



Update on Adiponectin KO Diabetic Mice and Development of Inducible Nox4 tg Mice

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Sharma

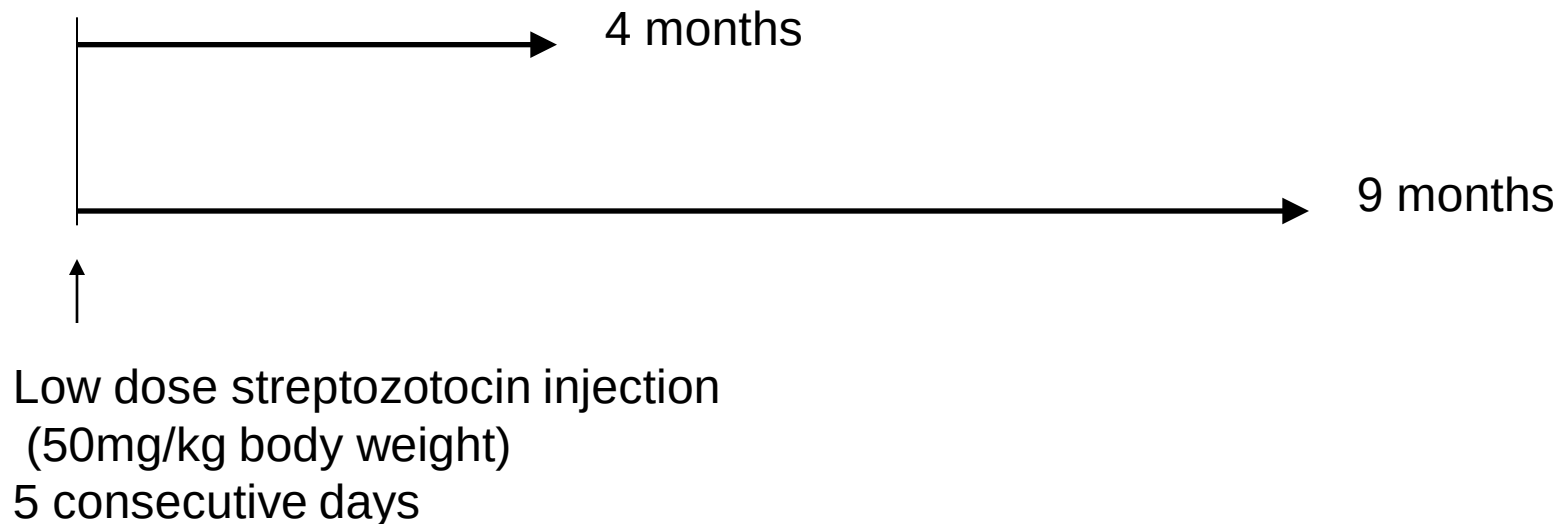
Center for Renal Translational Medicine
University of California, San Diego

Hypotheses

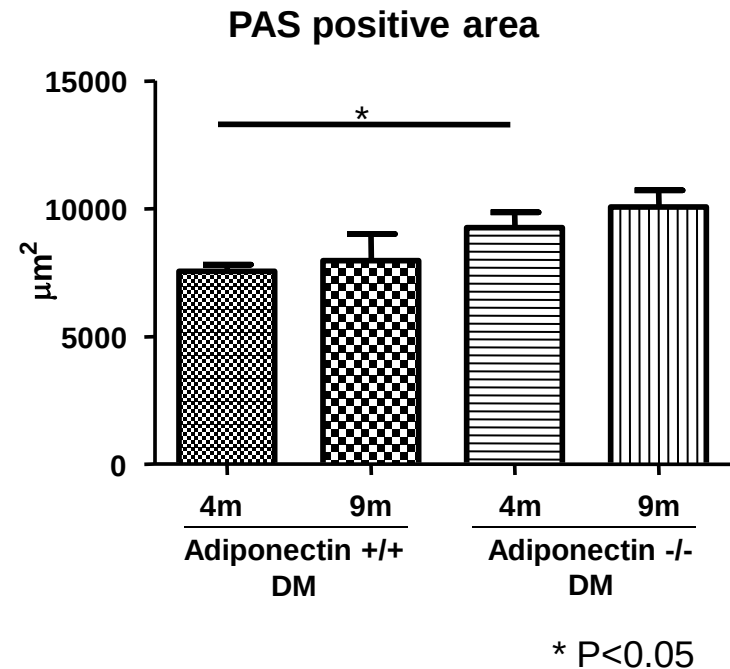
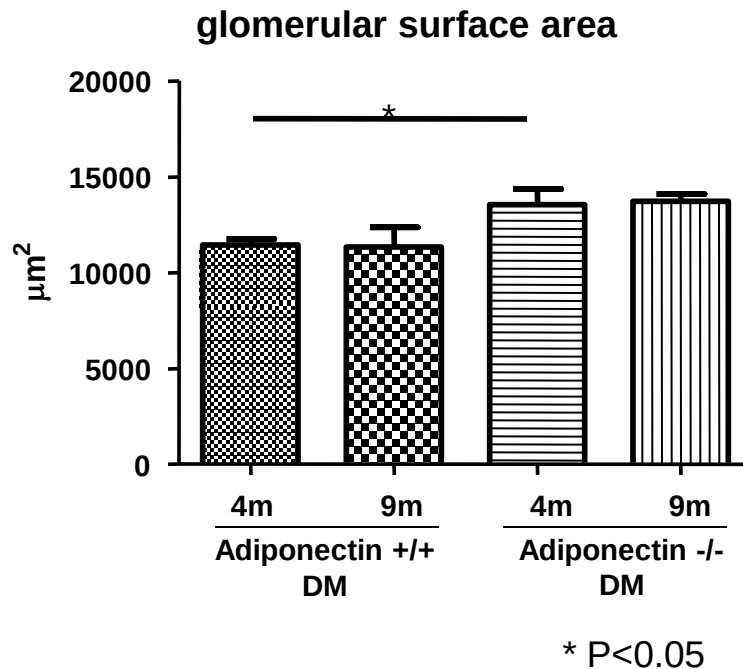
- Chronic deficiency of adiponectin in the diabetic state will exacerbate diabetic kidney disease
- Upregulation of podocyte Nox4 will lead to podocyte dysfunction

