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P115- TRPC6 binding to and activation of calpain regulates podocyte motility

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Mutations in transient receptor potential channel 6 (TRPC6) are associated with an inherited form of focal segmental glomerulosclerosis (FSGS). Upregulation of TRPC6 expression has also been identified in several acquired forms of proteinuric kidney disease. Despite widespread expression, patients with TRPC6 mutations do not present with any other pathological phenotype suggesting that this protein has a unique role within the target cell for FSGS, the kidney podocyte. Although most TRPC6 mutations are reported to cause changes in calcium dynamics, it is still unclear how these result in a podocyte specific phenotype. TRPC6 plays an important role in the motility and adhesion of podocytes. Here, we demonstrate that in a mouse model, knockout of TRPC6 results in increased glomerular permeability and alterations to the structure of the glomerular filtration barrier. Furthermore, we show that in comparison to wild type cells, TRPC6 knockout podocytes are less motile, more adhesive and have an altered actin cytoskeleton and lipid raft composition. Finally, we show that TRPC6 binds to ERK1/2 and the actin regulatory proteins caldesmon and calpain 1 and 2. Calpains are calcium dependent cysteine proteases which have critical functions in controlling the podocyte cytoskeleton and hence cell adhesion and motility via cleavage of paxillin, focal adhesion kinase (FAK) and talin. Knockdown or expression of the K874*, but not the gain of function G019S, disease causing mutant of TRPC6 results in the mislocalization of calpain and significant down-regulation of calpain activity leading to altered podocyte cytoskeleton, motility and adhesion. Importantly, our data suggests that the physical interaction between TRPC6 and calpain plays an important role in the podocyte, independent of TRPC6 channel activity.