

Products for  
IVF  
and  
Assisted Reproduction  
Related Procedures  
available from Cosmo Bio USA



**KYUDO:** Complete 2023 product catalog of the most widely utilized and trusted reagents and accessories for **RODENT** sperm cryopreservation and associated IVF procedures. Developed in conjunction with the Center for Animal Reproduction at Kumamoto University.



**Cosmo Bio brand** reagents and solutions for **RODENT** IVF-related procedures. First quality reagents packaged in volumes that may better fit your workflow and budget.



**Cosmo Bio brand** reagents and solutions for IVF-related procedures **PIG** IVF-related procedures.



**4BioDx:** “4MID” line of ELISA Kits measures proAKAP4, an emerging demonstrated reliable marker of functional sperm quality and male fertility. **Kits for 10 species** including agricultural livestock.

Cosmo Bio knows that consistent success in reproductive engineering depends on a myriad of high-spec, quality-controlled reagents as well as their proper handling and storage at each step along the supply chain. All of us at Cosmo Bio take great pride in the quality of our products and great care to insure that the materials you receive are always properly cared for.



**COSMO BIO USA**  
Great Reagents Drive Great Research

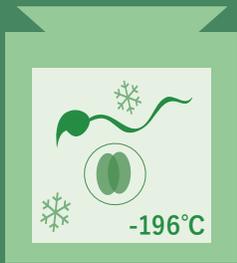
# KYUDO

"best reagents" for "best research"

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IVF

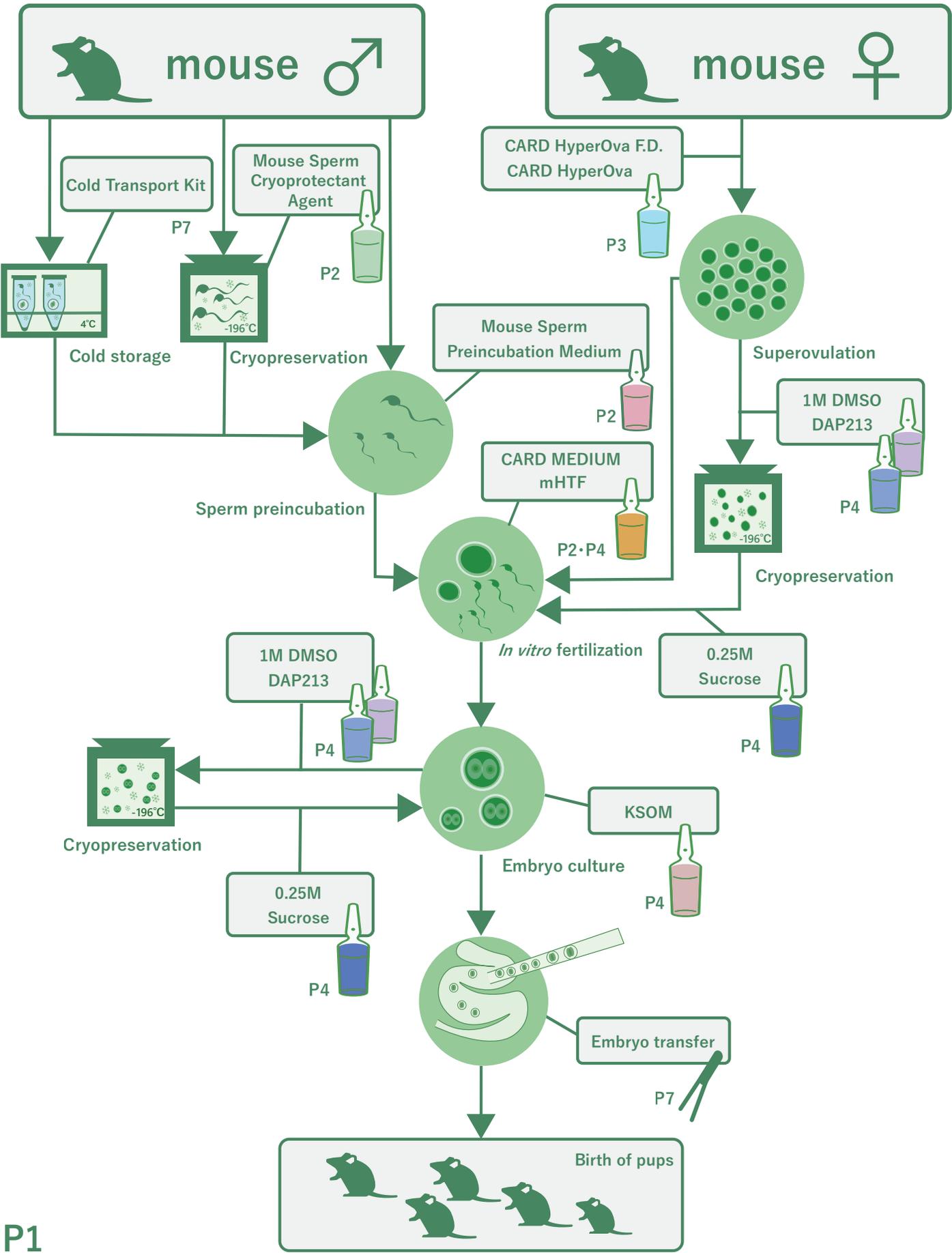


Cryopreservation



Embryo Transfer

# Cutting-edge technology brought to you

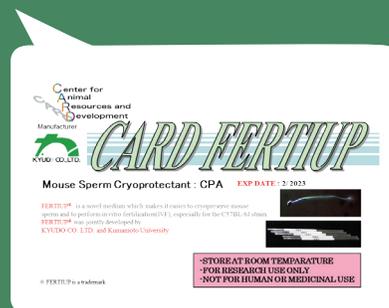


# — Reagents for Reproductive Technology of Mouse

## CARD FERTIUP Mouse Sperm Cryoprotectant Agent

CARD FERTIUP Mouse Sperm Cryoprotectant Agent mitigates freezing damage during sperm cryopreservation and improves *in vitro* fertilization rates using frozen-thawed sperm in numerous mouse strains, including the C57BL/6 strain. This agent also facilitates a massive reduction in costs associated with the upkeep of strains in comparison to the cost of live animal maintenance. (1)

\*Reference(p9):No.1

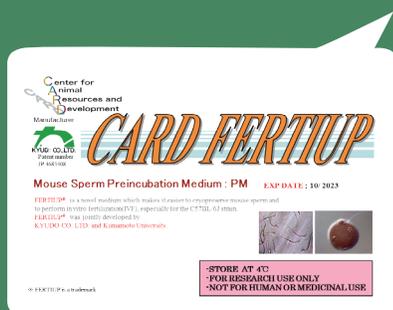


## CARD FERTIUP Mouse Sperm Preincubation Medium

CARD FERTIUP Mouse Sperm Preincubation Medium promotes capacitation of mouse sperm and improves *in vitro* fertilization rates using fresh, frozen-thawed and cold stored mouse sperm. (2)

\*Reference(p9):No.2

\*Patent(JP:4683408)

