mouse albumin promoter and mouse alpha-feptoprotein enhancer are integrated.

Methods: The pLive vector was cloned with SV40 T antigen gene, and then the plasmid was microinjected into pronuclear-stage zygotes from C57/B6 mice. The incorporation of the transgene among progenies was evaluated using PCR, and the SV40T gene positive animals were continuously crossed with normal mice. Histological confirmation of HCC was carried out in 24 weeks old transgenic mice.

Results: Multiple large tumor nodules were shown on the liver surface of the transgenic mice. In timorous tissues, poorly differentiated HCC were observed in H&E stains, AFP and Ki-67 were overexpressed in immunohistochemistry, suggesting HCC was successfully induced by SV40T antigen under organ specific promoter.

Conclusion: Our study demonstrated the establishment of transgenic mouse model of HCC using SV40T gene and pLive vector system and this model could be applied to evaluate the anti-tumor efficacy of several anti-cancer drugs or target therapy in HCC.

Keyword: Transgenic mouse, SV40T, HCC

PE-096
Cytochrome P450 epoxygenase 2C9 inhibitor sensitizes cancer stem cell apoptosis under hypoxia

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Background & Aims: Cancer stem cells (CSCs) are believed to be the key drivers in tumor growth and therapy resistance. Although hypoxia has been shown to induce tumor progression through activating multiple signals such as modifying angiogenesis and invasion and help maintain multiple normal stem cell population, its roles in CSCs are largely unknown. In this study, we tried to evaluate the role of hypoxia in the proliferation and therapy resistance of CSCs.

Methods: Side population (SP) cell analysis and sorting were used to detect subpopulations that function as CSCs. To find out specific signals activated in CSCs under hypoxic condition, we compared gene expression of hypoxic CSCs with that of normoxic CSCs using microarray. Huh-7 cell, a human hepatocellular carcinoma cell line, doxorubicin, cytochrome P450 epoxygenase 2C9 (CYP 2C9) inhibitor sulfaphenazole were used in this study. Cell growth and apoptosis were assessed using MTS assay, and apoptotic and kinase signaling pathways were explored by immunoblot analysis.

Results: Hypoxia induced CSC proliferation and microarray showed that the expression of CYP 2C9, which was known to be implicated in hypoxia-induced endothelial cell migration and angiogenesis, was significantly increased in hypoxic CSCs compared to normoxic CSCs. Treatment with doxorubicin induced CSC apoptosis via the activation of mitochondrial apoptotic signals, including caspase-9 activation, whereas hypoxic CSCs were less sensitive to doxorubicin-induced apoptosis. The CYP 2C9 inhibitor sulfaphenazole pre-treatment sensitized hypoxic CSCs to doxorubicin cytotoxicity, and this was attributed to more profound augmentation of JNK and caspase-9 activation.

Conclusion: These results indicate that hypoxia promotes CSC proliferation and hypoxia-inducible CYP 2C9 expression is responsible for doxorubicin resistance in CSCs under hypoxia. Thus, the selective interruption of CYP 2C9 may therapeutically be implicated in sensitizing CSCs to anti-cancer treatment, particularly in the advanced case exposed to hypoxic environment.

Keyword: Cancer stem cell, Side population, Cytochrome P450 epoxygenase 2C9 inhibitor, Doxorubicin, hypoxia

PE-097
Association of polymorphism in microRNA-196a-2 with occurrence of hepatocellular carcinoma

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Background: Single nucleotide polymorphisms (SNPs) in microRNA (miR)-196a-2 have been proposed to contribute to the susceptibility of various human cancers including gastric, breast and lung cancer in Asian population. We aimed to determine whether polymorphisms of miRNA-196a-2 could affect clinical outcomes of hepatitis B virus (HBV) infection in a Korean population.

Methods: Genotypings were performed in 1,439 Korean subjects having either past or present evidence of HBV infection: 404 spontaneously recovered (SR) subjects as controls and 1,035 chronic HBV carriers. Chronic HBV carriers were composed of 313 chronic hepatitis B (CH) patients, 305 liver cirrhosis (LC) patients, and 417 hepatocellular carcinoma (HCC) patients.

Results: By direct sequencing, the polymorphism rs12304647A>C in the pri-miRNA region of miR-196a-2 revealed significant minor allele frequency (0.210). rs12304647A > C SNP showed significantly susceptible effects to the occurrence of HCC in CH and LC groups (OR=0.70, p=0.005 in a codominant model; OR=0.63, p=0.001 in a recessive model), as results from logistic analyses. Cox relative hazards model adjusting for age (<40, 40-60, >60 years old), gender, HBV e antigen status, and LC revealed that rs12304647A>C retained its association with HCC occurrence in a codominant model (RH=1.14, p=0.05) and in a recessive model (RH=1.44, p=0.03). However, rs12304647 of miR-196a-2 had no association with the clearance of HBV.

Conclusion: The CC homozygotes of miR-196a-2 were more susceptible to HCC and showed earlier occurrence of HCC compared with the AA or AC genotypes among CH and LC