

全国高等学校教材



细胞生物学 实验教程（双语版）

主 审 李家大 夏赞贤

主 编 言惠文

副主编 叶 茂 武晶晶 周叶方 张树冰



人民卫生出版社
PEOPLE'S MEDICAL PUBLISHING HOUSE

全国高等学校教材

细胞生物学 实验教程

(双语版)

主 审 李家大 夏赞贤

主 编 言惠文

副主编 叶 茂 武晶晶 周叶方 张树冰

编 者 (按姓氏笔画排序)

文斗斗 (中南大学生命科学学院)

武晶晶 (中国医科大学基础医学院)

叶 茂 (湖南大学生物学院)

林建中 (湖南大学生物学院)

刘慕君 (中南大学生命科学学院)

周叶方 (中南大学生命科学学院)

孙曙明 (中南大学生命科学学院)

夏赞贤 (中南大学生命科学学院)

李宇晟 (中南大学湘雅医院)

唐璐璐 (中南大学生命科学学院)

李家大 (中南大学生命科学学院)

黄明敏 (湖南大学生物学院)

言惠文 (中南大学生命科学学院)

颜金鹏 (中南大学生命科学学院)

张树冰 (中南大学生命科学学院)

人民卫生出版社

图书在版编目(CIP)数据

细胞生物学实验教程:双语版:汉英对照/言惠文主编.
—北京:人民卫生出版社,2017
ISBN 978-7-117-25475-5

I. ①细… II. ①言… III. ①细胞生物学-实验-高等学校-教材-汉、英 IV. ①Q2-33

中国版本图书馆 CIP 数据核字(2017)第 276966 号

人卫智网	www. ipmph. com	医学教育、学术、考试、健康, 购书智慧智能综合服务平台
人卫官网	www. pmph. com	人卫官方资讯发布平台

版权所有,侵权必究!

细胞生物学实验教程(双语版)

主 编:言惠文

出版发行:人民卫生出版社(中继线 010-59780011)

地 址:北京市朝阳区潘家园南里 19 号

邮 编:100021

E - mail: pmph @ pmph. com

购书热线:010-59787592 010-59787584 010-65264830

印 刷:三河市潮河印业有限公司

经 销:新华书店

开 本:787×1092 1/16 印张:9 插页:2

字 数:225 千字

版 次:2018 年 1 月第 1 版 2018 年 1 月第 1 版第 1 次印刷

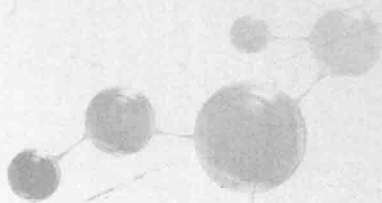
标准书号:ISBN 978-7-117-25475-5/R · 25476

定 价:32.00 元

打击盗版举报电话:010-59787491 E-mail: WQ @ pmph. com

(凡属印装质量问题请与本社市场营销中心联系退换)

前言



随着高校国际化交流的日趋频繁,专业课进行双语教学已成为了我国高校教育教学改革的一个重要内容。细胞生物学实验是临床医学各专业及生命科学相关专业的重要公共基础课程,对细胞生物学基本实验技能的掌握将奠定学生从事生命科学研究的重要基础,而目前“细胞生物学实验”双语教材比较缺乏,针对这一状况,作为从事该课程教学的一线教师,我们认为有义务在双语教材上有些作为,这即是我们出版这本教材的初衷。本细胞生物学实验教程(双语版)的编审作者全部为从事该课程教学的一线教师,100%的编写作者具有博士学位,73%的编写作者有海外留学经历,从而为本双语教材的质量保证奠定了较好的基础。

该教材的特点主要体现在两个方面,一是该教材的图片绝大部分为自己原创拍摄或制作,教师在编写过程中进行了广泛的文献、书籍及相关资料的查阅,特别注重增编了一些适宜本科学学生课外实践和学习的实用技术,为高校相关教学工作的开展和提质奠定了基础;另一方面,该教材为双语版,可方便学生在进行专业理论和技能学习的同时,又能进行专业英语的学习,为增强学生的国际交流能力奠定了基础,也使留学生能够更快更容易适应我们的教学。

该教材共编排有24个实验,其中实验1~3的内容与显微镜技术相关,实验4~5内容与细胞膜相关,实验5~6内容与细胞质相关,实验7~11内容与细胞核相关,实验12~13内容与细胞周期相关,实验14~18内容与细胞培养技术相关,实验19~24内容与分子生物学技术相关,各院校可结合本校实际情况进行选用,也可为学生从事课外科研创新活动提供较好参考和指导。