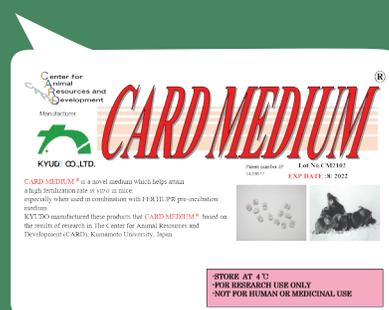


## CARD MEDIUM Mouse Fertilization Medium

CARD MEDIUM High-Performance Mouse IVF Medium brings about a massive improvement in the *in vitro* fertilization rates of numerous mouse strains, including the C57LB/6 and 129 strains. In fact, using CARD MEDIUM alongside CARD FERTIUP Mouse Sperm Preincubation Medium can yield *in vitro* fertilization rates of over 80% for frozen-thawed C57BL/6 mouse sperm. (3)

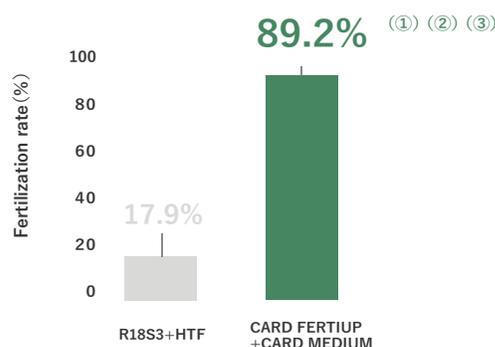
\*Reference(p9):No.3

\*Patent (JP:5439677|US:9,994,868 B2)



With CARD MEDIUM, we can even obtain fertilization rates of over 80% using frozen-thawed C57BL/6 sperm!

Fertilization rate of frozen-thawed C57BL/6 sperm



## CARD HyperOva Superovulation Reagent for Mouse

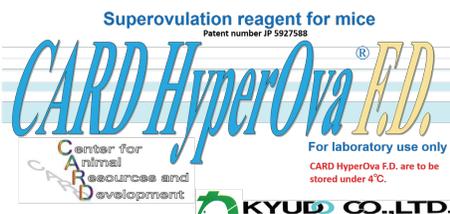
CARD HyperOva Mouse Superovulation Reagent enables us to obtain a much larger number of oocytes from females of numerous mouse strains than via conventional superovulation methods. CARD HyperOva may bring about a threefold or even fourfold reduction in the required number of donor mice. This contributes to a reduction in the number of animals per experiment, thereby upholding the 3 Rs Principle<sup>(4)</sup>

\*Reference(p9):No.4・No.5

\*Patent(JP:5927588 | US:10,500,275,B2 | EU:3260535)

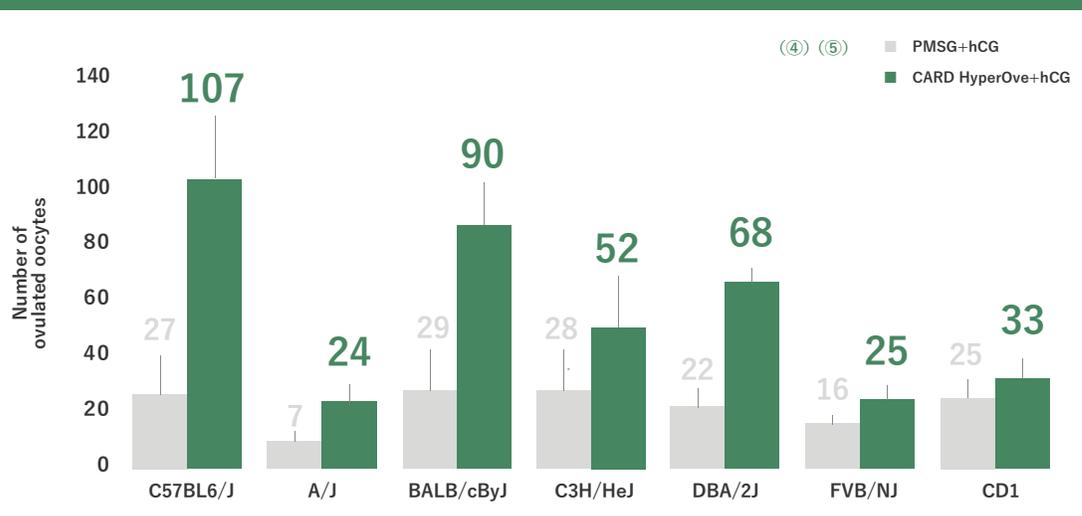


## CARD HyperOvaF.D. Superovulation Reagent for Mouse



CARD HyperOva F.D. retains the high performance of CARD HyperOva while also offering significantly improved storage stability through freeze drying. After preparing the reagent, cryopreserve it at a temperature at  $-20^{\circ}\text{C}$  or below, and then take as much of the reagent as you need, whenever you need it.<sup>(5)</sup>

Using CARD HyperOva, we can obtain three to four times more oocytes than conventional superovulation methods in C57BL/6 mice!



## CARD mHTF medium

CARD mHTF Medium is an *in vitro* fertilization medium. This medium enables superior *in vitro* fertilization rates and a high yield of 2-cell embryos.



## CARD KSOM

CARD KSOM is a culture medium for use on embryos obtained via *in vitro* fertilization. CARD KSOM contributes to superior development rates from 2-cell embryos to blastocysts.



## CARD 1M DMSO

CARD 1M DMSO is cryopreservation reagent utilized in simple vitrification. It is used in the cryopreservation of embryos and oocytes.



## CARD DAP213

CARD DAP213 is cryopreservation reagent utilized in simple vitrification. It is used in the cryopreservation of embryos and oocytes.

