# CRYOPRESERVATION TECHNOLOGY STATUS IN SOUTH AMERICA


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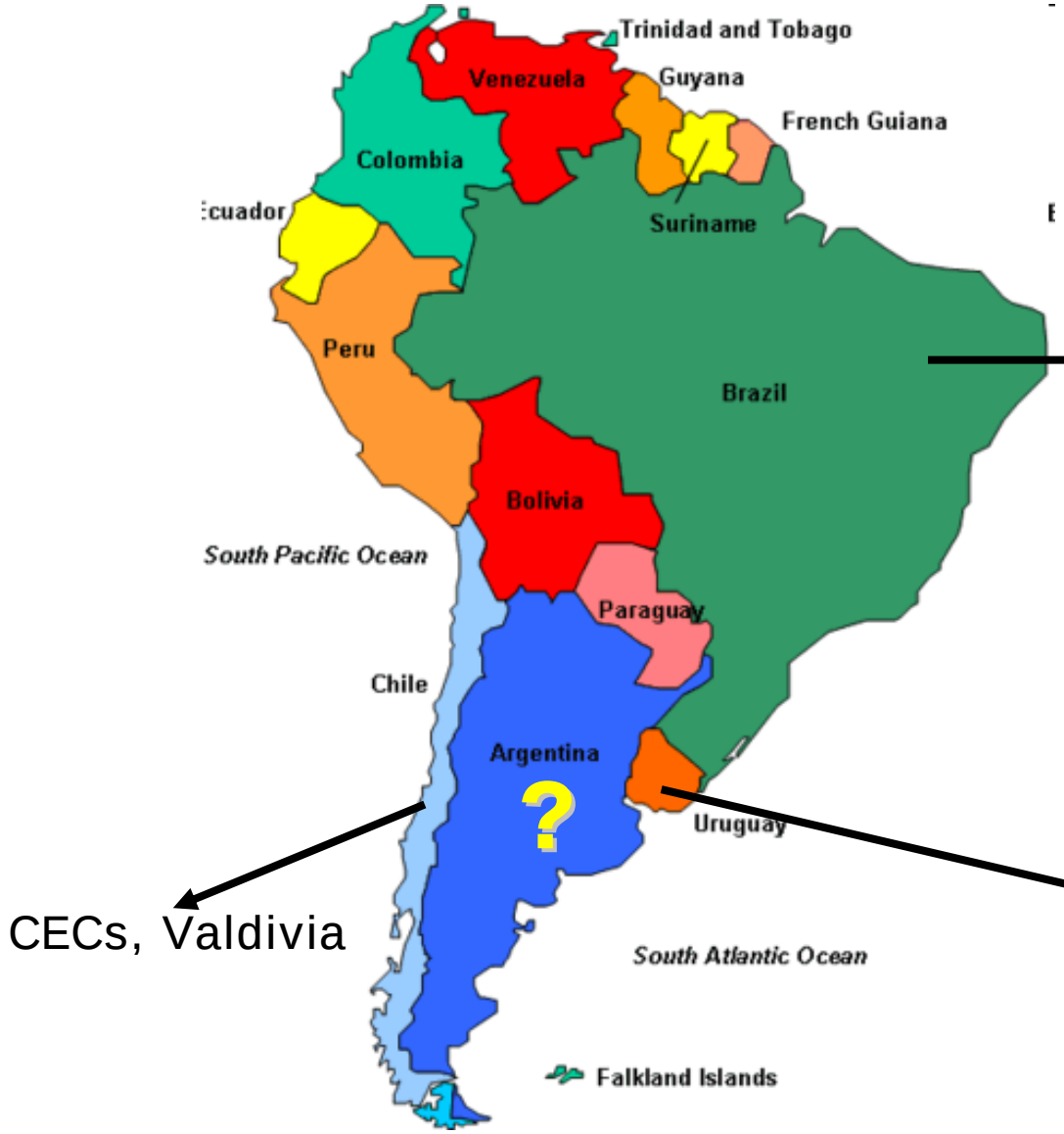
# CRYOPRESERVATION LABS




