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Original article

CYTOPROTECTIVE EFFECT OF LYOPHILIZED HUMAN UMBILICAL CORD BLOOD SERUM UNDER SIMULATED CYTOSTATIC TOXICITY IN VITRO

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Background. The high systemic toxicity of modern chemotherapeutic agents necessitates the search for effective and safe biogenic cytoprotectors. In biology, fetal animal sera are considered classic cytoprotectors. Umbilical cord blood serum is a promising analog of fetal sera, but its utilization requires the development of effective preservation methods.
Objective. The aim of this work was to evaluate the cytoprotective effect of lyophilized and frozen human umbilical cord blood serum.
Materials and Methods. The study was conducted on the L929 fibroblast cell line. Chemically induced toxicity was modeled by culturing the cells with doxorubicin (2 μg/mL), cisplatin (25 μg/mL), and cyclophosphamide (1mg/mL) for 24 hours. The cytoprotective effect of 10% cord blood serum, either frozen at -20°C or lyophilized, was investigated. Culture status was assessed using light microscopy, the MTT assay, and flow cytometry (Annexin V-FITC/7-AAD staining).
Results. The cytostatic agents significantly altered monolayer morphology, reduced cell viability, and initiated apoptosis (particularly doxorubicin and cisplatin). In the cisplatin-induced toxicity model, the introduction of both types of sera resulted in the complete neutralization of the toxic effect. Under doxorubicin exposure, the sera significantly reduced apoptosis levels and improved monolayer morphology. The in vitro effect of cyclophosphamide manifested as a moderate inhibition of proliferation, which was completely overcome by the addition of the sera. No statistically significant differences were found between the biological activities of the frozen and lyophilized sera.
Conclusions. Lyophilized umbilical cord blood serum fully retains the cytoprotective potential of native frozen serum. It effectively protects L929 cells from the toxic effects of chemotherapeutic drugs (especially cisplatin), which justifies further research into its potential application as a supportive therapy agent in oncology and regenerative medicine.

Keywords: freezing; lyophilization; serum; umbilical cord blood; metabolic activity; cell culture; doxorubicin; cisplatin; cyclophosphamide.

Introduction

Severe toxic manifestations of modern polychemotherapy regimens remain the main limiting factor in the treatment of cancer patients. These manifestations lead to dose intensity reduction and a deterioration in the patients' quality of life. While synthetic organoprotectors (such as amifostine or dexrazoxane) exist, their clinical utility is frequently limited by their narrow spectrum of activity, pharmacological incompatibility, or severe side effects of their own [1]. Consequently, searching for multicomponent biopreparations of natural origin capable of exerting multi-target antitoxic and cytoprotective effects on healthy tissues is an urgent task in oncopharmacology [2, 3].

The use of umbilical cord blood components, which are rich in fetal growth factors and regulatory peptides, opens up new perspectives for creating effective therapeutic support agents in oncology aimed at protecting normal cells from cytostatic damage. Biogenic multi-component preparations, particularly fetal sera and umbilical cord blood serum (CBS), present a promising alternative due to their naturally balanced cocktail of polypeptide growth factors, extracellular matrix proteins, and endogenic antioxidants that act synergistically through multiple cytoprotective pathways [4, 5]. Fetal bovine serum (FBS) and its human counterpart, human umbilical cord blood serum, which contains numerous biologically active components, are classic cytoprotectors and biostimulators in biology and medicine. Furthermore, proteomic profiling reveals that the molecular composition of human cord blood serum and fetal sera closely mirrors the secretome profile of mesenchymal stem cells [6], however CBS can be obtained, stored, and transported

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much more cost-effectively and easily than whole cells [7–10].

However, a critical barrier preventing the widespread clinical application of human CBS is its high vulnerability to degradation and the absolute requirement for an uninterrupted cold chain during storage and transport. Standard freezing (-20°C to -80°C) incurs significant logistical costs and risks freeze-thaw-induced protein denaturation over repeated cycles. While lyophilization (freeze-drying) offers an elegant solution for room-temperature preservation without the need for additional refrigeration equipment, it subjects delicate tertiary protein structures to severe dehydration stress, potentially compromising their bioactivity [11–13].

In previous studies, the authors demonstrated the preservation and high biological activity of CBS and umbilical cord extract after freezing and lyophilization, as well as their capacity to stimulate the adhesion, proliferative, and metabolic activity of L929 cells [14–16]. However, the cytoprotective effect of lyophilized umbilical cord blood serum under the influence of chemotherapeutic agents remains undetermined. Currently, there is a fundamental lack of quantitative comparative data on whether secondary dried human CBS retains its specific anti-apoptotic and metabolic cytoprotection against distinct classes of DNA-damaging and anti-metabolic chemotherapeutics. Addressing this missing link is essential to validate lyophilized CBS as a standardized, shelf-stable biotherapeutic platform for supportive oncology.

The aim of this work was to evaluate the cytoprotective effect of lyophilized and frozen human umbilical cord blood serum.

