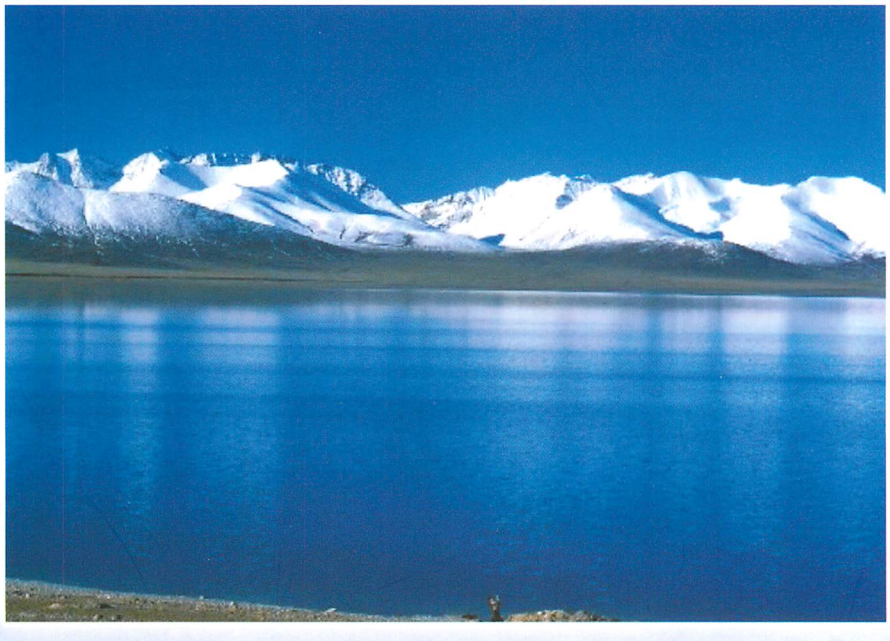
The Sixth Internatinal Symposium of Molecular Pathology

2010.8.20-8.21，Qinghai，the People's Republic of China



Hosted by

China Medical University

Kagoshima University

Qinghai University

The 5th International Symposium of Molecular Pathology

The Sixth Internatinal Symposium of Molecular Pathology
2010.8.20–8.21, Qinghai University, China

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General Information of the Sixth International Symposium of Molecular Pathology

Date: August 20-21, 2010

Venue: The symposium will be held in the meeting room of Medical College of Qinghai University

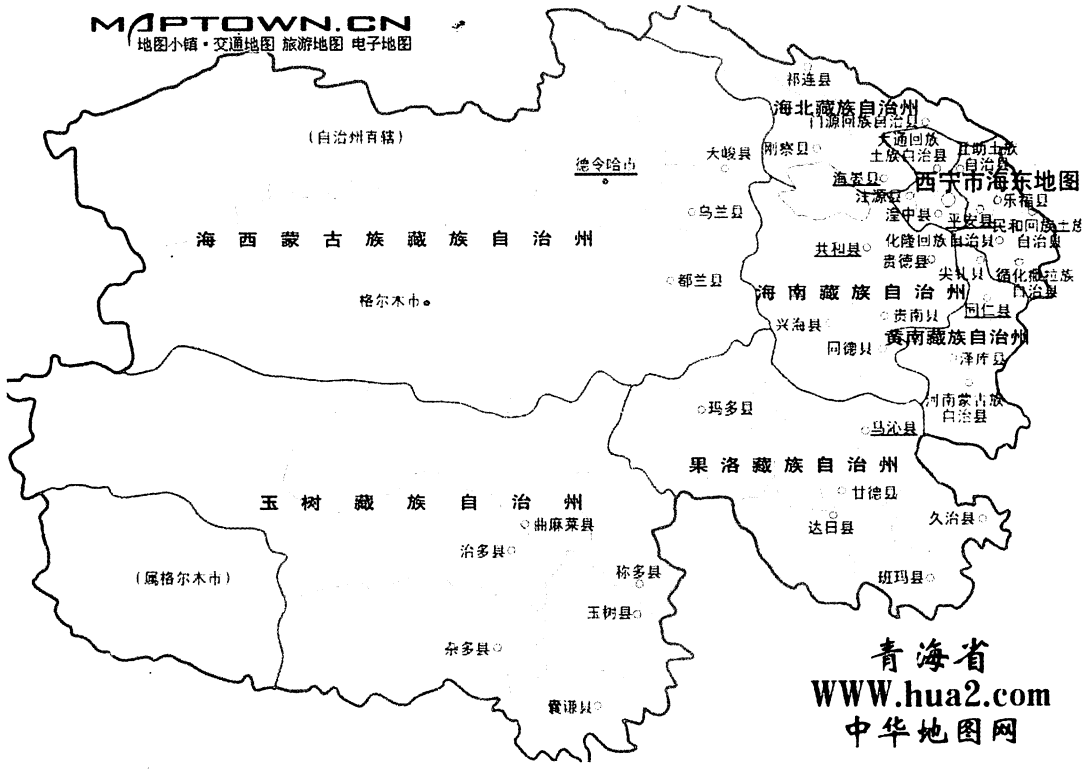
Language: The official language of the symposium is English

Wear: Informal

Registration: Most of the participants are pre-registered.People who want to attend the symposium must contact the Chinese side President of the symposium.

Special lecture: The presenter must finish the oral presentation within 20 min and leave at least 5 min for discussion.

Lecture: The presenter must finish the oral presentation within 7 min and leave at least 2 min for discussion.



Greeting

Ladies and Gentlemen,

On behalf of the Committee of the Sixth International Symposium of Molecular Pathology, I would like to use this opportunity to express my warm welcome all the foreign and domestic experts and scholars to this symposium. Also, I would like to express my sincere thanks to Oinhui University, Kagoshima University and Kanazawa University for their generous support to this symposium held in this beautiful city.

10 years ago, a group of scientists from China and Japan initiated the First International Symposium of Molecular Pathology. Since then, five symposiums have been organized successfully. Using this communication platform, we have established a long-term relationship not only in academic research and collaboration, but also in personal friendship. In addition, our joint-researches have covered a wide range of disciplines in molecular pathology, biochemistry, pharmacology and toxicology, and so on. It has been a wonderful decade. Today, we are meeting in this beautiful city for the Sixth International Symposium of Molecular Pathology, and we have noticed that more scientists around the world joint us and made the symposium a high-level academic discussion and personal communication. We wish all the participants will distribute and share their achievement and success during this symposium.

This symposium has also been sponsored by the College of Japanese Pathologists and China Medical Association. We are grateful to them for their kind supports.

On this memorable occasion, may I wish the Sixth International Symposium of Molecular Pathology in Qinghai a great success. To every participant to this symposium, I wish you happiness and good health.

Enjoy the symposium and enjoy your stay in Qinghai.

Thank you very much.

Enhua Wang
Dean, Basic Medical College, China Medical University
Chairman, Department of Pathology

Greeting

We celebrate the 6th meeting of our international symposium of molecular pathology (ISMP).

ISMP was organized by Keiichi Watanabe, Xinshan Jia and Eiichi Sato in order to interchange knowledge in the field of pathology between Japan and China and among the world in the future. ISMP had 4 meetings in China (Dunhuang (1998, First meeting)、Chengdu (2001, Second meeting)、Kunming (2004, Third meeting) and Urumqi (2006, Fourth meeting) and the last meeting in Japan (Kagoshima, 2007, Fifith meeting) when Chinese pathologists can travel in China and abroad as Chinese power became bigger even in the field of pathology. At the Kagoshima meeting, 27 Chinese and 68 Japanese participants attended.

From the Japanese standpoint, ISMP depends on friendship among Japanese and Chinese pathologists, on the personal friendship of Jia XS with people in Kagoshima, and on Japanese curiosity about old China, especially the Silk Road. From the Chinese standpoint, ISMP was the place where they can report their studies. In these ISMP meetings several research and educational trials were made, such as evaluation of pathological sense in Chinese young researchers and asking for the aid from Japan Bank for International Cooperation (JBIC). Financial and academic support from China Medical University and Kagoshima University is sending many active participants to ISMP meetings when many Japanese and Chinese participants attended ISMP meeting depending on so-called Scientific Research Founds.