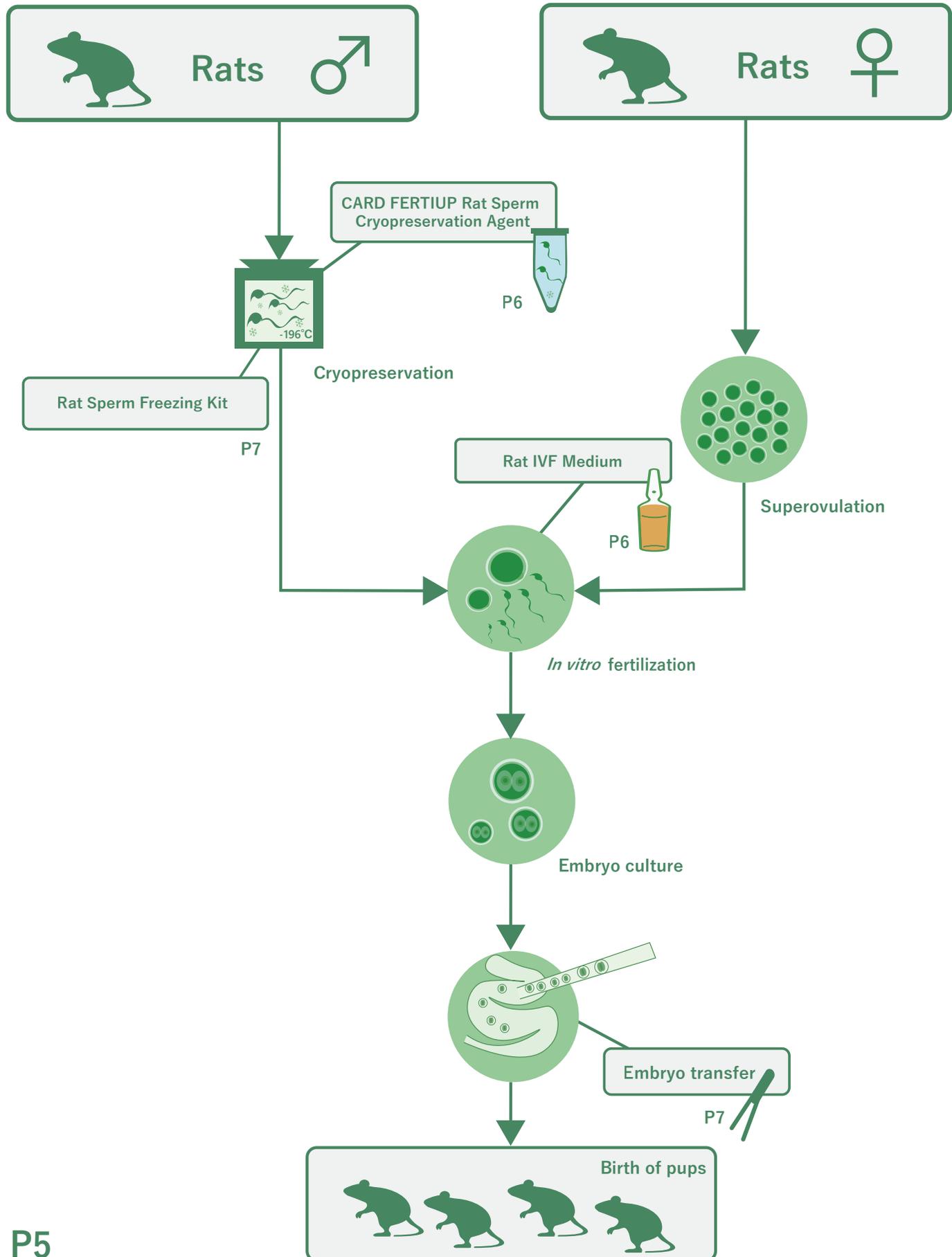


## CARD 0.25M Sucrose

CARD 0.25M Sucrose is used to warm vitrified embryos and oocytes via simple vitrification.



# Breaking new boundaries - Our new technology

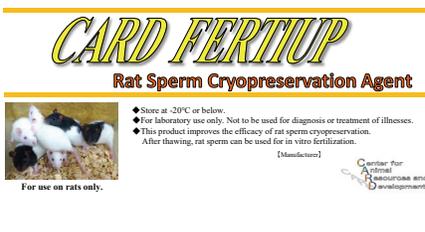


# — Reagents for Reproductive Technology of Rats

## CARD FERTIUP Rat Sperm Cryoprotectant Agent

CARD FERTIUP Rat Sperm Cryopreservation Agent is the world's first practical cryopreservation agent for use with rat sperm. Previously rat sperm cryopreservation was an extremely difficult task, but CARD FERTIUP Rat Cryopreservation Agent can now make it possible to cryopreserve rat sperm in an efficient manner. What's more, once thawed the rat sperm maintain a high motility and can be used in *in vitro* fertilization.

\*Reference(p9):No.6



**CARD FERTIUP**  
Rat Sperm Cryopreservation Agent

◆ Stored at -20°C or below.  
◆ For laboratory use only. Not to be used for diagnosis or treatment of illnesses.  
◆ This product improves the efficacy of rat sperm cryopreservation. After thawing, rat sperm can be used for *in vitro* fertilization.

For use on rats only.

Center for Animal Resources and Development

CARD Reproductive Engineering Media

**CARD Rat IVF medium**  
— A medium for rat *in vitro* fertilization



◆ Stored at 4°C or below.  
◆ For laboratory use only. Not to be used for diagnosis or treatment of illnesses. Contains no ingredients harmful to humans. Do not drink or apply to the human body.  
◆ This product is an IVF medium for use in rat reproductive engineering.

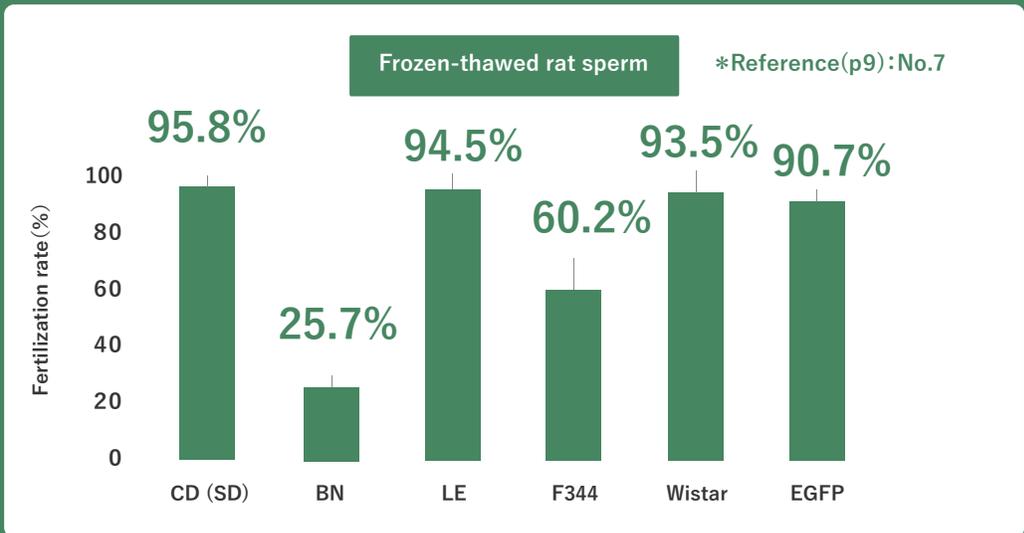
For use on rats only.

Center for Animal Resources and Development

## CARD Rat IVF Medium

CARD Rat IVF Medium is a novel medium developed specifically for use in rat *in vitro* fertilization. It offers superior *in vitro* fertilization rates, particularly when using frozen-thawed rat sperm.

We can obtain high fertilization rates using frozen-thawed sperm in several rats strains!



# — Reproductive Technology Tools for Mouse/Rat

## CARD Cold Transport Kit

The CARD Cold Transport Kit provides everything you need to safely and cheaply transport mouse cauda epididymides and 2-cell embryos at refrigerated temperatures. The transported cauda epididymides and embryos can be used at the destination institution to produce live mice via reproductive engineering techniques.



**Usage:**  
Transport of mouse cauda epididymides and 2-cell embryos at refrigerated temperatures.  
**Cold storage retention period:**  
Max. 72 hours  
(when using our cold storage solution for cauda epididymides)  
**Kit contents:**  
A cold storage box (A), cold packs (B; 4 x large, 2 x small), a Thermos bottle (C), paper boxes and packing material (D).