Materials and Methods

The standard L929 fibroblast cell line, widely utilized in pharmacological research, was chosen for the study. The cells were obtained from the Bank of Biological Objects of the Institute for Problems of Cryobiology and Cryomedicine of the National Academy of Sciences of Ukraine. Cultivation was performed in DMEM High Glucose medium (BioWest, France) supplemented with 10% FBS (Lonza, Germany) and 1% antibiotic-antimycotic (BioWest, France) at 37°C, 5% CO_2, and 100% humidity. During the experiments, FBS was excluded from the experimental groups due to its composition being analogous to CBS.

Doxorubicin (D), cisplatin (CP), and cyclophosphamide (CPH) were selected as model chemotherapeutic agents due to their distinct mechanisms of action and widespread use in cytoprotector screening [15, 17, 18]. Given the varying literature data regarding drug concentrations and exposure times for different cultures, a preliminary concentration-screening step was performed for standardization. The target concentration was defined as one that causes significant visual changes in the monolayer within 24 hours of cultivation (changes in shape, cell detachment, decreased confluence) without causing total cell death, thereby allowing for potential culture recovery. The chosen concentrations were: D – 2 μg/mL (3.452 μM), CP – 25 μg/mL (80 μM), and CPH – 1 mg/mL (3.58 mM).

CBS was obtained upon the informed consent of parturient women following uncomplicated pregnancy and delivery. Cord blood was collected into dry tubes, allowed to stand until a clot formed, and centrifuged at 2500 rpm for 15 minutes using a "CM-3M" centrifuge (Micromed, Ukraine). The supernatant was aliquoted into 1 mL cryovials (Corning, USA), frozen to -20°C (for storage) and -80°C (for lyophilization), and stored for no more than one month. Samples were thawed before use in a water bath at 37°C.

The cytoprotective effects of serum frozen at -20°C (FS) and lyophilized serum (LS) were investigated. Lyophilization of the pre-frozen (at -80°C) CBS was carried out in an "Alpha 1–2 LDplus" freeze-dryer (Martin Christ Gefriertrocknungsanlagen GmbH, Germany) according to a previously described two-stage method. In the first stage, the sample temperature was maintained up to -35°C at a pressure of 30 Pa for 6 hours, followed by a gradual temperature increase to +35°C at a rate of 1°C/h [14]. Prior to use, the lyophilized samples were reconstituted to their original volume with distilled water and sterilized by filtration.

Cells were cultured in media of the following compositions: 1- DMEM without growth factors (to assess the effect of the absence of stimulating components on the cell line); 2 - DMEM with a cytostatic agent without biostimulators; 3 -DMEM with a cytostatic agent and FS; 4 - DMEM with a cytostatic agent and LS. The concentration of CBS in the media was 10% of the total volume, corresponding to the standard concentration of FBS.

Assessment of the cell status under various cultivation conditions was performed using a complex of previously described methods [13, 15]. Cell morphology, size, and monolayer characteristics were evaluated via light microscopy. Cell viability was determined using the MTT assay with an "SM600" spectrophotometer (Utrao, China) [16]. Apoptotic changes were analyzed by flow cytometry with Annexin V-FITC (BD Pharmingen, USA) and 7-AAD (BD Pharmingen, USA) staining using a BD FACS Canto II flow cytometer (Becton Dickinson, USA) [15, 19].

For culture visualization, an inverted microscope "NIB-100" (Delta Optical, Czech Republic) equipped with a "Sigeta MCMOS 3100" camera (Sigeta, China) and "ToupView v3.7" software (Hangzhou ToupTek Photonics Co., Ltd, China) was used. Flow cytometry data were analyzed using FlowJo software (v10, BD Biosciences, USA). Statistical analysis of the results

was performed using the "Past v.3.15" software package (University of Oslo, Norway). Data are presented as the arithmetic mean and standard deviation (M±SD). The homogeneity of intra-group variances was verified using the built-in Leven's test. To determine the statistical significance of differences between the four experimental groups, a one-way analysis of variance (One-way ANOVA) was applied, followed by pairwise multiple comparisons using the post-hoc Tukey's test. Differences were considered statistically significant at $p < 0.05$. To stylistically refine and structure the text, the Gemini generative artificial intelligence model (Google, June 2026) was utilized. The final text was thoroughly reviewed and approved by the authors in compliance with scientific ethics.

The study protocols were reviewed and approved by the Bioethics Committee of the Institute for Problems of Cryobiology and Cryomedicine of the National Academy of Sciences of Ukraine (Protocol No. 6 of November 28, 2023).

Results

Morphological evaluation after thawing and subsequent passage revealed that cells cultured without the addition of sera exhibited a typical polygonal shape, tight intercellular contacts, and monolayer confluence close to 100% (Fig. 1).

When cultured in the presence of D without biostimulators, significant detachment of cells, reduced adhesion, and impaired spreading were observed (Fig. 2, A). Cultivation with D combined with either LS or FS showed improved cell adhesion and spreading; however, the monolayer density remained at approximately 50%, failing to reach full confluence (Fig. 2, B, C).