Fiocruz, BA
CECAL, RJ
CEDEME, SP
Centro de Bioterismo, SP
CEMIB, SP

CECs, Valdivia

UATE, MVD

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# REGIONAL EFFORTS

- Courses
  - Genetics of Laboratory Rodents 2008-2012 (IPMon)
  - Embryo & sperm cryopreservation 2010-2012 (CEMIB)
  - Sperm cryopreservation 2012 (CECs)
  - Yearly Postgraduate courses (IPMon)
- Meetings
  - 1st Symposium “Animal models and cryopreservation” 2010 (CEMIB)
  - 1st Colloquium “Animal Models & Cryopreserved Genetic Repository” 2012 (CEMIB)
  - XII Meeting of Brazilian Society of Laboratory Animal Science (SBCAL): “Paradigms of Laboratory Animal Science” 2012
- Network
  - Cryopreserved genetic repository (CEMIB)

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# UATE: IVF & Cryopreservation Lab

- ✓ IVF in mice and other spp (sheep and cattle)
- ✓ Sperm freezing in rodents
- ✓ Embryo slow freezing
- ✓ Embryo, oocyte and ovary vitrification

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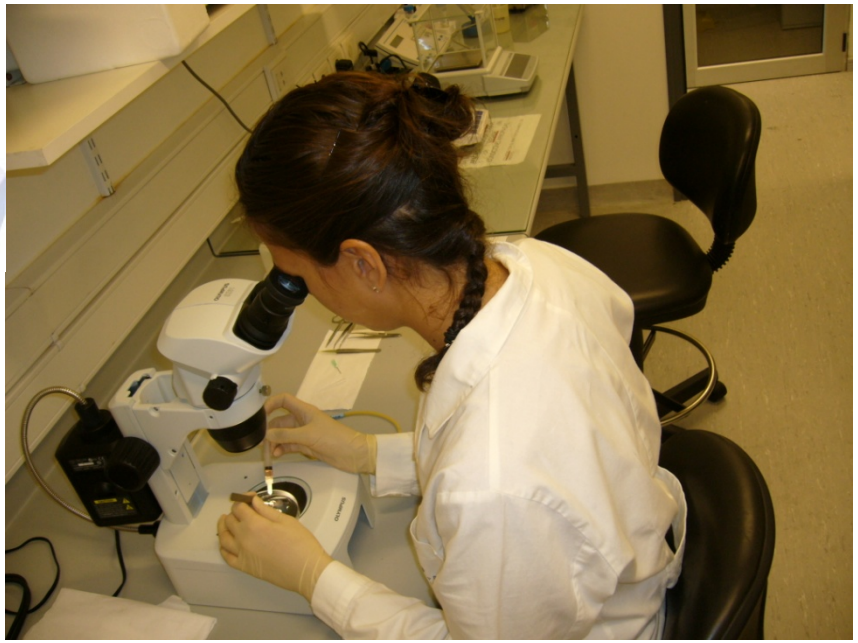
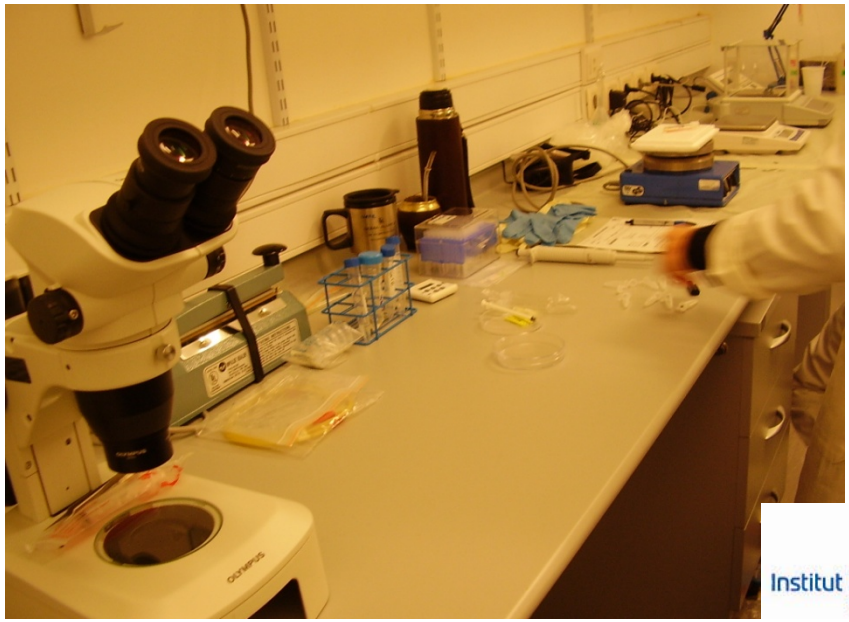
# UATE LABORATORY



