Title: Contextualizing Hepatocyte Functionality of Cryopreserved HepaRG® Cell Cultures

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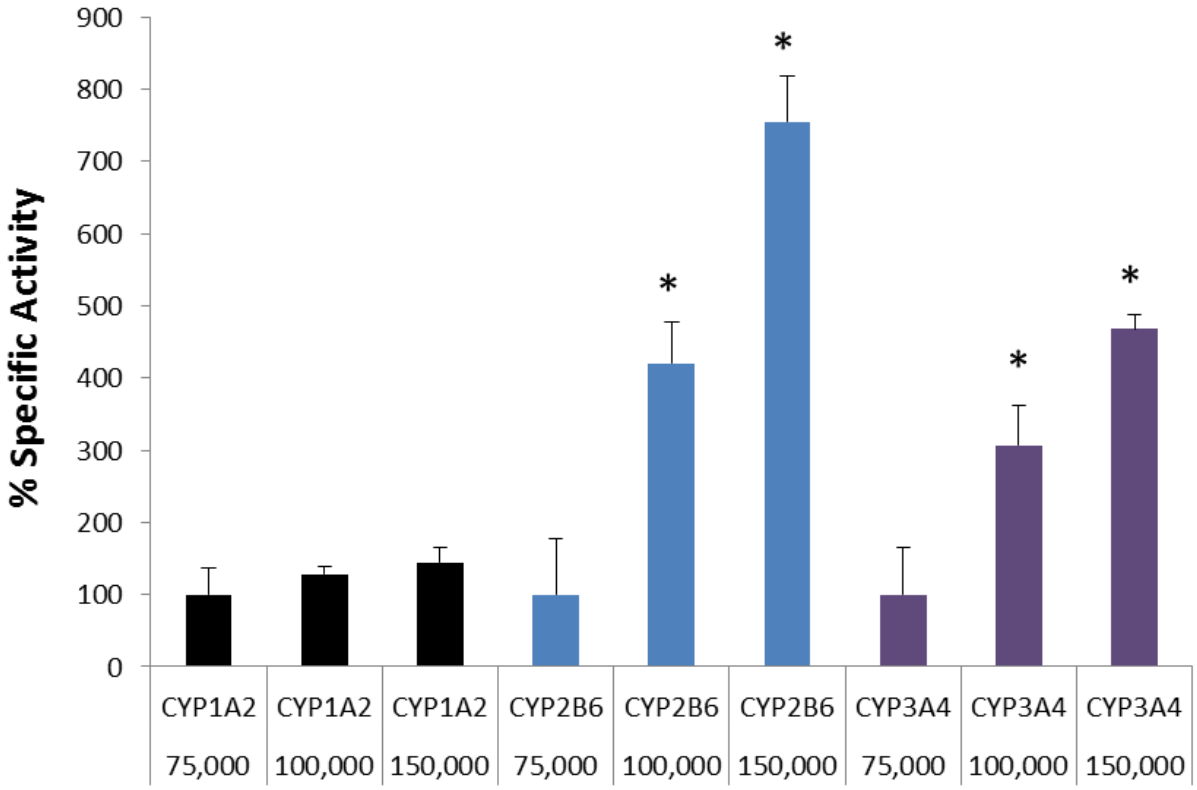
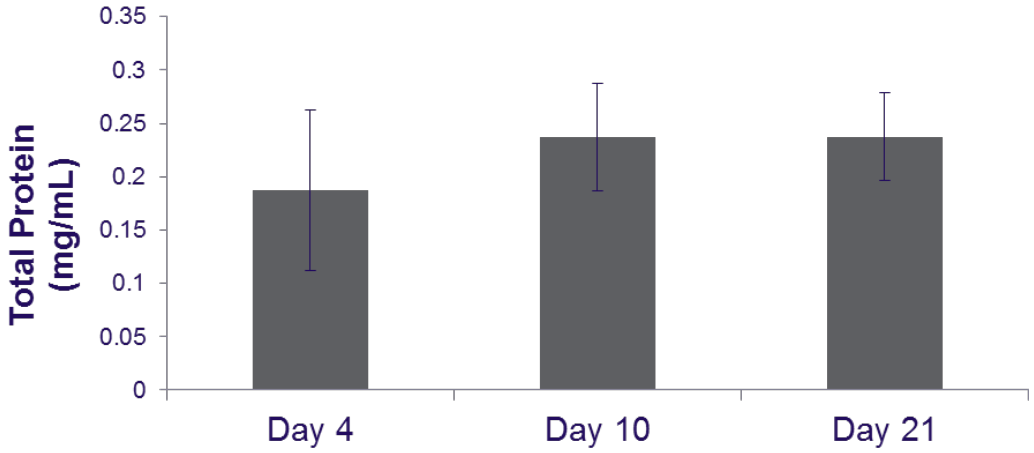


Supplemental Figure 1: Plating Cell Density Evaluation with Cryo-HepaRG. Cryo-HepaRG plating (seeding) cell densities (75,000, 100,000, and 150,000) were produced and evaluated via cell morphology observations and enzymatic activity measurements for CYP1A2 (phenacetin O-deethylase), CYP2B6 (bupropion hydroxylase), and CYP3A4/5 (testosterone 6ß-hydroxylase) enzymatic activities. Data represent the mean response of 3 independent wells and error bars reflect standard deviations of the mean responses. Asterisk (*) indicates statistically significant ($P<0.05$) using Tukey's pairwise comparison to 75,000 cells per well response.



Supplemental Figure 2: Total protein Assays with Cryo-HepaRG Cultures over Time to Evaluate Potential Cellular Proliferation. Due to the observed increases in enzymatic activity over time in culture with Cryo-HepaRG, total protein assays were employed after 4, 10, and 21 days in culture to assess the hypothesis that increased metabolic competence over time can be accounted for via cellular proliferation. Data represent mean responses of 3 independent well and error bars indicate standard deviations of mean responses. No statistically significant differences were observed using Tukey's pairwise comparison to the Day-4 control.