Extracorporeal blood circulation therapies: A comparative study on red blood cells of haemodialysis patients, therapeutic plasma exchange patients and healthy donors with advanced microscopes

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OBJECTIVES: Extracorporeal blood circulation (EBC) therapies are interventional methods of choice for the treatment of various diseases such as end-stage renal, autoimmune and neurological ones. During extracorporeal circulation blood cells can experience both mechanical stress and biochemical activation due to the incomplete biocompatibility of the physicochemical environment. In this work we comparatively studied, with two powerful microscopes, intact red blood cells (iRBCs) of patients subjected to EBC to explore new cellular diagnostic markers.

METHODS: We comparatively studied iRBCs coming from 7 haemodialysis (HD) patients, 7 therapeutic plasma exchange (TPE) patients and 7 healthy donors. The HD patients were subjected to standard 4-hour dialysis, thrice a week under various membranes (polysulfone, polyester-polymer alloy and ethylene-vinyl-alcohol copolymer) with Nikkiso® DBB-06 units. The TPE patients were subjected to a session that never exceeded 2 hours, under administration of colloid and crystalloid media (Human Albumin 5%, Hydroxethyl Starch 6% and saline NaCl 0.9%) with the Cobe® Spectra and Spectra Optia® units. The iRBCs refer to fresh RBCs that are deposited onto glass slides within 4 hours after collection without further treatment, such as washing with any kind of media (saline, phosphate-buffered saline etc); else the findings reported here are falsified. The iRBCs were studied with the Atomic Force Microscope (AFM) and Scanning Electron Microscope (SEM) that can morphologically survey both cells, at the micrometer scale (1 μm=10^4 m) and the cell membrane at the nanometer scale (1 nm=10^9 m) [1,2]. In particular, we studied iRBCs both prior (N=185 and 280 for HD and TPE patients, respectively) and after (N=172 and 278 for HD and TPE patients, respectively) the EBC session. A relevant sample of iRBCs (N=340) was studied for healthy donors.

RESULTS: Both AFM and SEM data revealed that the membrane of iRBCs has morphological abnormalities (MA) with typical size 100-1000 nm in HD patients, 100-2000 nm in TPE patients and 100-1000 nm in healthy donors, as shown in the representative Figures (1.a)-(1.b) (SEM) and (1.c)-(1.d) (AFM) for a TPE patient.

The occurrence of these findings in the healthy donors suggests that they possibly relate to the physiological aging of RBCs. In the patients, the change in the MA population during the EBC session is not statistically significant, with values 4% (p<0.05) and 17% (p<0.05) for the HD and TPE patients, respectively. When compared with the healthy donors, the MA population presents an increase that is statistically significant, 59% (p=0.0044) in HD patients, while it is not statistically significant, 20% (p>0.05) in the TPE patients, Figure (2). In the HD patients we observe a statistically significant correlation of MA population with the basic uremic markers such as Urea, Calcium, Phosphorous and Calcium-Phosphorous product (p<0.05), but not with creatinine (p>0.05).

CONCLUSIONS: The standard materials and methods employed today in EBC therapies exert minor mechanical stress and biochemical activation on the RBCs during the session in both HD and TPE patients, at least for the maximum duration of 4 hours and 2 hours, respectively studied here. The uremic environment degrades the membrane of RBCs in the HD patients probably accelerating their aging.