Our friend, Katsuyuki Aozasa, President of the Japanese Society of Pathology, acts to establish official exchange program in the field of Pathology between Japan and China. The official exchange program would include several aspects and it is a problem where our ISMP should be placed. Many Japanese pathologists point out that Chinese Pathology nowadays succeed in publication of many articles in famous Journals. Probably, the official invite speech in both Japan and China annual meetings of Society of Pathology may be only for the representatives. Here, we can expect that ISMP serves the space and time for many Japanese and Chinese pathologists to exchange their knowledge and experience under the official exchange programs of both societies of Pathology.

2010. 8.21

President (Japanese side) of 6th ISMP
Dr. Kazuhisa Hasui

Greeting

Distinguished Representatives and Guests,
Ladies and Gentlemen:

We are getting together now in Xining of Qinghai, well known as "the Summer City in China" for the Sixth International Molecular Pathology Seminar organized sponsored by China Medicine University, Japan Kagoshima University and Qinghai University Medical College. The conference is attended by experts and scholars from Japan, the USA and at home. I, acting as the vice conference chairman, would like to extend sincere welcome to all domestic and foreign experts or scholars to attend the international conference.

As you are aware, the molecular medical research is designed, by applying theories and methodologies of many basic disciplines such as molecular biology, cellular biology, pathology, genetics and bioinformatics, to reveal a causal molecular mechanism of cancer and infectious diseases which constitute major medical issues facing humanity

Molecular Pathology is a molecular science of pathology which applies molecular biotechnology to examine the happening and developing process of disease in terms of protein and nucleic acid level.

Disciplinary developments are always interrelated. The development of molecular pathology is invariably intertwined with that of molecular biology. With no exaggeration, all theories and techniques of molecular biology are applied and developed in the pathological research.

Examination of causes of some diseases in terms of genetics has paved a promising prospect for pathology. Detection of gene mutation, continuous analysis of gene, test of mRNA, hybridization of nucleic acids and PCR and other techniques are all used for detection, diagnosis of genetic, infectious, viral, parasitic and tumorous diseases, and investigation of disease-causing mechanisms and drug screening.

Furthermore, by chip technology, it is possible to develop immunologic chip and protein chip which are used for detecting on a large scale multifarious genetic expressions, correlations between proteins, protein and DNA, polymorphism and mutations of individual and group genes and for determining genotype inside human body. Genes are modified and transferred to certain cellular to achieve medical treatment.

In addition to the above research fields, molecular developmental biology, aging molecular biology, molecular neurobiology and tumor molecular biology, etc, are all fields closely related to molecular pathology in medical and life science researches.

Ladies and Gentlemen, all friends, I sincerely wish you have a big academic harvest at this conference and a great feel of the beauty of Qinghai in your spare time. Xining, the capital of Qinghai, which has a long history of around 2,100 years old, is the east gate of the Qinghai-Tibet

Plateau. Situated in the east of Qinghai, it is characterized by being surrounded by hills, three rivers converging, an altitude of 2,275 meters at downtown, and semi-arid continental highland climate. Located at the tertiary global level, Qinghai is the birthplace of the Yantze River, the Yellow River and Lancang River, the fatherland of thousands of mountains, and it is the most amazing rainbow after heavy rain, and it boasts the unique highland ethnic cultures.Within the radius of 200 kilometers of Xining, it enjoys the best of Qinghai tourism resource, namely, the Taer Lamasery, the Sun-moon Mountain, the Chahan River Forest Park, the Qongjia Forest Park, the Qinghai Lake with the Birds Island, the Base of Two Bombs, the Huzhu County Tu Ethnic Cultures, the Liuwan Porcelain Center, the Menda Mountain Lake, the Sala Ethnic Cultures, the Kanbula Landscapes, and the Haibai Jingyingtan Grassland, which are referred to as three golden sightseeing in Qinghai tourism.

We will have a good job at the conference organization and reception, showing our warm and sincere hospitality of highlanders to all of you here.

In conclusion, I wish all leaders, experts and scholars, and friends a good health! I wish the international conference a great successs! and once again I express my heartfelt thanks to all of you from far away to present at this conference in Qinghai.

Everyone, I wish you a great success for this conference! A pleasant stay in Qinghai!

Thank you!

Gerili,

The vice president of Qinghai University,

The Director of Altitude Medicine Research Center,

August, 2010

List of Chinese participants

Prof.	Enhua Wang（王恩华）	China Medical University
Prof.	Xianghong Yang（杨向红）	China Medical University
Prof.	Yifu Guan(关一夫）	China Medical University
Prof.	Yujie Zhao(赵雨杰）	China Medical University
Prof.	Qinchang Li（李庆昌）	China Medical University
Prof.	Jianhua Li（李建华）	China Medical University
Prof.	Ling Zhang(张玲）	Shenyang Medical College
Prof.	Yuchen Han(韩昱晨）	China Medical University
Prof.	Zeshi Cui(崔泽实）	China Medical University
Prof.	Yan Wang（王妍）	China Medical University
Prof.	Lan Luan（栾岚）	Shenyang No.8 Hospital
Prof.	Changqing Fang(方长青）	China Medical University
Prof.	Liying Hao(郝丽英）	China Medical University
Prof.	Haiping Zhang(张海萍）	Xiameng No.1 Hospital
Prof.	Zheng Zhu(朱正）	Longgang Center Hospital
Prof.	Rili Ge(格日力）	Qinhai Univ.China
Prof.	Yue Wu(吴岳）	Qinhai Univ.China
Prof.	Weng Yin(尹文）	The Forth Military Med.Univ
Prof.	Xiaofeng Hang(黄晓峰）	The Forth Military Med.Univ
Prof.	Yuhong Jing(景玉宏）	Lanzhou University
Prof.	Xunmi Zhao(赵勋蘼）	Guangxi Med. Univ.
Dr.	Juanzhi Chen(陈娟芝）	The South Med. Univ.
Prof.	Josef Prchal	University of Utah，USA
Ph.D student	Tatum Simonson	University of Utah，USA
Prof.	Luming Zhao	University of Utah，USA

List of Japanese participants

Prof.	Kazuhisa Hasui(莲井和久）	Kagoshima Univ.Japan
Prof.	Takami Matsuyama(松山隆美）	Kagoshima University
Prof.	Okumura Teruhisa（奥村晃久）	
Prof.	Eiichi Sato(佐藤荣一）	Kagoshima University
	Sato Yuriko（佐藤百合子）	
Prof.	Takahashi Kiyoshi(高橋潔）	Kumamoto University
	Taylor Reina	
Prof.	Ueda Yoshimichi(上田善道）	Kanazawa Med. University
	Ueda Satsuki(上田さつき）	
Prof.	Miyako Shimasaki(岛崎都）	Kanazawa Medical University

Scientific Program

2010.8.21

9:00 Opening

Greeting — Host: Yue Wu(吴岳)

Chairman： Rili Ge(格日力) — Qinghai Univ.China

Kazuhisa Hasui(莲井和久) — Kagoshima Univ.Japan

Enhua Wang(王恩华) — China Med.Univ.China

Photography of the symposium

10:30 SPECIAL LECTURE 1 — Chairman: Takahashi(高桥洁) Kumamoto University

Takami Matsuyama(松山隆美)

Kagoshima University

The pathological role of folate receptor beta-expressing macrophages in inflammatory diseases and cancers

11:00 SPECIAL LECTURE 2 — Chairman: Takami Matsuyama(松山隆美) Kagoshima University

Luming Zhou

University of Utah, USA

High-resolution DNA melting analysis in clinical research and diagnostics.

11:30 SPECIAL LECTURE 3 — Chairman: Eiichi Sato (佐藤荣一) Kagoshima University

Yifu Guan(关一夫)

China Medical University

Potential Applications of LNA to Gene Expression Regulation

12:00 Lunch Time

14:00 SPECIAL LECTURE 4 — Chairman: Yifu Guan(关一夫) China Medical University

Liying Hao(郝丽英)

China Medical University

The regulations of the CaV1.2 Ca2+ channels by calmodulin and Ca2+ in guinea-pig cardiac myocytes.

