南方科技大学 SOUTHERN UNIVERSITY OF SCIENCE AND TECHNOLOGY

课程详述

COURSE SPECIFICATION

以下课程信息可能根据实际授课需要或在课程检讨之后产生变动。如对课程有任何疑问，请联系授课教师。

The course information as follows may be subject to change, either during the session because of unforeseen circumstances, or following review of the course at the end of the session. Queries about the course should be directed to the course instructor.

1.	**课程名称 Course Title**	免疫学实验 Immunology Laboratory
2.	**授课院系 Originating Department**	医学院 School of Medicine
3.	**课程编号 Course Code**	MED327
4.	**课程学分 Credit Value**	2
5.	**课程类别 Course Type**	专业选修课/ Major Elective Courses
6.	**授课学期 Semester**	秋季/ Fall
7.	**授课语言 Teaching Language**	英文 English
8.	**授课教师、所属学系、联系方式（如属团队授课，请列明其他授课教师） Instructor(s), Affiliation& Contact （For team teaching, please list all instructors）**	刘依林，医学院，liuyl3@sustch.edu.cn Yilin Liu, School of Medicine, liuyl3@sustch.edu.cn
9.	**实验员/助教、所属学系、联系方式 Tutor/TA(s), Contact**	张婷，医学院，zhangt1@mail.sustech.edu.cn Ting Zhang, School of Medicine, zhangt1@mail.sustech.edu.cn
10.	**选课人数限额(可不填) Maximum Enrolment （Optional）**	

11. 授课方式 Delivery Method	讲授 Lectures	习题/辅导/讨论 Tutorials	实验/实习 Lab/Practical	其它(请具体注明) Other（Please specify）	总学时 Total
学时数 Credit Hours			64		64

12. 先修课程、其它学习要求 Pre-requisites or Other Academic Requirements	MED405 医学免疫学/ Medical Immunology 或/Or BIO405 免疫学/Immunology
13. 后续课程、其它学习规划 Courses for which this course is a pre-requisite	待定/To be determined
14. 其它要求修读本课程的学系 Cross-listing Dept.	待定/To be determined

教学大纲及教学日历 SYLLABUS

15. 教学目标 Course Objectives

本实验课是对《医学免疫学》（或《免疫学》）理论课程的补充，通过实验操作直观观察到免疫系统以及各组分的功能，以及与疾病的关系；理论联系实际，帮助学生巩固加深对免疫学理论知识的理解，掌握免疫学方向科研及临床检测的基本方法与先进技术。此外，本实验课程引入一种自身免疫性疾病的经典动物模型为研究对象开展综合实验，从表型、组织病理学、细胞与分子病理学等多方位全面剖析该疾病的发病原理与临床表现，使学生对医学免疫学研究领域有较为深入的认知；同时规范学生的实验操作技能，提高学生的批判性思维及独立分析问题、解决问题的能力，为学生的毕业设计项目及今后免疫学方向的科学研究打下坚实的基础。

As a supplementation to the theoretical course of <Medical Immunology> or <Immunology>, this laboratory course helps the students to better understand the functions of the immune system and each immune components through direct observation and experimentation, and to know their roles in human diseases. The course also delivers basic and most advanced techniques and technologies in the field of immunology research and clinical application. In addition, by using a classic animal model of an autoimmune disease, a comprehensive project is designed to investigate the pathogenesis and clinical manifestations of the disease from multiple aspects including phenotype assessment, histopathology, cellular and molecular pathology. The course helps the students to get a sense of immunology research, improves their experimental skills, and enhances their abilities of critical thinking and troubleshooting. By completion of this course, the students will be able to move smoothly to their final year project and future research work in immunology.

16. 预达学习成果 Learning Outcomes

本实验课程完成后，学生应掌握的实验原理与方法包括：抗原抗体反应，免疫电泳，以及巨噬细胞、自然杀伤细胞、T 细胞、B 细胞等各类免疫细胞的功能检测；通过大鼠被动皮肤过敏反应观察、了解 IgE 介导的 I 型超敏反应。此外，本课程以小鼠实验性自身免疫性脑脊髓炎为研究对象设计了综合实验，通过表型、组织病理学、细胞与分子病理学等多方位检测，涉及 HE 染色、免疫荧光染色、流式细胞学、ELISA、实时定量 PCR 等多项技术，使学生了解免疫学及生物医学领域科学研究的完整思路，提高学生的综合实验能力、批判性思维和独立解决问题的能力，激发学生对免疫学进一步学习和探索的热情。

