153 MECHANISMS DETERMINING AGGREGATION OF THE TRANSLATED HEMOGLOBINE MASS WITH EFFECT ON OXYGEN STIMULATING SIGNALS. T Sugita, P Payawong, RJ Aguirre, and RD Burg, Marston Bates Liver Research Center, Albert Einstein College of Medicine, Bronx, NY.

Transfused hepatocytes are permanently incorporated in liver parenchyma and retain normal function. Therapeutic strategies will benefit from a large transfused hepatocyte mass, which can be achieved when host hepatocytes are ablated. To define whether microbes could increase transfused hepatocyte numbers in the normal liver, we first analyzed the effect of partial hepatosplenectomy (PH), which induces coordinated release of growth factors and soluble signals. In one experiment, 2x10^7 transfused G26 HBV hepatocytes that produce serum HBsAg, as an excellent reporter of transfused hepatocyte mass, were injected intrasplenically into congenic C57BL/6 mice. After cell engraftment for 4 weeks, 50% PH was performed. However, instead of raising, serum HBsAg levels persistently decreased in recipients, suggesting no proliferation in transfused hepatocytes. Similarly, when hepatocytes were transfused into syngeneic C3H/HeJ mice, 24 days (D) post-transfusion, a 60% decrease in HBsAg levels occurred. The absence of hyperplasia in transfused cells was in agreement with our recent findings showing increased hepatocyte maturation, cell ploidy, and a decrease in cell number after PH. In conclusion, when hepatocytes were transfused into non-syngeneic recipients, no increase in hepatocyte mass occurred, and the cell number increased in the second week. This different hyperplastic response indicated that transfused cells escaped from hyperplastic signals affecting host hepatocytes when injected after PH. To determine whether growth factor stimulation could induce hyperplasia in transfused hepatocytes, we transplanted C3H/HeJ mice into syngeneic C3H/HeJ mice, which are highly growth factor-rich (SFBG, GeneTech Inc.) by systemic pumps for up to 1 week in mice, 4 weeks after transfusion of G26 HBV hepatocytes. Alternatively, HBV-infected hepatocytes were injected subcutaneously 1 day prior to transplantation to determine whether HBV-infected hepatocytes in syngeneic recipients would hyperplasia into transfused hepatocytes. The results of this study and the preliminary data suggest that transfused hepatocytes can proliferate in the normal liver in response to factors produced by host cells, growth factors, and no hyperplasia will be observed. Therefore, transfused hepatocytes will depend largely upon the rate of host hepatocyte loss and the mitogenic conditions in the recipient liver itself.

154 TRANSPLANTATION OFFULMINANT HAPLOID FAILURE RATS WITH A BIOARTIFICIAL LIVER (BAL) PROLONGS SURVIVAL AND IMPROVES HEPATOCYTE FUNCTION. D. Hase, J. Farnett, D. O'Meara, M. Alper, P. Farnett, S. An, H. Jones, Texas Tech University, Lubbock, TX; University of California at San Francisco, San Francisco, CA.

We have previously reported that rats in which fulminant hepatic failure (PHF) was induced by partial (50%) hepatectomy and right lobes necrosis died within 48 hrs without any signs of liver function (Regenolology 1996:24:124-9). We also found that PHF was induced by partial hepatectomy and that liver regeneration is potentiative to the PHF liver (Hepatology 1996:24:267A). In an attempt to determine if BAL treatment had improved survival and hepatic function, we injected 1x10^7 BAL fibroblasts into C57BL/6 mice on day 1. BAL fibroblasts were treated with the BAL (a group II control) or sham BAL. The data suggest that BAL fibroblasts can improve survival and liver function without significant side effects.

155 LONG-TERM CULTURE OF PRIMARY RAT HAPLOID CELL CULTURES IN A PACKED-BED BIOPROCESSOR USING FEBSAN-CELL DISCS. G Wang, A Strain, W Zhang, L Epstein, Y Chen, D Friedman, P Mc master, J Tzscholke, P Kaufmann, R & D Labs, New Brunswick Scientific, Edison, NJ; Liver Unit, Queen Elizabeth Hospital, Birmingham, UK; Lab of Cellular and Biochemical Toxicology, Rutgers University, Piscataway, NJ.

Primary hepatocytes following isolation from the liver lose many of their differentiated functions and survive for variable periods of time. The aim of this study was to determine the feasibility of adapting a large scale cell bioprocessor system previously used to culture CHO cells or hybridoma cells in batch, for use with long term stable functional primary hepatocyte cultures.

For hepatocytes (P 1 4x10^6) were attached to 0.5-mm polyethylene fibers housed in a 100 ml packed-bed basket within a 500 ml working volume. A reinculturing perfusion system with a gas phase and dissolved O2 control was used and cultures were maintained for up to 90 days. Viability was assessed by MTT assay, ornithine and albumin production. Other metabolic and biochemical parameters were monitored and morphology was assessed by EM.

In conclusion, cell attachment to discs was almost 100% after approx 2h MCT cell cultures indicated that viability at day 1 was stable at 100%. Long-term culture of hepatocytes was feasible and well maintained in this system. These data indicate that differentiated primary hepatocytes can be maintained in large scale cell bioprocessors for extended periods of time. The experimental design has numerous potential applications in the development of bioartificial liver systems as well as for long term drug metabolism and toxicity studies.