The addition of CP resulted in a marked decrease in monolayer confluence and adhesion, cell detachment, and a substantial amount of cellular debris in the culture (Fig. 2, D). The introduction of either FS or LS into the culture medium alongside CP led to an increase in adhesion, proper cell spreading, the absence of debris, and an increase in monolayer confluence (Fig. 2, E, F).

The addition of CPH did not disrupt cell adhesion or spreading, but the monolayer confluence did not exceed 70% (Fig. 2, G). When cultured with CPH and either of the sera, full confluence close to 100% was observed (Fig. 2, H, I).

In the cell viability analysis, it was found that the addition of D led to a significant decrease in viability compared to the control, whereas the addition of both sera increased this parameter, though not to the control level (Fig. 3, A). The addition of CP also caused a significant reduction in cell viability, which was completely neutralized by the addition of the sera (Fig. 3, B). CPH caused a significant decrease in viability; however, the viability values upon adding the sera did not differ significantly from either the control or the cells treated with CPH alone (Fig. 3, C).

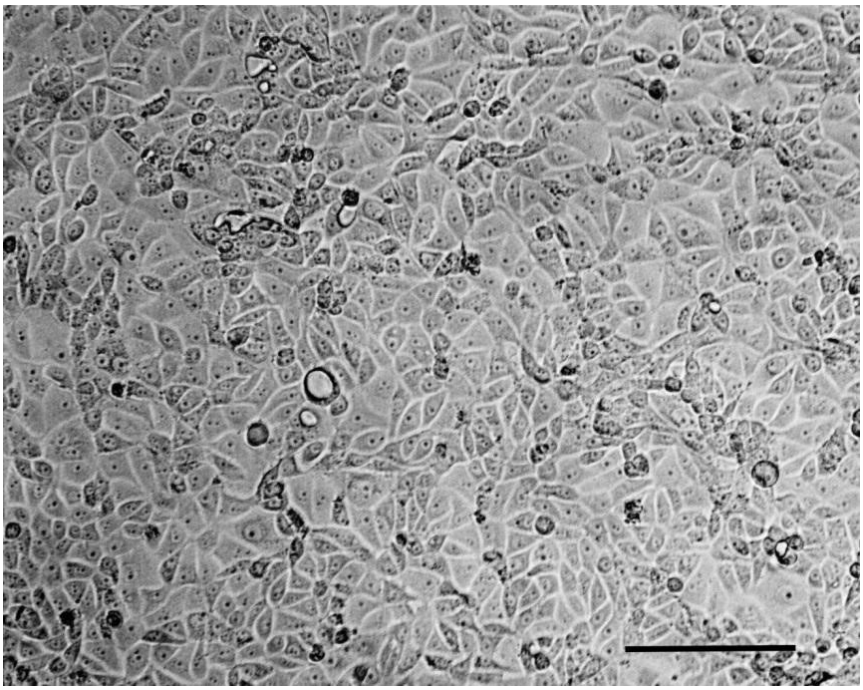


Figure 1. L929 cells after passage. Phase-contrast microscopy. Scale bar: 50μm

Flow cytometric analysis of apoptotic changes revealed that D induced significant apoptotic alterations, which were markedly reduced upon the addition of the sera, though they did not return to the control baseline (Fig. 3, D, G). Notably, the evaluation of early and late phases of apoptosis proved impossible because the cells exhibited intense, broad-spectrum fluorescence in the "red" channels of the flow cytometer after incubation with doxorubicin. This fluorescence overlapped with the 7-AAD spectrum and could not be resolved using the compensation function.

CP also induced significant apoptotic changes, which were completely neutralized after serum application (Fig. 3, E, H). CPH did not cause significant changes in the number of apoptotic cells in any of the groups (Fig. 3, F, I).

