Cell type: S16

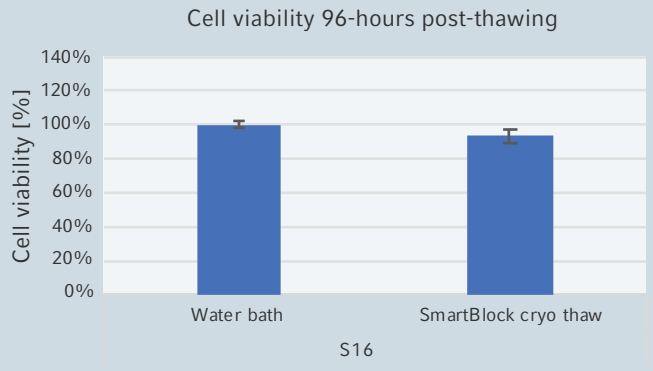
(ATCC no. CRL-2546)

Performance of the SmartBlock™ cryo thaw for cell thawing

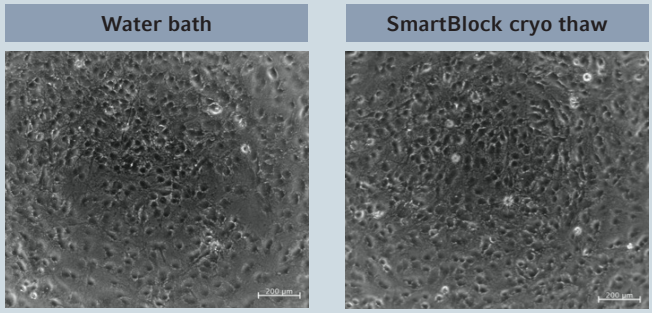
Freezing medium: 70 % Dulbecco's Modified Eagle's Medium (DMEM) (Invitrogen #21765-029), 20 % FBS Superior, (Biochrom AG #S 0615), 10 % DMSO (Roth #4720)

Procedure of freezing: Cells were aliquoted in Eppendorf CryoStorage Vials 2.0 mL (0030 079.485) at a concentration of $1x10^6$ /ml in 1 mL of freezing medium. Vials were slowly cooled down to -80 °C (cooling rate of 1 degree per minute) using a cryo freezing container (CoolCell LX, BioCision). After 24 hours the vials were transferred in liquid nitrogen (vapor phase).

Procedure of thawing: The vials where thawed using the program 'Thawing cells' (4 minutes, 500 rpm). Parallel thawing with a water bath was done. After thawing, vials were diluted with 10 mL medium, centrifugated and resuspended in 2 mL fresh medium and counted. Cells were seeded in 96-well cell culture plates and incubated at 37 °C with 5 % CO_2. After 96 hours, the cells morphology was analyzed by phase-contrast microscope and a CellTiter-Blue® Viability Assay (Promega, G8081) was done.



Bars represent average count of three wells



Representative Phase-contrast microscope images 96 h post-thawing (100× magnification)

The program 'Thawing cells' of the Eppendorf SmartBlock cryo thaw allows optimal thawing and homogeneous growth of S16 cells compared to the existing method (here: water bath).

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