### **RIKEN BRC CELL BANK Information Sheet**

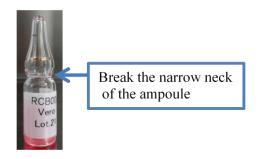
### **Caution**

Please always wear protective gloves and a face guard when handling frozen cells.

The ampoule or cryotube may burst rarely for any reason.

If you do not plan to cultivate for a while after receiving cells, please store the ampoule or cryotube in the vapor phase of liquid nitrogen or at lower than -140 degrees Celsius in a freezer.

### < How to open the glass ampoule >



- i. Protect your hands while breaking the glass using cotton soaked in 70% ethanol.
- ii. Hold the ampoule with one hand, with one thumb against the narrow top section.
- iii. Hold the bottom of the ampoule firmly while pushing the top section away from you.

  Applying light pressure should easily separate the top from the ampoule.







## [Thawing procedure] (For both adherent and/or non-adherent cell lines)

Please check the data sheet before starting cultures (especially basal medium containing L-glutamine or not), and if you find attached instructions, read it carefully.

$\downarrow$	Add 5ml culture medium into 15ml conical tube.
	Pick up the ampoule or cryotube and transfer it into water bath (37 degrees C) immediately.
	Swirl the ampoule or cryotube in water bath till there is a small amount of frozen remains. (Please don't wait till frozen materials are thawed entirely.)
<b>\</b>	Wipe the ampoule or cryotube with 70% ethanol.
	Open the ampoule or cryotube carefully and transfer cell suspension to 15ml conical tube prepared.
<b>\</b>	Centrifuge at 1,000 rpm, for 3 minutes, at room temperature.
<b>\</b>	Remove supernatant and re-suspend cell pellet with 5ml fresh culture medium.
$\downarrow$	Centrifuge at 1,000 rpm, for 3 minutes, at room temperature.
	Remove supernatant and re-suspend cell pellet with appropriate amount of fresh culture medium.
$\downarrow$	Seed cell suspension into <u>two</u> 60mm culture dishes or <u>two</u> T-25 flasks.  *However, please seed cells in adequate culture vessels depending on freezing cell number, cell viability or cell adhesion efficiency.
$\downarrow$	Observe cells next day, and if high rate of dead cells or low adhesion rate is observed, do NOT change the medium or subculture cells.

If you have any questions, please contact us by e-mail (cellqa.brc@riken.jp) immediately.

### [Request for publication]

We request you to send us a reprint of your publication.

We seek to share all data available on the Biological Resources in the RIKEN BRC.

# [Information]

RIKEN BRC CELL BANK

http://cell.brc.riken.jp/en/

# [Contact us]

Order: cellbank.brc@riken.jp

Technical support : cellqa.brc@riken.jp

Payment : brc-front@brc.riken.jp