

CURRICULUM VITAE

Surname Name: Zheng
Given Name: Lishu
Sex: Female
Citizenship: P.R.China
Address: State Key Laboratory for Molecular Virology and Genetic Engineering,
National Institute for Viral Disease Control and Prevention,
Chinese Center for Disease Control and Prevention
100 Ying Xin Jie, Xuan Wu Qu,
Beijing 100052, P. R. China
Tel: 86-10-83549762 Fax: 86-10-63541221
E-mail: zhenglishu2006@yahoo.com.cn

EDUCATION

2003.9-present Ph.D. Candidate (Ph.D. degree will be granted in July,2006)
(Pathogenic biology)
State Key Laboratory for Molecular Virology and Genetic Engineering
National Institute for Viral Disease Control and Prevention,
Chinese Center for Disease Control and Prevention
1998.9-2001.7 M.S (Toxicology)
China Medical University
1991.8-1996.7 B.S. (Preventive Medicine)
Shenyang Medical College

WORK EXPERIENCE

2001.7-2003.8 Major: Virology ,Molecular Biology and Epidemiology
(Research Assistant)
National Institute for Viral Disease Control and Prevention,
Chinese Center for Disease Control and Prevention
1996.8-1998.8 Major: Disease Control and Epidemiology ; Disease Statistic Analysis.
(Public Health Doctor)
Anti-Epidemic Station of Dongling District in Shenyang Liaoning
Province.

RESEARCH EXPERIENCE

1. High-level expression and purification of influenza virus M2 protein in *E. coli* . and detection of its antigenicity

M2 gene from influenza virus A/PR/8/34 infected MDCK cells was cloned, and deleted its transmembrane domain (26-43) in order to obtain high level expression. Then the mutated M2 (sM2) was subcloned into expression vector pET30a, and the recombinant plasmid pET30a-sM2 was highly expressed in *E.coli* BL21 induced by IPTG. The sM2 fusion protein was purified with the Ni²⁺ chelating sepharose. Sera collected from the mice that were immunized with the purified fusion protein could bind to influenza virus infect MDCK cells testified by immunofluorescence experiment. This indicated that the expressed sM2 protein possesses M2 antigenicity.

2. Expression, purification and characterization of hemolytic activity of the recombinant *Listeria monocytogenes* Listeriolysin O

The hlyA gene of LLO was amplified by PCR and compared with the other 9 hlyA genes in GenBank showing 3 amino substitutions most. The constructed LLO expression plasmid pET30a-hlyA lacking the amino acid signal sequence was induced by IPTG in *E.coli* strain BL21DE3 and obtain recombinant fusion protein with high purity after being purified with the Ni²⁺ chelating sepharose. The immunity of the fusion protein is satisfactory determined with Western blot and the hemolysin showed its maximum hemolytic activity 1.41×10^4 HU/mg at pH5.5 tested with human erythrocyte. The fusion protein LLO with sufficient immunity and hemolytic activity can be useful for mechanism investigation and immune applications of LLO in the future.

3. Protection of mice against influenza A virus challenge by vaccination with modified sM2 and LLO protein

Female BALB/c mice 6 weeks of age were immunized with 10ug influenza modified sM2 and/or 10ug LLO protein by the intraperitoneal route. Three inoculations were administered at three-weekly intervals. Three weeks after final vaccination, mice were subjected to sublethal 0.5LD50 and lethal 10LD50 homologous A/PR/8/34 (H1N1) and heterologous A/Sydney/5/97 (H3N2) influenza viruses challenge. Lung homogenates were prepared at 7 days after challenge and titrated in MDCK cells for virus infectivity. The data suggested that vaccination of mice with sM2 protein resulted in a significant reduction in virus shedding by day 7 post-infection, which is sufficient to protect mice from death following lethal infection with either homologous or heterologous viruses. Furthermore, co-immunization with LLO could enhance viral clearance from the lungs of infected mice because of increased CTL activity testified by ELISPOT assay.

