Preliminary design of a passive-cooling dry ice transport box for long distance shipment of a cryopreservation patient

Introduction / Abstract

The transportation of cryopreserved patients is a complex topic. Adequate cooling needs to be ensured, specific cooling rates should be reached, while limiting the duration at temperatures known to favor ice crystal nucleation. In the past, international transport was done at a stable -78.5 °C (dry ice).

Here we propose a transport box design that allows cooling of the patients during transport. The design permits multi-day transport durations without temperature increases and without holding the body at static temperatures.

Prototype designs have been tested with a simulated body (80 kg) and between 69 to 83 kg of dry ice as a coolant, archiving continuously decreasing temperature for 164:55h and 185:15 h, respectively, before the dry ice was used up and temperatures started to increase.

Our data suggests that transportation while cooling is feasible, allows for sufficient safety margins, and can be done without complex technology.

This data should be considered preliminary. Further evaluation is needed to understand which option is best for different situations and what is feasible, especially in last minute cases. Some options are: transport at higher temperatures for short durations, transport at a static -78.5 °C (dry ice), transport while cooling but potentially not reaching critical cooling rates, using equilibrium vitrification solutions, transport at cryogenic temperatures over land or via dry shippers by plane, etc.

Materials and Methods

Design of Transport box

Multiple layer transport boxes designed for readily available construction material (size: 210 cm x 80 cm x 70 cm). For the outside to inside (vertically) the box is composed of the following lawyers

1. 12mm thick medium-density-fibre (MDF) boards
2. exterior insulation layer of 100mm
3. space/emergency blanket, silver layer towards the exterior
4. secondary exterior insulation layer of 20mm
5. third exterior insulation layer of 20mm
6. space/emergency blanket, silver layer towards the exterior
7. Coolant / Dry Ice
8. space/emergency blanket, silver layer towards the exterior
9. First interior insulation layer of 20mm
10. space/emergency blanket, silver layer towards the exterior
11. Second interior insulation layer of 20mm
12. space/emergency blanket, silver layer towards the exterior
13. Simulated body
14. space/emergency blanket, silver layer towards the exterior
15. secondary exterior insulation layer of 20mm
16. space/emergency blanket, silver layer towards the exterior
17. exterior insulation layer of 100mm
18. 12mm thick medium-density-fibre (MDF) boards

Laterally the box from outside to the center is composed of:

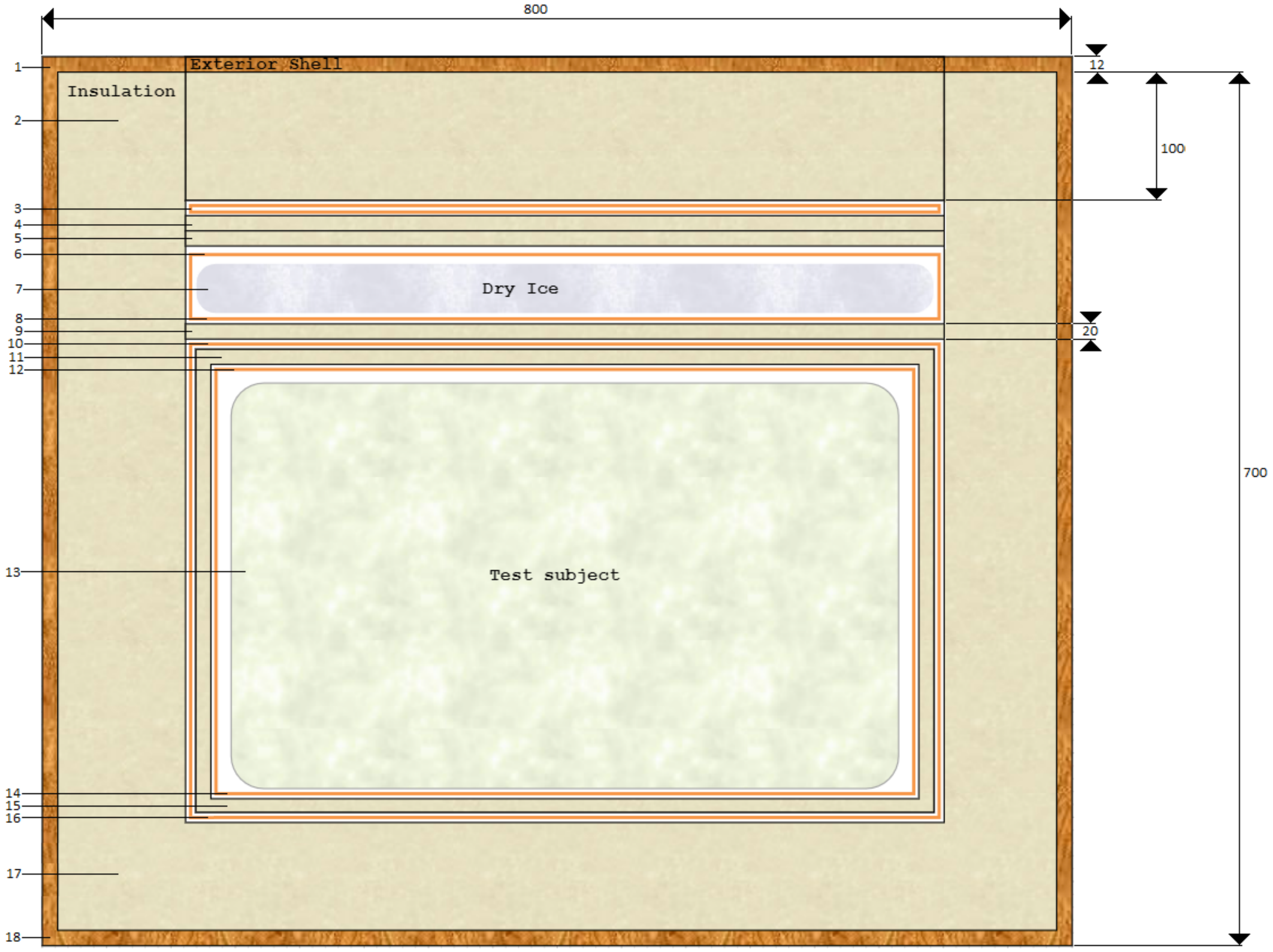
- 12mm thick medium-density-fibre (MDF) boards
- exterior insulation layer of 100mm
- space/emergency blanket, silver layer towards the exterior

  At the height of the dry ice there are no further layers. At the height of the test subject there is an additional layer of
- insulation of 20mm and space/emergency blanket
- silver layer towards the exterior
- dry ice

Every layer of space blanket/emergency blanket has a thickness of 12 microns and every insulation has a thermal conductivity of 0,036 W/mK).

Rifts between the insulation material are filled up with insulation foam.

MDF boards are assembled into a box with 4.8cm x 2.4cm lumber slats and standard construction screws. Primary insulation layer is glued to the MDF boards with construction glue. Gaps, edges and seams are filled in with standard insulation foam.



The box design allows for virtually any starting temperature and end temperature over different durations by altering the thickness of insulation.

Coolant / Dry Ice

As a coolant 80kg dry ice nuggets (diameter 1.6cm) are used. Nuggets are equally spread over the surface of the space blanket between the exterior and interior insulation.

Simulated Body

As a Simulated Body we used a combination of 108% v/w modified VM-1 cryoprotectant and a biophantom solution.

At the first experiment we used 80.00 kg of “bio phantom solution” and at the second experiment we used 24,43 kg of cryoprotectant and 56,66 kg of “bio phantom solution” .

108% v/w cryoprotectant

Water	31,4	%
Ethylene glycol	31,8	%
DMSO	25,7	%
9x m RPS-2	11,1	%
Glucose	7,9	%
KCl	2,1	%
TRIS	0,9	%
HCl	0,6	%