**Kit contents:**  
A Styrofoam box (A), sperm cryopreservation straw holding support (foam float (B) and plate (C))

## CARD Rat Sperm Freezing Kit

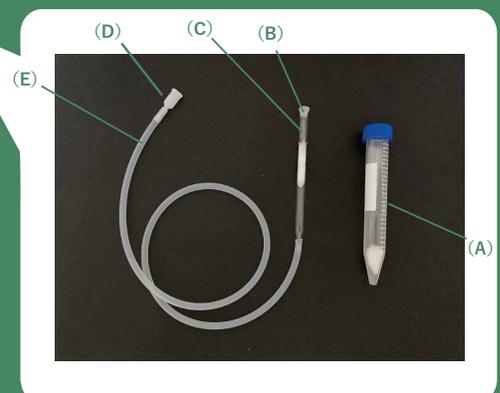
CARD Rat Sperm Freezing Kit is a useful kit for producing high-quality frozen rat sperm using CARD FERTIUP Rat Sperm Cryopreservation Agent. It allows users to correctly carry out the cooling process during the sperm cryopreservation operation.

## Embryo Manipulation Instrument Kit

Our embryo manipulation instrument kit utilizes a glass capillary to ensure the uniform transfer of embryos.

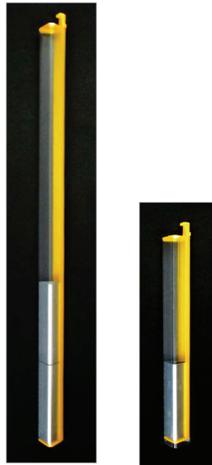
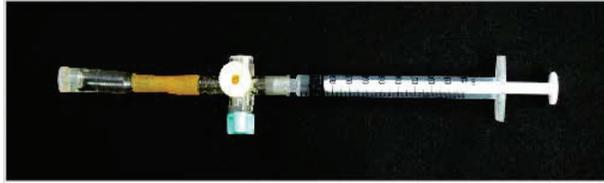
**Set contents:** Glass capillaries for embryo transfer (20 pieces; dry-heat sterilized) (A), a connector (B), a Pasteur pipette (C), a mouthpiece (D), a silicon tube (E)

\* We will send customers all equipment required for embryo transfer.



## Straw Connector

A straw connector is used for sucking up sperm suspended in CARD FERTIUP Sperm Cryopreservation Agent into sperm straws.



## Triangular Cassette Long (W278mm) Triangular Cassette Short (W145mm)

Cassettes used when cryopreserving straws containing sperm suspension in liquid nitrogen. We offer two types of cassette: a long 278mm cassette, which can store up to around 30 straws, and a short 145mm, which can store up to around 15 straws.

## Sperm Freezing Straws (10 pieces)

This is a 0.25mL straw used for sperm storage. Our straws enable the storage of sperm in liquid nitrogen for long periods of time. All straws have been sterilized using EOG.



## Freezing Canister

A freezing canister for use in sperm cryopreservation. Using a float guarantees a cooling rate conducive to mouse sperm cryopreservation on liquid nitrogen, which in turn enables the storage of higher quality frozen sperm.

# About Purchases from Outside of Japan

Our reproductive technology reagent products are used by research institutions around the world.



Distributed worldwide by Cosmo Bio USA  
**COSMO BIO USA**  
 Great Reagents Drive Great Research

2792 Loker Avenue West, Suite 101  
 Carlsbad, CA 92010 USA  
<https://www.cosmobioussa.com/>  
[support@cosmobioussa.com](mailto:support@cosmobioussa.com)  
 +1 858-334-3928

## Technical support

If you have any questions, queries or issues concerning our reagents for use in mouse/rat reproductive technology, please feel free to contact us using the contact details below.

Our veteran technicians are on hand to help solve any problems that you may have.



[web\\_req@kyudo.co.jp](mailto:web_req@kyudo.co.jp)



No.	References
1	Takeo T, Nakagata N. Combination medium of cryoprotective agents containing L-glutamine and methyl- $\beta$ -cyclodextrin in a preincubation medium yields a high fertilization rate for cryopreserved C57BL/6J mouse sperm. <i>Laboratory Animals</i> , 44(2), 132-137, 2010
2	Takeo T, Hoshii T, Kondo Y, Toyodome H, Arima H, Yamamura K, Irie T, Nakagata N. Methyl-beta-cyclodextrin improves fertilizing ability of C57BL/6 mouse sperm after freezing and thawing by facilitating cholesterol efflux from the cells. <i>Biology of Reproduction</i> , 78(3):546-51, 2008
3	Takeo T, Nakagata N. Reduced glutathione enhances fertility of frozen/thawed C57BL/6 mouse sperm after exposure to methyl-beta-cyclodextrin. <i>Biology of Reproduction</i> , 85(5), 1066-1072, 2011
4	Takeo T, Nakagata N. Immunotherapy using inhibin antiserum enhanced the efficacy of equine chorionic gonadotropin on superovulation in major inbred and outbred mice strains. <i>Theriogenology</i> , 86(5), 1341-1346, 2016
5	Takeo T, Nakagata N. Superovulation using the combined administration of inhibin antiserum and equine chorionic gonadotropin increases the number of ovulated oocytes in C57BL/6 female mice. <i>PLoS One</i> , 10(5), 2015
6	Nakagata N, Mikoda N, Nakao S, Nakatsukasa E, Takeo T. Establishment of sperm cryopreservation and in vitro fertilisation protocols for rats. <i>Scientific Reports</i> , 10, 93, 2020
7	Takeo T, Nakao S, Mikoda N, Yamaga K, Maeda R, Tsuchiyama S, Nakatsukasa E, Nakagata N. Optimized protocols for sperm cryopreservation and in vitro fertilization in the rat. <i>Lab animal</i> , 51, 256-274, 2022

# Products List

Reagents for Rat Reproductive Technology	STANDARD	Storage Temp.
CARD FERTIUP Rat Sperm Cryoprotectant Agent	1.5 mL	-80°C to -20°C
CARD Rat IVF Medium	1.0 mL	4°C

Reagents for Mouse Reproductive Technology	STANDARD	Storage Temp.
CARD FERTIUP Mouse Sperm Cryoprotectant Agent: CPA	0.5 mL	18 to 28°C
	1.0 mL	
CARD FERTIUP Mouse Sperm Preincubation Medium: PM	0.5 mL	4°C
	1.0 mL	
CARD MEDIUM Mouse Fertilization Medium	1 kit	4°C
CARD FERTIUP PM (0.5mL) and CARD MEDIUM Mouse Fertilization Medium Kit	1 set	4°C
CARD FERTIUP PM (1.0mL) and CARD MEDIUM Mouse Fertilization Medium Kit	1 set	
CARD HyperOva Superovulation Reagent for Mouse	0.6 mL	-80°C to -20°C
	1.0 mL	
CARD HyperOva F.D. Superovulation Reagent for Mouse	1 kit	4°C
	5 kit	
CARD mHTF Medium	2.0 mL	4°C
	5.0 mL	
CARD KSOM	2.0 mL	4°C
	5.0 mL	
CARD 0.25M Sucrose	2.0 mL	4°C
	5.0 mL	
CARD 1M DMSO	1.0 mL	4°C
CARD DAP213	0.5 mL	4°C
	1.0 mL	

Reproductive Technology Tools for Mouse/Rat	STANDARD	Storage Temp.
Straw Connector	1 unit	-
Triangular Cassette Short (W145mm)	10 units	-
Triangular Cassette Long (W278mm)	10 units	-
Sperm Freezing Straws (10 pieces)	10 units	-
CARD Cold Transport Kit (*The storage temp. of cold storage solution for cauda epididymis)	1 kit	4°C*
Embryo Manipulation Instrument Kit	1 kit	-
Freezing Canister	1 unit	-
CARD Rat Sperm Freezing Kit	1 kit	-

Mouse/Rat Reproductive Engineering Reagents are intended for research use only and are not intended for diagnostic or therapeutic purposes. FERTIUP, CARD MEDIUM, and CARD HyperOva are registered trademarks. All information is current as of October 2022.

