and IL-1 β were significantly decreased in the fraction 1 group, fraction 2 group ($P<0.01$), fraction 3 group and fraction 4 group ($P<0.05$); level of MDA in liver was reduced in all the treatment groups ($P<0.05$); level of SOD in liver was significantly increased in the fraction 1 group, fraction 2 group ($P<0.01$), fraction 3 group and fraction 4 group ($P<0.05$). HE staining shows that the pathological condition was alleviated in all the treatment groups.

Conclusion: The fraction 1 group, fraction 2 group, fraction 3 group and fraction 4 group show protective effect in the ConA induced acute liver injury, which might be achieved via the anti-inflammatory and anti-oxidant mechanism.

P14

Leonurine: the gospel of stroke

Qiuyan Zhang¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R. China

Stroke is the second most common cause of death, and is the primary cause of serious long-term disability, and also produces an immense burden on the health care and national economies. Ischemic strokes occupy about 80% of all stroke. Using a rat model of transient middle cerebral ischemia occlusion (tMCAO) for 90 minutes, we have investigated the effect of treatment with leonurine at 0.5, 2 and 6 hours after tMCAO. At the same time, we used edaravone as the positive control. We measured the infarct size, water content and neurological deficits. We found that leonurine can significantly reduce the infarct volume and neurological deficits. The same phenomenon was found in water content experiment. We also found that leonurine has the effect on blood-brain barrier (BBB) disruption by measuring the Evans blue leakage. In conclusion, the post-treatment with leonurine can significantly reduce the infarct size, water content and BBB permeability. These findings are not

enough, we will find out the deeper mechanism.

P15

Quercetin3-O-(6''-O- α -L-rhamnopyransoyl)- β -D-glucopyranoside-7-O- β -D-glucopyrano-side (QG30) isolated from *Zanthoxylum bungeanum* protects HepG2 cells from cholesterol overloading induced by sterol through promoting the expression of SREBP-2

Liangjie Zhong¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R. China

Background: In our previous studies, we found n-butanol fraction isolated from *Zanthoxylum bungeanum* could regulate the lipid metabolism. Then we wanted to find out which substance has the lipid-lowing effect. Finally, we used the biological activity guided purification to isolate a flavonoid glycoside from *Zanthoxylum bungeanum*, namely: Quercetin3-O-(6''-O- α -L-rhamnopyransoyl)- β -D-glucopyranoside-7-O- β -D-glucopyranoside (QG30), which was first found from *Zanthoxylum bungeanum*. In this study, we investigated possible cholesterol-lowing effect and mechanisms of QG30 in HepG2 cells.

Methods: HepG2 cells, pretreated with different concentrations of QG30 or quercetin, were incubated with 13 μ M 25-hydroxycholesterol and 130 μ M cholesterol to make intracellular cholesterol over-loading. The intracellular total cholesterol and free cholesterol were detected to evaluate the lipid-lowering effects of QG30. The expression of sterol regulatory element binding protein-2 (SREBP-2), low density lipoprotein receptor (LDLR), Proprotein Convertase Subtilisin Kexin 9 (PCSK9), the ATP binding-cassette (ABC) half-transporters ABCG5 and ABCG8 were evaluated by Western blot assay. The uptake of LDL labelled with 1,1'-Diocetadecyl-3,3,3,3'-tetramethylindocarbocyanineperchlorate (Dil-LDL) in HepG2 cells was assessed by flow cytometry to determine the activity of LDLR. Quantitative RT-PCR was performed to detect the gene expression of SREBP-2. To confirm the role of SREBP-2 on the cholesterol-lowing effect of QG30, 4-(2-Aminoethyl) benzenesulfonyl fluoride hydrochloride (AEBSF) was used to inhibit the processing of SREBP-2. Then the intracellular total cholesterol, the expression of SREBP-2 and LDLR were determined.