“bio phantom solution”

water	41,3	%
glycerin	31,3	%
ethylene glycol	25,7	%

Data Acquisition

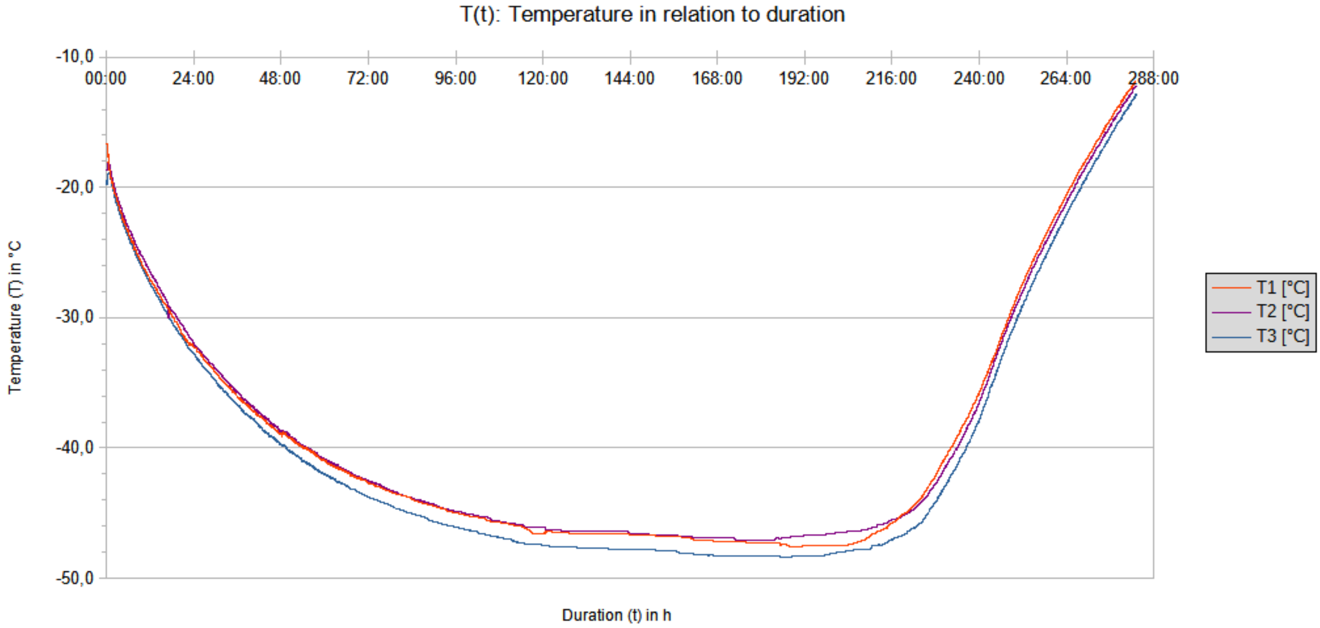
Four K-type thermocouple probes and a 4-channel data logger was used for data acquisition. Three thermocouples were positioned in the simulated body and the fourth probe was inside the other insulation layer. Raw data was saved in csv format and plotted in Excel.

Results

Experiments were started with the simulated body pre-cooled to -18.4°C and -18.6°C, respectively. After between around 176 hours (7.3 days) and 169 hours (7 days) temperatures reached the minimum and started to increase again.

<u>Test 1:</u>

Starting temperature:	-18.4 °C
Amount of Dry Ice:	68.52 kg
Thermocouple probe location 1-3:	Test subject
Minimum temperature:	-47.6 °C (average of Thermocouple 1-3)
Estimated average ambient temperature:	24.5°C
Minimum temperature reached after:	175h and 55min