14:30 SPECIAL LECTURE 5 Chairman: Yue Wu (吴岳)
Qinghai University

Yoshimichi Ueda(上田善道)
Kanazawa Medical University
Relationship of aquaporin 1, 3 and 5 expression in lung cancer cells to cellular differentiation, invasive growth and metastasis potential.

15:00 LECTURE 6 Chairman: Yuchen Han(韩昱晨)
China Medical University

Kazuhisa Hasui(莲井和久)
Kagoshima University
Immunohistochemical analysis of oxidative modified DNA (8-OHdG) in Chinese nasopharyngeal lymphomas.

15:10 LECTURE 7 Chairman: Qinchang Li (李庆昌)
China Medical University

Haiyan Wang(王海燕)
Qinhai University
Study on Cartilage Induced With Adipo-mesenchymal Stem Cells

15:20 LECTURE 8 Chairman: Kazuhisa Hasui(莲井和久)
Kagoshima University

Yuchen Han(韩煜晨)
China Medical University
EHMT1 interact with MT1h and regulate histone H3K9 methylation to inhibit proliferation and movement.

15:30 LECTURE 9 Chairman: Liying Hao(郝丽英)
China Medical University

Miyako Shimasaki(岛崎都)
Kanazawa Medical University
Involvement of aquaporin 1 and 5 in metastasis potential of human osteosarcoma.

17:00 Dinner Party.

CATALOG

The pathological role of folate receptor beta-expressing macrophages in inflammatory diseases and cancers

Takami Matsuyama, Taku Nagai and Kazuhisa Hasui.
Department of Immunology, Graduate School of Medical and Dental Sciences,
Kagoshima University, Kagoshima City, Japan

The folate receptor beta(FR) is expressed restrictedly on macrophages and functions as the receptor for oxidized folates. It has been rare to observe FR -expressing macrophages in normal human tissues. Previously, we and other groups reported that FR -expressing macrophages were present in rheumatoid arthritis synovium and produced TNF-α . Thus, FR -expressing macrophages were considered to belong to M1 macrophages which promote inflammation. Meanwhile, alternatively activated macrophages which play a role in resolution of inflammation are designated as M2 macrophages. M2 macrophages observed in tumor tissues promote the angiogenesis and suppress T cell functions. The molecular and functional characterization of these polarized macrophages is a current topic of investigation. However, the link between FR -expressing macrophages and other inflammatory diseases or cancers is poorly understood. In this study, we examined the distribution of FR -expressing macrophages in interstitial pneumonia, atherosclerosis and cancers. Furthermore, to explore the pathological roles of FR -expressing macrophages in these diseases, we developed a recombinant immunotoxin (consisting of the Ig heavy chain Fv portion of anti-mouse FRβ mAb with Pseudomonas exotoxin A and the Ig light chain Fv portion of anti-mouse FRβ mAb) to deplete FR -expressing macrophages. The immunotoxin was the IC50 of 10ng-100ng/ml in the cytotoxicity on various FR -expressing cell lines and reduced their TNF-α and NO production. FR -expressing macrophages were observed in lesions from inflammatory diseases and cancers. The immunotoxin depleted FR -expressing macrophages in lesions of rheumatoid arthritis, interstitial pneumonia and glioma models. and reduced the disease activity of rheumatoid arthritis and interstitial pneumonia models, and the tumor growth of the glioma model.

These results suggest that although FR -expressing macrophages are M1 macrophages in inflammatory lesions, they appear to be M2 macrophages in tumor tissues. The targeting of FR expressing macrophages may be promising in the treatment of inflammatory diseases and cancers In addition, the imaging of FR -expressing macrophages in these diseases may be useful as a noninvasive indicator for disease activity of inflammatory diseases and cancers with poor outcome.

Genetic cloning and expression studies of neuroglobin in high altitude hypoxic adaptation species-Plateau pika (Ochotona curzoniae)

BAI Zhenzhong, HAN Shufen, YANG Yingzhong, CAO Yue, MA Lan, LIU Shou, GE Rili(Research Center for High Alt itude Medicine,Qinghai University Medical School,Xining 810001, CHINA)

Abstract: For identification of the neuorglobin genes coding sequences molecular cloning and examination of the tissues expression spectrums and showing the hypoxic adaptations mechanisms in Plateau Pika

Methods: Extracting the total RNA, cloning Ngb coding sequences cDNA with reverse-transcrip-

tion RT-PCR, capturing and confirming the cer tain sequences with DNA sequencing Examination the tissues mRNA expressions spectrums of neuroglobin with In suit hybridization ISH technology In addition, semi-quantitative RT-PCR and western-blot technologies were being used to analysis of the rela-tive neuroglobin mRNA and protein expression amounts in various tissues with Plateau Pika

Results: cloning and sequencing results confirmed it was the neruoglobin gene coding sequence of Plateau Pika with Blast analysis; and the ISH results exhibited the neuroglobin gene expressions spectrums indicate widely distributed with higher amount in brain tissues of Plateau Pika Additionally, neuroglogin mRNA also expressed in the other tissues such as testis and adrenal glands besides brain tissues

Conclusion: we can speculate that Ngb might play an important role in the adaptations of the plateau Pika under the high altitude environment Furthermore, native Tibetan species neuroglobin genes will also present fundamental evidence to investigate high altitude adaptable related genetics study in the futur. □

Key words: Gene cloning; Gene Expression; Neuroglobin; Plateau pika

Potential Applications of LNA to Gene Expression Regulation

Xiaoyang Zhao, Bo Liu, Jin Yan and Yifu Guan

Department of Biochemistry and Molecular Biology, China Medical University, Shenyang, China 110001

Abstract: -D-Locked nucleic acid (LNA) is a novel nucleotide analogue having a methylene bridge between O-2' and C-4' atoms of the ribose. This unusual structure 'locks' the ribose in the 3'-endo/N-type conformation which is dominant in A-form DNA and RNA. Our previous studies have demonstrated that LNA does not only increase the duplex stability through high binding affinity of the complementary hybridization, but also can enhance the discrimination capability against the mismatches. Recently, to explore further its usefulness in biotechnology and bioengineering, we have conducted investigations on LNA's application in gene expression regulation. G-quadruplex is a unique secondary structure of DNA and RNA. G-quadruplex structures are selected through SELEX procedure and demonstrate many valuable behaviors: easy preparation, high target recognition specificity, high binding affinity, and wide range of layouts. A typical model of G-quadruplex is thrombin binding aptamer (TBA). TBA is a 15-mer nucleotide with a unimolecular antiparallel G-quadruplex. To enhance its stability, we have made modifications of some G residues with LNA. When it is modified with single LNA, its structure stability is increased and its characteristic of single strand, lateral anti-parallel structure keeps unchanged. Multiple LNA modifications, however, causes the G-quadruplex undergo conformational changes or collapsed. These structural changes are confirmed by CD, UV and fluorescence spectroscopic data as well as EMSA results. After consulting the promoter sequences of oncogenes, we proposed a model to use the G-quadruplex as a molecular element for regulating the gene expression. The selected LNA modification allows us to control the conversion between the G-quadroplex structure and DNA duplexes. This discovery could enhance our ability to regulate the gene expression and lead to the discovery of

new target for cancer treatment.

The regulations of the CaV1.2 Ca^{2+} channels by calmodulin and Ca^{2+} in guinea-pig cardiac myocytes

Li-ying Hao[1], Dong-yun Han[2,3], Guo Feng[1,2], Etsuko Minobe[2], Masaki Kameyama[2]

[1] Department of Pharmaceutical Toxicology, School of Pharmaceutical Sciences, China Medical University, Shenyang 110001, China. lyhao@mail.cmu.edu.cn; [2] Department of Physiology, School of Medicine and Dental Sciences, Kagoshima University, Kagoshima 890-8544, Japan. kame@m.kufm.kagoshim-u.ac.jp; [3] Department of Pharmacology, School of Pharmacy, Shenyang Pharmaceutical University, China.handy60@163.com.