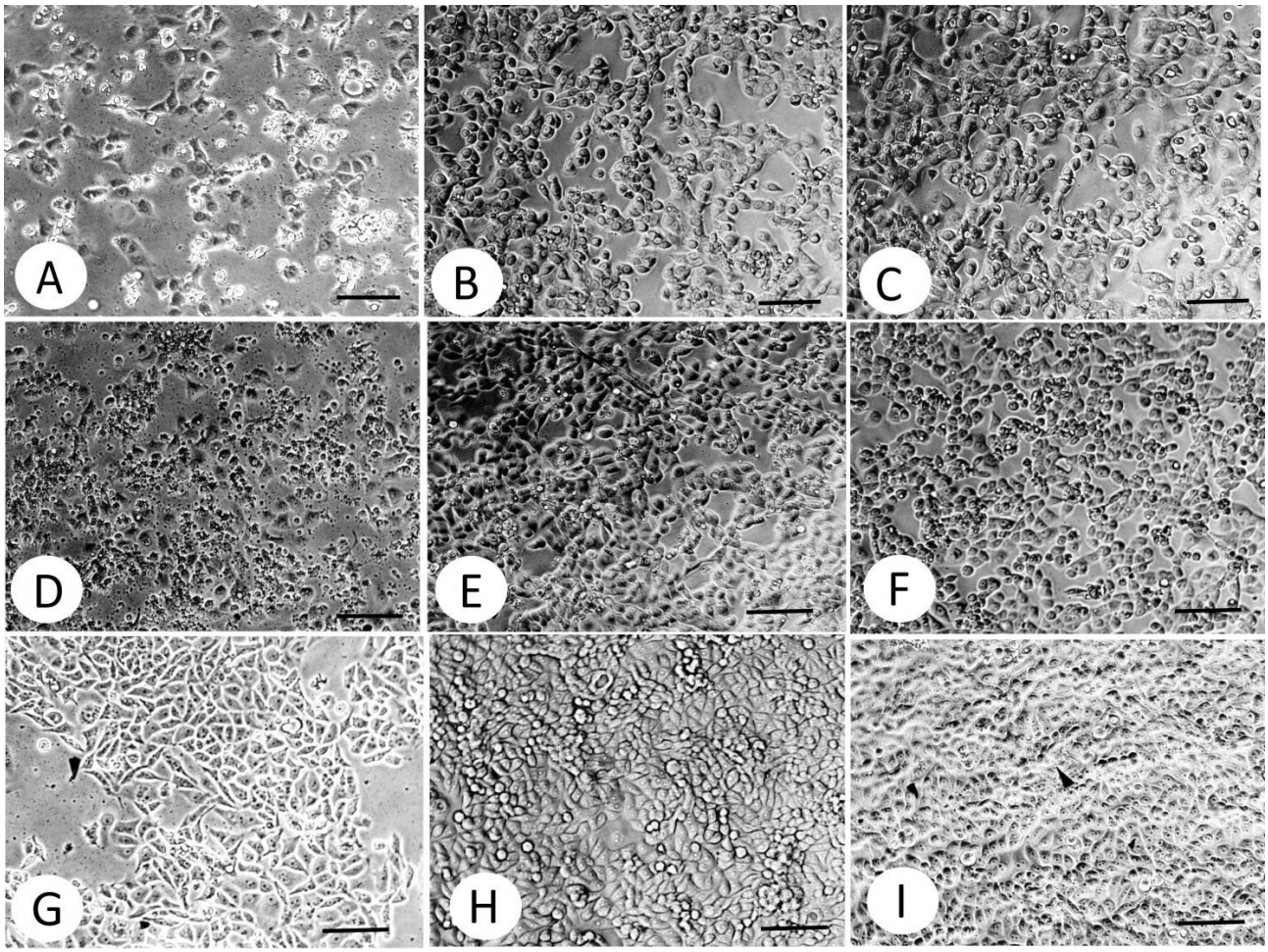


Figure 2. L929 cells under cytotoxic stress in different cultivation media. A – doxorubicin; B – doxorubicin with FS; C – doxorubicin with LS; D – cisplatin; E – cisplatin with FS; F – cisplatin with LS; G – cyclophosphamide; H – cyclophosphamide with FS; I – cyclophosphamide with LS. Phase-contrast microscopy. Scale bar: 50μm