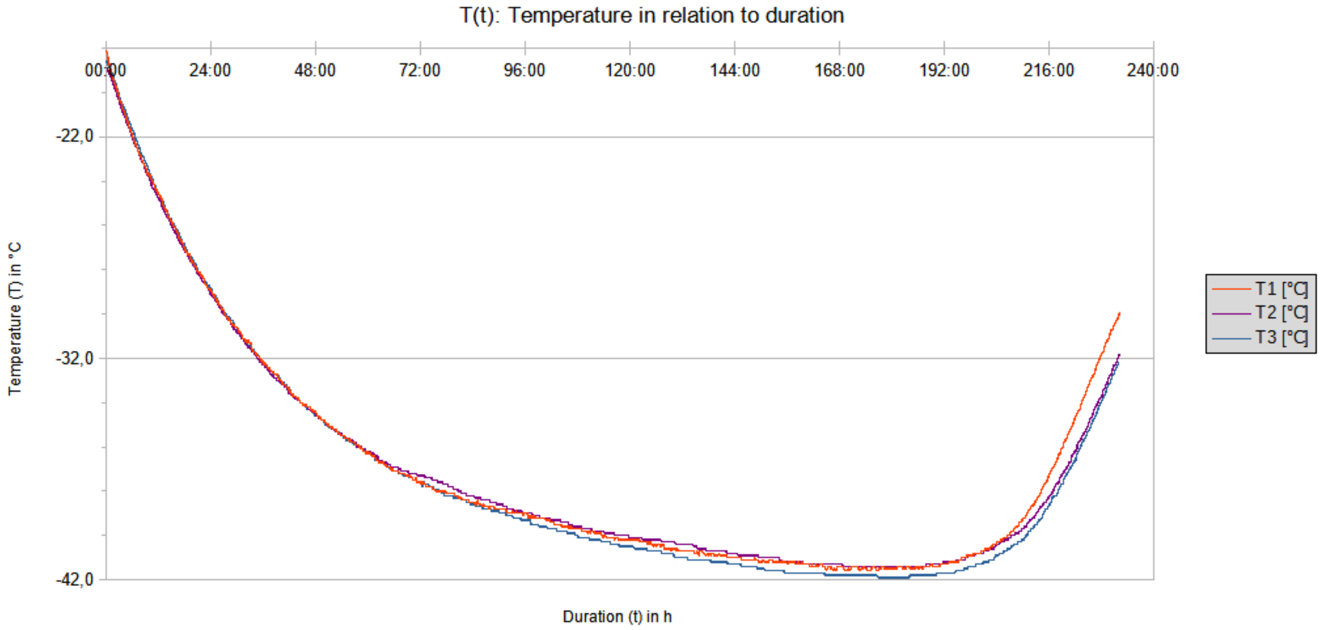
<u>Test 2:</u>

Starting temperature:	-18.6 °C
Amount of Dry Ice:	83.44 kg
Thermocouple probe location 1-3:	Test subject

Minimum temperature: -41.6 °C (average of Thermocouple 1-3)

Estimated average ambient temperature: 24.5°C

Minimum temperature reached after: 169 h



Different position of dry ice:

Positing the dry ice under the Simulated Body (position 10) is not recommendable. In one pre-test we encountered large temperature gradients inside the insulated transport box and even increasing temperatures in the simulated body WHILE dry ice was still available. Positioning the dry ice above the body archives advantageous thermodynamics inside the transport box.

Different dry ice form factor:

Increasing the size of the dry ice from micropellets (1.5mm) to pellets (3.0mm) or nuggets (1.6cm) improves handling but does not seem to have an impact on cooling rate or safe duration.

Discussion

Historically, during long-distance transportation patients destined for cryopreservation have been cooled with dry ice (-78.5 °C). Cooling with dry ice keeps the body in a temperature range (≈ -60

°C to -80 °C) that is known to be ideal for ice crystal nucleation, which should be avoided as much as possible.
Different solutions to these problems have been proposed such as cooling below the ice crystal nucleation temperature range with an electrical cooling system or liquid nitrogen-based cooling boxes as well as transportation while the body is still being cooled down and has not yet reached a static temperature.
Cooling to below ice crystal nucleation temperature range is technologically very complex which makes it very challenging to initiate transportation without delay should a last-minute case happen. Furthermore, air transport is significantly limited if any liquid nitrogen cooling is involved.
If field cryopreservation and cooling with dry ice during transportation is done, it is of vital importance that no temperature increase occurs after initial cooling.
Due to that, transportation while the cooling process is still ongoing has been avoided as it was not clear if sufficiently large safety margins can be achieved to avoid these potential temperature increases, e.g. due to delays in the shipment procedure.

To test if sufficiently large safety margins can be achieved, we conducted all experiments under unfavorable conditions with ambient temperatures between 21 °C and 26 °C. Similar conditions should only rarely occur during real-life transportation as cargo is regularly stored in air conditioned environments and cargo holds in planes maintain temperature well below 20° C.

If additional safety margins are required, the body can be pre-cooled to below the -18.4 °C to -18.6 °C range we used in these experiments and additional dry ice can be used.
For maximal security transportation should be done in cooled transport containers that likely increase the safe duration relevantly.

Please note: it is not yet known which transport option is the most preferable under different situations and expected transport durations.