the structures of distant bacterial homologs. DISCUSSION/SIGNIFICANCE OF IMPACT: We are using these experimentally verified structures and functional data to answer questions about the mechanism of ferroportin iron transport, structural dynamics and the significance of mutations in ferroportin seen in different populations, especially the Q248H mutation found in Africans and black Americans with moderate to high prevalence.

4369

Reprogramming of vascular smooth muscle cells to multipotent progenitor cells contributes to progression of atherosclerosis*

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OBJECTIVES/GOALS: Our lab previously identified a population of vascular smooth muscle (SMC)-derived progenitor cells (AdvSca1-SM) which expand robustly in response to disease and can differentiate into multiple cell types. We now aim to define the role of these AdvSca1-SM cells in atherosclerotic plaque progression. METHODS/STUDY POPULATION: Goal one uses SMC lineage tracing mice and a model of atherosclerosis to track reprogramming of SMCs to AdvSca1-SM cells in the setting of disease. Arteries are analyzed using flow cytometry and immunofluorescence to quantify changes in number of mature SMCs and AdvSca1-SM cells. Goal two uses AdvSca1-SM lineage tracing mice with high cholesterol-induced atherosclerosis and plaque neovascularization. Arteries are analyzed to quantify expansion of AdvSca1-SM cells, subsequent re-differentiation into mature SMC, endothelial cells, or macrophages, and contribution to plaque neovascularization. Mechanistic findings from both goals are being investigated in diseased human coronary arteries. RESULTS/ANTICIPATED RESULTS: Flow cytometry from SMC lineage tracing mice revealed a 7- to 13-fold expansion of AdvSca1-SM cells in carotid arteries ($p<0.001$) and aortas ($p = 0.03$) after 6 weeks of western diet; no differences in macrophage numbers were observed. Additional SMC and AdvSca1-SM cell lineage tracing mice are on atherogenic diets to assess early and advanced atherosclerosis. We predict that AdvSca1-SM cells will contribute to macrophage accumulation as well as plaque neovascularization in the setting of severe atherosclerosis. Translational relevance of mechanisms driving SMC reprogramming and AdvSca1-SM cell contribution to plaque progression are being applied to studies of diseased human coronary arteries. DISCUSSION/SIGNIFICANCE OF IMPACT: Our data suggest a role for AdvSca1-SM cells in atherosclerosis. Ongoing work will clarify the mechanisms driving plaque-associated AdvSca1-SM expansion and define the ultimate fates of these cells. *In vivo* modulation of this process could provide the basis for future anti-atherosclerotic therapies. CONFLICT OF INTEREST DESCRIPTION: AD - CCTSI TOTTS TL1TR002533; SL - 18POST34030397 from the American Heart Association; AJ – no conflicts; KS - 1F31HL147393 from the National Heart, Lung, and Blood Institute, NIH; MM – no conflicts; RT – no conflicts; KSM – no conflicts; RAN - R01CA236222 from the National Cancer Institute, NIH, and 2018-03 from the Lungevity Foundation; and MCMW-E - R01 HL121877 from the National Heart, Lung, and Blood Institute, NIH, and 25A8679 from the Chernowitz Foundation.

4307

Role of Pre-pregnancy Uterine Natural Killer Cells in Human Embryo Implantation

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OBJECTIVES/GOALS: Human placentation requires complex coordination between maternal and fetal cell types but remains incompletely understood. **We hypothesize that uterine natural killer (uNK) cells, an immune cell type that increases in abundance during the implantation window, is essential for appropriate implantation and placentation.** METHODS/STUDY POPULATION: We plan to examine stromal cell (SC) decidualization, spiral artery remodeling, and EVT invasion, processes vital for early pregnancy establishment, in the presence or absence of secretory phase uNK cells. Fetal extravillous trophoblasts (EVTs) will be isolated from first trimester pregnancy tissue; maternal SCs, endothelial cells (ECs) and uNK cells will be obtained from secretory phase uterine tissue. SCs will be placed in monoculture and coculture with uNK cells and prolactin will be measured to evaluate decidualization. To study EVT invasion, we will utilize our novel "implantation-on-a-chip" device to determine how addition of uNK cells affects EVT migration through a collagen-matrigel matrix. In this system, we will also examine spiral artery remodeling with or without uNK cells via TUNEL staining. RESULTS/ANTICIPATED RESULTS: We anticipate that uNK cell addition to SCs will lead to a significant increase in SC prolactin levels, suggesting a role of uNK cells in endometrial decidualization. In vitro, we expect the addition of uNK cells will increase EC apoptosis and promote EVT invasion. DISCUSSION/SIGNIFICANCE OF IMPACT: Although decidual NK cells are known to participate in placentation, the role of pre-pregnancy uNK cells is unknown. uNK cell involvement in processes important for the earliest stages of pregnancy would provide a potential marker for abnormal placentation and offer avenues for intervention to decrease placentation associated perinatal morbidity.

4121

The beneficial, anti-fibrotic effects of chemokine receptor 2 and 5 antagonists on fat-exposed mouse primary hepatic stellate cells (pHSCs)

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OBJECTIVES/GOALS: Non-alcoholic steatohepatitis (NASH) is a leading cause of cirrhosis in the world for which no anti-fibrotic therapies exist. We hypothesized that BMS-22 and maraviroc (MVC), chemokine receptor 2 (CCR2) and 5 (CCR5) antagonists, respectively, would diminish the fibrogenic activity of "fat-exposed" murine pHSCs. METHODS/STUDY POPULATION: pHSCs were isolated from livers of 6 week old male mice following 4 weeks on a NASH-inducing choline-deficient high fat diet (CDAHFD, "fat-exposed") or standard diet (SD) and passaged *in vitro*. Early passage (6-12) pHSCs were plate-adhered and TGF-b-treated (10ng/mL) to maximally activate their pro-fibrogenic genes, *collagen 1α1*

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