Are you having trouble storing your transgenic mouse or rat strains?

Are you struggling with maintenance, upkeep or breeding?

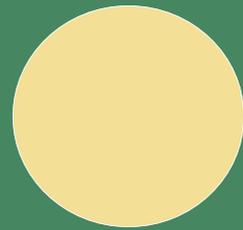
We can help.

We at Kyudo Co, Ltd. provide a wide range of reagents for use at every stage of mouse and rat reproductive technology, from superovulation and *in vitro* fertilization through to the freezing of sperm and embryos.

These high quality reagents were developed in collaboration with the Center for Animal Resources and Development, Kumamoto University. We manufacture and sell the reagents under license to customers worldwide.

Using reproductive engineering techniques alongside our reagents contributes to a significant reduction in the number of animals required per experiment, thereby following the 3R Principle.

Kyudo Co., Ltd



# Frequently Asked Questions (FAQ)

## General Questions

**Q1** Is it possible to download the online manual for reproductive engineering-related reagents?

**A1** You can download it from our homepage. Please refer to the URL below.  
[https://www.cosmobiousa.com/content/document/cosmo-bio-ltd/10167\\_KYD\\_CARDmanual\\_english.pdf](https://www.cosmobiousa.com/content/document/cosmo-bio-ltd/10167_KYD_CARDmanual_english.pdf)

**Q2** Do the products contain substances harmful to the human body?

**A2** No, the products do not contain substances harmful to the human body.

**Q3** Are SDS (Safety Data Sheet) available for the products?

**A3** Yes, SDS (Safety Data Sheet) is available for each reproductive engineering-related reagent product. Please send us a request by e-mail ([support@cosmobiousa.com](mailto:support@cosmobiousa.com))

**Q4** Can the products be used for therapeutic purposes in humans or animals?

**A4** No, our reproductive engineering reagents are intended for research use only. They are not intended for diagnostic or therapeutic use.

**Q5** There is a foreign object in the product, can I exchange it?

**A5** Yes, please send us an inquiry with the photo of the condition and we will replace them.

**Q6** After using the product, is it possible to dispense the remaining amount and store it at low temperature (freeze, refrigerate)?

**A6** We cannot guarantee the quality after cold storage, and do not recommend storage at low temperatures.

**Q7** Please tell me how to store mouse and rat reproductive engineering-related reagents and expiration dates.

[Preservation method and expiration date]

Please use by the indicated expiration date

1. Reproductive engineering-related reagents for mice

- CARD FERTIUP mouse sperm cryopreservation medium: room temperature (18-28°C), 18 months after production date

**A7** • CARD FERTIUP mouse sperm preculture medium: refrigerated (2-10°C), 18 months after production date

- CARD MEDIUM<sup>®</sup> : Refrigerate (2-10°C), 18 months after production date

- CARD HyperOva<sup>®</sup> : Frozen (-80°C), 36 months after production date

- CARD HyperOva<sup>®</sup> FD: Refrigerate (4°C), 24 months after production date

2. Reproductive engineering-related reagents for rats

- CARD FERTIUP Rat Sperm Cryopreservation Solution: Frozen (-20°C or below), 24 months after production date

- CARD Rat IVF Medium: Refrigerated (4°C), 18 months after production date

**Q8** Can reproductive engineering-related media be used immediately after delivery?

**A8** Filter sterilization is not required unless specified in the usage instructions. Please use it after gas equilibrium etc. according to the usage of each product.

# Frequently Asked Questions (FAQ)

## CARD FERTIUP<sup>®</sup> and CARD MEDIUM<sup>®</sup> (Mouse)

### Questions regarding the CARD FERTIUP<sup>®</sup> Products

**Q1** We were able to obtain a high fertility rate for the C57BL/6 strain of mouse via *in vitro* fertilization using cryopreserved spermatozoa treated with CARD FERTIUP<sup>®</sup> Mouse Cryoprotectant/Mouse Preincubation Medium. Does the media increase the *in vitro* fertility rate of other strains too?

**A1** When mouse sperm cryopreservation medium/mouse sperm pre-incubation medium is used together, it is effective in ameliorating the fertility rates in many commercially available mouse strains.

**Q2** After opening the product, can it be subdivided and frozen?

**A2** We cannot guarantee the quality after freezing, so we do not recommend this method of storage.

**Q3** Can I use left over CARD FERTIUP<sup>®</sup> Mouse Cryoprotectant if I transfer the medium into tubes and store it at refrigerated temperatures?

**A3** Once opened, use up the ampoule as quickly as possible. Please purchase an ampoule size which you can use up in one occasion. Please note that the cryoprotectant ampoule should be stored at room temperature as it contains high doses of saccharides and skim milk, which may sink to the bottom of the ampoule after refrigerating or freezing.

**Q4** There are crystals/precipitants in my un-opened CARD FERTIUP<sup>®</sup> Mouse Cryoprotectant.

**A4** In rare cases, crystals may precipitate due to changes in ambient temperature (low temperature). Incubate at 37°C for 30 minutes to 1 hour, and stir to melt the crystals. There is no problem with cryopreservation performance after thawing.

**Q5** Liquid paraffin is used in the usage instructions for the CARD FERTIUP<sup>®</sup> series; doesn't the paraffin have adverse effects on sperm?

**A5** Liquid paraffin does not adversely affect sperm suspension (sperm suspended in storage solution).

**Q6** After preincubating cryopreserved sperm with CARD FERTIUP<sup>®</sup> Mouse Preincubation Medium, what is the sperm concentration when carrying out *in vitro* fertilization using an IVF medium?

**A6** Typically, the sperm concentration in cryopreserved sperm ranges from 100,000~200,000 sperm/ $\mu$ L. When introducing freeze-thawed sperm suspension (10  $\mu$ L) into Mouse Sperm Preincubation Medium (90  $\mu$ L), the sperm concentration is diluted tenfold. This means that the sperm concentration in the preincubation medium is 10,000~20,000 sperm/ $\mu$ L. Thereafter, the preincubated sperm suspension (10  $\mu$ L) is further diluted tenfold when it is introduced into IVF medium (90  $\mu$ L). This results in a sperm concentration in the IVF medium of 1,000~2,000 sperm/ $\mu$ L. Finally, the ratio of motile sperm in after freeze-thawing is around 20%, which means that the motile sperm concentration in the IVF medium is 200~400 sperm/ $\mu$ L.