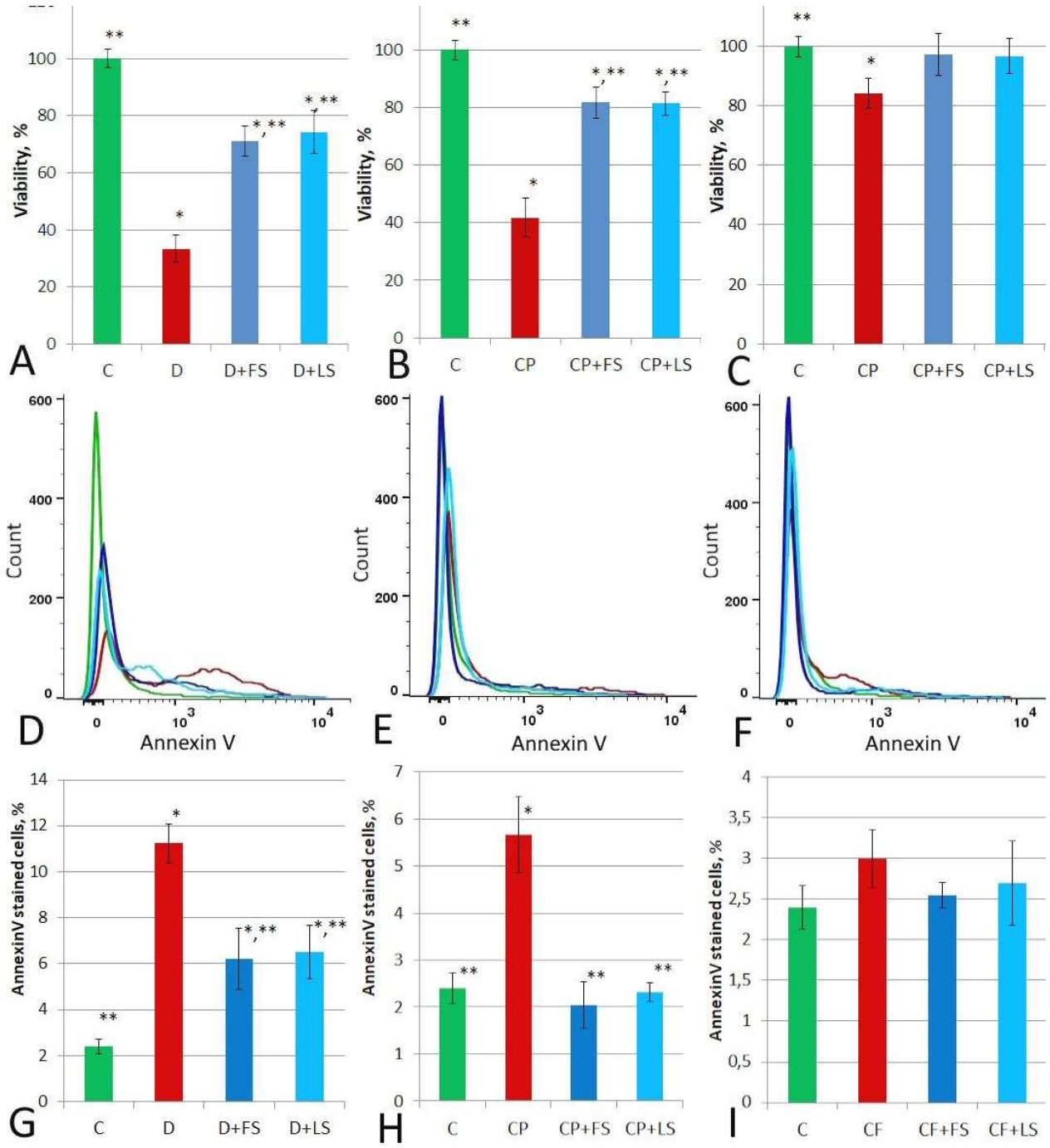


Figure 3. Toxicity parameters of L929 cells in different cultivation media. A–C – cell viability according to the MTT assay (M±SD); D–F – representative flow cytometry histograms with Annexin V; G–I – percentage of cells with apoptotic changes (M±SD). Green – cells without cytostatic agent (Control); red – cytostatic agent without serum; dark blue – cytostatic agent with FS; light blue – cytostatic agent with LS. A, D, G – doxorubicin models; B, E, H – cisplatin models; C, F, I – cyclophosphamide models. Statistical significance: * - $p < 0.05$ vs. control; ** - $p < 0.05$ vs. cytostatic without biostimulators (Tukey's post-hoc test)

Discussion

To critically evaluate the protective breadth of human CBS, we intentionally selected a panel of three chemotherapeutic agents-doxorubicin, cisplatin, and cyclophosphamide - representing fundamentally distinct classes of cytotoxicity (anthracyclines, platinum-based coordination complexes, and oxazaphosphorine alkylating agents, respectively). This selection allowed us to challenge the biosystem across different cell death execution modes: severe reactive oxygen species generation with topoisomerase II inhibition (doxorubicin), direct intra-strand DNA cross-linking with p53 cascade activation (cisplatin), and passive cyto-

static proliferation arrest (cyclophosphamide in non-metabolizing systems).

The choice of the L929 mouse fibroblast line served as a standardized, highly reproducible connective tissue model. In supportive oncology, non-transformed stromal and connective tissue cells are critical secondary targets of systemic chemotherapy toxicity, manifesting clinically as mucositis, tissue fibrosis, and impaired healing. Evaluating monolayer integrity, cell adhesion loss, metabolic activity (via MTT assay), and membrane phospholipid asymmetry inversion (via Annexin V/7-AAD profiling) provided a robust multi-parametric read-out capable of differentiating between reversible cytostasis, necrotic detachment, and programmed apoptotic death.

The results obtained demonstrate a distinct cytoprotective effect of both FS and lyophilized LS on L929 fibroblasts under simulated chemically induced toxic stress. Both FS and LS exhibited identical biological activity. This indicates the full preservation of the cytoprotective and regulatory molecules of CBS after freeze-drying, rendering LS a highly promising form for long-term storage and clinical application.

The varying patterns of viability and cell morphology recovery under the influence of CBS are evidently related to the specific primary mechanisms of cytotoxicity characteristic of the selected chemotherapeutic agents.

Doxorubicin is an anthracycline antibiotic that initiates cell death through DNA intercalation, inhibition of topoisomerase II, and the generation of reactive oxygen species, leading to profound oxidative stress and apoptosis [20]. In our study, doxorubicin caused pronounced cell detachment, reduced viability, and the triggering of apoptosis. The addition of FS and LS substantially improved the adhesive properties of L929 cells and reduced apoptosis levels, yet failed to reach intact control values. This partial rescue profile is strongly consistent with reports by other researchers [21, 22], who tested stem cell secretomes against anthracycline toxicity in cancer and non-cancer cells. This can be explained by the fact that doxorubicin causes irreversible double-strand DNA breaks. It is highly probable that CBS components (specifically fibronectin, vitronectin, and growth factors) [7] are capable of supporting the survival and adhesion of cells with partial damage but cannot completely counteract the direct genotoxic effect of D.