By completing this laboratory course, the students should be familiar with the principles and experiment methods of antigen-antibody interaction, immunoelectrophoresis and various functional assays on macrophages, natural killer cells, T cells and B cells. The students will observe and understand IgE-mediated type I hypersensitivity in a rat model of type

l passive cutaneous anaphylaxis. In addition, by using a mouse model of experimental autoimmune encephalomyelitis (EAE), the students will learn the clinical manifestations and pathogenesis of the disease through phenotype assessment, histopathology, cellular and molecular pathology involving a variety of techniques such as HE staining, immunofluorescent staining, flow cytometry, ELISA and real-time PCR. Based on this comprehensive project, the students will get ideas of how to conduct a scientific project in the field of immunology or biomedicine. The course also provides the students with an opportunity to improve their experimental skills, enhances their abilities of critical thinking and troubleshooting, and inspires them to further study and explore immunology.

17. **课程内容及教学日历 （如授课语言以英文为主，则课程内容介绍可以用英文；如团队教学或模块教学，教学日历须注明主讲人）**
Course Contents (in Parts/Chapters/Sections/Weeks. Please notify name of instructor for course section(s), if this is a team teaching or module course.)

Module 1: Basic Immunology Experiments

Lab 1: Course Introduction and Animal Training (4 hours)

Introduction of Immunology Laboratory course, laboratory safety guidelines, course requirement and assessment criteria; Training of animal (mouse and rat) grasping techniques and basic operations including subcutaneous injection, intraperitoneal injection, intravenous injection.

Lab 2: Antigen-Antibody Interaction and Immunoelectrophoresis (4 hours)

To learn the principles and clinical applications of antigen-antibody interaction, and to perform ABO blood typing and double agar diffusion assay; To demonstrate immunoelectrophoresis, a techniques combining double immunodiffusion and electrophoresis.

Lab 3: Macrophage Phagocytosis Assay (4 hours)

To learn the biological activity of phagocytosis; To enrich macrophages in the peritoneal fluid and to induce macrophage phagocytosis with chicken red blood cells; To obtain macrophage-enriched cells, make a smear and stain with Gimsa-Wright's staining solution; To observe the morphological changes of macrophages during the process of phagocytosis by light microscopy, and to assess phagocytic activity.

Lab 4: NK Cell Activity Assay (4 hours)

To understand the killing mechanism of NK cells; To stimulate NK cell activity by co-incubation with YAC-1 cells, and to evaluate the activity of NK cells by determining the release of lactate dehydrogenase from dead YAC-1 cells.

Lab 5: Isolation of T and B Cell using Magnetic Dynabeads (4 hours)

To learn about the principle and applications of immunomagnetic separation; To isolate T cells and B cells from mouse thymus and spleen using anti-CD3 and anti-CD19 Dynabeads, respectively; To count the numbers of the isolated cells using a hemocytometer, and to compare the abundance of T/B cells in the immune organs.

Lab 6: T Cell Activation I -- Proliferation Assay (4 hours)

To recapitulate the process of T cell activation; To observe the morphology of activated/non-activated mouse peripheral and splenic T cells stimulated with phytohemagglutinin; To assess the proliferation rate of activated T cells by MTT assay.

Lab 7: T Cell Activation II -- ELISpot Assay (4 hours)

To learn about the principle and application of ELISpot assay; To determine the level of secreted IFN-γ by activated T cells by ELISpot assay for the assessment of T cell activation.

Lab 8: Hemolytic Plaque Assay (4 hours)

To understand the principle of hemolytic plaque assay for the evaluation of antibody production by B cells and the involvement of complement-mediated cell lysis; To perform hemolytic plaque assay using mouse splenocytes, sheep RBCs and guinea pig serum, to observe hemolytic plaques and to evaluate antibody producing ability of B cells.

Lab 9: Rat Type I Passive Cutaneous Anaphylaxis (4 hours)

To recapitulate biology of hypersensitivity/allergy; To induce passive cutaneous anaphylaxis in a ovalbumin-immunized rat; To evaluate the degree of hypersensitive reaction by injecting Evan's blue via tail vein and measuring blue plaques on the rat skin.

Module 2: A Comprehensive Project Based on an Autoimmune Disease Model

Lab 10: Introduction to an EAE model, Animal Behavior Assessment and Preparation of Spinal Cord Sections (4 hours)

To introduce a mouse model of experimental autoimmune encephalomyelitis (EAE) and how this model represents a human disease -- multiple sclerosis; To demonstrate the method of EAE induction in mouse; To assess the severity of EAE mice according to the scoring criteria of clinical signs such as limp tail and/or hind limb paralysis; To dissect mouse spinal cord, prepare cryo blocks, and make cryosections.