**Q7** I would like to download the instruction manual for the CARD FERTIUP<sup>®</sup> series. Where can I download them from?

**A7** They are available on our website, [https://www.cosmobiousa.com/content/document/cosmo-bio-ltd/10167\\_KYD\\_CARDmanual\\_english.pdf](https://www.cosmobiousa.com/content/document/cosmo-bio-ltd/10167_KYD_CARDmanual_english.pdf)

**Q8** Is there a method to confirm whether the freezing and thawing of sperm has been correctly carried out?

**A8** 1. To confirm the quality of sperm after freeze-thawing, preincubate the sperm in preincubation medium after freezing and judge according to the number of active motile sperm at the edge of the drop.  
2. For final confirmation, carry out *in vitro* fertilization of unfertilized oocytes and confirm that you have attained at least a 30% fertility rate. When checking the motility of the sperm before freezing, dilute the suspension of sperm in medium such as CARD mHTF or PB1 medium.  
(We carry out IVF during lot checks to check and confirm that all of our products on sale attain an *in vitro* fertility rate of 30% or above for C57BL/6 mice)

**Q9** How about legacy stock of cryopreserved spermatozoa?

**A9** Please Check here. Rescue In Vitro Fertilization Method for Legacy Stock of Cryopreserved Spermatozoa

**Q10** In vitro fertilization does not work as well as we thought. Sperm and eggs have no problem but the embryos died the next day. What kind of problems can be considered?

The quality of liquid paraffin could be a problem that effects in vitro fertilization rates, and arresting embryogenesis . Moreover, liquid paraffin gradually degrades after opened. Please use as promptly as possible after opening. We recommend using the following products.

**A10** Manufacturer name: Nacalai Tesque  
Grade: Specially Prepared Reagent for tissue culture  
Name of product: Paraffin liquid  
Model number: 26137-85

## Questions regarding CARD MEDIUM<sup>®</sup>

**Q1** For how many months after the manufacture date is CARD MEDIUM<sup>®</sup> warranted for?

**A1** The warrantee period for CARD MEDIUM<sup>®</sup> is 18 months. This warrantee period may increase in the future, depending on the results of investigations and reviews.

**Q2** Is it possible to attain a high *in vitro* fertility rate without using CARD MEDIUM<sup>®</sup> with CARD FERTIUP<sup>®</sup> series?

**A2** No. The CARD FERTIUP<sup>®</sup> series works with sperm and CARD MEDIUM<sup>®</sup> with oocytes to raise the fertility rate. Use both products simultaneously to attain a fertility rate of 80% or more even with C57BL/6 strain mouse.

**Q3** Can high *in vitro* fertilization rates be obtained with strains other than C57BL/6 mice?

**A3** Yes, high *in vitro* fertilization rates can be obtained in various mouse strains.  
[Reference link] DOI: 10.1016/j.theriogenology.2016.04.076  
(Link: <https://www.sciencedirect.com/science/article/abs/pii/S0093691X16301194?via%3Dihub>)

**Q4** In vitro fertilization was performed with CARD MEDIUM<sup>®</sup> , and embryonic development beyond the 2-cell stage did not progress well. The medium used for in vitro culture is KSOM medium or Whitten medium. What problems could have slow down the development of the embryo?

The following problems are possible.

**A4** 1. The incubator temperature and carbon dioxide gas concentration  
2. The in vitro culture medium; If prepared by yourself, there may be problems with the distilled water or ingredients. Please consider using "CARD KSOM".  
3. The mineral oil that coats the medium: If old, the embryo development may be inhibited.

## Frequently Asked Questions (FAQ)

# CARD FERTIUP® Rat Sperm Cryopreservation Agent and CARD Rat IVF MEDIUM

### General Questions regarding CARD FERTIUP® Rat Sperm Cryopreservation Agents

Q1	How should I store CARD FERTIUP® Rat Sperm Cryopreservation Agent? By when should I use it?
A1	<ol style="list-style-type: none"><li>1. Please store CARD FERTIUP® Rat Sperm Cryopreservation Agent in a freezer at or below -20°C.</li><li>2. Use before the best before end usage date which is two years (24 months) from the manufacture date.</li></ol>
Q2	After opening a vial of CARD FERTIUP® Rat Sperm Cryopreservation Agent, can I aliquot the contents into smaller portions and use it for cryopreservation?
A2	No. CARD FERTIUP® Rat Sperm Cryopreservation Agent is sold in 1.5mL measures, and one vial (1.5mL) is to be used per rat cauda epididymis. Please follow the usage instructions and use one vial (1.5mL) of CARD FERTIUP® Rat Sperm Cryopreservation Agent per rat cauda epididymis.
Q3	Does CARD FERTIUP® Rat Sperm Cryopreservation Agent contain any ingredients which are harmful to humans?
A3	No. It does not contain ingredients which are harmful to humans.
Q4	There appears to be a foreign object in my vial of CARD FERTIUP® Rat Sperm Cryopreservation Agent. Can I get a replacement?
A4	Kyudo carries out thorough quality control; however, if you have contamination in your vial of CARD FERTIUP® Rat Sperm Cryopreservation Agent, we would be happy to replace your vial with a new one.

### General Questions regarding CARD Rat IVF Medium

Q1	How should I store CARD Rat IVF medium? By when should I use it?
A1	<ol style="list-style-type: none"><li>1. Please store CARD Rat IVF medium in a refrigerator at 4°C.</li><li>2. Use before the best before end usage date of CARD Rat IVF medium is 1.5 years (18 months) from the manufacture date.</li></ol>
Q2	After opening a vial of CARD Rat IVF medium, can I aliquot into smaller portions and use it for cryopreservation?
A2	No. After freezing, we cannot guarantee the quality of the sperm.
Q3	Does CARD Rat IVF medium contain any ingredients which are harmful to humans?
A3	No. It does not contain any ingredients which are harmful to humans.
Q4	There appears to be a foreign object in my vial of CARD Rat IVF medium. Can I get a replacement?
A4	Kyudo carries out thorough quality control; however, if your vial is contaminated upon receipt, we certainly will replace your vial with a new one.

# Frequently Asked Questions (FAQ)

## CARD HyperOva<sup>®</sup> and CARD HyperOva<sup>®</sup> FD

### Questions regarding CARD Hyperova<sup>®</sup>

**Q1** After opening the CARD HyperOva<sup>®</sup>, can it be subdivided and stored frozen?

**A1** No. We do not recommend refreezing after thawing because we cannot guarantee the quality.

**Q2** I didn't ovulate much. Is it defective?

**A2** For C57BL/6J mice, administration is recommended between 26 and 30 days of age (birth date is considered as 0 days of age). Due to the nature of the product, the effects of this product may not be fully obtained at certain ages. In addition, the optimum age in weeks may differ depending on the strain. For special strains and ages, please contact us individually.

**Q3** A sediment is deposited on the product. Are there any problems with using it?

**A3** In rare cases, ingredients may precipitate. Precipitates are not the main component of this product. Avoid deposits during use.

