THE EXPRESSION OF HEAT SHOCK PROTEINS IN CELLS FROM SAPHENOUS VEINS – THE ROLE OF INCUBATION PROCEDURE

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Since saphenous vein was used in 1967 by Falvaro and Effler for coronary by-pass grafting, it has been exploited as a routine and basic biological material in cardiac revascularisation. Its preparation and collection prior to revascularisation influence the physiological properties of venous by-pass grafting. High expression of the inducible heat shock protein 70 kDa (HSP72) enhances resistance against proteotoxic stresses and is a marker of cell stress.

Saphenous veins were taken from five patients. 4cm long segments were cut off from each proximal part, and were then kept under standard conditions (medium 199, 37ºC) for 1h.

Every part was divided into eight 5mm long pieces and placed in different solutions at different temperatures (suitable for a given solution) for 1h incubation:

1. Cardioplegia, 4ºC
2. 0.9% NaCl, 20ºC
3. 0.9% NaCl, heparin (50 u/ml), 20ºC
4. 0.9% NaCl, heparin and papaverine (0.4mg/ml), 20ºC
5. 0.9% NaCl and blood from the patient 1:1 (heparin 3mg/kg b.m.), 20ºC
6. 0.9% NaCl and blood from the patient 1:1 (heparin 3mg/kg b.m.), 37ºC
7. medium 199, 44ºC – positive control condition
8. medium 199, 37ºC

After this, the segments were incubated under standard conditions for 4h, frozen to -140ºC, and RNA and soluble proteins were isolated using the TRIZOL (Gibco) method. Semiquantitative RT-PCR (Enhanced Avion RT-PCR kit, Sigma) was used for the expression of indicite HSP 70. Western-blot analysis (anti HSP 70 Stress Gen) was used to visualise HSP 70 translation.

Differences in the expression of HSP70 mRNA in particular patients were observed. Heat and mechanical injury enhanced the expression of HSP70. Heparin and papaverine decrease mRNA transcription in saphenous vein tissue.

This work was supported by KBN grant No. 4 P05C 006 17.