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*BioTechniques* 46:550-552 (June 2009)

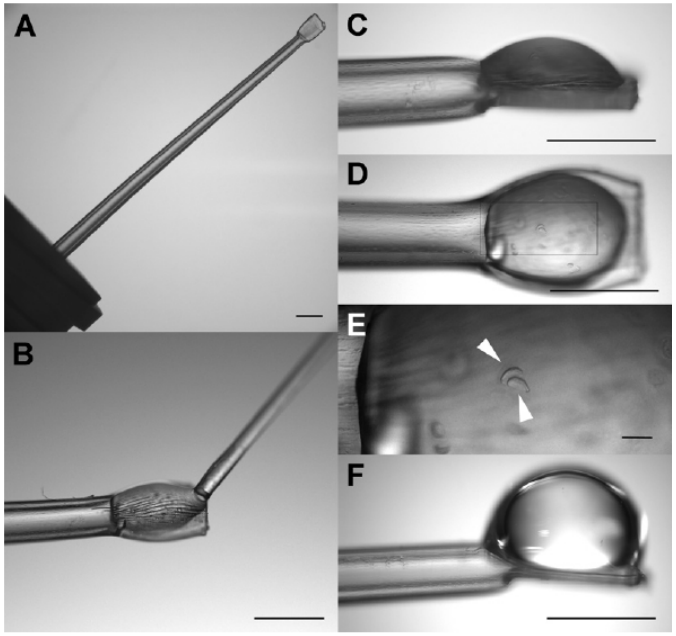
# Mouse embryo cryopreservation utilizing a novel high-capacity vitrification spatula

Wai Hung Tsang and King L. Chow

*BioTechniques Protocol Guide 2010* (p. 55)

# Cryopresevation of mouse embryos with a vitrification spatula

Wai Hung Tsang and King L. Chow




Easy to handle, ultra-fast cooling, store samples in a closed system. Highest embryo storage capacity.

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# SLOW FREEZING vs VITRIFICATION

- KO mice (MyD 88 -/-)

- Three groups:
  - 1) Slow freezing (Renard & Babinet, 1984)
  - 2) Vitrification spatula
  - 3) Control

- Evaluation of recovery, survival and development rates at 24 and 48 h after thawing/warming.

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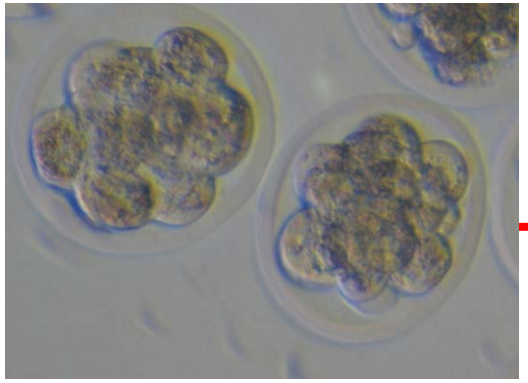
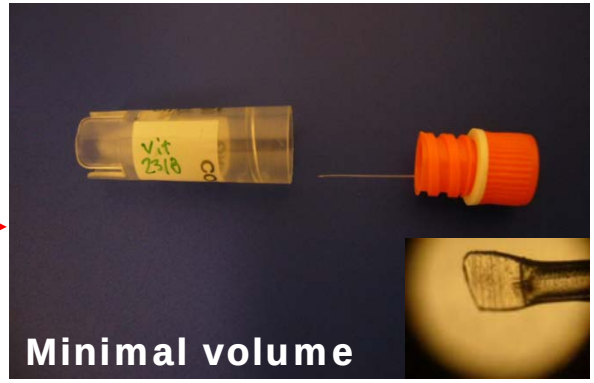
# METHODOLOGY