The most pronounced protective effect of FS and LS was recorded in the cisplatin model. Cisplatin forms intra- and inter-strand DNA crosslinks and activates the p53-dependent apoptosis pathway [23, 24]. The introduction of LS and FS to the culture medium resulted in the complete neutralization of cisplatin toxicity: cell viability recovered to control levels, adhesion and monolayer morphology were restored, and cellular debris was significantly reduced. The obtained effect is similar to the action of MSC [25,26]. The high efficacy of CBS may be attributed to the presence of antioxidant enzymes and cytokines (including TGF-β, FGF, and IGF-1) within its composition [5, 7]. These factors are capable of activating intracellular DNA repair systems and blocking caspase cascades.

Cyclophosphamide is an alkylating agent that requires in vivo metabolic activation by cytochrome P450 enzymes to form active metabolites [27]. Since metabolic activation is minimal in an in vitro system using the L929 line, the recorded effect (a moderate decrease in viability and a reduction of confluence to 70% without apoptosis activation) reflects the action of the unmodified cyclophosphamide itself. This action likely has the character of a temporary cytostatic block and the inhibition of proliferation.

Synthetic cytoprotective agents, such as N-acetylcysteine, amifostine, and dexrazoxane, have long been evaluated for mitigating drug-induced toxicity. In contrast, studies evaluating mono-target synthetic antioxidants under platinum stress report significantly lower protective magnitudes. For instance, Sancho-Martínez et al. [28] demonstrated that N-acetylcysteine primarily shifts cisplatin-induced cellular necrosis toward apoptosis by scavenging reactive oxygen species (ROS), but fails to prevent direct DNA adduct formation or fully restore metabolic viability, providing only partial 20-35% protection at therapeutic platinum concentrations. Similarly, Prieto González et al. [29] observed that while amifostine reduces ROS-mediated DNA strand breaks, its protective plateau is quickly reached, leaving persistent genotoxic damage.

The high cytoprotective activity of both frozen and lyophilized umbilical cord blood serum is based on its unique multicomponent composition. Human CBS contains higher concentrations of anti-inflammatory cytokines, neurotrophic factors, and endogenous antioxidants acting synergistically [7, 10]. Serum proteins ensure rapid spreading of fibroblasts, which is critical for their survival, since the loss of contact with the substratum triggers programmed cell death (anoikis) in adherent cell lines. CBS growth factors activate signaling pathways, suppress pro-apoptotic proteins of the Bcl-2 family, and block the activation of effector caspases.

Regarding property preservation during lyophilization: the absence of statistically significant differences between the effects of FS and LS proves that the selected two-stage freeze-drying regimen (-35°C at 30 Pa followed by secondary drying at +35°C) does not cause denaturation of protein growth factors and cytoprotective molecules, which is consistent with similar studies [11, 13].

Thus, LS has a cytoprotective effect similar to the action of MSCs and their secretomes, while being much easier to obtain and store, which makes it prom-

ising for further application in overcoming the complications of chemotherapy.

Conclusions

Thus, lyophilized human umbilical cord blood serum fully retains the biological potential of frozen serum and is capable of effectively protecting connective tissue cells from the destructive effects of chemotherapeutic drugs (exerting a particularly pronounced effect against cisplatin). This substantiates the prospects of its further investigation as a supportive therapy and regenerative medicine agent aimed at reducing the manifestations of local and systemic toxicity of cytostatics.

Interests disclosure

The authors declare no conflicts of interest.

References

[1] Hensley ML, Hagerty KL, Kewalramani T, Green DM, Meropol NJ, Wasserman TH, Cohen GI, Emami B, Gradishar WJ, Mitchell RB, Thigpen JT, Trotti A, von Hoff D, Schuchter LM. American Society of Clinical Oncology 2008 Clinical Practice Guideline Update: Use of Chemotherapy and Radiation Therapy Protectants. J Clin Oncol. 2009;27:127–45. DOI: 10.1200/JCO.2008.17.2627

[2] Scotté F, Taylor A, Davies A. Supportive Care: The “Keystone” of Modern Oncology Practice. Cancers. 2023;15:3860. DOI: 10.3390/cancers15153860

[3] Englisch A, Eissler K, Dannehl D, Englisch J, Hartkopf AD, Brucker SY, Grischke EM, Volmer LL, Engler T. Impact of chemotherapy dose capping on treatment intensity and survival in early breast cancer patients with high body surface area. Arch Gynecol Obstet. 2026;313:75. DOI: 10.1007/s00404-026-08317-2

[4] Obeagu EI. Umbilical cord blood: a comprehensive review of protective and restorative properties in clinical applications – a narrative review. Ann Med Surg. 2025;87:6618–25. DOI: 10.1097/MS9.0000000000003823

[5] Buchheiser A, Liedtke S, Looijenga LHJ, Kögler G. Cord blood for tissue regeneration. J Cell Biochem. 2009;108:762–8. DOI: 10.1002/jcb.22320