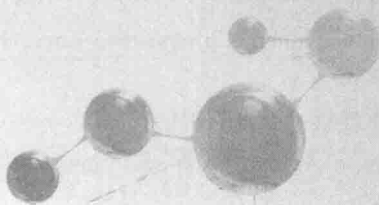
在该教材项目的立项和编写过程中,得到了中南大学、出版社、同仁和学生的支持,中南大学给予了该教材精品教材项目支持,遗传杂志社授权使用了减数分裂不同时期图片,中南大学的邬玲仟教授特别提供了人类染色体核型分析相关照片,刘水平等老师提供了拍照需要的相关装片,研究生潘艺文、赵铭日两位同学协助老师做了大量资料查找、基础编写和编辑工作,本科生陈镜颖参与了化学结构式的编辑,还有其他领导、老师和同学都给予了无私的帮助和关心,在此我们一并表示最诚挚的谢意!

虽然编者一直在努力将本教材做好做精,但也难免有疏漏和不当之处,希望读者多多包涵,并在使用过程中发现问题后及时反馈您的宝贵意见,以便我们在未来能够将本教材做得更好、更加完善。

编者

2017年9月

目 录



实验一 光学显微镜的结构和使用	1
Experiment 1 The Structure and the Usage of an Optical Microscope	5
实验二 细胞形态和细胞器的观察	10
Experiment 2 Observation of Cell Morphology and Organelles	13
实验三 显微测量	16
Experiment 3 Microscopic Measurement	18
实验四 细胞膜的通透性	20
Experiment 4 Permeability of Cell Membrane	22
实验五 聚乙二醇(PEG)诱导的细胞融合	25
Experiment 5 Cell Fusion Induced by Polyethylene Glycol(PEG)	27
实验六 线粒体的活体染色	29
Experiment 6 Supravital Staining of Mitochondria	31
实验七 动植物细胞骨架的制备和观察	33
Experiment 7 The Preparation and Observation of Cytoskeleton of Animal Cells and Plant Cells	35
实验八 Feulgen 染色法测定 DNA	38
Experiment 8 Identify DNA by Feulgen Staining	40
实验九 毛囊细胞核仁组织区银染	43
Experiment 9 Silver Staining Technique for Nucleolus Organizer Region of Hair Follicle Cells	45
实验十 人类染色体标本制备	48
Experiment 10 Human Chromosome Preparation	50

实验十一 人类染色体常规核型分析	52
Experiment 11 Analysis of Conventional Human Chromosome Karyotype	55
实验十二 动、植物细胞有丝分裂的观察	59
Experiment 12 Observation of Mitosis in Animal Cells and Plant Cells	61
实验十三 蝗虫精巢减数分裂的观察	64
Experiment 13 Observation of Meiosis in the Grasshopper Testis	67
实验十四 动物细胞原代培养	71
Experiment 14 Primary Culture of Animal Cells	73
实验十五 细胞的传代培养	75
Experiment 15 Cell Subculture	77
实验十六 细胞计数和死活细胞的鉴定	80
Experiment 16 Cell Counting and Identification of Dead and Alive Cells	83
实验十七 MTT 和 CCK-8 检测法	86
Experiment 17 MTT and CCK-8 Detection	89
实验十八 植物愈伤组织的诱导	93
Experiment 18 Induction of Plant Callus	96
实验十九 TRIzol 法提取 RNA 和 DNA	100
Experiment 19 RNA and DNA Extraction Using TRIzol Reagent	103
实验二十 琼脂糖电泳法鉴定 DNA	106
Experiment 20 Identify DNA with the Method of Agarose Gel Electrophoresis	108
实验二十一 聚合酶链式反应(PCR)	110
Experiment 21 Polymerase Chain Reaction(PCR)	113
实验二十二 脂质体介导转染法	116
Experiment 22 Liposome-mediated Transfection	118
实验二十三 DNA 重组技术	120
Experiment 23 DNA Recombination Technology	125
实验二十四 蛋白质聚丙烯酰胺凝胶电泳及 western blot 分析	130
Experiment 24 Polyacrylamide Gel Electrophoresis And Western Blot for Proteins	134