**Q4** Is CARD HyperOva<sup>®</sup> effective against hyperovulation in rats?

**A4** This product is manufactured based on mouse antigens. The effect of superovulation in rats has been confirmed to some extent, though not as much as in mice. In the case of rats, we administer 0.4-0.8mL up to 5-week-old rats, and 48-56 hours later, hCG (15-30IU/head) is administered, and eggs are collected the next morning. Please note that the reactivity differs depending on the rat strain

**Q5** Is CARD HyperOva<sup>®</sup> a mixture of equine CG and goat anti-inhibin antiserum?

**A5** Yes, it is mixed.

**Q6** Tell me more about the horse gonadotropin.

**A6** The horse gonadotropin in CARD HyperOva<sup>®</sup> is purified from a horse born and raised in Japan.

**Q7** What is the country of origin for the goat?

**A7** The goat is a male Saanen goat born and raised in Japan.

**Q8** Did the goat co-mingle with any other livestock?

**A8** No. Only with other goats.

**Q9** Is the antibody unprocessed goat serum?

**A9** The goat serum is diluted 2x with physiological saline. The horse gonadotropin is also dissolved in saline.

**Q10** For the goat anti-mouse inhibin antiserum, what is the source of the immunogen?

**A10** The immunogen is a synthetic peptide.

**Q11** I can see a clump/precipitate/sediment in the vial.

**Q12** Such imperfections do not diminish the effectiveness of CARD HyperOva<sup>®</sup>. Please avoid the clumps and use only the solution.

**Q13** Can the product in 1ml vials be refrozen after being thawed if we don't need all of it at the moment?

**A13** Once the 1ml vial is opened, please use it to administrate intraperitoneally all at once. 0.1mL - 0.2 mL can be administered to each mouse.

**Q14** Will the product work after storage at 4°C for one week instead of -20°C ?

**A14** We cannot guarantee the quality under this storage condition. The CARD HyperOva may lost activity and we cannot recommend using it.

# Frequently Asked Questions (FAQ)

## CARD HyperOva<sup>®</sup> and CARD HyperOva<sup>®</sup> FD

### Questions regarding CARD Hyperova<sup>®</sup> FD

**Q1** Does the CARD HyperOva<sup>®</sup> FD have an expiration date?

**A1** Yes. As of January 2020, it is two years from the manufacture date. Please use it within the expiration date.

**Q2** What is the difference between conventional CARD HyperOva<sup>®</sup> and CARD HyperOva<sup>®</sup> FD?

**A2** CARD HyperOva<sup>®</sup> FD is manufactured with a new formulation suitable for lyophilization. Conventional products needed to be stored at -20°C to -80°C, but this product can be stored at 4°C.

**Q3** Are there any difference in the superovulation performance of CARD HyperOva<sup>®</sup> FD compared to conventional products?

**A3** Please expect the same superovulation results as the conventional CARD HyperOva<sup>®</sup>

**Q4** What should I do for long-term storage after thawing CARD HyperOva<sup>®</sup> FD?

**A4** Aliquot and store at or below -20°C. Please thaw before use. The superovulation effect has been confirmed to be maintained for about 3 months after the product is thawed or frozen.

# Frequently Asked Questions (FAQ)

## Others

Q1	Frozen mouse sperm has been sent from the United States, and we are considering in vitro fertilization. Is it possible to perform in vitro fertilization using cryopreserved mouse sperm frozen in a method other than the CARD method?
A1	For in vitro fertilization methods using sperm frozen by methods other than the CARD method, please refer to the online manual of the Kumamoto University Life Resource Research and Support Center (CARD).
Q2	IVF didn't go as well as expected. The sperm and egg are fine, but the embryo dies. What are the possible causes?
A2	<p>The quality of liquid paraffin is one of the reasons why IVF fails and embryos stop developing. We recommend using the following liquid paraffin products. In addition, we have experienced that the quality of liquid paraffin deteriorates after opening. Please use as soon as possible after opening.</p> <p>Manufacturer name: Nacalai Tesque Standards: Special reagent for tissue culture Product name: Liquid paraffin Model number: 26137-85</p>

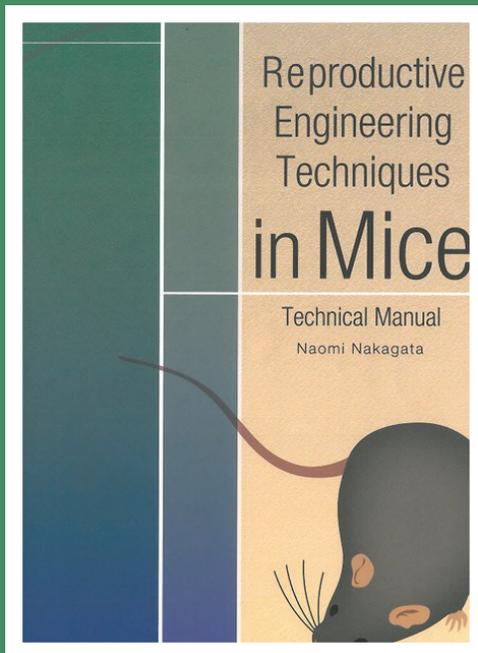
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**by Naomi Nakagata (3rd Edition)**

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REPRODUCTIVE ENGINEERING TECHNIQUES IN MICE

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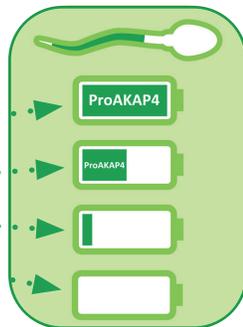
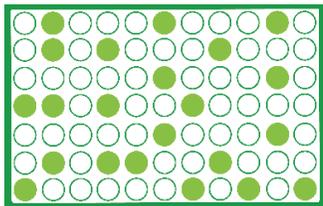
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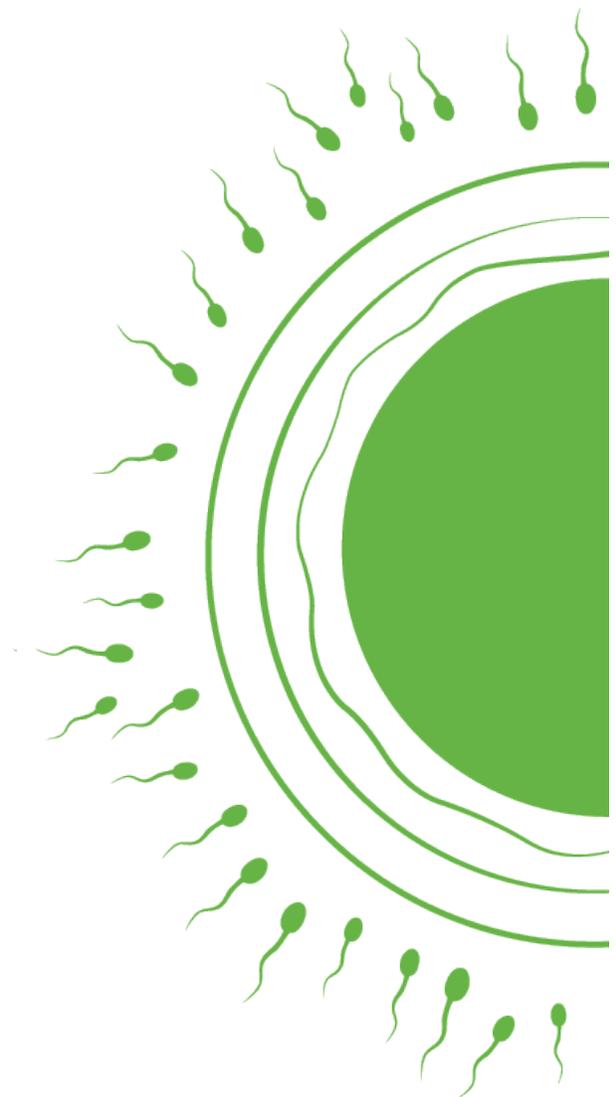
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# PRODUCT LIST



