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Features of a stem cell's cryopreservation

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Abstract

Stem cells are the unique source for the renewal of all kinds of tissue. The umbilical cord blood (UCB) is well known to be a rich source of stem cells with practical and ethical advantages, but it can be received only at child-birth. That is what raised the question about the preservation of stem cells from cord blood.

To solve this problem, cryopreservation in liquid nitrogen is used. The storage of biopreparations—after freezing—is performed in cryobanks in liquid nitrogen with cryoprotectant. DMSO is used as a cryoprotectant to prevent cell destruction at low temperatures. The received cells are poured into cryobags (20 ml) or cryoampules (4.5 ml), frozen with DMSO, and placed in liquid nitrogen (-196°C) for long-term storage. The process of cryoconservation consists of three stages: cooling from +20°C to +1°C using Coolmix (Biosafe, Switzerland), then freezing from +1°C to -100°C using Planer (UK), and finally transferring into storage with a temperature of -196°C.

For optimal freezing conditions, Planer controller freezer (Planer, UK) was used. The freezing program has been optimized for stem cells and the speed of freezing is from 1 to 3 K per minute, which avoids cell damage.

To avoid cross contamination, all samples of cord blood are placed in a specialized Dewar quarantine vessel until the results of an analysis on infections are received. Then samples are removed to "clear" Dewar vessels. There are currently more than 150 samples in the cryobank at the Stem Cell Bank Pokrovski.

According our results, after unfreezing the cell decreases only by $21.35 \pm 2.64\%$ ($p < 0,001$). The duration of UCB samples stored in a frozen state was 1 day and 1, 6, and 12 months; there was no difference in viability that depended on the storage duration. Therefore, the selected method of cryoconservation is rather effective and suitable for stem cells from umbilical cord blood. As a result, at the Stem Cell Bank Pokrovski the method of cord blood cryopreservation was optimized and adopted successfully.

Keywords: cryopreservation, stem cell, controller freezer, viability, crystallization point