滴片，涂片制作实验报告

一、实验器材及试剂

1、实验器材

名称	厂家	型号
移液枪	大龙仪器	7010101009
吸头	Wanwu	G82010
烤箱	天津市莱玻瑞仪器设备有限公司	GFL-230
离心机	DRAGONLAB	D3024R
载玻片	Wanwu	
正置光学显微镜	日本尼康	NIKON ECLIPSE E100

2、主要实验试剂

试剂名称	厂家	货号
固定液	Wanwu	G1101

二、实验步骤

1、涂片的制作：

血涂片制作：

1.1.取新鲜血液选用石蜡用的干净的防脱玻片，用移液枪吸取 10ul 的血液，滴加在玻片的一侧；再取一片辅助玻片（也可选用盖玻片），用其宽边靠近血液，会发现血液会沿着接触边缘向两边晕开，当两边都要到达边缘时，以 45 度的角度用手轻轻的稳健且匀速的把辅助玻片推向另一侧进行涂片。

1.2 自然晾干后，用甲醇固定 15min，水洗晾干。

2.细胞悬液滴片制作:

2.1 取细胞悬液，2800rpm　4℃离心 5min，弃上清液，根据底部沉淀的细胞量加入 2ml 甲醇固定。若肉眼看不见细胞沉淀时，用 3000rpm，4℃离心 10min。

2.2 取用甲醇重悬的细胞悬液，2800rpm　25℃离心 5min，弃上清液，根据底部沉淀加入 PBS：①若肉眼看不见细胞沉淀时，用 3000rpm，25℃离心 10min，依然无沉淀时，留取底部约 0.2ml 的液体，再加入 0.5ml 的 PBS 混匀后用移液枪吸取 200ul，滴于提前用组化笔画好的小圆圈中（3cmX2cm 的椭圆）；②若细胞沉淀量很少，绿豆大小时，弃上清，留取底部沉淀，加入 0.5mlPBS,用移液枪吹打混匀后，吸取 200ul，涂于提前用组化笔画好的小圆圈中（3cmX2cm 的椭圆)；③若细胞沉淀量很多，黄豆大小或更大时，弃上清，留取底部沉淀，加入 2mlPBS, 用移液枪吹打混匀后，吸取 150ul，涂于提前用组化笔画好

的大圆圈中（最大长边约 5cm，最大短边约 2cm 的椭圆），在显微镜下观察细胞量的多少，若还是过厚，再用 PBS 对倍稀释该细胞悬液，直至在显微镜下观察到细胞量涂布均匀。

2.3 用枪将细胞悬液铺满整个圆圈，涂片放置自然晾干。

合肥万物

Experimental report of making drip and smear

1.Experimental equipment and reagents

1.1 Experimental equipment

name	factory	model
Pipette	Dalong Instruments	7010101009
Tip	Wanwu	G82010
oven	Tianjin LaiBoRui Instrument Equipment Co., Ltd.	GFL-230
Centrifuge	DragonLab	D3024R
Glass slide	Wanwu	
Upright optical microscope	Nikon Japan	Nikon Eclipse E100

1.2 Main experimental reagents

Reagent name	factory	Article number
Fixative	Wanwu	G1101

2. Experimental procedure

1. Smear production: blood smear production:

1.1 A clean anti-stripping slide with paraffin is used for fresh blood ,10 ul of blood is absorbed with a pipette gun and added to one side of the slide. A slide (or cover glass) is added to the slide. Close to the blood with its wide side, the blood will faint along the edge of contact. When both sides reach the edge, gently apply a steady and uniform hand to the other side at 45 degrees.

1.2 After natural drying, fixed with methanol for 15 min, to wash to dry.

2. Production of cell suspension droplets

2.1Take the cell suspension, centrifuge at 2800rpm and 4℃ for 5min, discard the supernatant, add 2ml of methanol according to the amount of cells precipitated at the bottom to fix it. If the cell pellet is not visible to the naked eye, centrifuge at 3000rpm and 4℃for 10min.

2.2Take methanol and resuspend The cell suspension is centrifuged at 2800rpm and 25℃for 5min. Discard the supernatant and add PBS according to the bottom pellet:

①If the cell pellet is not visible to the naked eye, centrifuge at 3000rpm and 25C for 10min. Liquid, add 0.5ml of PBS and mix well, pipette 200ul with a pipette, and drop it into a small circle (3cmX2cm oval) drawn in advance with a grouping pen;

②If the amount of cell sediment is small, when the size of mung bean is, discard Supernatant, leave bottom sediment, add 0.5ml PBS, mix with pipette gun, mix 200ul, apply to small circle (3cmX2cm oval) drawn in advance with grouping pen;

③If there is a lot of cell sediment, soybean When the size is larger, discard the supernatant, leave the bottom sediment, add 2ml PBS, mix with a pipette gun, draw 150ul, and apply it to a large circle drawn in advance with a grouping pen (maximum long side is about 5cm, maximum (The ellipse with a short side of about 2cm), observe the amount of cells under the microscope.If it is still too thick, then dilute the cell suspension with PBS until the cell amount is uniformly observed under the microscope.

3. Use a gun to spread the cell suspension over the entire circle and place the smear to dry naturally.

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