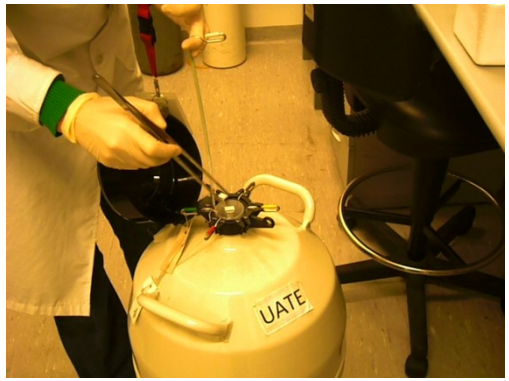


8-cell embryos


**Pre-vitrification (30s)**
10% DMSO
10% EG




**Vitrification (30s)**
15% DMSO
15% EG
60% Ficoll + sucrose




**Warming**
0.5 M Sucrose (2 min)
0.25 M Sucrose (2 min)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# PRELIMINARY RESULTS

## Slow freezing vs. vitrification spatula

| | Recovery rate | Survival rate | Development rate 24 h | Development rate 48 h |
|---|---|---|---|---|
| Slow freezing | 84.9% (45/53)[a] | 84.4% (38/45)[a] | 73.7% (28/38)[a] | 52.6% (20/38)[a] |
| Vitrification | 96.4% (134/139)[b] | 99.3% (133/134)[b] | 95.5% (127/133)[b] | 92.5% (123/133)[b] |
| Control | ---- | ---- | 98.9% (87/88)[b] | 94.3% (83/88)[b] |

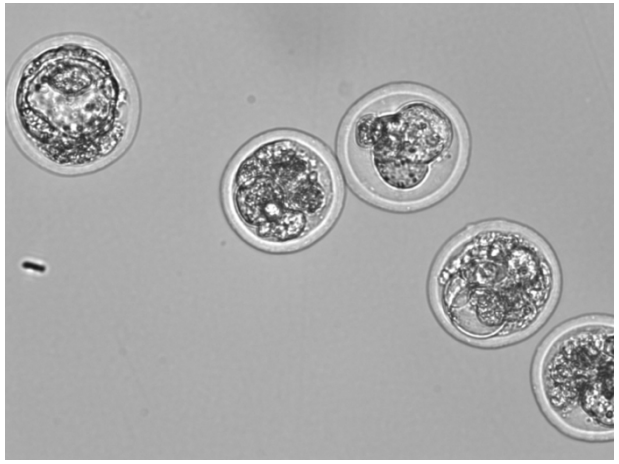
a vs. b, $P<0.05$

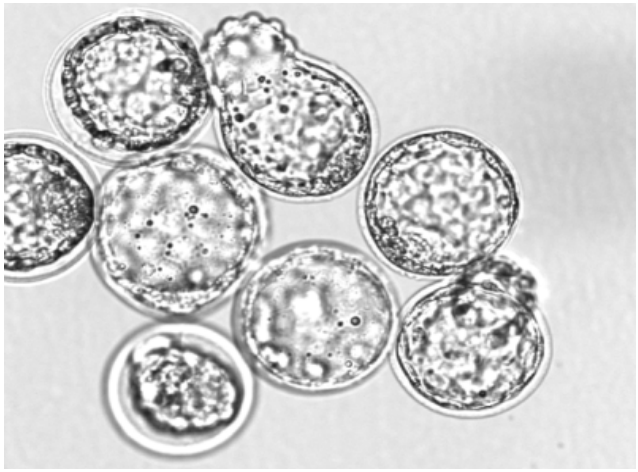
# PRELIMINARY RESULTS

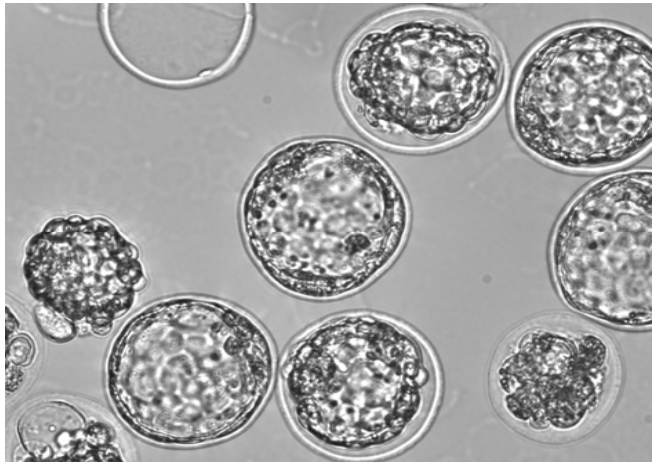
## Slow freezing vs. vitrification spatula

Efficiency rate (survival 48 h/frozen or vitrified embryos)

| Slow freezing | Vitrification | Control |
|---|---|---|
| **37.7%** (20/53)[a] | **88.5%** (123/139)[b] | **94.3%** (83/88)[b] |







a vs. b, $P<0.05$

# CONCLUSIONS

- Several transgenic & KO models in our region.
- High standard cryopreservation technology is available in South America.
- Regional repository willing to be operative.
- Regional efforts to spread knowledge (courses, meetings).