[6] Vizoso FJ, Eiro N, Cid S, Schneider J, Perez-Fernandez R. Mesenchymal stem cell secretome: toward cell-free therapeutic strategies in regenerative medicine. Int J Mol Sci. 2017;18:1852. DOI: 10.3390/ijms18091852

[7] Maharajan N, Cho GW, Choi JH, Jang CH. Regenerative therapy using umbilical cord serum. In Vivo. 2021;35:699–705. DOI: 10.21873/invivo.12310

[8] van der Valk J. Fetal bovine serum—a cell culture dilemma. Science. 2022;375:143–4. DOI: 10.1126/science.abm1317

[9] Subiran C, Kristensen SG, Andersen CY. Umbilical cord blood–derived platelet-rich plasma: a clinically acceptable substitute for fetal bovine serum? Fertil Steril. 2021;115:336–7. DOI: 10.1016/j.fertnstert.2020.10.027

[10] Prokopyuk VYu, Karpenko VG, Shevchenko MV, Safonov RA, Pasieshvili NM, Lazurenko VV, Prokopyuk OS. Experience in clinical application of cryopreserved placental derivatives: cells, tissue, membranes, extract, and cord blood serum. Innov Biosyst Bioeng. 2020;4:168–76. DOI: 10.20535/ibb.2020.4.3.215215

[11] Samyuktha AP, Ramesh Y, Chandra YPC, Penabaka V. A review of lyophilization. GSC Biol Pharm Sci. 2025;30:007–21. DOI: 10.30574/gscbps.2025.30.2.0035

[12] Izutsu K. Applications of freezing and freeze-drying in pharmaceutical formulations. In: Iwaya-Inoue M, Sakurai M, Uemura M, editors. Survival strategies in extreme cold: desiccation adaptation mechanisms and their applications. Singapore: Springer; 2018. p. 371–83. DOI: 10.1007/978-981-13-1244-1_20

[13] Prado NO, Lalli LA, Blanes L, Zanette DL, Aoki MN. Lyophilization of molecular biology reactions: a review. Mini Rev Med Chem. 2023;23:480–96. DOI: 10.2174/1389557522666220802144057

[14] Hoidina VS, Posokhov YO, Kot YG, Nakonechna OA, Prokopiuk VY. The impact of different lyophilization regimes on the protein composition of human cord blood serum. Biophys Bull. 2025;54:71–82. DOI: 10.26565/2075-3810-2025-54-05

[15] Kaverinska AI, Lazurenko VV, Zhelezniakov OY, Prokopiuk VY. Anti-apoptotic effect of umbilical cord cryoextract and its lyophilized form on L929 cells. Regul Mech Biosyst. 2024;15:980–4. DOI: 10.15421/0224144

[16] Kaverinska A, Shevchenko N, Osetsky A, Sukhodub L, Lazurenko V, Zhelezniakov O, Prokopiuk V. The effect of lyophilized and frozen umbilical cord cryoextract on L929 cell culture. Innov Biosyst Bioeng. 2025;9:3–12. DOI: 10.20535/ibb.2025.9.1.313718

[17] Kaludjerović GN, Miljković D, Momcilović M, Djinović VM, Mostarica Stojković M, Sabo TJ, Trajković V. Novel platinum(IV) complexes induce rapid tumor cell death in vitro. Int J Cancer. 2005;116:479–86. DOI: 10.1002/ijc.21080

[18] Gut I, Danielová V, Holubová J, Soucek P, Klucková H. Cytotoxicity of cyclophosphamide, paclitaxel, and docetaxel for tumor cell lines in vitro: effects of concentration, time and cytochrome P450-catalyzed metabolism. Arch Toxicol. 2000;74:437–46. DOI: 10.1007/s002040000141

[19] Prokopiuk V, Lupan M, Grygorova G, Posokhov Y, Maksimchuk P, Tryfonyuk L, Shevchenko N, Demchenko L, Kot K, Kurmangaliyeva S, Havranek O, Seminko V, Onishchenko A, Kot Y, Yefimova S, Tkachenko A. Erythrotoxicity of iron-doped cerium oxide nanoparticles is mediated by eryptosis. Toxicol Lett. 2026;421:111922. DOI: 10.1016/j.toxlet.2026.111922

[20] Radeva L, Yoncheva K. Doxorubicin toxicity and recent approaches to alleviating its adverse effects with focus on oxidative stress. Molecules. 2025;30:3311. DOI: 10.3390/molecules30153311

[21] Serras AS, Camões SP, Antunes B, Costa VM, Dionísio F, Yazar V, Vitorino R, Remião F, Castro M, Oliveira NG, Miranda JP. The secretome of human neonatal mesenchymal stem cells modulates doxorubicin-induced cytotoxicity: impact in non-tumor cells. Int J Mol Sci. 2021;22:13072. DOI: 10.3390/ijms222313072

[22] Yang Z, Yan F, Yuan J, Yang M, Wang J, You C, Ren K. Human umbilical cord mesenchymal stem cell-derived exosome ameliorate doxorubicin-induced senescence. Exp Cell Res. 2025;447:114450. DOI: 10.1016/j.yexcr.2025.114450

