

# Vitrification

## VitriBlast™ Kit 510(k)

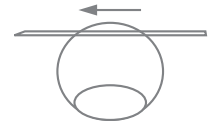
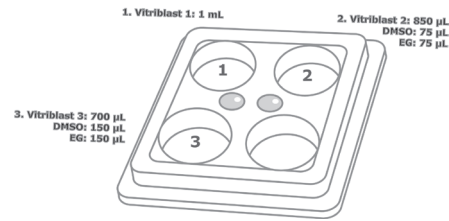


## ThermoBlast™ Kit 510(k)

# How to use VitriBlast™

## Reagents and Equipment

- VitriBlast™ kit
- Device for vitrification
- Inverted microscope
- Culture dishes (NUNC 4-well)
- Liquid nitrogen reservoir
- Stopwatch or timer
- Sterile pipettes
- CO<sub>2</sub> Incubator
- Liquid nitrogen
- Heated stage



## Selection of appropriate vitrification device

Use a legally marketed device indicated for use in blastocyst vitrification procedures. Use a closed system to prevent the potential risk of viral contamination using open systems where the sample comes in direct contact with liquid nitrogen. The device needs to meet the following rate of cooling: minimum 1.800°C/min (High security straw)

## Directions for Use

### Vitrifying Blastocysts with VitriBlast™

Work on a heated stage at all times when manipulating the

blastocyst. Do not let the blastocyst remain exposed to the microscope light during incubations.

1. Label the 4 well culture dish with the patient ID and each well with each solution number
2. 1 hr prior to use or the day before; remove the DMSO from the refrigerator, and let liquify in RT
3. Prepare a 4 well culture dish by adding 1 mL of VitriBlast 1 to the first well
4. Add 850µL of VitriBlast™ 2, 75µL of DMSO and 75µL EG respectively to the second well. Mix thoroughly
5. Add 700µL of VitriBlast™ 3, 150µL of DMSO and 150µL EG respectively to the third well. Mix thoroughly

8. Transfer the punctured and collapsed blastocyst to VitriBlast™ 1. Incubate **1.5-2 minutes** on the heated stage

9. Transfer the blastocyst to VitriBlast™ 2 by aspirating VitriBlast2 into the pipette tip, collect the blastocyst and transfer to VitriBlast™ 2 (well 2). Incubate for exactly 2 minutes on the heated stage

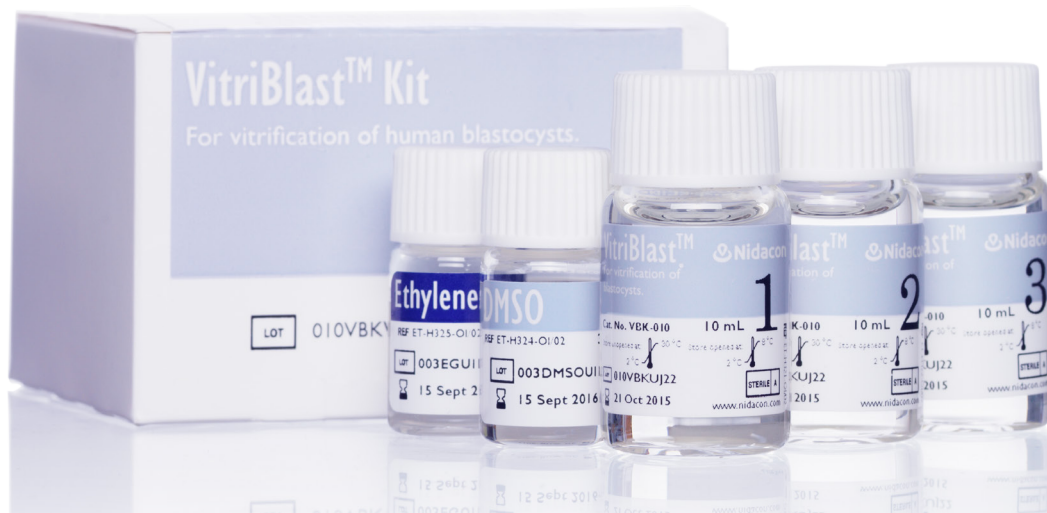
10. During the 2 minute incubation; prepare 2 x 10µL drops of VitriBlast™ 3 in the middle of the dish (see diagram). At the correct time, move the blastocyst by aspirating VitriBlast™ 3 from the well into the pipette tip, collect the blastocyst from solution 2 in the second well, and transfer it to solution no 3 in the 10µL droplet

11. Load blastocyst onto vitrification device in the smallest volume of VitriBlast™ 3 possible

12. The blastocyst must remain in VitriBlast™ 3 for **30-45 seconds**, including the time on the device

13. Plunge quickly into liquid nitrogen

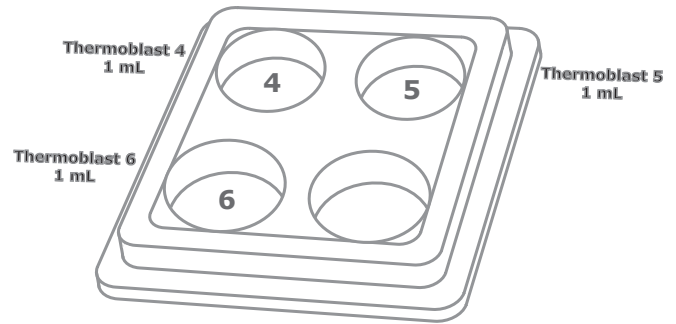
14. Transfer to storage in liquid nitrogen. Do not let the blastocyst come in contact with room tempered air during transfer



# How to use ThermoBlast™

## Reagents and Equipment

- ThermoBlast™ kit
- Sterile pipettes
- Culture dishes (NUNC 4-well)
- CO<sub>2</sub> Incubator
- Stopwatch or timer
- Heated stage
- Inverted microscope



## Directions for Use

### Warming Vitrified blastocysts with ThermoBlast™

1. Label a 4 well culture dish with the patient ID and each well with each solution number
2. Prepare the culture dish by adding 1 ml of ThermoBlast™ 4 into the first well, 1 ml ThermoBlast™ 5 to the second well and 1mL of ThermoBlast™ 6 to the third well
3. Incubate at 37°C in 5-6% CO<sub>2</sub> for **30 minutes**
4. Immerse the part of the device, containing the blastocyst in the surface of solution 4. Allow the blastocyst to fall off.  
Identify its presence in the well and incubate for **2 minutes** on the heated stage. Note that 2 minutes is for the total incubation time.
5. Transfer the blastocyst to ThermoBlast™ 5. Incubate for **3 minutes** on the heated stage
6. Transfer the blastocyst to ThermoBlast™ 6. Incubate for **5 minutes** on the heated stage
7. Transfer to culture medium
8. For a correct evaluation, wait 1-4 hrs before transfer, in order to allow the blastocyst to reexpand





# A part of something big

## ***We are important***

In a natural conception there are two parents involved in the process of creating life. In ART we have "several parents" and we all have an influence and are equally important. We are a part of creating life. We have an in-built meaning and purpose because we are doing a job that makes a difference. We all have the same aim and it is a joint effort.

## ***"A part of something big"***



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