L’administration de 10-4M de DHLA dans le liquide de perfusion des cœurs isolés n’a pas conduit à une amélioration de la dysfonction contractile cardiaque qui caractérise la séquence d’IR. En revanche, dans nos expériences réalisées in vitro sur GR, en présence de DHLA, l’efflux potassique extracellulaire, qui signe la dégradation précoce des membranes érythrocytaires par le stress oxydant, a été diminué de manière significative et dose-dépendante. Le DHLA était plus efficace que l’AL aux mêmes concentrations et offrait une protection plus importante que celle du Trolox.

En conclusion, il apparaît que, dans nos conditions expérimentales, le myocarde ischémique n’est pas sensible au traitement par 10-4M de DHLA. En revanche, les membranes érythrocytaires sont protégées de manière très significative par ce composé antioxydant. Nos résultats ouvrent des perspectives quant à un possible bénéfice de l’utilisation du DHLA ou de l’AL dans certaines pathologies où le stress oxydant intéresse principalement le territoire vasculaire (diabète).

Jeudi 2 avril 2009, de 15h30 à 17h00
D — ANGIOPENSE, FACTEURS DE CROISSANCE, CELLULES PROGENITRICES

D001
PROKINETICIN RECEPTOR SIGNALING IN CARDIOVASCULAR FUNCTION: FOES OR FRIENDS?
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Prokineticins are secreted peptides that activate two receptors. We investigated the role of each receptor in heart by generating transgenic (TG) mice overexpressing PKR1 or PKR2 in cardiomyocytes. TG-PKR1 displayed an increased number of epicardial-derived progenitor cell (EPDCs), capillary density and vessels, without inducing any cardiomyocyte abnormalities. Coculturing EPDCs with H9c2 cardiomyoblasts overexpressing PKR2 upregulates prokineticin-2 as a paracrine factor that promotes EPDC differentiation into endothelial and smooth muscle cells, mimicking our transgenic model. Exogenous prokineticin-2 induces neonatal and adult EPDC differentiation. These prokineticin-2 effects were abolished in EPDCs from mice with PKR1-null mutation, indicating involvement of PKR1. This study provides a novel insight for possible therapeutic strategies aiming at restoring pluripotency of adult EPDCs to promote neovascularization by induction of cardiomyocyte PKR1 signaling. TG-PKR2 exhibit increased hypertrophic gene expressions and increased left ventricular end-systolic and diastolic diameters without cardiac dysfunction at the age of 24 weeks. TG hearts exhibit abnormal endothelial cell shape and ultrastructure, changed cellular distribution of a tight junction protein ZO-1, and vascular leakage in heart. The application of media conditioned by H9c2 cardioblast cells overexpressing PKR2 disturbed ZO-1 localization in H5V endothelial cells, mimicking the TG model. These findings showed that cardiomyocyte PKR2 signalling leads to eccentric hypertrophy in an autocrine regulation and impaired endothelial integrity in a paracrine regulation.

Our findings should facilitate the discovery of specific agonist and antagonist targeting PKR1 and PKR2 for possible use of treatment of ischemic heart diseases.

D002
SONIC HEDGEHOG INDUCES ANGIOGENESIS VIA RHO KINASE–DEPENDENT MMP-9 AND OSTEOPONTIN EXPRESSION
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Rationale and Objectives — The morphogen Sonic Hedgehog (Shh) is known to promote neovascularization in adults via indirect induction of pro-angiogenic cytokine expression by fibroblasts. Direct effects of Shh on endothelial cell (EC) function and angiogenesis, however, have not been characterized. Accordingly, we performed a series of in vitro and in vivo studies to evaluate the direct effects of Shh on EC and to investigate certain mechanisms by which Shh modulates angiogenesis.

Methods and Results — Our data disclose that Shh promotes capillary morphogenesis (tube length on Matrigel increased to 271±50% of the length in untreated cells), induces EC migration (191±35%) and increases EC expression of matrix metalloproteinase 9 (MMP-9) and osteopontin (OPN), which are shown to be essential for Shh-induced angiogenesis both in vitro and in vivo. Shh effects in ECs, however, occur while Gli-dependent transcription is not modulated. Furthermore, our studies show that changes in gene expression and EC migration are mediated by Shh induction of Rho. The Rho dependence of Shh-induced EC angiogenic activity is documented in vitro using dominant negative constructs for RhoA and Rho kinase (ROCK), showing that RhoA and ROCK blockade attenuates Shh induced migration and tube formation, as well as the Shh induced expression of MMP-9 and OPN. In vivo in the mouse corneal angiogenesis model pharmacologic inhibition of ROCK blocks Shh induced angiogenesis, confirming the ROCK dependence of this process. Furthermore, in MMP-9 and OPN null mice Shh induced angiogenesis also blocked, indicating that Shh induced angiogenesis is also dependent on MMP-9 and OPN expression.

Conclusion — These data elucidate an entirely novel, “non-classical” pathway by which Shh directly modulates EC phenotype and angiogenic activity.

D003
ANGIOGENIC EFFECTS OF ADENOSINE INVOLVE REGULATION OF VASCULAR ENDOTHELIAL GROWTH FACTOR AND SOLUBLE VASCULAR ENDOTHELIAL GROWTH FACTOR RECEPTOR -1
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Background — Vascular Endothelial Growth Factor (VEGF) and Hypoxia Inducible Factor-1α (HIF-1α) are both positive regulators of angiogenesis. The soluble form of VEGF receptor 1 (sVEGFR1 or