Conclusions 1) MRA and adjunctive PTCA result in significant increases in coronary lumen dimensions, basal flow velocity and hyperemic peak coronary flow velocity. 2) Failure to increase coronary flow reserve after MRA and adjunctive PTCA is due to proportional increases in basal and hyperemic flow velocity and not to impaired coronary artery flow from microembolization.

995-22 The Effect of Perioperative Storage Solutions on the Long Term Vein Graft Function and Morphology

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It has been shown that suboptimal preparation of a vein graft prior to its insertion results in immediate morphological and functional damage to the endothelial cells but not to the underlying smooth muscle cells. However, little is known if such perioperative injury to the vein grafts may influence the subsequent development of intimal hyperplasia and smooth muscle cell contractility. This study examines the influence of storage in saline or Ringer's lactate on the development of intimal hyperplasia and vasomotor function in experimental vein grafts. Twenty-six NZW rabbits had a carotid vein bypass graft performed after the veins had been immersed (15 minutes) in either heparinized saline (Sal; n = 13) or Ringer's lactate (RL; n = 13) and each group was harvested after 28 days for either histology (n = 8) or functional studies (n = 5; four 5 mm rings/graft). Saline storage of the vein graft resulted in a 38% increase in the thickness of the intimal hyperplasia (113 ± 2 vs. 83 ± 2 μm, Sal vs. RL; mean ± SEM, $p < 0.05$) without a change in medial thickness (87 ± 5 vs. 86 ± 8 μm, Sal vs. RL; $p > 0.05$). There was no difference in the sensitivity to norepinephrine, serotonin and bradykinin between the two sets of vein grafts. The maximal contractile forces to serotonin and bradykinin were increased in the saline compared to Ringer's lactate stored vein grafts.

	Saline	Ringer's	p-value
Norepinephrine	0.88 ± 0.12	1.57 ± 0.20	<0.05
Serotonin	1.23 ± 0.15	0.37 ± 0.13	<0.01
Bradykinin	2.08 ± 0.11	0.52 ± 0.07	<0.01

Values are the standardized maximal contractile force (maximal contraction / contraction to 60 mM KCl) expressed as mean ± SEM.

Saline storage of the vein graft results in the increased development of intimal hyperplasia with an overall enhanced contractility but without changes in agonist sensitivity. This study places further emphasis on the need for good perioperative care of the vein bypass graft because it results not only in the previously documented short term problems but also in long term structural and contractile changes which may contribute to decreased graft patency.

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Wednesday, March 22, 1995, 9:00 a.m.–11:00 a.m.
Ernest N. Morial Convention Center, Hall E
Presentation Hour: 9:00 a.m.–10:00 a.m.

996-8 In Vivo Adenovirus-Mediated Gene Transfer via the Pulmonary Artery of Rats

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Gene transfer into the pulmonary vasculature may be a powerful technique both for the investigation of pulmonary pathophysiology and for the development of genetic therapies for pulmonary vascular disease. To evaluate the potential for in vivo pulmonary arterial gene transfer, we infused adenoviral vectors into the left pulmonary artery of Sprague-Dawley and cotton rats. Access to the left pulmonary artery was obtained either by a percutaneous, transcatheter approach, or via a thoracotomy and pulmonary arteriotomy. When using the thoracotomy approach, both the pulmonary arterial inflow and the pulmonary venous outflow were occluded during vector infusion and throughout a subsequent 20 minute dwell period. The success of gene transfer was assessed by staining for evidence of recombinant gene expression in lungs excised at time points ranging from 48 to 72 hours after virus infusion. *Results:* Using the surgical technique, pulmonary gene transfer was successful in 15% of surviving Sprague-Dawley and 30% of surviving cotton rats. Percutaneous pulmonary gene transfer was not successful. In those rats with pulmonary gene transfer, 1–8% of total pulmonary cells expressed the recombinant gene. Recombinant gene expression was found in endothelial cells (0.2–18% of total transduced cells), smooth muscle cells (0–3%), macrophages (1–7%), airway epithelial cells (2–50%), and alveolar epithelial cells (38–94%). Studies investigating the low rate of successful gene transfer in individual animals suggested that insufficient physical contact of the virions with pulmonary cells was the likely etiology. *Conclusion:* In vivo gene transfer into the rat pulmonary vasculature can be accomplished with adenovirus vectors. Pulmonary arterial infusion of the vectors results in low level endothelial cell transduction, with higher levels of gene transfer into nonvascular pulmonary cells.