实验一

光学显微镜的结构和使用

《实验目的》

1. 辨识光学显微镜的各部分结构并熟悉其性能。
2. 学习光学显微镜的使用方法。

《实验准备》

1. 器材 普通光学显微镜。
2. 其他 毛发交叉装片、镜纸、吸水纸、香柏油、镜头清洗剂(酒精:乙醚=3:2)。

《实验原理》

显微镜的最重要的功能部件是物镜和目镜,物镜和目镜相当于凸透镜,被检标本放在物镜下方,光线通过标本后可在物镜的上方形成一个放大的倒立实像,目镜透镜将这实像放大并形成虚像,通过调焦观察,眼睛可以看到这虚拟的图像(图 1-1)。样品的总放大倍数可运用下列公式进行计算:物镜放大倍数 $\times$ 目镜放大倍数=总放大倍数。

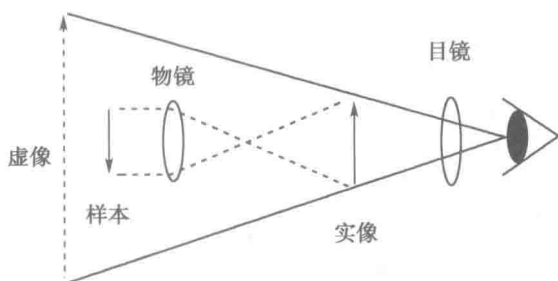


图 1-1 显微镜放大原理示意图

《实验步骤》

1. 认识显微镜的基本结构 显微镜的结构由三个部分组成,即机械部分、光学部分和照明部分(图 1-2)。

(1) 机械部分:

镜座:位于最底部,是显微镜的基座,用以稳定显微镜,装有照明光源或反光镜。

镜臂:为弯弧形,是使用者取用显微镜时握持的部位。

镜柱:是连接镜臂与镜座的短柱,具支持显微镜的作用。

镜筒:连于镜臂,其上端装有目镜,下端装有物镜转换器。目镜的镜筒有单筒和双筒两种,双筒之间的距离可以调节。

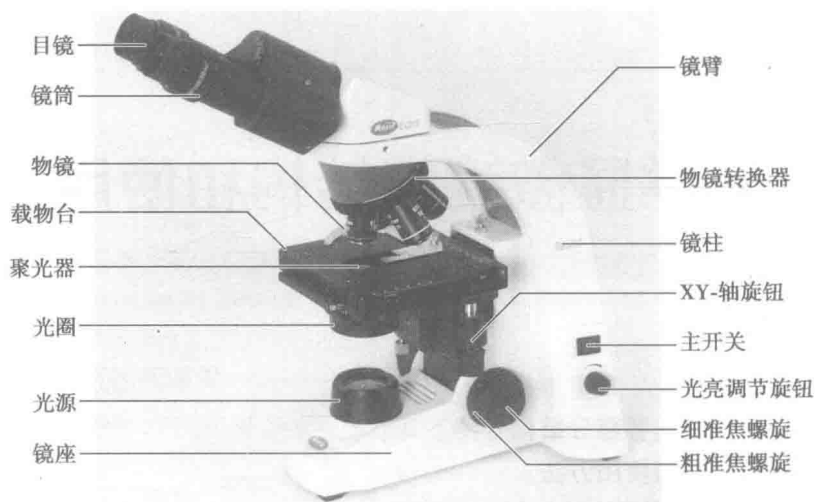


图 1-2 普通光学显微镜

粗(细)准焦螺旋:位于镜臂的上端(直立式镜筒)或下端(倾斜式镜筒),可调节标本与物镜之间的距离。粗准焦旋钮是大旋钮,它可以帮助我们迅速调节镜头和样品之间的距离,通常在低倍镜观察时使用。细准焦旋钮是小螺旋,用于高倍镜或油镜观察时的精确调焦。

物镜转换器:物镜转换器是一个凹形的圆盘,有 3~4 个物镜螺旋口,可以安装不同放大倍数的物镜。旋转物镜转换器时,即可更换物镜。

载物台:载物台位于镜臂前方,为方形或圆形的平台,用于安放载玻片,其中央有圆形的通光孔。在载物台上装有压片夹,通过 XY-轴旋钮可调节样本装片前后左右移动,使我们更容易找到目标。在压片夹上附着有纵、横游标尺,用以确定样本位置。

(2) 光学部分:

目镜:安装在镜筒的上端,每台显微镜通常配置 2~3 个不同放大倍数的目镜,最常使用的是“10×”(数字表示放大倍数)的目镜,目镜可安放指针。

物镜:安装在物镜转换器上,是显微镜最主要的光学部件,每台显微镜通常配置 3~4 个不同放大倍数的物镜,各物镜的长短不同,一般是越短的物镜,其放大率越低;同样,越长的物镜,其放大率越高。“4×”或“10×”者为低倍镜,“40×”者为高倍镜,“100×”者为油镜。另外,显微镜的物镜上还刻有镜口率,如 0.25 等,数字越大,其分辨率越高。

(3) 照明部分:

聚光器:聚光器位于镜台的下面,由 2~3 个透镜组合而成,其作用是将光线汇集成束,集中到所要观察的样本上。聚光器的一侧有一螺旋,可升降聚光器以调节光线之强弱。

光圈:位于聚光器的下端,可调控进入聚光器光束的大小。其外侧有一可移动小柄,可以根据需要开大或关小以调节光量。

反光镜或光源:反光镜位于聚光镜的下方,可向各方向转动,其中一面为平面镜,另一面为凹面镜。凹面镜聚光作用强,在光线较弱时使用;平面镜只有反射作用,在光线较强时选用。现在很多显微镜用照明光源代替反光镜,可以通过光亮调节旋钮进行亮度调节。

2. 显微镜的使用方法 以右手握住镜臂,左手托住镜座的方式将显微镜从镜箱内取

出,轻轻地平放在实验台上,检查各部件完整后继续操作。使用前用纱布清洁机械部分,用镜纸以螺旋方式从里向外清洁镜头部位。

(1) 低倍镜的使用:

对光:开灯,旋转粗准焦旋钮,使载物台下降到适宜位置;旋转物镜转换器,使低倍镜对准载物台中央的通光孔,打开光圈并使聚光器上升到适当位置,调节反光镜或调光旋钮,使视野中的光线亮度达到最适宜状态,观察过程中应注意两眼同时睁开。

置片:将毛发交叉玻片(盖玻片朝上)置放于载物台,用压片夹固定位置,并将要观察的毛发交叉部位移至载物台中央的通光孔上。

调焦:从侧面注视低倍镜镜头,旋转粗准焦旋钮,让镜筒缓慢下降或载物台缓慢上升,至低倍镜镜头将接近标本玻片为止(注意两者不能相碰,以免损坏镜头或装片),然后通过目镜观察,转动粗准焦旋钮(方向不可转反),使物镜与装片的距离慢慢远离,直到视野中出现可见物象为止,使用细准焦旋钮使物像调至清晰,用移动推片器将毛发交叉点移至视野中央。若调焦过程中,镜头与玻片的距离超过1cm还未见到物像,则按上述步骤重新操作。

(2) 高倍镜的使用:

1) 先用低倍镜观察视野,找到毛发交叉,移至视野中央并调焦直至清晰

2) 然后换用高倍镜进行局部观察。通过目镜观察视野,此时只能使用细准焦旋钮进行微调,直至看到清晰的物像出现为止。

3) 有时从低倍镜换用高倍镜时,会发生高倍物镜碰触到玻片而无法转换到位的情况,此时不能强硬转动,应检查是否玻片放反、是否玻片过厚以及物镜是否松动等情况。排除问题后再重新进行转换。

(3) 油镜的使用:

1) 在高倍镜下将需要进一步放大的目标物像移至视野中央,由于油镜所需光线较强,故往往光圈开至最大,将光线调至最合适强度。

2) 移开高倍镜,在标本玻片上滴一滴香柏油作为介质,缓慢转换油镜,使镜头浸入油滴中,使用细准焦旋钮进行上下微调,即可看清物像。

3) 如果需要重新操作,切不可直接转用非油镜物镜,以免这些物镜粘上油滴。此时应用擦镜纸擦拭去除标本玻片上的油,然后用擦镜纸蘸少许镜头清洗剂轻擦,再用干净的镜纸擦拭洁净后,遵循从低倍镜、高倍镜、油镜的顺序进行重新操作和观察。

4) 油镜使用完毕后,调升镜头并转至一边,用镜纸洁净镜头和标本玻片。

5) 如果是无盖玻片的标本,则可采用拉纸法去除载玻片上的油,即先用一小片镜纸盖在油滴上,再往纸上滴几滴镜头清洗剂,趁湿将纸往外拉,如此反复几次即可将玻片上的油去除。

6) 当你使用完毕,清除镜头、载物台等上的油污。

《《注意事项》》

1. 必须双手取用显微镜,一手紧握镜臂,一手平托镜座,轻柔平稳安放。
2. 使用时须按步骤小心缓慢操作,不要让镜头触碰到盖玻片或装片。
3. 使用高倍镜观察液体标本时,一定要加盖玻片,以免高倍镜受到污染和腐蚀。
4. 显微镜应放在清洁、干燥、无震动、无腐蚀性气体存在的房间。

