flow, was mildly increased at 1 hr (2.0±2.3), then increased over the first week peaking at day 4 (5.8±2.6), then declined rapidly (0.9±0.7 at day 28). MB signal peaked earlier in normalized blood flow and blood volume, were not detected until day 14 (0.5±0.11 and 0.8±0.09, for blood flow and volume at 28 days). MB signal peak also preceded an increase in isometric muscle tissue P2O5 (normalized values of 0.24±0.06 and 0.47±0.10, at 1 hr and 28 days, respectively).

We conclude that CEU with microbubbles targeted to endothelial αvβ3 integrins can be used to non-invasively assess angiogenic responses in skeletal muscle. These results suggest that targeted CEU imaging of endothelial markers of angiogenesis may potentially be used for assessing intrinsic and therapeutic angiogenesis prior to changes in perfusion.

Thomas R. Porter, Derek Knapp, Lucia Venneri, Joseph Oberdorfer, John Lof, Patrick Iversen, Feng Xie, University of Nebraska Medical Center, Omaha, NE, AVI BioPharma, Inc., Oxnard, CA

BACKGROUND: Although perfluorocarbon containing albumin microbubbles (PESDA) can bind large quantities of antisease (AS) to the c-myc protein which promotes intimal hyperplasia, it is unknown how much c-myc synthesis wherein the intracoronary (IC) stent or balloon injury is actually suppressed by this intravenous (IV) targeting technique in the early period following vascular injury. To examine this, we performed high spatial phase-contrast imaging of AS to c-myc uptake and Western Blot studies of c-myc protein synthesis in corona arteries from eight pigs 50 minutes following IC stent and balloon injury (two vessels per pig). Pigs were treated with either direct IC anti-c-myc (4 milligrams), or the same dose of anti-c-myc IV bound or unbound to PESDA. IV PESDA containing anti-c-myc was given in the presence or absence of transcatheter 1 megahertz ultrasound (TTU) (pulsed wave at 0.6 W/cm2). RESULTS: C-myc protein synthesis in the injured coronary arteries (normalized to control vessels) was significantly lower when pigs were given IV anti-c-myc bound to PESDA irrespective of whether TTTU was concomitantly delivered (TABLE). Suppression of c-myc synthesis was comparable to direct IC injection. CONCLUSION: These data confirm that simply binding anti-c-myc to IV PESDA is a non-invasive method of targeting therapeutic genes to selective sites of IC balloon or stent injury and suppressing the formation of the c-myc protein which mediates intimal hyperplasia and restenosis.

*p<0.05 compared to other groups (ANOVA)

Direct IC AS IV AS/PESDA IV AS/PESDA IV AS+TTU

<table>
<thead>
<tr>
<th>c-myc protein ratio</th>
<th>0.94±0.26</th>
<th>0.88±0.11</th>
<th>0.88±0.28</th>
<th>2.11±0.28*</th>
</tr>
</thead>
<tbody>
<tr>
<td>anti-c-myc uptake (nanograms)</td>
<td>13±15</td>
<td>24±3</td>
<td>31±6</td>
<td>29±38</td>
</tr>
</tbody>
</table>

9:45 a.m.

802-3 Improvement in Epicardial Recanalization Rates With Transcranial Therapeutic Ultrasound and Intravenous Microbubbles Containing Ligands Which Attach to the Glycoprotein Ilb/Ilia Receptor on Activated Platelets

Thomas R. Porter, Joseph Oberdorfer, Lucia Venneri, John Lof, Feng Xie, University of Nebraska Medical Center, Omaha, NE

BACKGROUND: Intravenous (IV) perfluorocarbon containing microbubbles (PCMB) and transcranial ultrasound (TTU) were used to target PCMB to platelet receptors on platelet rich plasma. In addition, we examined the potential of using TTU to target PCMB to the c-myc protein. We hypothesized that TTU could target PCMB to the c-myc protein which promotes intimal hyperplasia. We examined the potential of using TTU to target PCMB to the c-myc protein which promotes intimal hyperplasia. We examined the potential of using TTU to target PCMB to the c-myc protein which promotes intimal hyperplasia. We examined the potential of using TTU to target PCMB to the c-myc protein which promotes intimal hyperplasia. We examined the potential of using TTU to target PCMB to the c-myc protein which promotes intimal hyperplasia. We examined the potential of using TTU to target PCMB to the c-myc protein which promotes intimal hyperplasia.

METHODS: We created a brain tumor model where 10^5 UI87MG cells derived from a human glioblastoma tumor cell line were embedded in gel and injected intracranially in athymic rats. Animals were sacrificed after either 14 or 28 days to obtain samples for each of the control groups. Control rats (n=6) were injected with gel alone. Targeted CEU imaging was performed 15 min following i.v. injection of control microbubbles (MB) or αvβ3 targeted microbubbles (MBT) bearing the disintegrin echistatin on their surface. Cerebral perfusion was assessed by CEU during continuous infusion of non-targeted microbubbles. A corresponding brain slice was processed for immunochemistry.

RESULTS: Tumors were -4-fold larger at day 28 compared to day 14 (p<0.05). Perfusion was detected within all tumors and was characterized by low microvascular blood velocity. On histology, 15-50 μm neovessels were abundant within the tumors and stained positively for endothelial PECAM-1. Dense staining for αvβ3 on the endothelium was found within tumors, especially at the outer margin, whereas minimal staining was seen in control regions. On CEU, MB signal intensity in gel tumor injection sites was low and similar to that in the contralateral hemisphere. Both at 14 and 28 days, MB signal in tumors was significantly (p<0.01) greater than that for MB in normal control regions, or for MB within the tumor. MB signal in tumors was greater at 28 versus 14 days (p<0.05), and was greatest at the peripheral margins of the tumors. MB intensity within tumor correlated well with microvascular blood volume derived from CEU perfusion imaging (r=0.83, p<0.01).

Conclusions: CEU with microbubbles targeted for endothelial αvβ3 can be used to assess tumor angiogenesis. These results have important implications for developing methods for early detection of primary or metastatic disease, or for developing novel anti-angiogenic therapies with microbubble delivery systems.

Raffi H. Bekeredjian, Shuyuan Chen, Peter A. Frenkel, Paul A. Grayburn, Ralph V. Snodin, University of Texas Southwestern Medical Center at Dallas, Dallas, TX

We have previously shown that ultrasound targeted microbubble destruction (UTMD) can augment expression of an adenoviral reporter in the heart. We now show that this method of delivery selectively delivers to non-invasive strategies (pancreas and brain) and can be extended to plasmid vectors. Recombinant adenoviruses or plasmids containing expression constructs of beta-galactosidase and luciferase were incorporated into albumin-coated perfluoropropane-filled microbubbles during their preparation. These bubbles were infused into the internal jugular vein of rats and destroyed with ultrasound while passing through the target organ. Organ activity was measured every 4 hours and analyzed for reporter gene activity. Luciferase activity in organs targeted with adenovirus was 104 times higher than in control organs. In contrast, liver activity was even higher. Histological examination revealed transgene-derived beta-galactosidase activity in subsets of brain neurons and pancreatic islets. Luciferase transfection with plasmids showed highly specific gene expression in the heart, 10-fold lower than with adenovirus, but with negligible activity in liver (figure). We conclude that ultrasound targeted microbubble destruction may be used to selectively deliver therapeutic genes to the heart, pancreas, and brain.

10:15 a.m.