Abstract: L-type Ca^{2+} channel (CaV1.2) is regulated by Ca^{2+}, which referred to as Ca^{2+}-dependent facilitation (CDF) and inactivation (CDI). Calmodulin (CaM) is a ubiquitous Ca^{2+}-binding protein that works as a Ca^{2+} sensor for both positive and negative modulations. In present study, we examined the effects of Ca^{2+} and CaM on Ca^{2+} channels in guinea-pig cardiac myocytes with the inside-out patch configuration of patch-clamp technique. When a free $[Ca^{2+}]$ was controlled at 0.1 ?M, CaM (0.15, 0.7, 1.4, 2.1, 3.5 and 7.0 ?M) + ATP (2.4 mM) induced channel activities of 27%, 98%, 142%, 222%, 65% and 20% relative to the control activity, respectively, showing a bell-shaped relationship. With increasing $[Ca^{2+}]$, the bell-shaped curve of CaM was shifted to the left side (lower concentration side). However, this shift was abolished when wild-type CaM was replaced with CaM mutants, CaM1234, for $[Ca^{2+}]$ up to 0.25 ?M, indicating that this shift was induced by Ca^{2+} and mediated by Ca^{2+}/CaM complex formation. We further investigated the concentration- and Ca^{2+}-dependent effects of CaM mutants, CaM12 and CaM34, in which Ca^{2+} binding to its N- and C-lobes was eliminated respectively. The concentration-response curves of CaM12 and CaM34 (0.7-10?M) were both bell-shaped, similar to that for wild-type CaM. However, there was no obvious leftward shift of the curves by increasing $[Ca^{2+}]$, suggesting that both functional lobes of CaM were necessary for the Ca^{2+}-dependent shift. We suggests that both apoCaM (Ca^{2+}-free CaM) and Ca^{2+}/CaM could induce facilitation and inactivation of CaV1.2 Ca^{2+} channel activities, and that Ca^{2+} could accelerate CaM-dependent facilitation (CaMDF) and inactivation (CaMDI). Both N- and C-lobes of CaM are required for the Ca^{2+}-dependent regulations of CaV1.2 Ca^{2+} channels. This work is supported by the grants from the Japan Society for the Promotion of Science and the National Natural Science Foundation of China (No.30670761, No.30870907).

Relationship of aquaporin 1, 3 and 5 expression in lung cancer cells to cellular differentiation, invasive growth and metastasis potential.

Yoshimichi Ueda*1, Yuichiro Machida*1*2, Miyako Shimasaki*1, Katsuaki Sato*1, Tsutomu Sakuma*2 and Shogo Katsuda*1

Department of Pathophysiological and Experimental Pathology *1 and Thoracic Surgery*2, School of Medicine, Kanazawa Medical University, Ishikawa, Japan

Abstract: An oncogenic capacity of aquaporins (AQPs), transmembrane channels for water, was recently proposed. This study seeks to elucidate the involvement of AQP1, 3 and 5 in the develop-

ment and progression of lung cancer. Expression analyses of AQP1, 3 and 5 by immunohistochemistry, western blot and laser-captured microdissection / real time RT-PCR in 160 lung cancers of various histologic subtypes showed that AQP1, 3 and 5 were expressed in tumor cells of 71, 40 and 56%, of lung cancers, respectively. AQPs expressions were frequent in adenocarcinomas (ADCs), while AQP1 and 5 were negative in squamous cell carcinomas. Bronchioloalveolar carcinoma (BAC) cells exhibited an apicolateral AQP1 and apicolateral or basolateral AQP3 localization in non-mucinous type, and apical AQP1 and 5 and basolateral AQP3 expression in mucinous type, which corresponded to AQPs expression of non-neoplastic lung tissue. Basolateral AQP5 expression was acquired during tumorigenesis of non-mucinous BAC. In contrast, invasive ADC tumor cells, either with fibroblastic reaction or papillary growth in the alveolar space, overexpressed AQP1 and 5 with loss of subcellular polarization and with an intracytoplasmic distribution. Overexpression of AQP1 correlated with high postoperative ADC metastasis ratios and unfavorable disease-free survival rates ($p=0.031$). We conclude that expression patterns of AQP1, 3 and 5 in lung cancer cells are mostly associated with cellular differentiation. However, the expression of AQP1 and 5 is up-regulated in invading lung cancer cells, particularly in ADCs, and the overexpression of AQP1 with loss of subcellular polarization is suggested to be involved in their invasive and metastatic potential.

Immunohistochemical analysis of oxidative modified DNA (8-OHdG) in Chinese nasopharyngeal lymphomas.

Hasui K[1], Jia XS[2], Aozasa K[3], Eizuru Y[1], Matsuyama T[1], Sato E[4]

1) Graduate School of Medical and Dental Sciences, Kagoshima University
2) Department of Pathology, China Medical University
3) Department of Pathology, Osaka University
4) Kagoshima University

Most nasopharyngeal lymphomas are Epstein-Barr virus (EBV)-associated NK/T cell lymphomas (NKTCL) revealing peculiar autophagic cell death (JECH 2009, 49: 9). Environmental factors such as pesticide effect on their onset (Int J Cancer. 2007, 120:406) and G/C to T/A point mutations were observed (Cancer Sci. 2003, 94:297). In order to see oxidative stress-related modified DNA in NKTCL, this study investigated guanine oxidation in nasopharyngeal lymphomas by means of immunohistochemistry of 8-hydroxyguanosine (8-OHdG). Oxidative stresses induce cell necrosis, oxidation of nuclear acids often makes guanine to be more Fapy G and less 8-OHdG. Respiratory and squamous epithelia showed nuclear stain of 8-OHdG when serous and mucous glandular epithelia indicated also nuclear stain of 8-OHdG in a half and less than half lesions. Nuclear stain of 8-OHdG was seen in most squamous carcinomas but less dysplasia. Inflammatory lymphocytes were negative for 8-OHdG. But most nasopharyngeal lymphomas showed cytoplasmal and nuclear stain of 8-OHdG. There were two kinds of cell-decreasing growth patterns of these lymphoma cells. One showed increase of 8-OHdG-positive nuclei and cell debris with enhanced autophagy labeled by LC3 when the other revealed massive increase of 8-OHdG-positive cell debris with autophagic

cell death densely labeled by LC3. Consequently, oxidative stresses effect on the nasopharyngeal mucosa when endogenous reactive oxygen spices (ROS) oxidate guanine in carcinomas and lymphomas. In the lymphomas enhanced autophagy propel accumulation of Fapy G disturbing DNA synthesis and autophagy-related cell death and EBV-related autophagy enhancement showed autophagic cell death with 8-OHdG-positive cell debris. It is sure that oxidative stresses effect on nasopharyngeal lymphomagenesis while endogenous ROS yield point mutations in their genes.

The Role of ET-1 and iNOS on Hypoxic Stress-induced Gastric Mucosal Lesion in Rats

Liu Shou Yang huilian Bai zhenzhong Li Su Jin guoen Ge rili

Abstract: Objective To explore the expression and change of endothelin-1 (ET-1) and inducible nitric oxide synthase(iNOS) induced by hypoxia-stress at high altitude of gastric mucosal of SD rat.

Methods: Sixty SD rats were randomly separated the control group and the experimental groups. The experimental groups, as well as the acute and chronic hypoxia models of SD rats were made in Kekexili National Natural Reserve Areas. The control group is Xining group. Semi-quantitative RT-PCR and Western blot were used to test the expression levels of ET-1 and iNOS and one-way ANOVA of SPSS software was used for statistics.

Results: There were the significant increased expressions levels in both mRNA and protein for the hypoxic stress group versus the control ones. We also observed the ET-1and iNOS levels higher expressed in the gastric mucosal lesions, maintaining this increased trends for the periods, with the peak occurring at the third day and seventh days, grandly downing forwards for the hypoxic groups in the later days, the concentrations consistent with the baselines in the control group. RT-PCR results indicated the ET-1 and iNOS expression levels consistent with the Western-blot imprinting results. But the ET receptor families showed A- type receptor higher expressed than B-types. Those indicated there were differences of those cytokines in the acute compared with the chronic hypoxic and control group except for the expression levels of the iNOS.