5. 保持清洁,勿使污物、尘埃、手指等接触光学部分。
6. 镜内零件不得任意拆出,或与他镜调换。
7. 当你使用完毕后,抬高物镜或降低载物台,移除装片。一定要用擦镜纸对光学表面部分进行彻底清洁。
8. 使用后将物镜返回至低倍镜,并将显微镜放回其存放位置(盒子里)。

《《思考题》》

1. 如果装片倒置放置于载物台上进行显微观察,会发生什么后果?
2. 为什么用低倍镜观察时要调弱光线?

(言惠文 夏赞贤)

Experiment 1

The Structure and the Usage of an Optical Microscope

《《 Objectives 》》

1. Identify the parts of an optical microscope and be familiar with their functions.
2. To learn how to use an optical microscope.

《《 Preparation 》》

1. **Apparatus** Ordinary optical microscope.
2. **Others** Hair-crossed slide, lens paper, bibulous paper, cedar oil, lens-cleaning solution (alcohol : ethylether = 3 : 2).

《《 Theory 》》

The most important functional parts of a microscope are objectives and eyepieces. The objectives and eyepieces of an optical microscope are equivalent to the convex lens. When the samples are placed below the objective lens, as the light is transmitted through a specimen, an inverted and magnified real image are formed above the objective lens. This real image which is magnified by ocular lens and then a virtual image is formed. The virtual image can be seen by your eyes through your focusing and observation (Figure 1-1). Total magnification of the specimen can be calculated by the following formula: Magnification by the Objective Lens $\times$ Magnification by the Ocular Lens = Total Magnification

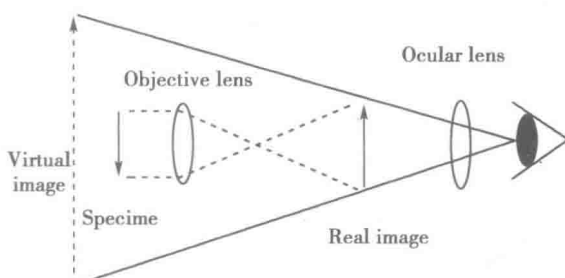


Figure 1-1 Schematic diagram of microscope magnifying principle

《《 Procedures 》》

1. **Identify the structure of an optical microscope** An optical microscope is composed of three groups: the mechanical parts, the lighting parts and the optical parts (Figure 1-2).

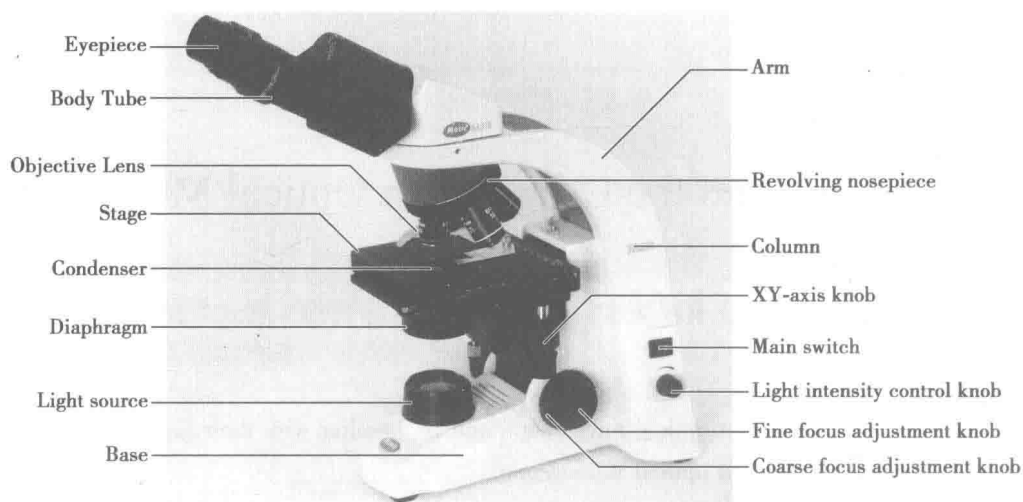


Figure 1-2 Ordinary optical microscope

(1) The mechanical parts:

Base: Located at the bottom of a microscope. It is equipped with a light source or a reflector and used to stabilize the microscope.

