4 ALTERATIONS IN POLYPEPTIDE AND GENE EXPRESSION DURING IN VITRO CYTODIFFERENTIATION OF SMOOTH MUSCLE CELLS. G. Liau and P.J. Wirth, Laboratory of Molecular Biology, Holland Laboratory, Rockville, MD 20855 and Laboratory of Experimental Carcinogenesis, National Cancer Institute, Bethesda, MD 20892.

When proliferating cultured smooth muscle cells (SMC) become quiescent, they express higher levels of the differentiation markers: smooth muscle (SM) specific isoactin and SM myosin. We have examined by two-dimensional (2-D) gel electrophoresis the expression of isoactin and other polypeptides in this *in vitro* model for the partial differentiation of SMC. Of the approximately 1,000 cellular polypeptides resolved by this method, only a discrete set of polypeptides exhibit an increased level of expression. Fifteen polypeptides have increased expression of between 5- to 20-fold. In addition, 2 polypeptides which are clearly expressed in quiescent SMC are undetectable in proliferating SMC. Other cellular polypeptides are expressed in equivalent amounts or are less expressed in quiescent SMC. The increased expression of certain polypeptides could be due to increased gene expression since increases in specific mRNA species are detected. The mRNA level for the alpha 1 (III) collagen and the alpha 1 (IV) collagen genes increased 5-fold in quiescent SMC. By contrast, mRNA for the alpha 2 (I) collagen and fibronectin are expressed at the same level in proliferating and quiescent SMC.

5 A NOVEL PHENOTYPE-TRANSFORMING PROTEIN ISOLATED FROM INJURED AORTIC SMOOTH MUSCLE CELLS. R.M. Hysmith and P.J. Boor, Department of Pathology, Chemical Pathology Division, University of Texas Medical Branch, Galveston, TX, USA.

We have found that when aortic smooth muscle cells (ASMCs) in a contractile phenotype are exposed to sublethal concentrations of the cardiovascular toxin allylamine, cells change to a synthetic phenotype (evidenced by ultrastructural morphologic characteristics including increased rough endoplasmic reticulum and decreased contractile elements) and then undergo a surge in cell proliferation. From the conditioned culture medium of allylamine-injured ASMCs, we have purified a 29-k Da cationic protein (FC-29) which has profound effects on the phenotypic expression of ASMCs. At concentrations $\geq$ 30 ng, FC-29 will change ASMCs from a contractile phenotype to a synthetic phenotype - as demonstrated by a loss of α-actin and by morphologic transformation to fibroblast-like cells. This transformation is apparent within 8 hrs after exposure to 30 ng of FC-29, and is irreversible after exposure to FC-29 concentrations $\geq$ 60 ng. At concentrations $\geq$ 100 ng, FC-29 will induce proliferation of ASMCs in a contractile phenotype. However, FC-29 has no apparent proliferative effect on ASMCs expressing a synthetic phenotype. When ASMCs of a synthetic phenotype are exposed to FC-29, there is a pronounced increase in differential "nodules" formation. FC-29 has no proliferative effect on aortic endothelial or adventitial fibroblast cells. When FC-29 was compared with PDGF, we found that PDGF would not cause either phenotypic changes or induce proliferation of ASMCs in a contractile state.

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6 TGF-β INCREASES C-SIS mRNA IN ENDOTHELIAL CELLS AND STIMULATES THE PRODUCTION OF EC-DERIVED MITOGENS FOR SMOOTH MUSCLE CELLS. Andrew C. Nicholson, Timothy A. McCaffrey, Babette B. Weksler, and David P. Hajjar, Departments of Pathology and Medicine, Cornell University Medical College, New York, NY.

Transforming growth factor-β (TGF-β) is a bifunctional regulator of growth which has been shown to inhibit the growth of endothelial cell (EC) and smooth muscle cell (SMC) monolayers, while stimulating the anchorage independent growth of SMC. Platelets are the major site of TGF-β storage and contain similar concentrations of TGF-β and platelet-derived growth factor (PDGF). TGF-β (5ng/ml) was added to human umbilical vein EC maintained for 24 hours in serum free media. Steady state levels of c-sis mRNA (which codes for the B-chain of PDGF) in cells exposed to TGF-β were twice that of control cells as measured by densitometric scans of Northern blot autoradiograms. Conditioned media (ECCM) from TGF-β treated EC stimulated more ^{3}H-thymidine uptake in target bovine aortic SMC than ECCM from control EC. Platelet degranulation at sites of endothelial injury may stimulate SMC growth directly through the release of PDGF and indirectly by TGF-β induction of endothelial-derived mitogens.