

Cryopreservation of Mammalian Oocytes and Embryos

Toni Köbli
Thermo Fisher Scientific
Karlsruhe, Germany

Key Words:

- Cryopreservation
- Oocytes
- Embryos

Abstract

Cryopreservation of bovine embryos has become a standard method of commercial embryo transfer. The process requires a transportable unit that offers all the advantages of a large-scale, high-performance laboratory cryostat used for research purposes. Thermo Fisher Scientific offers several cryostats that are ideal for this application.

The Process

Embryos (preferably morulae or blastocysts) are packed in paillettes and submerged in the -4 to -6°C cooling bath of the temperature control unit. After 5 minutes, seeding is initiated by touching the paillettes with tweezers cooled in liquid nitrogen. After another 5 minutes, the embryos are cooled gradually to an insertion temperature of -32°C . A cool-down rate of $0.3^{\circ}/\text{min}$ is achieved via a programmable controller. After reaching -32°C , the paillettes are plunged directly into liquid nitrogen to ensure the fastest possible chilling.

The process has long been established as a common method for deep freezing laboratory animal embryos. In recent years, it has been used in the field with great success for the cryopreservation of bovine embryos. Further modifications have led to the use of cryopreservation for deep-freezing equine embryos as well as the unfertilized embryos of mice and rabbits. These more sensitive samples are packed in paillettes and placed in the cooling bath at $+15^{\circ}\text{C}$. After 5 minutes the samples are cooled down to -6°C at a cooling rate of $1^{\circ}\text{C}/\text{min}$.

The remaining steps in the cryopreservation process are identical to those described earlier.

The Thermo Fisher Scientific Solution

Thermo Fisher offers two separate units with different cooling rates needed for either sensitive or hardier samples. Each unit is equipped with a programmer so the operator can select the desired cooling rate. The bath temperature and/or program sequence is displayed on a large LCD for easy reference. Each unit also has RS232C and RS485 serial interfaces and a timer for added convenience.

The paillette rack allows up to 30 samples to be submerged in liquid nitrogen at once. All paillettes are dropped into the container by rotating the rack. Handling is simple.

Thermo Scientific HAAKE PII2 C35P Refrigerated Circulator
Order number:
441-0351 for 230V/50Hz and
441-0352 for 115V/60Hz
Cool down rates:
 $+15$ to -6°C : $0.5^{\circ}\text{C}/\text{min}$.
 -6 to -32°C : $1.0^{\circ}\text{C}/\text{min}$.



Thermo Scientific HAAKE PII2 C35P Refrigerated Circulator

Thermo Scientific HAAKE PII2 C50P Refrigerated Circulator
Order number:
441-0501 230V/50Hz and
441-0509 220V/60Hz
Cool down rates:
 $+15$ to -6°C : $1.5^{\circ}\text{C}/\text{min}$.
 -6 to -32°C : $2.5^{\circ}\text{C}/\text{min}$.

Technical Data		PII2 C35P	PII2 C50P
Working Temperature Range	C	-35 to 200	-50 to 200
Temperature Accuracy	K	0.01	0.01
Cooling Capacity at $+20\text{C}/-20\text{C}$	W	400/150	850/500
Heating Capacity 230V/115V	kW	2.0/1.2	2.0/-
Bath Opening/Depth	cm	22 x 14 / 15	22 x 14 / 15
Bath Volume	L	8	8
Total Dimensions	cm	40 x 51 / 71	40 X 51 / 71
Weight	Kg	40	46
Ambient Temperature Range	C	5 to 40	5 to 40

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Benelux
Tel. +31 (0) 76 5 79 55 55
info.tc.nl@thermofisher.com

China
Tel. +86 (21) 68 65 45 88
info.tc.china@thermofisher.com

France
Tel. +33 (0) 1 60 92 48 00
info.tc.fr@thermofisher.com

Germany
Dieselstr. 4,
76227 Karlsruhe
Tel. +49(0)721 40 94-444
info.tc.de@thermofisher.com

India
Tel. +91 (22) 27 78 11 01
info.tc.in@thermofisher.com

United Kingdom
Tel. +44 (0) 1785 81 36 48
info.tc.uk@thermofisher.com

USA
Tel. 603 436 9444
info.tc.us@thermofisher.com

www.thermo.com/tc

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