Arm: Shape like arc. Users are supposed to grasp the arm to hold a microscope.

Column: Attached to the base and arm. It supports the microscope.

Body Tube: Attached to the arm. It is equipped with eyepieces on the top and revolving nose-piece at the bottom. It is classified into monocular or binocular, the distance between the two tubes of a binocular can be adjusted.

Coarse (fine)-focus adjustment knob: It is located above (vertical tube) or below (inclined tube) the arm, which is designed to adjust the distance between the specimen and the objective lens. Coarse-focus adjustment knob is the larger knob, which can rapidly help adjust the distance between the lens and the specimen-for focusing, mostly using under the low-powered lens. Fine-focus adjustment knob is the smaller knob for precise focusing under the high-powered lens or oil-immersion lens.

Revolving Nosepiece: It is a concave disk with 3 or 4 spiral ports of objective lens. The objectives of different magnifications can be switched by rotating the revolving nosepiece.

Stage: Located in front of the arm. It is a square or a circular platform designed to place the slide. There is a circular aperture in the center. The specimen holder is installed on the stage. Slide specimen can be moved around through adjusting the XY-axis knob so that the target can be located easily. Vertical and horizontal vernier caliper is attached to the specimen holder to determine the position of the specimen.

(2) The optical parts:

Eyepiece: It is installed on the upper end of the body tube. There are 2 or 3 different magnification eyepieces and the “10×” (Figures show the magnification) is the most frequently used. Pointer can be placed in the eyepiece.

Objective lenses: As the most important component of a microscope, they are installed on the

revolving nosepiece. Each microscope is usually equipped with 3 to 4 different magnification of the objectives. The length of the objectives is different. Generally, the shorter objectives provide lower magnification, and the longer ones have higher magnification. The tubes with low-power lens are engraved with "4×" or "10×". The tube with high-power lens is engraved with "40×". The tube with oil-immersion lens is engraved with "100×". In addition, the tube is also engraved with "0.25" representing Numerical Aperture (NA in short). The bigger the number is, the higher resolution will be.

(3) The lighting parts:

Condenser: It is under the stage and consists of two or three lenses which can gather the light to the specimens. There is a knob on the side of the condenser. We can control the intensity of the light by adjusting the position of the condenser.

Diaphragm: It is beneath the condenser and capable of adjusting the size of the light beam. Its outside movable small handle is the controlling part which regulates the amount of light as required.

Reflector or Light Source: Below the condenser, there is reflector which can rotate in all direction. One of its sides is plane and the other is concave. The focusing effect of the concave mirror is strong and it is used under the weak light condition usually. The plane mirror possesses reflection effect and it is often chosen under stronger light condition. Many microscopes use a light source instead of reflector, which the brightness can be adjusted by the light intensity control knob.

2. The usage of an optical microscope You should grab the arm of a microscope with your right hand and support the base with left hand, and place it gently on the work bench. Then you could continue your work after checking each part of the microscope. You should clean the mechanical parts with gauze and clean the lens with lens paper in a spiral motion starting at the center of the lens and working outwards before using.

(1) The usage of low-power lens:

Adjusting light: Turn on the light. Revolve coarse adjustment knob and let the stage down slightly to the proper position. Align the low-power lens with the center hole of the stage by rotating the revolving nosepiece. Open the diaphragm, raise the condenser to the proper height and adjust the reflector or the light regulating knob for optimum light. You should open your both eyes during the observation process.

Placing the slide: Place a hair-cross slide (the side with glass upward) on the stage and clamp it with stage clips. Then center the hair-cross slide over the aperture of the stage.

Focusing: Observe from the side, revolve coarse-focus adjustment knob to let the low-power lens down, or raise the stage up slowly until the low-power lens is close to the slide (do not let these two touch, lest damage the lens or slide). Then look through the eyepiece, revolve coarse-focus adjustment knob (notice the direction of rotation) and let the objectives and the slide far away slowly until a visible image appears. Use the fine-focus adjustment knob to get a clear image and let the hair-cross point at the center of view field. During your focusing, if you could not see the image when the distance between the objective and a slide is more than 1 cm, you may do it again according to the steps above.

(2) The usage of high-power lens:

1) First, observe with low-power lens. Find the hair cross from the view field. Let the hair-cross in the center of your view field and get focusing.

2) Then switch to the high-power lens for local observation. Observe the view field through eyepiece. In this step, only the fine-focus adjustment knob can be used for focusing.

