44.

EFFECTS OF PRESERVATION SOLUTIONS ON BLOOD

Piercarmine Panzera**,Luigi Greco*, Maria T. Rotelli*,Vito Lavolpe**, Anna M. Salerno**, Antonella Gentile*, Giorgio Catalano**, Giuseppe Cicco**, Vincenzo Memeo** [1]

1. INTRODUCTION

Preservation solutions in organ transplantation are designed to satisfy three principles: they have to rapidly wash out the blood and cool the organ, prevent cell swelling and interstitial oedema formation, and prevent excessive cellular acidosis [1, 2].

University of Wisconsin (UW) solution has been considered the gold standard for liver graft preservation. A new preserving solution, Celsior solution (CS)[3], is now available. This is an extracellular type of solution, initially used successfully in heart transplantation[4], and more recently shown to be an effective alternative to UW solution also in kidney[5], pancreas[6], lung[7], small intestine[8] and liver[9] transplantation.

Although clinical and experimental studies show a similar effectiveness in organ preservation, the Celsior solution formula theoretically promises some advantages. Its low potassium content should avoid glomerular capillary contraction due to calcium-associated contracture during kidney transplantation or endothelial impairment and biliary tract damage, that seems to occur in liver transplantation after a lengthy cold ischemia time using UW solution[10, 11].

CS contains oxygen radical scavengers, with reduced glutathione and mannitol and histidine, that might prevent oxidative injury caused by post-reperfusion free radicals, while the rapidly oxidized glutathione in shelf-stored UW solution definitely lacks this important property[11].

1
* Department of General Surgery and Liver Transplantation
Faculty of Medicine , University of Bari
** CEMOT
Centre of research in Hemorheology, Microcirculation and Oxygen Transport
University of Bari

Another advantage of the Celsior formula is that it does not contain macromolecules. The presence of a fairly high concentration of macromolecules in the suspension medium is known to be associated with red blood cell (RBC) aggregation[12].

It has been demonstrated that the Hydroxyethyl Starch (HES) contained in UW solution (50g/L) causes erythrocyte aggregation[13, 14]. The formation of erythrocyte rouleaux during organ perfusion might affect the blood flush-out in the microcirculation, ultimately affecting organ preservation and increasing the risk of primary dysfunction after transplantation[15].

The effect of UW solution on RBC deformability is still unclear. Some authors have shown a stiffening effect of UW solution on RBC[16], but this has not been confirmed by others [17].

The aim of this study is to investigate whether the CS solution presents any hemorheological advantage compared with UW solution.

2. MATERIALS AND METHODS

2.1. Preparation of Blood Samples

Venous blood samples were obtained from 17 healthy individuals aged 25 to 31 years by vein puncture from an antecubital vein and anticoagulated with EDTA. RBC were separated from the blood by centrifugation at 1400 *g* for 10 min and then resuspended in autologous plasma at a hematocrit of 38%. Then the blood was admixtured in vitro at 4°C with the UW or the CS at the following mixing ratios: blood: UW=3:1; 6:1; blood:CS=3:1; 6:1. Pure blood and blood diluted with Saline solution (SS) (blood:SS=3:1; 6:1) were used as controls. All the admixtures were prepared immediately before use. For the RBC deformability determination, 200 µl of the different admixtures were diluted in 5 ml of a polyvinylpyrovidone solution. The experiments were done at 22°C.

2.2. Determination of RBC Aggregation

RBC aggregation was quantified using a laser-assisted optical rotation red cell analyser (LORCA RR Mechatronics, Hoorn, The Netherlands)[17, 18]. This instrument consists of a laser light, a rotating thermostated cup and a video camera connected to an ellipse-fit computer program. Using the ektacytometric principle it quantifies the aggregation. The blood was brought under a shear rate of 500sec^{-1}, after which the shear was stopped at t=0. The backscattered intensity from the blood layer was measured for 120 sec after shear stop. The intensity decreased with erythrocyte aggregation. We considered the following parameters: the Aggregation Index (AI) and the time necessary to obtain 50% of the final aggregation ($t_{1/2}$).

2.3. Determination of Erythrocyte Deformability

To investigate RBC deformability, a dedicated software applied to LORCA was used, measuring the diffraction pattern of the RBC under various shear stresses in the range of 0.3 to 30 Pa. The Elongation Index (EI) was the parameter used to express RBC deformability at several shear stresses. An increased EI indicates greater cell deformability.