## 4MID® Kits : Anti-proAKAP4 ELISA kits

Reference	Designation	Specificity	Price
<b>4VDX-18K2</b>	Pig 4MID® Kit	Pig proAKAP4	request a quote
<b>4VDX-18K3</b>	Horse 4MID® Kit	Horse proAKAP4	request a quote
<b>4VDX-18K4</b>	Bull 4MID® Kit	Bovine proAKAP4	request a quote
<b>4VDX-18K5</b>	Dog 4MID® Kit	Dog proAKAP4	request a quote
<b>4VDX-18K6</b>	Rabbit 4MID® Kit	Rabbit proAKAP4	request a quote
<b>4VDX-18K7</b>	Ram 4MID® Kit	Ram proAKAP4	request a quote
<b>4BDX-18K8</b>	Mouse 4MID® Kit	Mouse / Rat proAKAP4	request a quote
<b>4VDX-18K9</b>	Goat 4MID® Kit	Goat proAKAP4	request a quote
<b>4VDX-19K10</b>	Cat 4MID® Kit	Cat proAKAP4	request a quote
<b>4VDX-19K11</b>	Camel 4MID® Kit	Camel proAKAP4	request a quote

Reference	Antibody	Reactivity	Price
<b>4BDX-1901</b>	Anti-proAKAP4 (clone 5H2) - 100 µg	Goat, Camel, Cat, Dog, Pig, Rabbit	request a quote
<b>4BDX-1901S</b>	Anti-proAKAP4 (clone 5H2) - 20 µg	Goat, Camel, Cat, Dog, Pig, Rabbit	request a quote
<b>4BDX-1701</b>	Anti-proAKAP4 (clone 6F12) - 100 µg	Horse, Bull, Ram, Camel, Mouse	request a quote
<b>4BDX-1701S</b>	Anti-proAKAP4 (clone 6F12) - 20 µg	Horse, Bull, Ram, Camel, Mouse	request a quote
<b>4BDX-1602</b>	Anti-AKAP4 (clone 7E10) - 100 µg	Horse, Bull, Mouse, Rat, Camel	request a quote
<b>4BDX-1602S</b>	Anti-AKAP4 (clone 7E10) - 20 µg	Horse, Bull, Mouse, Rat, Camel	request a quote
<b>4BDX-1805</b>	Anti-AKAP4 (clone 10F8) - 100 µg	Dog, Rabbit, Pig, Ram, Cat, Goat	request a quote
<b>4BDX-1805S</b>	Anti-AKAP4 (clone 10F8) - 20 µg	Dog, Rabbit, Pig, Ram, Cat, Goat	request a quote

All our monoclonal antibodies are validated for Western-Blot, Immunohistochemistry, Immunofluorescence and Flow cytometry applications.

We sell 4BioDx products directly or through our distributor Cosmo Bio USA. Please contact us at [contact@4biodx.com](mailto:contact@4biodx.com) or contact them at [support@cosmobiousa.com](mailto:support@cosmobiousa.com)

All our **products are sold for in vitro use only**, not to be administered to humans or animals and not to be used as diagnostics nor therapeutics.



**COSMO BIO USA**  
Great Reagents Drive Great Research

## Cosmo Bio Brand Media and Solutions for RODENT and PIG IVF-Related Procedures

### for MOUSE and/or RAT

	<b>Size</b>	<b>Cat. No.</b>
<b>DAP213 (DMSO, Acetamide, Propylene Glycol)</b>	10 x 1 ml	CSR-R-T073
<b>DMSO 1M</b>	10 x 2 ml	CSR-R-T072
<b>HTF (Human Tubal Fluid)</b>	10 x 2 ml / 10 x 5 ml	CSR-R-B070 / CSR-R-B071
<b>M2</b>	10 x 2 ml / 10 x 5 ml	CSR-R-M083 / CSR-R-M084
<b>Modified Krebs-Ringer Bicarbonate (mKRB) medium</b>	100 ml	CSR-R-M193
<b>Mouse KSOM (Potassium Simplex Optimized Medium)</b>	10 x 2 ml / 10 x 5 ml	CSR-R-B074 / CSR-R-B075
<b>mR1ECM (modified Rat 1-cell Embryo Culture Medium)</b>	10 x 2 ml / 10 x 5 ml	CSR-R-M191 / CSR-R-M174
<b>mWWM (modified Whitten's Medium)</b>	10 x 2 ml / 10 x 5 ml	CSR-R-B080 / CSR-B081
<b>P10 (Propylene Glycol in PB1)</b>	10 x 2 ml	CSR-R-P186
<b>PB1</b>	10 x 2 ml / 10 x 5 ml	CSR-R-P185 / CSR-R-P138
<b>PEPeS (Percoll in PB1)</b>	10 x 1 ml	CSR-R-P187
<b>R18S3 (Medium for Mouse Sperm Cryopreservation)</b>	10 x 1 ml	CSR-R-R082
<b>Rat KSOM (Potassium Simplex Optimized Medium)</b>	10 x 1 ml	CSR-R-R148
<b>SPB1</b>	10 x 2 ml / 10 x 5 ml	CSR-R-S187 / CSR-R-S188
<b>Sucrose 0.25M</b>	10 x 2 ml / 10 x 5 ml	CSR-R-Y077 / CSE-R-Y078



### for PIG (defined media and solutions)

	<b>Size</b>	<b>Cat. No.</b>
<b>Basic Culture Kit for Porcine IVF Embryo Production</b>	1 kit	CSR-CK029
<b>Complete Culture Kit Porcine IVF Embryo Production</b>	1 kit	CSR-CK030
<b>POE-CM (Porcine Oocyte/Embryo Collection Medium)</b>	5 x 100 ml	CSR-CK020
<b>POM (Basic Medium For Porcine Oocyte Maturation)</b>	3 x 25 ml	CSR-CK021
<b>HP-POM (High Performance Porcine Oocyte Maturation Medium)</b>	3 x 25 ml	CSR-CK022
<b>dbcAMP-100X (dbcAMP Concentrate, 100x Solution)</b>	500 ul	CSR-CK027
<b>SEM-5X (Porcine Semen Dilution Medium)</b>	25 ml	CSR-CK026
<b>PFM (Porcine Fertilization Medium): CSR-CK023</b>	2 x 100 ml	CSR-CK023
<b>PZM-5 (Porcine Embryo Development Medium)</b>	3 x 25 ml	CSR-CK024
<b>PBM (Porcine Blastocyst Medium)</b>	2 x 25 ml	CSR-CK025