3) Sometimes when switching the low-power lens to high-power lens, the high-power objective lens will touch the specimen and cannot move to the right position. You are not allowed to switch violently in this situation. You should check whether the slide is placed inversion, or the slide is too thick, or the objective lens is loose. Switch it again after excluding these problems.

(3) The usage of oil-immersion lens:

1) Move the image which needs to be magnified further to the center of the view field under high-power lens. Owing to the stronger light required under oil-immersion lens, you should open the diaphragm to its maximum and adjust the light to appropriate intensity.

2) Once get a clear image under the high-power lens, move the lens away and drip a drop of cedar oil on the slide, switch the oil-immersion lens slowly so that the lens is immersed into the oil, and obtain your clear image by rotating fine-focus adjustment knob slightly.

3) If you need to re-operate, you should not use non-oil lens objective directly lest the objectives sticking with oil. In this situation, you may use lens paper to wipe oil on the slide, and dip a little lens-cleaning solution on the lens paper to wipe the slide slightly, then use another clean lens paper to wipe it again. Finally, you may re-operate and observe by following the procedure of using low-power lens, high-power lens and oil-immersion lens.

4) After using oil-immersion lens, you should raise and rotate the revolving nosepiece to let the lens go aside. Use the lens paper to clean the lens and slide.

5) If the specimen is with no coverslip, you may remove the oil of the specimen by the pulling-paper method, that is, covering the oil with a small piece of lens paper at first, dropping lens-cleaning solution on the paper and then pulling out the paper immediately. Repeat several times to remove the oil completely.

6) When finished, clean the oil off the lenses, stage, etc.

《《Precautions》》

1. Carry the microscope with your both hands, one hand grasping the arm of the microscope and another supporting its base. Gently place the microscope and keep balance without any tremors.

2. Following the procedure step by step carefully and slowly. Never allow the lens to touch the coverslip or the slide.

3. When using high-power lens to observe the liquid specimen, you must use coverslip to prevent the lens from being polluted and corroded.

4. The microscope should be placed in a clean, dry room with no corrosive gases.

5. Keep clean and do not let dirt, dust or fingers contact the optical components.

6. The components of optical microscope shall not be removed arbitrarily or be replaced with

other objectives.

7. When finished, raise the objective or lower the stage, remove the slide. Be sure to use lens paper cleaning the optical surface of the microscope thoroughly.

8. Rotate to low-power lens after use. Return the microscope to its storage place (inside a box).

《《 Questions 》》

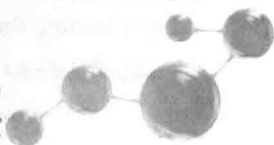
1. What will happen when an inverted slide is placed on the stage during your microscopic observation?

2. Why should you reduce the light when using low-power lens?

(Yan Huiwen Xia Zanzian)

实验二

细胞形态和细胞器的观察



《实验目的》

1. 掌握光镜下细胞的形态特点。
2. 观察细胞器的基本形态和结构。
3. 学习生物绘图的方法。

《实验准备》

1. 器材 普通光学显微镜。
2. 材料 镜纸、吸水纸、香柏油、镜头清洗剂(酒精和乙醚的混合物)。
3. 用于形态观察的装片 兔卵巢切片,人精涂片,蛙(或蝾螈)表皮细胞,平滑肌细胞,神经细胞分离装片。
4. 用于细胞器观察的装片 猫的脊神经节细胞,大白鼠胰腺细胞,马蛔虫子宫装片。

《实验原理》

细胞是生物体的基本结构单位。细胞有两种类型,真核细胞和原核细胞,真核细胞包含细胞核而原核细胞不含核。真核细胞的基本结构包括细胞膜、细胞质、细胞核和细胞壁(植物)。细胞形态多样,原核细胞比真核细胞更小、更简单。真核细胞和原核细胞都有细胞器,但原核细胞的细胞器通常是更简单且无膜包被。真核细胞中有各种细胞器,如线粒体、高尔基复合体、中心体等。经过细胞固定及特异染色处理后,可在光学显微镜下观察到细胞形态及其细胞器。

《实验步骤》

1. 细胞形态的观察

(1) 圆形(球形)细胞:首先在低倍镜下观察兔卵巢切片标本。在视野中可以看到,卵巢皮质(卵巢的外周部分)有一些圆形(球形)的,呈淡红色的卵细胞(见书末彩图 2-1)。这些细胞比普通细胞大,是处于不同发育时期的卵泡,如原始卵泡,初级卵泡,次级卵泡和成熟卵泡。原始卵泡由颗粒细胞包围卵母细胞形成,当颗粒细胞达到一层以上便形成初级卵泡,次级卵泡透明带明显,成熟卵泡的卵泡腔很大,卵丘很明显。在高倍镜下观察,可以看见卵细胞的外周有一圈红色的透明带,在透明带的外周围绕有放射冠。