Results: Administration of QG30 dose-dependently suppressed the cholesterol overloading, and extension of the QG30 exposure time promoted this effect. Compared with quercetin, QG30 showed the stronger inhibiting effect on cholesterol accumulation. Pretreatment of QG30 dose-dependently increased the expression of SREBP-2, LDLR, ABCG5 and ABCG8, nevertheless decreased the expression of PCSK9. QG30 promoted the gene expression of SREBP-2, which was agreed with the change in SREBP-2 protein. The effect of LDL-c on reducing LDL receptor activity was reversed by QG30. After inhibiting SREBP-2 mature by AEBSF, the cholesterol-lowing and the promoting effects of QG30 on expression of SREBP-2 and LDLR were abolished.

Conclusion: We demonstrated that QG30 inhibited the cholesterol accumulation in HepG2 cells, promoted the cholesterol efflux through ABCG5/8 and regulated the cholesterol metabolism via SREBP-2/LDLR pathway.

P16

S-Propargyl-cysteine (SPRC) diminishes mitochondrial dysfunctions in heart failure

Dan Wu¹, Yizhun Zhu¹

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R. China

Aims: One of the important factors of heart failure (HF) is mitochondrial dysfunction. Growing evidences indicate that damage of mitochondria causes mitochondrial pathway apoptosis and the loss of cardiomyocytes, which are significant in the process of HF. At the meantime, the injury of mitochondrial respiratory chain results in a decrease of cellular ATP level in HF. Hydrogen sulfide (H₂S) is the third gasotransmitter, which presents a wide range of cell function in the body. S-Propargyl-cysteine (SPRC) is water-soluble H₂S endogenous donor which has protective effect on acute myocardial infarction in rats. The aim of this study was to investigate how SPRC diminishes mitochondrial dysfunctions in HF.

Methods: HF in C57BL/6 mice (male, 6-8 weeks old) was induced by Isoprenaline (7.5 mg/kg), which was administered for 3 weeks, once a day by subcutaneous injection. Hydrogen peroxide (H₂O₂) 200μM was used to induce Myocardial oxidative damage in H9c2 cells. H&E stain and masson trichrome stain were used to determine the histopathological change. Cell viability assay was used to determine the protective effect of SPRC in vitro. Caspase activity assay was used to determine the apoptosis level. Lipid peroxidation, antioxidant enzymes, ATP level, mitochondrial ΔΨm measurement and mitochondrial respiratory chain complexes (I-IV) measurement were used to determine the mitochondrial function. Western blot was used to determine the expression levels of Bcl-2, Bax, pro-Caspase 3/9 and PKCε.

Results: H&E stain and masson trichrome stain showed a significant loss of myocardium and increased fibrosis in isoproterenol group compared with the vehicle group. After SPRC (10, 25 mg/kg) treatment, the level of myocardial fibrosis was significantly reduced. SPRC (10 - 50 μM) were found to increase cell viability significantly, which reduced by H₂O₂ in H9c2 cells. SPRC (10, 25 mg/kg) significantly decreased the level of apoptosis in mice compared to the model group. The same result was observed in SPRC (10 - 50 μM) treated H9c2 cells. The level of mitochondrial glutathione (GSH) and superoxide dismutase (SOD) was recovered by SPRC treatment and the content of mitochondrial lipid peroxidation (LPO) was decreased compared with H₂O₂ group. The content of ATP and the activity of mitochondrial respiratory chain complexes were significantly increased in SPRC treated grouped which were severely reduced in H₂O₂ group. The mitochondrial membrane potential was significantly reduced and the calcium swelling was markedly decreased in the SPRC groups. SPRC induced translocation of PKCε to mitochondria, which reduced by H₂O₂.

Conclusion: This study suggests the therapeutic ability of SPRC in HF through the protective effect of mitochondrial dysfunction.