4. Cloning, Expression and Identification of Adhesion Gene hpaA of *Helicobacter pylori*

A prokaryotic expression system of *H.pylori* hpaA gene with high efficiency was established successfully. The expression HpaA fusion protein with satisfactory immunity can be used as candidate antigen in *H.pylori*.

5. Peptide inhibitors of human influenza A neuraminidase from phage display 12 peptides library.

Virus in culture supernatants from fully cytopathic cultures of MDCK cells was cleared of cellular debris by centrifugation at 1000rpm for 10 min, solubilized with the addition of Nonidet P-40 to a final concentration of 0.1% and used without further modification as the source of enzyme. Then peptides specially inhibiting the activity of neuraminidase were selected from the 12 peptides library using 3 round screening. The selected peptides were synthesized by solid phase peptide synthesizer and can inhibit the activity of neuraminidase.

6. Expression, purification and antiviral activities of a new recombinant human interferon- λ 2

According to preferred codons used in *E. coli*, the highly-expressed human interferon- λ gene was designed, synthesized and cloned into expression vector pBV220 and transferred to *E.coli DH5 α* . The recombinant plasmid pBV220-huIFN- λ was highly expressed in *E.coli DH5 α* and the expressed product existed in inclusion body containing about 15% of total somatic protein. FN- ϵ protein was purified by CM FF column and size exclusion chromatography and reached purity above 90%. The anti-viral activity of the product is about 1.5×10^6 IU/mg on WISH/VSV test system. It possessed more stringent species specificity and similar anti-HBV activity as compared with interferon- α 2b.

7. Expression of Recombinant Human IFN2 β 1a cDNA in CHO Cells

Mix recombinant plasmids pRCPCMV2IFN β DNA and pSV2dhfr2DNA (used as a selective marker) at a ratio of 3 : 1 and transfect into CHO-kldhfr cells by liposome technique. Screen the single clones grown in thymine-free selective medium for the pressure amplification of IFN2 β gene. Subject the screened clones to large scale culture, and collect the culture liquid for the purification by a series of procedures. The daily yield of IFN2 β secreted by the cells resistant to 0.6 μ mol/L MTX was 10^5 unit/ 10^6 cells. The specific activity of human IFN2 β expressed in CHO cells reached 2.2×10^8 IU/mg after purification. In conclusion, A CHO cell strain suitable for the expression of human IFN2 β was established

LABORATORY SKILLS

1. MOLECULAR BIOLOGY

Isolation and purification of DNA and RNA from tissue and culture cell; PCR and RT-PCR; Gene cloning; DNA sequencing; Vector construction; Gene mutation (deletion and site-direction); DNA and RNA transfection (liposome and electroporation); Gene expression in prokaryotic and eukaryotic system; SDS-PAGE assay; DNA and RNA hybridization; Computational Analysis for Protein and Nucleic Acid (Sequences were aligned with DNASTar and phylogenetic trees were drawn by using PHYLIP and ClustalX)

2. BIOCHEMISTRY

Chromatography techniques (including gel-filtration chromatography, ion-exchange chromatography, affinity chromatography), ultracentrifugation

3. VIROLOGY

Virus isolation, virus biological identification

4. CELL BIOLOGY

Mammalian Cell Culture, Mammalian Cell Transfection, Reporter Gene Assay, Immunohistochemistry

5. IMMUNOLOGY

ELISA, western-blotting, preparation of monoclonal antibody

6. EPIDEMIOLOGY AND STATISTICS

Familiar with epidemiology research methods. Able to do statistics analysis by biosoftware: SPSS, EPI2000, SAS. Automated sequencing and analysis.

