Identifying and treating *ROBO1*$^{-ve}$/*DOCK1*$^{+ve}$ prostate cancer: An aggressive cancer type highly prevalent in African-American patients

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Grant Funding: R01CA193739

Metastasis is the principal cause of death in prostate cancer (CaP) patients. The prognosis is very poor for metastatic CaP. On comparison, African-American patients exhibit high mortality rates than Caucasian counterparts. The failure of therapies for metastatic CaP is observed very high in patients belonging to African-American race. There is a need to identify new target-based therapies which could be either (i) beneficial to patients regardless of race or (ii) treat high-risk patients such as African-Americans who fail standard therapies compared to Caucasians. This requires identification of molecular pathways which are either (i) preferentially active in African-Americans or (ii) highly active in both races. We posit that the loss of *ROBO1* in advanced stage CaP contributes to the disparity observed between African-Americans and Caucasian. We conducted a comprehensive analysis of race-based primary and metastatic tumors for ROBO1 and its downstream targets (RAC1, DOCK1, E-Cadherin, and β-catenin). We show that ROBO1 expression is progressively lost with the increasing stage of CaP in African-American patients. Next, in order to explore the underlying mechanism that leads to *ROBO1* loss, we analyzed matching tumor DNA from patients and found that the *ROBO1* promoter is methylated in the majority of African-American cases in agreement with the CaP stage. In addition, we found that ROBO1 loss is associated to increasing RAC1 activation in tumors and representative cell models. We show that loss of ROBO1

results in increased DOCK1 activity that in turn induces Rac1 signaling in CaP cells. We hypothesized that targeting DOCK1/Rac1 network could be an ideal strategy to treat metastatic CaP, particularly in *ROBO1*$^{-/-}$ cases. To test the hypothesis, we tested the therapeutic efficacy of a selective inhibitor of DOCK1/Rac1 interaction in ROBO1^{+ve} and ROBO1^{-ve} CaP cells. We observed that DOCK1-inhibitor treatment significantly decreased the RAC1 activation and β-catenin activity in metastatic CaP cells. Notably, inhibition of DOCK1/Rac1 was observed to significantly decrease the motility and invasiveness of aggressive metastatic CaP cells. Furthermore, DOCK1-inhibitor significantly blocked the potential of metastatic CaP cells to extravasate (cross-over) through the endothelial lining (constituted of HUVEC cells).

These data suggested the significance of DOCK1/Rac1 pathway in a metastasis of CaP particularly in African-Americans. We conclude that the ROBO1/RAC1 ratio in tumor biopsies could be developed as an early biomarker of metastasis in African-American patients. We suggest that DOCK1-inhibitor therapy would be more beneficial to patients who exhibit *ROBO1*$^{-}$ tumor, and African-American patients are such as group who likely to benefit this therapy.