精子获能液使用说明书 （小鼠专用）

产品	货号	规格	用途
c-TYH	72021	2mL	小鼠精子获能液

【主要组成成分】

胚胎移植用水、双抗，酚红，β-环糊精等

【储存条件及有效期】

1、保存：4℃，避光保存

2、有效期：6个月，开口后未使用的试剂，请及时用封口膜密封，并于24h内使用完毕

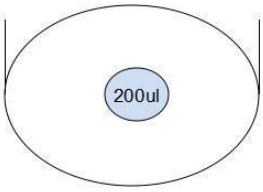
【小鼠要求】

雄鼠，建议3-6月龄，繁殖笼的雄鼠使用前至少单放3天，非繁殖笼的库存雄鼠，实验前先试配后至少单放一周。

【使用方法】

1、体外受精（新鲜精子）

1.1 在35mm皿中做一个200μL c-TYH滴，覆盖矿物油后，在培养箱中预平衡30min。



c-TYH 精子获能皿

1.2 麻醉雄鼠或脱颈椎法处死雄鼠后，打开腹腔迅速分离出两侧的附睾尾和输精管。 将附睾尾放于滤纸上尽可能去除脂肪和毛发和血渍。

1.3 用一次性1mL 注射器轻轻扎4-6下附睾尾，挤出“精子团”，用显微镊挑出“精子团”放入到之前平衡好的c-TYH中获能。将获能皿放置于培养箱中获能30min。

1.4 获能结束后，从c-TYH获能滴边缘吸取活力较好的精子，加到含有卵子的受精滴中，根据精子的密度和活力情况，加精子的量建议5-8μL左右。

2、体外受精（复苏精子）

2.1 用在35mm皿中做一个90μL c-TYH滴，覆盖矿物油后，在培养箱中预平衡30min。

2.2 冷冻麦管里地精子在37℃中水浴10min解冻精子，之后用移液器吸取解冻后的精子加入到c-TYH获能滴中。37℃、5%的CO2培养箱中获能30min。

2.3 获能结束后，从c-TYH获能滴边缘吸取活力较好的精子，加到含有卵子的受精滴中，根据精

子的密度和活力情况，加精子的量建议10μL左右。

【注意事项】

1、精子采集时避免血液、脂肪、毛发等组织进入获能滴内。

2、精子获能皿在培养箱内平衡30min。

3、吸取精子的角度需要倾向并且缓慢轻柔，避免损伤精子尾巴。

4、若遇到精子密度较稀，在不追溯亲代的情况下，可以将不同鼠的精子采集到同一受精滴。

Sperm capacitation medium (Mouse-specific)

Product	Item No.	specification	Application
c-TYH	72021	2mL	Sperm capacitation

Composition

Water for embryo transfer, Penicillin-Streptomycin, Phenol Red, β-Cyclodextrin

Storage conditions and expiration date

1. Storage Temperature: 4℃,Protect from light.

2. Expiration date: 6 months. If the reagent is not used after opening, please seal it with sealing film in time and use it within 24 hours.

Mouse

12- to 24-week-old males, male mice in breeding colonies should be left alone for at least 3 days before use, and male mice in no- breeding colonies should be left alone for at least one week after trial mating before the experiment.

Methods

1. In vitro fertilization-fresh sperm

1.1 A 35-mm culture dish with a 200 μL drop of c-TYH covered with oil was prepared and equilibrated for at least 30 min in the incubator.

1.2 Collect tail of epididymis from a male killed via cervical dislocation or anesthetized, and place them on a flter paper. Remove the fat and blood from the epididymides.

1.3 A 1 mL syringe was used to gently punctured 4-6 times in the tail of the epididymis, Squeeze tail of the epididymis by the hands and help the "sperm clumps" outflow.Transfer of sperm clumps using microforceps for placement in the c-TYH for 30min in the incubator.

1.4 After sperm capacitation, pick up sperm suspension from the edge of the dropwas added to the HTF dishes containing oocytes.

Depending on the density and viability of the sperm, it is recommended to add about

5-8 μL of sperm.

2. IVF-cryorecovered sperm

2.1 A 35-mm culture dish with a 90 μL drop of c-TYH covered with oil was prepared and equilibrated for at least 30 min in the incubator.

2.2 Pick up the frozen straw from the liquid nitrogen tank, and place it in a 37 ℃water bath for 10min.

2.3 Slowly aspirated sperm from straw and added it to the pre-equilibrate Sperm capacitation dish, capacitated in an incubator for 30min. pick up 10μL of sperm suspension from the edge of the dropwas added to the HTF dishes containing oocytes.

Cautions:

1. Avoid blood, fat and hair and tissue from entering capacitation drops during Sperm capacitation medium.
2. Place the sperm capacitation dish in an incubator equilibrate for at least 30 min.
3. The angle of sperm aspiration needs to be inclined and gentle to avoid damaging the sperm tail.
4. Without tracing back the parental generation, if the sperm is low densities , the sperm of different strains mice can be collected into the same fertilization drop for improve the efficiency of IVF.