LANGUAGE QUALIFICATION

CET-6, good ability to communicate with others in English, both verbal and written

PUBLICATIONS

1. **ZHENG Lishu**, DUAN Zhaojun, PENG Fuwang, et al. Expression, purification of influenza virus M2 protein and immune protection in mice. Chinese J Exp Clin Virol. 2006 (in press)
2. Qu xiaowang, Qi zhengyu, Duan zhaojun,---**Zheng lishu**, Hou yunde. Human bocavirus infection among hospitalized children with acute respiratory tract disease in China. Chinese Journal of Virology. 2006,21(1):21-26
3. **ZHENG Li-shu**, LI Wu-ping, WANG Gang, et al. Expression, Purification and Characterization of Hemolytic Activity of the Recombinant *Listeria monocytogenes* Listeriolysin O. Chin J Biological. 2006,19 (3), 66-69
4. **ZHENG Lishu**, DUAN Zhaojun. Development of A influenza virus M2 vaccine. Chin J Virol. 2006 (review, in press)
5. **Zheng Lishu**, Yi Zuoan, Li Wuping, et al. Cloning ,Expression and Identification of Adhesion Gene hpaA of *Helicobacter Pylori*. Chin J Biological. 2005,18 (2), 89-92
6. Wang gang, Li wuping, Zhang chenghai, Yi zuoan, **Zheng lishu**, Zhang hui, Duan zhaojun, Hou yunde. Expression, purification and antiviral activities of a new recombinant human interferon- λ 2. Chinese J Exp Clin Virol. 2005,19(3), 232-235
7. Fang Zhao-yin, ZHANG Li-jie, TANG jing-yu, ZHANG Qing, HU Hai-kuan, XIE Hua-ping, **ZHENG Li-shu**, Duncan Steels, Paul Kilgore, Joseph Bresee, Eric Hummelman, Xi Jiang, Roger Glass. Rotavirus diarrhea among children in Lulong county, Hebei province, China. Chinese Journal of Virology. 2005,21(1):21-26
8. LI Wu-ping, YI Zuo-an, ZHANG Cheng-hai, WANG Gang, **ZHENG Li-shu**. Expression of Recombinant Human IFN2 β 1a cDNA in CHO Cells. Chin J Biological.2004,17(5):290-293
9. **Zheng Lishu**, Fang Zhaoyin. Rotavirus Vaccine: present and future. Foreign Medical Science. 2003,10(1):1-4 (review)
10. **Zheng Lishu**, Jin Yihe, Jin Cuihong, et al. Experimental Study of BPA and β -HCH on Estrogenic Activity of Mice. CHINAPUBLIC HEALTH. 2002,18(8):922-924
11. **Zheng Lishu**, Jin Yihe, Zhang Yinghua. The Effects of Nanoparticles TiO₂ and 17 β -E₂ on Uterine Organ Coefficient and POD Activity in Mice. Chinese J Ind Med. 2002, 15 (2): 83-84
12. **Zheng Lishu**, Tong Zhili, Zhang Qing, et al. Analysis of Relationship Between Rotavirus G Serotype and Diarrhea Severity in Children. Chinese Journal of Disease Control and Prevention. 2002, 6 (2): 149-150
13. Lu Xiaobo, **Zheng Lishu**, Lv Xiangzhneg, et al. Effect on Bcl-2、Fas Protein Expression in Hippocampus of Rat Offspring Exposed to Lead. CHINAPUBLIC HEALTH. 2002,18(3):291-292
14. Lu Xiaobo, Lv Xiangzhneg, **Zheng Lishu**, et al. The experimental study on apoptosis of Hippocampus and Cortex Cells in Rat Offsprings Exposed to Lead. Chinese J Ind Med. 2002,15(1):19-21
15. **Zheng Lishu**, Zhang Qing, Xie Huaping, et al. Investigation of Rotavirus Diarrhea Among Children in Lulong County 1999-2000. Chin J Pediatr. 2002,40(9):555

16. **Zheng Lishu**, Zhang Qing, Tang Jingyu. The Relationship between Breeding Methods and Rotavirus Diarrhea among Children under 1 Year Old. *Chinese J Exp Clin Virol.* 2002,16(1):68
17. Jin Cuihong, Lv Xiangzheng, **Zheng Lishu**, et al. The Determination and Analysis of the Levels of Blood and Bone Lead in 32 Children. *J Chin Med Univ.* 2001,30(6):442-446.