Effect of preconditioning of porcine hepatocyte during spheroid culture on following human plasma treatment

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Abstract

The performance of bioartificial liver (BAL) system primarily depends on the functional activities of the immobilized hepatocytes. One of the most promsing technologies retaining their functional activities is to use hepatocyte spheroids, which is known superior to single hepatocytes. In this study we tried to find out the role of preconditioning medium during spheroid culture and the effect on following treatment with human plasma.

The isolated porcine hepatocytes were inoculated into Petri dish at a density of 0.5 million cells/ml. At that time the hepatocyte cultures were preconditioned with twelve culture media consisting of different amino acid/vitamin compositions. All media contained 3.5 microM of hydrocortisone, 10 microg/ml of insulin, 10 microg/ml of transferrin, 10 ng/ml of sodium selenite, and 10 ng/ml of epidermal growth factor. After 24 hours of suspension culture, media were discarded and 1 mM NH_4Cl-added fresh frozen human plasma was added and treated for 24 hours.

The cell viability and functional activities after spheroid formation were very variable depending on each preconditioning medium. Hepatocyte spheroid cultures with amino acid/vitamin-fortified media showed not only high viability (by MTT, LDH assay) but also high functional activities (of ammonia removal, albumin synthesis). This phenomenon continued after human plasma treatment for 24 hours. So it suggested that preconditioning with a specific amino acid and vitamin composition can significantly affect the behavior of hepatocyte *in vitro*.