Role of Antioxidants to Increase Survival and Viability of Cryopreserved Cord Blood Hematopoietic Progenitor Cells

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Hematopoietic progenitor cells (HPCs) have been used in practical medicine as an efficient therapy for diseases of various genesis [K. Ballen, 2013]. The antioxidants (AO) supplement to cryoprotective medium [H. Cheng, 2014] may improve the survival and viability of cord blood (CB) HPCs after cryopreservation.

The research aim was to determine the role of antioxidants in improving the survival and viability of cryopreserved CB HPCs. The fraction of nucleated cells isolated using the Polyclurcin was mixed with 25% DMSO up to the final concentrations of 5; 7.5 and 10% in the sample. The media were supplemented with following antioxidants: ascorbic acid (AA) in 0.05; 0.1; 0.15 and 0.2 mM concentrations; N-acetyl-L-cysteine (AC) in 5; 10 and 30 mM and glutathione in concentrations of 0.5; 1, 3 and 5 mM. Cryopreservation was carried out using the programmable freezer (Cryoson, Germany) with 1–3 deg/min rate down to –80°C, followed by immersion in liquid nitrogen.

Analysis of survival and viability of CB HPCs, cryopreserved with different concentrations of DMSO and without AO, demonstrated the reduction of these parameters in all the samples. The lowest cell loss was herewith observed in case of 7.5 and 10% DMSO, where the amount of CD45+CD34+7AAD–-cells was up to 65% and 40% did after transferring the cells to the conditions close to physiological ones.

A comparative analysis of AO revealed that AA in 0.1 and 0.15 mM concentrations and AC (10 and 15 mM) combined with 7.5% and 10% DMSO provided the survival of up to 80% of HPCs after cryopreservation, however after transferring to the conditions close to physiological ones this index was about 45–50% and 50–55% (with AA and AC use, respectively) as compared to the indices of AO free samples. The glutathione, supplemented in 1 and 3 mM concentration to cryoprotective medium with 7.5 and 10% DMSO enabled to preserve up to 90% of HPCs in a viable state after cryopreservation, and up to 70% of cells after transferring to the conditions close to physiological ones, that was 30% higher than the AO free samples. It should be noted that the results of HPCs cryopreservation in the solutions with 5% DMSO and the mentioned above efficient concentrations of AO were at the level of the data, obtained with 7.5 and 10% DMSO concentrations with no AO.

Thus, the AO supplement to cryoprotective medium, especially glutathione, can improve the CB HPCs resistance to cryopreservation factors, thereby testifying to an efficient application of this substance in developing the protocols for CB HPCs low temperature storage.