Conclusion: Our results demonstrated the increased ET-1 synthesis in the gastric mucosal, which caused down regulations of the blood supply for the genesis of stress-induced gastric mucosal lesion once they exposed in the acute hypoxia. In the mean time, temporally up-regulations of the iNOS at the initial stress periods to deepen the unbalanced pathological exchanges of the stressed gastric mucosal lesions in rats.

Key words: Hypoxia Stress; Gastric mucosal lesion ; Endothelin-1; Inducible nitric oxide synthase

EHMT1 interact with MT1h and regulate histone H3K9 methylation to inhibit proliferation and movement

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Abstract: Euchromatin histone methyltransferase 1 (EHMT1) was previously identified as one of the interaction proteins of metallothionein 1h (MT1h) in human lung cancer cDNA library by yeast two hybrid system. In this study, we showed that EHMT1 binds with MT1h. In yeast two hybrid,

pBD-MT1h and pAD-EHMT1 were co-transformed into yeast strain AH109, and the yeast colonies grew in a SD-leu/-his/-ade/-trp high-stringency medium, and showed positive α -galactosidase activity. Coimmunoprecipitation of MT1h and EHMT1 from lung cancer cell line A549 verified the interaction of MT1h and EHMT1 in vivo. In vitro, GST-MT1h fusion protein binds with EHMT1 in a cell-free system. Co-localization showed MT1h and EHMT1 co-localize in the nucleus, this also suggested except for cytoplasm expression, MT1h also express in the nuclear.

The expression of MT1h induced the increase of histone methyltransferase activity, and also higher histone lysine H3K9 tri-methylation. MT1h expression inhibited cell cycle and cell migration by functional analysis.

We conclude that MT1h interaction with EHMT1 may play a role in histone methylation, leading to tumor suppression.

Key words: MT1h, H3K9, DNA methylation, lung cancer

Involvement of aquaporin 1 and 5 in metastasis potential of human osteosarcoma.

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Osteosarcoma frequently metastasizes and causes miserable end result. However, little is known on its exact mechanism. Involvement of aquaporins (AQPs), transmembrane channels for water, in the aggressive growth of malignant tumor was recently disclosed in several carcinomas. This study aims to elucidate the involvement of AQP1 and 5 in metastasis potential of human osteosarcoma (OS). Western blot analyses of AQP1 and 5 in 5 human OS cell lines and 10 tumor tissues showed AQP1 expression in all 5 cell lines and 7 OS tissues, and AQP5 in 4 cell lines and 6 tissues. Immunohistochemistry in non-neoplastic skeletal tissue revealed membranal localization of AQP1 in active osteoblasts, while AQP5 was rarely expressed. In biopsied OS tissues, AQP1 was positive in tumor cells, with either membranal or cytoplasmic pattern, of 22/25 (88%) and AQP5, with cytoplasmic pattern, in 17/25 (68%). AQP1 expression was frequent in an osteoblastic subtype. Immunostaining of excised tumors showed the enhanced expression of AQP1 at the invasion front including filopodia. The cytoplasmic overexpression of AQP1 both in biopsy samples and at the invasion front of resected tumors correlated with high postoperative metastases ($p=0.004$, 0.035) and unfavorable survival rates ($p=0.0025$, 0.044). The former was proved an independent prognostic factor in multivariate-analysis. Furthermore, studies in a metastasis model using HT1080 cells of various metastasis potentials transplanted to nude mice revealed increased AQP1 and 5 expressions in cells with high metastatic ability compared with those with low one. We conclude that membranal expression of AQP1 in OS cells is most likely associated with cellular differentiation, but that the overexpression of AQP1 and 5 in OS cells seems to be involved in their invasive and metastatic potential.

MicroRNA Hsa-miR-125a-5p induces apoptosis by activating p53 in lung cancer cells

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Background: The mature microRNA hsa-miR-125a-5p is derived from the 5' end of pre-miR-125a. Although hsa-miR-125a-5p is dysregulated in some tumours, its specific roles in lung cancer cell apoptosis is still unknown. To study its function, we examined the effects of hsa-miR-125a-5p on apoptosis in lung cancer cells and investigated its probable regulatory mechanism.

Methods and Results: We showed that hsa-miR-125a-5p expression was lower in different lung cancer cell lines than in Human bronchial epithelial (HBE) cells by qRT-PCR. In gain-of-function experiments, we found that hsa-miR-125a-5p suppressed proliferation and induced apoptosis in A549 cells by MTT and flow cytometry, respectively. In addition, wild-type p53 mRNA and protein expression was increased by hsa-miR-125a-5p over-expression. Moreover, blocking wild-type p53 attenuated the effect of hsa-miR-125a-5p on apoptosis. In loss-of-function experiments, wild-type p53 mRNA and protein expression was decreased by blocking hsa-miR-125a-5p. The effect of hsa-miR-125a-5p inhibitor on apoptosis was also weakened by blocking wild-type p53.

Conclusion: Taken together, these data suggest that hsa-miR-125a-5p induces apoptosis via a p53-dependent pathway in human lung cancer cells. These results provide new insight into the roles of the miR-125a family in lung cancer.

Expression and Clinicopathologic Significance of Human Achaete-scute homolog 1 in Pulmonary Neuroendocrine Tumors

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Abstract: Background and objective Human Achaete-scute homolog 1 (hASH1) gene is a basic-helix-loop-helix transcription factor, plays a critical role in development of the central nervous systems, adrenal medullary chromaffin cells, thyroid C cells, and pulmonary neuroendocrine cells. During development in vertebrates, ASH1 gene is usually temporary and expressed in embryonic pulmonary neuroendocrine cells in early differentiation stage, the emergence of terminal differentiation markers its expression tended to silence. The aim of this study is to determine the normal lung tissue and various types of lung tumors hASH1 gene expression, to analyze whether its expression was correlated with pulmonary neuroendocrine markers, and to explore the possibility of hASH1 as clinical pathological markers in the neuroendocrine tumors compared with previous neuroendocrine tumor markers.

Methods: hASH1, Chromogranin A, Synaptophysin and CD56 expression was examined in lung tumor specimens (lung inflammatory pseudotumor, squamous cell carcinoma, adenocarcinomas, large cell carcinoma, typical carcinoids, atypical carcinoids, large cell neuroendocrine carcinomas

and small cell lung carcinoma and corresponding normal lung specimens using immunohistochemistry (S-P method). Western blot and reverse transcription polymerase chain reaction (RT-PCR) assay were applied to detect the expressions of hASH1 protein and mRNA in lung cancer tissues.

Results: hASH1 expression was detected in 2/16 (12.5%) typical carcinoids, 15/20 (75%) atypical carcinoids, 6/10 (60%) large cell neuroendocrine carcinomas and 31/40 (77.5%) small cell lung carcinoma, respectively, but not in any normal lung tissue (0/10), lung inflammatory pseudotumor(0/49), squamous cell carcinoma (0/30), adenocarcinomas (0/30) or large cell carcinoma (0/20). There was a significant difference in hASH1 expression incidence between typical carcinoids and atypical carcinoids ($P<0.01$), but not in large cell neuroendocrine carcinomas and small cell lung carcinoma ($P>0.05$). hASH1 expression correlated very closely with Chromogranin A, Synaptophysin and CD56 expression ($P<0.05$).

Conclusion: hASH1 is a new kind of highly specific markers of pulmonary neuroendocrine tumours, and may be applied to clinical pathology diagnosing of the pulmonary neuroendocrine tumors.

Clinicopathological Features of 10 Cases of Atypical Lipomatous Tumor/Well-differentiated Liposarcoma

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Abstract: **Objective**: To explore the clinicopathological characteristics and pathol ogical differential diagnosis of atypical lipimatous tumor(ALT)/well-differentiated liposarcoma(WDL).

Methods: Ten cases of ALT/WDL were reported, with review of relevant literature.

Results: Three of cases recurred. All cases are well circumscribed but not encapsulated and they resemble ordinary lipoma grossly. In histopathologic examination, the atypical cells may concentrate on the fibrous strands that traverse the adipose tissue lobules or be scattered among the mature adipocytes and lipoblasts can be seen. Two cases showed other heterologous differentiated elements.