P17

Evaluating the potential for drug-medicinal plant interactions in Jamaica

David Picking and Rupika Delgoda

Natural Products Institute, University of the West Indies, Mona, Kingston 7, Jamaica

Three surveys conducted by our laboratory have concluded the existence of a high prevalence of concomitant use of medicinal plants with prescription medicines in Jamaica. 80% of those on prescription medicines visiting pharmacies were also self-medicating with medicinal plants, with only 18% of physicians aware of such practices. Armed with the knowledge of medicinal plants most commonly used in combination therapy, we investigated the inhibitory impact of 13 such plant infusions/decoctions on the activity of human cytochrome P450 (CYP) enzymes (CYPs 2D6, 3A4, 1A1, 1A2, 2C19/1B1) using an *in-vitro* fluorimetric assay. Following control experiments for intrinsic fluorescence and interference with assays, 4 extracts were found to display potent ($IC_{50} \leq 9.9 \mu\text{g/ml}$) inhibition of at least one of the enzymes examined, as exemplified by the selective inhibition by *Cinnamodendron corticosum* against CYP1A2 (IC_{50} of $7.8 \pm 1.8 \mu\text{g/ml}$). 49 out of 65 interactions examined demonstrated moderate to weak inhibition ($\geq 10 \mu\text{g/ml}$) e.g., *Petiveria alliacea* and *Croton linearis*, moderately or weakly inhibited all enzymes screened, indicating an unlikely clinical manifestation for these extracts. Active phytochemical analysis from two select plants identify that quassinoids found in *Picrasma excelsa* are responsible for the mixed type competitive inhibition observed (quassin $K_i = 10.8 \mu\text{M}$), while nine key phytochemicals, including lignans analysed from *Hyptis verticillata* did not contribute to the potency of the extract, indicating likely synergistic effects. Such *in-vitro* evaluations have been key in identifying extracts needing clinical examinations.

P18

Cardioprotective effects from Leonurine on ischemia/reperfusion rats and molecular mechanisms

Fenfen Ma, Yi-zhun Zhu¹

1. Department of Pharmacology, School of Pharmacy, Fudan University, Shanghai, P.R. China

Leonurine, a Chinese medicinal herb, is reported to be myocardial protection. However, its role in cardioprotection remains largely unknown. Our recent study aimed to investigate the effects of preconditioning on myocardial ischemia/ reperfusion (MI/R) injury and to test the possible mechanisms. Rats were treated with leonurine or saline once time. Afterward, all the animals were subjected to 30min of myocardial ischemia followed by 2h of reperfusion. Leonurine preconditioning significantly improved cardiac function following MI/R. Meanwhile, leonurine reduced infarct size, plasma creatine kinase(CK), lactate dehydrogenase (LDH) and malondialdehyde(MDA) activities and myocardial apoptosis at the end of reperfusion in rat hearts. Moreover, leonurine preconditioning significantly inhibited superoxide generation, and increased phosphorylated Akt(p-Akt) and phosphorylated eNOS(p-eNOS) expression, and enhanced superoxide dismutase activity in I/R hearts. In cultured cardiomyocytes, leonurine showed concentration - dependent inhibitory effects on cardiomyocyte apoptosis induced by hypoxia/ re-oxygenation. These effects were associated with an increase in phosphorylated Akt(p-Akt) and phosphorylated eNOS(p-eNOS) , ratio of Bcl-2/Bax and a decrease in caspase-3 activation. In conclusion, leonurine prevents cardiomyocyte apoptosis by elevating phosphorylation of eNOS and phosphorylation of AKt. These findings provide potential benefits of ensuring *leonurine* preconditioning, as a strategy to prevent ischemic heart disease. In all, leonurine offers a potential therapeutic approach to prevent the progression of myocardial ischemia/ reperfusion.

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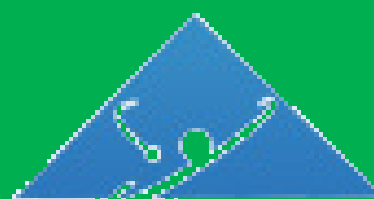
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