996-9 Immune-mediated Response to Adenovirus Affects the Expression of Genes Delivered to Adult Rat Myocardium

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Direct injection of adenovirus (Ad) has been suggested as an efficient method for *in vivo* gene transfer into the myocardium and as a potential tool for gene therapy of myocardial disease. However, previous studies demonstrated that this method is limited by a sharp decline of gene expression after 1 week. To test the hypothesis that an immune-effector mechanism is involved in this decline we compared the results following injection of Ad-5 carrying the β-galactosidase(β-gal) gene into the left ventricular myocardium of athymic nude (NR; n = 7) versus Sprague-Dawley (SD; n = 7) rats. Volumes of 25–50 μl of a 1.0 × 10^9 PFU/ml solution were injected. Hearts were harvested and stained for β-gal activity at 30–35 days. β-gal activity was scored on a 1– 4+ rating system based on the number of stained cells observed per high power field (hpf).

	4+ >50 Cells/hpf	3+ >25 Cells/hpf	2+ >10 Cells/hpf	1+ 0–5 Cells/hpf
NR n = 7	5	0	2	0
SD n = 7	0	0	0	7

Score 4+ was significantly more frequent among NR vs. SD hearts (5 of 7 vs. 0 of 7; $p = 0.02$). Further, an inflammatory response was limited to the epicardium in NR as compared to SD in which there was also an intense inflammation with mononuclear cell infiltration and collagen deposition in the myocardium. The present model provides efficient gene expression for at least 35 days without significant inflammatory reaction. Our data suggests that an immune-mediated response to Ad can severely effect the expression of genes delivered by this virus.

996-10 A Novel Approach to Identifying mRNAs Differentially Expressed Under Hemodynamic Pressure Overload During Sheep Fetal Heart Development

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The adaptive response to pressure overload is different in neonatal hearts with congenital lesions than in adult hearts with acquired lesions. In adult hearts this response is characterized by an altered pattern of gene expression. Little is known about the changes in gene expression that occur in neonates. We have used a novel approach to examine these changes in expression of messenger RNAs in an *in vivo* model of fetal lambs heart after induced acute pressure overload. In ewes (which normally carry twin gestations) the main pulmonary artery of one fetal lamb was banded *in utero* for one hour to produce an acute pressure overload on the right ventricle (RV) while the twin underwent a sham operation. Total RNA was isolated and reverse transcribed from both the banded and control RV. The resulting cDNAs were amplified by the polymerase chain reaction technique using generalized oligonucleotide primers and the products displayed on sequencing gels. Comparative analysis of the differentially expressed cDNA bands from each condition revealed both up-regulation and down-regulation of several cardiac genes in the banded fetal hearts compared with the control lamb hearts. No morphological changes in the cardiac tissues were observed during this same time period. Eighty (300–500bp) differentially expressed cDNAs were identified. Twenty-three of these were analyzed by Northern blot and four were confirmed as being differentially expressed. Two of these were sequenced, identifying the 3'-untranslated regions which had no identifiable homology to the sequences in GENEBANK. This study reports on using an *in vivo* fetal lamb model to study changes in gene expression induced by hemodynamic pressure overload using a technique that identifies all differentially expressed messages.

Conclusion: This is the first study to examine alterations in fetal cardiac gene expression to hemodynamic overload in a large mammalian model.