Lab 11: HE Staining of EAE Spinal Cord Sections (4 hours)

To introduce the principle of HE staining and the histopathological characteristics of EAE; To perform HE staining on the cryosections prepared in the last laboratory, and to observe the stained sections by light microscopy.

Lab 12: Immunofluorescent Staining (4 hours)

To learn the principle and application of immunofluorescent staining; To stain myelin basic protein and Iba-1 (a microglial activation marker) on the sections prepared in Lab 10 using respective primary antibodies and fluorophore-labeled secondary antibodies; To learn how to use an inverted fluorescent microscope and to observe the staining outcomes.

Lab 13: Flow Cytometric Analysis of Different T Cell Subsets in EAE (4 hours)

To introduce the working principle of flow cytometer, and to recapitulate different T cell subsets; To isolate single cells from fresh spinal cord of an EAE mouse, and to stain for CD4+IL-17+ (Th17) or CD4+IFN-γ+ (Th1) cells, followed by flow cytometry; To learn how to set gates, make compensation and analyze flow cytometry data.

Lab 14: Evaluation of Cytokine Production by ELISA (4 hours)

To learn the principle of and application of ELISA assay, and to introduce the features of cytokine production during EAE development; To obtain serum and spinal cord homogenates from an EAE mouse and to perform an ELISA assay in order to determine the levels of IL-17 and IFN-γ.

Lab 15: Gene Expression Analysis by Real-Time PCR (4 hours)

To be familiar with the principle and data analyzing method of real-time PCR; To extract total RNA from EAE mouse spinal cord, and to perform real-time PCR for identification of altered gene expression in EAE.

Lab 16: Review and Presentation (4 hours)

The students will do group presentations on the experiment design, data interpretation and future work plan of the EAE project.

Section	Topic	Hours
1	Course Introduction and Animal Training	4

2	Antigen-Antibody Interaction and Immunoelectrophoresis	4
3	Macrophage Phagocytosis Assay	4
4	NK Cell Activity Assay	4
5	Isolation of T and B Cell using Magnetic Dynabeads	4
6	T Cell Activation I -- Proliferation Assay	4
7	T Cell Activation II -- ELISpot Assay	4
8	Hemolytic Plaque Assay	4
9	Rat Type I Passive Cutaneous Anaphylaxis	4
10	Introduction to an EAE model, Animal Behavior Assessment and Preparation of Spinal Cord Sections	4
11	HE Staining of EAE Spinal Cord Sections	4
12	Immunofluorescent Staining	4
13	Flow Cytometric Analysis of Different T Cell Subsets in EAE	4
14	Evaluation of Cytokine Production by ELISA	4
15	Gene Expression Analysis by Real-Time PCR	4
16	Review and Presentation	4

教材及其它参考资料 Textbook and Supplementary Readings

教材：

1. Leukocytes: Methods and Protocols, Robert B. Ashman, New York, Humana Press, 2012.

2. Experimental Autoimmune Encephalomyelitis in the Mouse, Stephen D. Miller, William J. Karpus, and Todd S. Davidson, Current protocols in immunology, May 2007, Vol.Chapter 15, pp.Unit 15.1.

3. 免疫学基础与病原生物学，卢芳国， 范虹， 科学出版社，2013.

课程评估 ASSESSMENT

19.	评估形式	评估时间 Time	占考试总成绩百分比	违纪处罚 Penalty	备注 Notes

Type of Assessment		% of final score		
出勤 Attendance				
课堂表现 Class Performance		10		
小测验 Quiz				
课程项目 Projects		30		Lab Notebook
平时作业 Assignments		30		Lab Report
期中考试 Mid-Term Test				
期末考试 Final Exam				
期末报告 Final Presentation		30		Group Oral Presentation
其它（可根据需要改写以上评估方式） Others (The above may be modified as necessary)				

20. 记分方式 **GRADING SYSTEM**

☑ A. 十三级等级制 **Letter Grading**
☐ B. 二级记分制（通过/不通过） **Pass/Fail Grading**

课程审批 REVIEW AND APPROVAL

21. **本课程设置已经过以下责任人/委员会审议通过**
This Course has been approved by the following person or committee of authority

本课程已经医学院教学副院长张文勇教授审核通过。