V02

精子细胞 BWW 培养基储存液

货号：G2586
规格：500mL
保存：2-8℃, 有效期 1 年。

产品介绍：

正常精液是一种混合物，在射精时由睾丸和附睾的分泌物及悬浮其中的精子与前列腺、精囊腺和尿道球腺的分泌物混合而成，最终射出的混合物是一种粘稠的液体。精子分析的方法有很多，其中可通过培养进行检测。

精子细胞 BWW 培养基储存液主要由氯化钠、氯化钾、氯化钙、硫酸镁、酚红等组成，不含葡萄糖、丙酮酸钠、BSA 等以及抗生素，是一种旨在用于广谱动物和人体精子细胞获能处理的常用营养液。

操作步骤：(*仅供参考*)

1、取干净的精液样本室温放置，使之充分液化。
2、制备精子细胞(仅供参考，不是必须步骤)：
1）上泳法：取一个无菌的 15ml 锥底离心管，加入 1ml 液化的精液，在其上方轻轻加入 1.2mlearle 形成液层，将离心管倾斜 45°，以增加精液和培养液的接触面积，37℃孵育 1 小时。轻轻将试管竖直，吸取最上层 1ml 培养液，其中包含高活力的精子。加入 8ml 增补的 Earle 培养液稀释，300～500g 离心 5 分钟，弃上清。加入 Earle 培养液 0.5ml 重新悬浮细胞，用于精子密度或功能的评估。
2）非连续密度梯度法：取一个无菌的锥底离心管，加入 Percoll。轻轻加入 3ml40%的 Percoll 于 Percoll 液面上，小心操作，不要打乱两种液体的界面。轻轻加入 1-2ml 精液于梯度溶液上，弃上清。将管底的精子团重新悬浮于的 Earle 培养液中，离心，弃上清。加入 1mlEarle 培养液，重新悬浮。
3、将含有精子细胞的离心管置于 37℃含 5%CO2、95%空气的细胞培养箱中孵育。如无上述培养箱，可将离心管密封加盖，置于 37℃的普通培养箱内孵育。在孵育过程中，大多数活动的精子从精浆中游离到覆盖上面的培养液内。
4、离心精子悬液，使精子细胞密度接近于 10×10^6/ml，将精子重悬于精子细胞 BWW 培养基中，并在 37℃含 5%CO2、95%空气的细胞培养箱内孵育。如无上述培养箱，可将离心管密封加盖，置于 37℃的普通培养箱内孵育。在孵育过程中，将试管 20°倾斜。

注意事项：

1、注意无菌操作，尽量避免污染。
2、如无 Earle 培养液和增补的 Earle 培养液，可用精子细胞 BWW 培养基代替。
3、为了您的安全和健康，请穿实验服并戴一次性手套操作。

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Sperm BWW Stock Solution

Cat: G2586
Size: 500mL
Storage: 2-8℃, valid for 1 year.

Introduction

Normal semen is a kind of mixture, which is composed of the secretion of testis and epididymis and the sperm suspended in it and the secretion of prostate gland, seminal vesicle gland and bulbar gland of urethra during ejaculation. The resulting mixture is a viscous liquid. There are many methods of sperm analysis, which can detect by culture.

Sperm BWW Stock Solution is mainly composed of sodium chloride, potassium chloride, calcium chloride, magnesium sulfate, phenol red and so on. It do not contain glucose, sodium pyruvate, BSA and antibiotics. It is a common nutrient solution for capacitative treatment of broad-spectrum animal and human sperm cells.

Protocol(*for reference only*)

1. Take a clean semen sample and place it at room temperature for 30-60min to make it fully liquefied.
2. Prepare sperm cells (*for reference only, not necessary step*):
   1) Upper stroke method: take a sterile 15ml conical bottom centrifuge tube and add 1ml liquefied semen, then gently add 1.2ml earle above the semen to form a liquid layer, tilt the centrifuge tube in 45°to increase the contact area of semen and culture solution. Incubate it at 37℃ for 1 hour. Gently erect the test tube and suck the top 1ml of culture solution, which contains highly active sperm. Add 8ml of supplemental Earle culture solution to dilute, centrifugate in 300-500g for 5 min, and discard the supernatant. Add 0.5ml Earle culture solution and resuspend the cells for the evaluation of sperm density or function.
   2) Discontinuous density gradient method: take a sterile conical bottom centrifuge tube and add Percoll, then gently add 3ml 40% Percoll onto the liquid surface, operate carefully to avoid disrupting the interface between two liquids. Gently add 1-2ml semen to the gradient solution and discard the supernatant. Resuspend the sperm mass at the bottom of the tube in Earle culture solution, centrifuge and discard the supernatant. Add 1ml Earle culture solution and resuspend.
3. Incubate the centrifuge tube with sperm cells in a cell incubator containing 5% CO_2 and 95% air at 37 ℃. If there is no such incubator, can seal and cover the centrifuge tube, then incubate it in an ordinary incubator at 37 ℃. In the process of incubation, most of the motile sperm dissociate from seminal plasma to the culture medium covered above.
4. Centrifuge the sperm suspension to make the sperm cell density close to 10×10^6/ml. Resuspend the sperm cell in BWW Culture Medium and incubate in a cell incubator containing 5% CO_2 and 95% air at 37 ℃. If there is no such incubator, can seal and cover the centrifuge tube, then incubate it in an ordinary incubator at 37 ℃. During incubation, tilt the tube in 20°.

Note

1. Pay attention to aseptic operation and try to avoid pollution.
2. If there is no Earle culture solution or supplementary Earle culture solution, can replace with Sperm BWW Culture Medium.
3. For your safety and health, please wear experimental clothes and disposable gloves.