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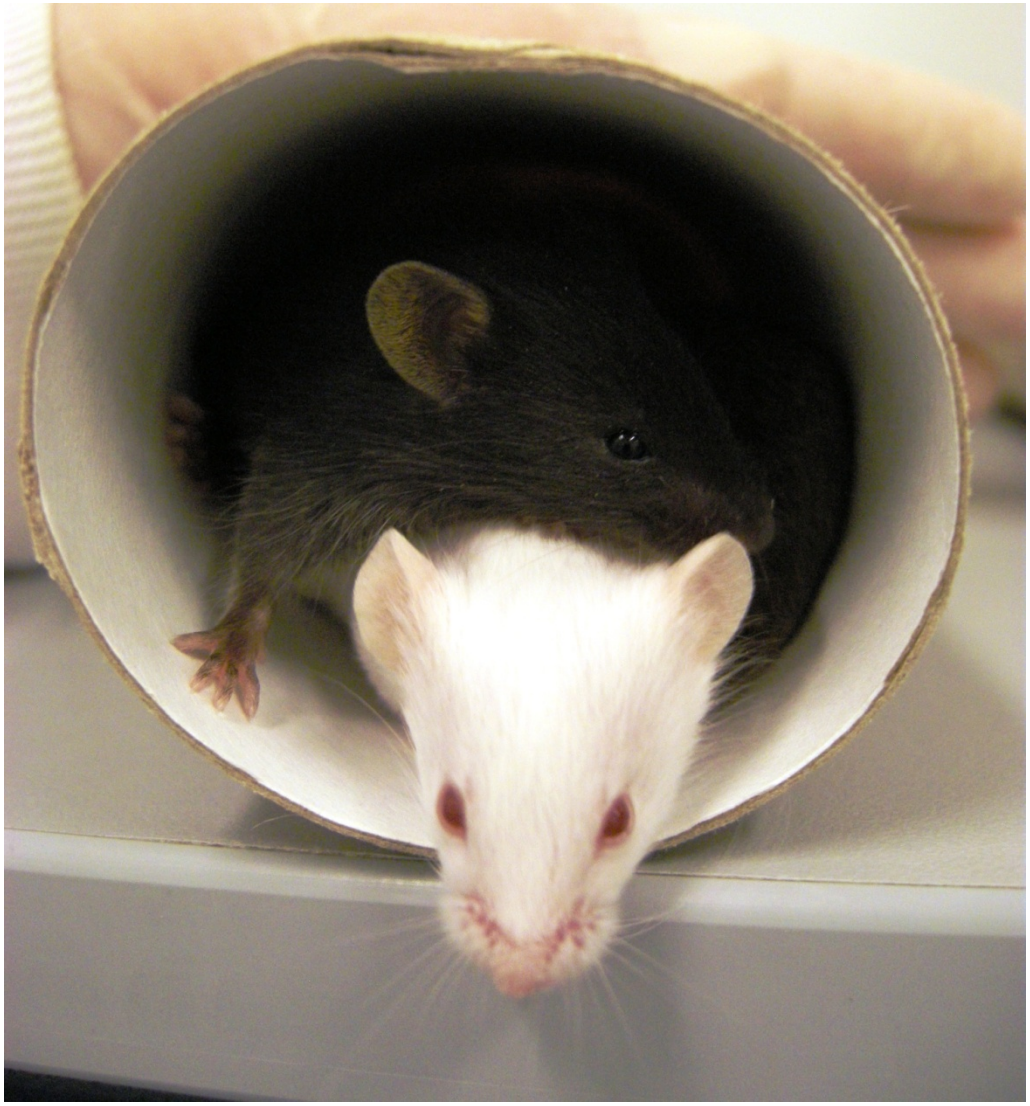
# MUCHAS GRACIAS



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# Cryotop vs. spatula

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