Conclusion: The behavior of ALT is substantially different depending on their location ALT may recur but there are no tumor-related deaths. WDL have a very high incidence of recurrence even dedifferentiated and some of the patients die as a result.ALT/WDL shows a variety of histopathologic phenotype and needs to be distinguished from other tumors arising from lipocyte or others.

Relationship between expressions of heparanase and GA-binding protein in Human Breast Cancer

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Objective: To observe the expressions of heparanase, GA-binding protein in breast carcinorma

and study their roles in tumor.

Methods: A total of 74 paraffin-embedded tissue specimens of breast carcinoma and 20 cases of nearby normal tissue were examined by immunohistochemical S-P method. The data were analyzed by SPSS statistical software.

Results: Immunohistochemistry showed that heparanase, GA- binding protein were distributed in cytoplasm and nuclear of tumor cells, incontrast to their nonexpression in normal breast tissue. GABP-positive cases had a higher positive rate of heparanase than GABP-negative cases. A significant correlations were found among the expressi- ons of heparanase, GA-binding protein and clinical stage, pathological stage, lymph node metastasis and clinical prognosis.

Conclusion: Expressions of heparanase and GA-binding protein are significantly associated with breast carcinoma invasion and metastasis.We suppose that GA- binding protein up-regulate heparanase.

Key Words: breast cancer, heparanase, GA-binding protein, immunohistochemistry

CCL19/CCR7 regulate the Expression of Heparanase via Sp1 in Lung Cancer A549 cell

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PURPOSE: Chemokine CCL19 and its receptor CCR7 play an important role in the homing of lymphocytes. CCR7 overexpression was found to be associated with tumor invasion.But the mechanism that CCR7 promote tumor cells invasion is unclear.In this study, we added CCL19 to A549 cell exogenously and activated CCR7 to detect the expression of Sp1 and Heparanase(Hpa) and cell invasiveness.

Methods: We added CCL19 to A549 cell exogenously and used RT-PCR , Western Blot analysis, Chromatin immunoprecipitation (ChIP) and Transwell to detect the expression of Sp1 and Hpa, whether Sp1could bind to the promoter of Hpa or not, and cell invasiveness respectively

Results: Treatment human lung adenocarcinoma A549 cells with recombinant human CCL19 obviously increased the expression of Sp1 and heparanase mRNA and protein. After blocking CCR7 and the expression of Sp1 respectively, the expression of Sp1 and heparanase mRNA and protein decreased, while decreased expression of heparanase was not affected by CCL19. ChIP analysis demonstrated CCL19 enhanced the DNA binding of Sp1 to the promoter of heparanase .After incubation with CCL19, the invasion ability of A549 cell increased in comparison the control group . After blocking CCR7 and Sp1 respectively, the invasion ability decreased. Migration assay showed that the invasion ability of A549 cell did not change.

Conclusions: CCL19 can activate CCR7 to up-regulate the expression of Sp1 and promote the transcription of heparanase mRNA and cell invasiveness in lung adenocarcinoma A549 cell .

Effects of angiostatin on HUVECs in vitro and its mechanism

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Introduction and Objective: Angiostatin is endogenous tumor angiogenesis inhibitor, which affect vascular endothelial cells, block the proliferation and migration, and reduce tumor angiogenesis. Mechanism of antiangiogenic action of angiostatin has not explained fully so far, the cell signaling pathway of angiostatin acting on the vascular endothelial cells has not been proven. We investigate the relationship between expression of angiostatin and tumor angiogenesis in the tumor tissue and the regulation of angiogenesis factor VEGF link in the body material, animal and in vitro cell culture experiments at three levels. These researches may provide some theoretical references and foundational understandings for the prevention and treatment of tumors.

Methods: Primary cells culture; MTT colorimetric assay; Trypan blue stain cells counting; Establishing of tumor microvascular model; Flow cytometry; and western blot.

Results: (1)MTT assay results shows: Different concentrations of angiostatin act in human umbilical vein endothelial cells after 24 h, OD490 absorbance values were lower than the control group, its inhibiting action on the proliferation of endothelial cells with increasing concentrations of angiostatin increased, compared with the control group differences were significant ($P < 0.01$). And along with extension of acting time, the proliferation of human umbilical vein endothelial cells inhibited significantly, its absorbance values were lower than the control group. Measurement results show that: when 72h, the maximum inhibition rate reached 91.37 percent, compared with the control group the difference was significant ($P < 0.01$).(2)Flow test shows: from 4 mg/L angiostatin on, apoptosis began, and to 16 mg/L angiostatin, there is a clear peak of apoptosis, and increased with the dose, apoptosis peak is gradually strong.(3)Test of cavity formation in endothelial cells: After human umbilical vein endothelial cells were cultured in Tail plastic of mice and induced by tumor supernatant, endothelial cells of morphology have been observed from scattered in the state to gathered, gradually cord-like dendritic formation, 12h reticular formation obvious. In cultured human umbilical vein endothelial cells after 3 h (vascular network has not formed before) by adding angiostatin, endothelial cells have not formed network structure. When endothelial cells were cultured for 24h (vascular network has been formed) by adding angiostatin, we discovered 8h endothelial cells decrease in the number of formating networks and became round, and gradually gathered, 12h blood vessels network structure formed by endothelial cells have been ruptured, and disappear after fracture, the remnants of the endothelial cells form cells lumped together.(4)Flow detection expression of integrin $\alpha v \beta 3$ in endothelial cells: FCM results showed, the average fluorescence density expression in control group is 6.76 ± 2.19, pure bFGF group is 42.3 ± 4.41, Angiostatin low dose group is 24.8 ± 4.26, the high-dose group of angiostatin is 2.34 ± 1.16. Comparing groups, there is a relatively obvious statistical significance. This results indicate that angiostatin restrained the expression of integrin $\alpha v \beta 3$ in endothelial cells by bFGF induction.(5) Western Blot detection Angiostatin role after the angiogenesis pathway : After reaction of endothelial cells and spicific signal pathway inhibitors Akt, P38, FAK protein expression levels are relatively low, bFGF stimulated slightly higher. At the same time in the endothelial cells after treated with Angiostatin, expression levels of NF-?Bp65 and Survivin increased with the dose of Angio-

statin reduced.

Conclusions: The expression of Angiostatin in human NSCLC correlate with the clinical tumor stage, differentiation and lymph node metastasis. 2. Angiostatin can restrain the growth on A549 lung carcinoma in mice transplanted tumor and inhibit neovascularization.3. Angiostatin can inhibit the proliferation of human vascular endothelial cell incubated in vitro and induce its apoptosis, and restrain blood vessel cavity formation induced by tumor in vitro. 4. Angiostatin related with inhibition of endothelial cell proliferation or induction of apoptosis and the role of the integrin avβ3 expression and Survivin, NF-κB.

Expression of TRAF1 and its Relationship with TRAF2 in the Different Metastatic Potential Breast Cancer Cell Lines

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Abstract: BACKGROUND & OBJECTIVE: The expression and role of Tumor necrosis factor receptor-associated factor 1 (TRAF1) in breast cancer is unclear. This research was to study the expression of TRAF1 and the relationship of TRAF1 and TRAF2 in the different metastatic potential breast cancer cell lines.

Methods: immunocytochemistry staining and western blot were used to detect the expression of TRAF1. Co-immunoprecipitation was used to study the relationship between TRAF1 and TRAF2.

Results: In comparison with the normal, weakly and/or moderate metastatic counterparts, TRAF1 was upregulated in highly metastatic potential breast cancer cell lines ($P<0.05$), while the quantity of TRAF1 in combination with TRAF2 was downregulated($P<0.05$).

Conclusions: As the metastatic potentiality of breast cancer cell lines increases, the expression of TRAF1 would be stronger while the quantity of TRAF1 in combination with TRAF2 would be smaller. It suggests that TRAF2 can reduce the depressant effect through the decreased combination between TRAF1 and TRAF2 and then to promote the invasion and metastasis of breast cancer.

Key words: Breast cancer; Tumor necrosis factor receptor-associated factor 1 (TRAF1); Neoplasm mestastasis

Expression and Role of OCT3/4 in the Regeneration of Rat Trachea Epithelium Cell in Vivo

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This work was supported by National Natural Science Foundation of China (grant 30972966 to X.-S.J)

Objectives: To explore the role of the OCT3/4 in regeneration of rat tracheal epithelium.