(2) 蝌蚪形细胞:将人精子涂片置于低倍镜下观察,视野中看到人精子为蓝色的小点,其形状很难看清。转换至高倍镜观察,可以看到细胞形状呈蝌蚪形(见书末彩图 2-2),细胞头部椭圆形且着色较深,内有细胞核,细长的部分为尾部。

(3) **多边形细胞**:在低倍镜下观察蛛螯(或蛙)表皮装片(见书末彩图 2-3),你会看到上皮细胞就像墙上的砖样紧密相连。细胞核呈深染的圆形或椭圆形结构,细胞质围绕细胞核。细胞膜深染,分隔各个细胞。

(4) **梭形细胞**:以平滑肌细胞示例(见书末彩图 2-4)。多数平滑肌细胞由呈长梭形或长纺锤形的单核细胞构成,相互交错,密集排列。核位于细胞中央,呈杆状或长椭圆形。在低倍镜下观察平滑肌分离装片标本,视野中可见平滑肌细胞染成红色。

(5) **星形细胞**:在低倍镜下观察(见书末彩图 2-5),可以看到神经细胞细胞呈星形,深蓝色,从胞体发出轴突。

2. 细胞器的观察

(1) **高尔基复合体**:光镜下高尔基复合体是位于细胞核附近的一些网状结构,用浸银法易于显示。细胞经过镀银法染色后,细胞质和高尔基复合体着色,细胞核不着色。

取猫的脊神经节细胞切片标本,观察高尔基复合体(见书末彩图 2-6)。低倍镜下,视野中可以观察到许多黄色的神经细胞,这些细胞呈圆形或不规则形状,界限明显。在视野中找出轮廓清晰的目标部位,转换至高倍镜进行观察,可以看到细胞核为无色区,周围细胞质被染成黄色。有些细胞核中可以看到黄色的核仁。在细胞核的周围,有一些被染成黄褐色或棕黑色的颗粒状、斑块状及卷曲的线状结构。它们共同组成细网状的结构,分布在胞质中,这就是高尔基复合体。有时候在视野中会看到某些细胞中高尔基复合体就好像散布在整个细胞中,这可能是没有经过细胞核的切面。

(2) **线粒体**:线粒体是一些大小不一的球状、棒状或细丝状颗粒。因其形态可发生变化,还可呈环状、线状、哑铃状、分杈状、扁盘状或其他形状。

1) 劳弗式快蓝染色法:将大白鼠胰腺细胞(劳弗式快蓝染色)的切片放置于载物台上,在低倍镜下找到细胞,然后转至高倍镜观察(见书末彩图 2-7)。在高倍镜下可以看见线粒体被染成蓝绿色,也可以观察到被染成紫红色的酶原颗粒,在细胞中呈堆状分布。

2) 苏木精染色法:将大白鼠胰腺细胞(苏木精染色)的切片放置于载物台上,在显微镜下观察(见书末彩图 2-7),线粒体为蓝灰色的粒状或线状结构,但细胞界线模糊不清。

(3) **中心体**:将马蛔虫子宫装片放置在显微镜载物台上,在低倍镜下找到并观察处于分裂中期的细胞,然后切换到高倍镜(见书末彩图 2-8)。视野中可看到,细胞为不均质的蓝色,其中染色体和中心体染色较深,为蓝黑色。染色体位于细胞中央,为杆状,中心体位于细胞质的两极,为蓝黑色粒状。两个中心粒之间的丝状结构是纺锤丝。在视野中,有时候只能观察到细胞一极的中心体。

3. **生物绘图** 根据显微镜所观察到的细胞或细胞器的形态,进行生物绘图,生物绘图基本要求如下:

(1) 图像力求真实,准确。

(2) 图的大小、位置分配及各部分结构比例要恰当。

(3) 主要运用点和线进行生物绘图,点:主要表现非膜相结构,反映光线的明暗强弱及结构的疏密程度,点要圆润,不能拖尾;线:主要表现膜相结构,轮廓线条要均匀,不能反复涂抹。

(4) 图的下方注明图的标题,图中各结构名称用尺引出平行线,平行线末端在同一直线上,结构名称注于线的末端。