[23] Dasari S, Tchounwou PB. Cisplatin in cancer therapy: molecular mechanisms of action. Eur J Pharmacol. 2014;740:364–78. DOI: 10.1016/j.ejphar.2014.07.025

[24] Elmorsy EA, Saber S, Hamad RS, Abdel-Reheim MA, El-Kott AF, AlShehri MA, Morsy K, Salama SA, Youssef ME. Advances in understanding cisplatin-induced toxicity: molecular mechanisms and protective strategies. Eur J Pharm Sci. 2024;203:106939. DOI: 10.1016/j.ejps.2024.106939

[25] Ibrahim MA, Khalifa AM, Mohamed AA, Galhom RA, Korayem HE, Abd El-Fadeal NM, Abd-Eltawab Tammam A, Khalifa MM, Elserafy OS, Abdel-Karim RI. Bone-marrow-derived mesenchymal stem cells, their conditioned media, and olive leaf extract protect against cisplatin-induced toxicity by alleviating oxidative stress, inflammation, and apoptosis in rats. Toxics. 2022;10:526. DOI: 10.3390/toxics10090526

[26] Simovic Markovic B, Gazdic M, Arsenijevic A, Jovicic N, Jeremic J, Djonov V, Arsenijevic N, Lukic ML, Volarevic V. Mesenchymal stem cells attenuate cisplatin-induced nephrotoxicity in iNOS-dependent manner. Stem Cells Int. 2017;2017:1315378. DOI: 10.1155/2017/1315378

[27] Pulkova NV, Zyrina AN, Mnafki NA, Kuznetsova IM. Microfluidic chip as a tool for effective in vitro evaluation of cyclophosphamide prodrug toxicity. Bull Exp Biol Med. 2022;173:146–50. DOI: 10.1007/s10517-022-05510-6

[28] Sancho-Martínez SM, Prieto-García L, Prieto M, Fuentes-Calvo I, López-Novoa JM, Morales AI, Martínez-Salgado C, López-Hernández FJ. N-acetylcysteine transforms necrosis into apoptosis and affords tailored protection from cisplatin cytotoxicity. Toxicol Appl Pharmacol. 2018;349:83–93. DOI: 10.1016/j.taap.2018.04.010

[29] González EAP, Fuchs AG, Sánchez GS. Amifostine (WR2721) confers DNA protection to in vivo cisplatin-treated murine peripheral blood leukocytes. Dose Response. 2009;7:dose-response.08-026.Prieto. DOI: 10.2203/dose-response.08-026.Prieto

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ЦИТОПРОТЕКТОРНА ДІЯ ЛІОФІЛІЗОВАНОЇ СИРОВАТКИ КОРДОВОЇ КРОВІ ЗА УМОВ МОДЕЛЮВАННЯ ЦИТОСТАТИЧНОЇ ТОКСИЧНОСТІ IN VITRO

Актуальність. Висока системна токсичність сучасних хіміопрепаратів зумовлює потребу в пошуку ефективних та безпечних біогенних цитопротекторів. Класичними цитопротекторами в біології є фетальні сироватки тварин. Сироватка кордової крові є перспективним аналогом фетальних сироваток застосування якої потребує розробки ефективних методів зберігання.
Метою роботи було визначення цитопротекторної дії ліофілізованої та замороженої сироватки кордової крові.
Матеріали та методи. Дослідження проведено клітинах лінії L929. Хіміоіндуковану токсичність моделювали шляхом культивування з доксирубіцином (2 мкг/мл), цисплатином (25 мкг/мл) та циклофосфамідом (1 мг/мл) протягом 24 годин. Досліджували цитопротекторний ефект 10% замороженої до –20 ºС та ліофілізованої сироватки кордової крові. Оцінку стану культури проводили за допомогою світлової мікроскопії, МТТ-тесту та проточної цитометрії (забарвлення AnnexinV-FITC/7-AAD).
Результати. Встановлено, що цитостатики значно змінювали морфологію моношару, знижували життєздатність клітин та ініціювали апоптоз (особливо доксирубіцин та цисплатин). У моделі з цисплатином введення сироваток призвело до повного нівелювання токсичного ефекту. При дії доксирубіцину сироватки значущо знижували рівень апоптозу та покращували морфологію моношару. Ефект циклофосфаміду in vitro мав характер помірного гальмування проліферації, що повністю долалося додаванням сироваток. Статистично значущих відмінностей між біологічною активністю замороженої та ліофілізованої сироваток виявлено не було.
Висновки. Ліофілізована сироватка кордової крові повністю зберігає цитопротекторний потенціал нативної замороженої сироватки. Вона ефективно захищає клітини лінії L929 від токсичної дії хіміопрепаратів (особливо цисплатину), що обґрунтовує перспективність її подальшого дослідження як засобу супровідної терапії в онкології та регенеративній медицині.

Ключові слова: заморожування; ліофілізація; сироватка; кордова кров; метаболічна активність; культура клітин; стовбурові клітини; регенеративна медицина; доксирубіцин; цисплатин; циклофосфамід.