Materials and methods: An in vivo model of rat tracheal epithelial regeneration using 5-fluorouracil (5-FU) was developed, to induce injury. Expression levels of Oct3/4 were examined using microscopically observed immunofluorescence, and cell morphological changes were observed using HE staining and electron microscope, during the recovery process.

Results: Oct3/4 were not detectable in normal tracheal epithelium. After treatment with 5-FU, the normally proliferating tracheal epithelium desquamated and only a few cells in G0 phase of the cell cycle were left on the basement membrane and Oct3/4 could be observed at this time. Thereafter, the number of Oct3/4-positive cells increased gradually. When the cells differentiated into ciliate cells, mucous cells or basal cells, and restored pseudostratified mucociliary epithelium, the number of Oct3/4-positive cells decreased and gradually disappeared.

Conclusions: G0 phase cells with resistance to 5-FU damage expressed Oct3/4. This indicated that these cells were undifferentiated, but had the ability to terminally differentiate into downstream-type cells. They possessed stem cell properties. The results are consistent with Oct3/4-expressing cells being considered as tracheal stem cells.

Key words: OCT3/4, cell reprogram, differentiated, 5-FU

Expression and Role of Oct3/4 in Wound-repair Process of Rat Lung Induced by Bleomycin

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Abstract: Introduction A concept of a correlative genome Oct3/4、Sox2、Nanog for embryonic stem (ES) cells is expressed only in the ES cells, but not in mature somatic cells. Some researchers had already used retroviras transduction with three genes: Oct3/4、Sox2、Nanog into fibroblast and directly induced somatic cells into a pluripotent stem cell state. Xinshan Jia et al have developed an ex vivo model of rat tracheal epithelial regeneration using 5-fluorouracil (5-FU) to induce tissue injury and found the G0 phase cells which resistance to 5-FU could express Oct3/4、Sox2、Nanog, indicating tracheal epithelium cells reprogrammed into stem cells.When these cells differentiated into ciliate cells, mucous cells, basal cells, the number of Oct3/4-, Sox2- and Nanog- positive cells decreased and gradually disappeared.

Objective: Observe the wound-repair process in vivo of rat lung epithelium induced by bleomycin and explore the expression of Oct3/4 and its dynamic regularity to testify whether epithelium cells reprogrammed into stem cells. Methods An in vivo model of rat lung epithelial regeneration using bleomycin was developed, to induce injury and expression levels of Oct3/4 was dynamically examined using Western blot analysis, microscopically observed immunofluorescence, and cell morphological changes were observed using HE staining, during the recovery process. Using double-positive cells of alveolar type II cell-specific marker prosurfactant apoprotein-C (SP-C) and Clara cell-specific marker CC10 to locate bronchioalveolar stem cells, and PCNA-negative cells were G0 phase cells.

Results: Oct3/4 was not detectable in normal lung alveolar epithelium. 1.After treatment with bleomycin 1 day , the normally alveolar epithelium began necrosis and decreased and only a few cells in G0 phase of the cell cycle were left and immunofluorescence indicated Oct3/4 could be observed at this time. After 2 days, the number of G0 phase cells still exist and Oct3/4 -positive cells

were more than the first day. When the cells differentiated into mature, they began to generation and recover from the injury and the number of Oct3/4-positive cells decreased and gradually disappeared but fibrosis was proliferated. The number of Oct3/4-positive cells was more than the bronchioalveolar stem cells 2.Western blotting analysis showed that there were different Oct3/4 levels at different times after introduction of bleomycin which in accordance with the change of immunofluorescene, Oct3/4 was minimally detected after treatment with bleomycin for 1day, reaching a maximal level at 2 days after the introduction of bleomycin, and then decreased over time. Until 5 days after the treatment of bleomycin, very low Oct3/4 was detected. Conclusions :The remained G0 phase cells with resistance to bleomycin damage expressed Oct3/4 and reprogrammed into stem cells. These cells were undifferentiated, but had the ability to terminally differentiate into downstream-type cells and repair the injuries. They possessed stem cell properties.

Key words: Lung Stem cell; Oct3/4;injure; repair;bleomycin.

Changes of Histone deacetylase 1 and the roles of Trichostatin A (TSA) during Regulated Growth of Tracheal Stem Cells in the Rat Regeneration

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Introduction: we have previously constructed a tracheal injury model induced by 5-fluorouracil (5-FU)ex vivo in rats and idendified bronchial stem cells exist in the G0 cells. However, the mechanism that regulate tracheal stem cell proliferation and differentiation is still unkown. Histone deacetylase 1 has been knows as a switch of cell from proliferation to differentiation. To determine the function of HDAC1 during regeneration after injury of rat tracheal stem cell, we observe the changes in space-time expression and explore the relationship between HDAC1 and tracheal stem cell proliferation and differentiation. In addition, TSA is an organic compound selectively inhibits mammalian histone deacetylase (HDAC). We want to observe the role of TSA after injury of tracheal stem cells.

Materials and Methods: 1. Preparation of tracheal stem cells regeneration model. 2. HE dyeing and Immunohistochemistry were used to observe the morphological changes during tracheal epithelium regeneration. 3. Western blot was used as quantitative analysis to observe the changes of tracheal cells.

Results: 1. Morphological Changes in Tracheal Epithelium: The tracheal epithelium desquamated after 5-FU treatment. The residual were the tracheal epithelium at 3,6,9,12,24,48h after removing 5-FU, and found the morphological of the cells changed extreme flat ,cuboidaland pseudostratified mucocilary gradually. Whereas, the tracheal cells after TSA treatment, this progress can be see slow down obviously. 2.Expression of HDAC1 in Tracheal Epithelium:HDAC1 localized in nucleus. Results of immunohistochemistry showed the expression of HDAC1 was negative in normal cell and increased gradually till 24h, then decreased slightly at 48h. In addition, the expression of

HDAC1 after the treatment of TSA showed that this progress has been inhibited. 3.Changes in HDAC1protein Level : Result of western blot indicated that there was no detectable level of HDAC1 protein in normal tracheal epithelium and was elevated slightly at 3 , 6h after the removel of 5-FU, and reached a top at 24h, then decreased slightly at 48h..And after TSA treatment the progress was slowed down slightly.

Conclusions: Our work revealed the expression of HDAC1 was few during tracheal stem cell proliferation and increased obviously when differentiation occurs, and the differentiation progress can be slow down after TSA treatment. We conclude that HDAC1 function as a molecular switch of tracheal epithelial stem cells during regeneration after injury.

5-FU induces the expression of transcriptional factor OCT3/4 and increases the number of side population cells in lung adenocarcinoma SPC-A1

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Objective: To observe expression of OCT4 and SOX2 of transcription factor and the percentage of side population (SP) cells when lung adenocarcinoma SPC-A1 exposed to 5-FU .Properties of SP cells were evaluated by their invasion potential and tumorigenic potential.

Methods: Proliferation inhibition was detected by MTT assay, acording to which IC50 was caculated. Expression levels of stem cell markers were examined by immunofluorescence and Western-Blot after treatment of 5FU for 0h 24h 48h at IC50. Additionally, SP cells were sorted using FACS. Properties of SP cells were evaluated by transwell, colony-forming experiment and tumor formation in an animal model.

Results: 5-FU can greatly inhibate the proliferation of SPC-A1 , the IC50 of which was 100μ M. The exposure to 5FU can stimulate the expression of stem cell markers and increase the percentage of SP cells.SP cells have a greater invasive potential and a greater ability to form tumor in vivo.

Conclusion: SP cells exist in human lung adenecarcinoma cell lines and the percentage could incresse after the treatment of 5-FU.

Key words: Lung cancer cells 5-FU Cancer stem cell OCT4 Side population cells

Bone marrow Derived Mesenchymal Stem Cells Promote the Expression of Cdk4 in Pancreatic Islets in Vitro

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Introduction: We had reported that transplanting rat bone marrow mesenchymal stem cells (MSCs) can mitigate the diabetic syndromes and reduce the high blood glucose. The mechanism

still needs to be elucidated. Cdk4 gene is thought to regulate the regeneration and proliferation of pancreatic islet β cells. So whether MSCs promote the regeneration of islet through the pathway of Cdk4 need to be elucided.

We cocultured the pancreatic islets or insulin secreting cell line INS1 with MSCs in vitro and observed the influence of the MSCs to the expression of Cdk4 in the pancreatic islets and INS1. The aim of our study is to explore the potential mechanism of MSCs to promote the regeneration of the islets β cells in vitro.

Method: 1. Isolation of rat pancreatic islets and MSCs in vitro. 2. Coculture of pancreatic islets or INS1 with MSCs in separate room of one transwell. 3. Detect of the expression of cell proliferation gene Cdk4 in rat pancreatic islets and INS1 by real-time PCR.

Results: The Cdk4 gene of pancreatic islets and INS1 was high expressed by 2.281 and 1.869 folds respectively after coculturing pancreatic islets or INS1 with MSCs for 5 days.

Conclusion: The bone-marrow derived mesenchymal stem cells promote the regeneration of the pancreatic islet via the pathway of Cdk4.

Key words: Bone marrow mesenchymal stem cells, pancreatic islets, Cdk4.

The Expression and Significance of MMP–2 and VEGF in Lung AdenocarcinomaCell

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[Abstract] Objective: To investigate the expression of matrix metallo- proteinase-2 (MMP-2) and vascular endothelial growth factor (VEGF) in pleural effusion and sputum cytological specimen of the patients with lung adenocarcinoma and the role during invasion and metastasis of the lung cancer.

Methods: The expressions of MMP-2 and VEGF in 264 cases of paraffine cytological specimen which come from pleural effusion and sputum were detected by immuncytochemistry method.

Result: The positive rate of MMP-2 was71.7% in adenocarcinoma cells of pleural effusion 、16.7% in atypical hyperplasia cells and 39.1% in adenocarcinoma cells of sputum; it showed no expression in hyperplastic epithelial cells from benign pleural effusion. The positive rate of MMP-2 in tumor cells of malignant pleural effusion was significantly higher than that in atypical hyperplastic cells or hyperplastic epithelial cells of the pleural effusion and adenocarcinoma cell in sputum($P<0.05$).The incidence of VEGF expression was 89.1% in pleural effusion adenocarcinoma cells、33.3% in atypical hyperplasia cells compared to 47.8% in adenocarcinoma cell of sputum; it showed no expression in hyperplastic epithelial cells of benign pleural effusion. The positive rate of VEGF in tumor cell of malignant pleural effusion was significantly higher than that in atypical hyperplastic cells 、hyperplastic epithelial cells of the pleural effusion and tumor cell in sputum($P<0.05$). Moreover, the positive rate between MMP-2and VEGF had positive correlation($r=0.867$, $p=0.049$).

Conclusion: The high expressions of MMP-2 and VEGF in tumor cells from pleural effusion of the patients with lung cancer may have relationship with tumors invasion and metastasis.Moreover, the unity of MMP-2 and VEGF inmmunecytochemistry is helpful for pathological diagnosis of adenocarcinma cells of lung cancer.

Key words: MMP-2; VEGF; Lung adenocarcinoma cell

女性盆腔 Ewing /PNET 肿瘤一例及文献复习

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本文综合应用免疫组织化学染色和分子生物学检测确诊了一例 33 岁女性患者右侧盆腔原始外周神经外胚层瘤 / 龙文氏肉瘤（Ewing sarcom/ primitive neuroectodermal tumour Ewing /PNET）。Ewing /PNET 是指包括骨和软组织 Ewing 肉瘤、Askin 瘤和外周原始神经外胚瘤在内的一组肿瘤。现认为起源于神经内胚层。作者结合文献探讨了 Ewing /PNET 与腹腔内促纤维增生性小圆形细胞肿瘤（IADSRCT）、神经母细胞瘤胚胎性横纹肌肉瘤、恶性淋巴瘤、小细胞未分化癌、恶性外胚层间叶瘤以及可能发生转移此处的卵巢小细胞肿瘤：卵巢肺型小细胞癌，分为两型：高钙型和肺型，卵巢高钙型小细胞癌相鉴别。

Dishevelled-1 and Dishevelled-3 Affect Cell Invasion Mainly Through Canonical and Noncanonical Wnt Pathway, Respectively, and Associate With Poor Prognosis in Nonsmall Cell Lung Cancer

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Abstract: Dishevelled (Dvl) family proteins are overexpressed in nonsmall cell lung cancer (NSCLC), but the correlation between Dvl overexpression and patient prognosis is not clear. The underlying mechanisms of Dvl-1 and Dvl-3 promoting lung cancer cell invasion require further research. We used immunohistochemistry to assess the presence of Dvl-1, Dvl-3, b-catenin, and p120ctn, and compared their expression to the prognosis in 102 specimens from NSCLC patients. We also examined the effect of Dvl-1 and Dvl-3 on Tcf-dependent transcriptional activity, as well as on the invasiveness in A549 and LTEP-a-2 lung cancer cells. The results showed that Dvl-1 correlated to the abnormal expression of b-catenin, while Dvl-3 correlated to p120ctn. Both Dvl-1 and Dvl-3 were related to the poor prognosis of patient. Dvl-1 overexpression enhanced the Tcf-dependent transcriptional activity and b-catenin expression significantly. However, Dvl-3 had little effect on the Tcf-dependent transcriptional activity and b-catenin expression, which was accompanied by p38 and JNK phosphorylation. Furthermore, the invasiveness of Dvl-3-enhanced cells was inhibited by p38 and JNK inhibitors. Exogenous expression of both Dvl-1 and Dvl-3 increased the p120ctn protein expression, while only Dvl-3 upregulated p120ctn mRNA. We conclude that both protein and mRNA of Dvl-1 and Dvl-3 are overexpressed in NSCLC in a manner related to poor prognosis. Dvl-1 may affect the biological behavior of lung cancer cells mainly through b-catenin (canonical Wnt pathway), while Dvl-3 mainly through p38 and JNK pathway (noncanonical Wnt pathway).

Key words: dishevelled; b-catenin; Wnt; p120ctn; lung cancer

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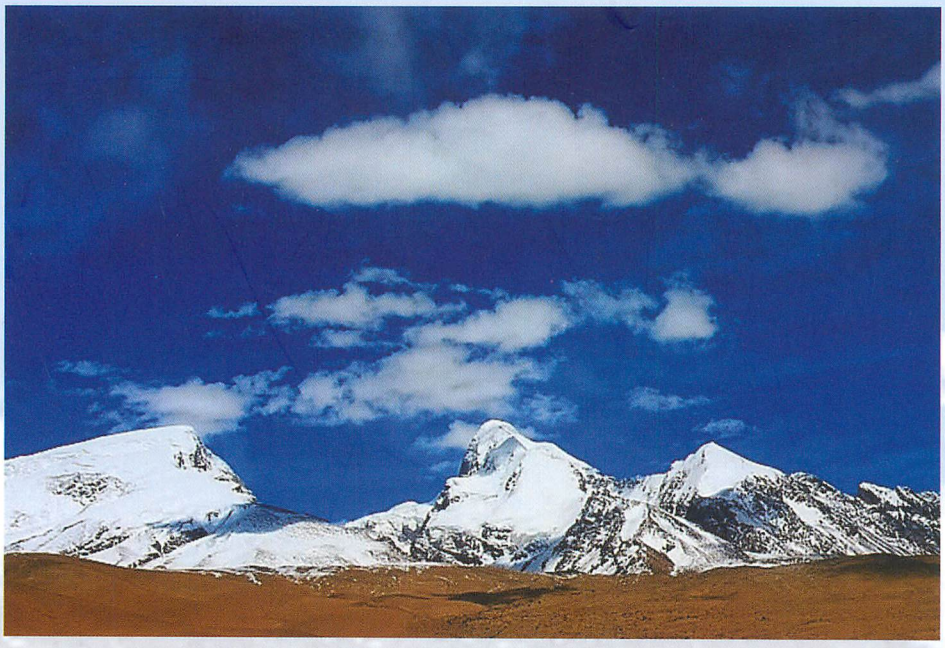

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