Study Design

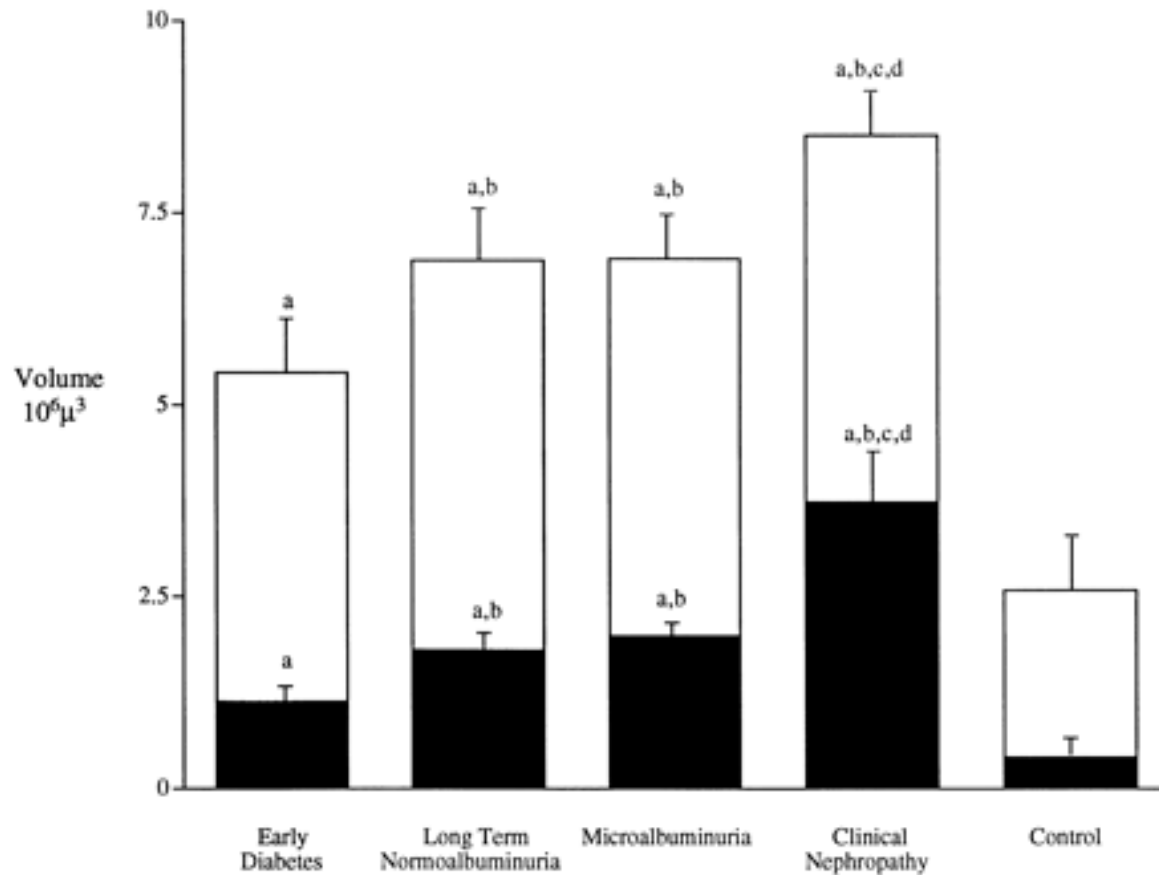
- Adiponectin $^{+/-}$ and $^{-/-}$ mice
- Renal function
- Morphometry
- Immunohistological study



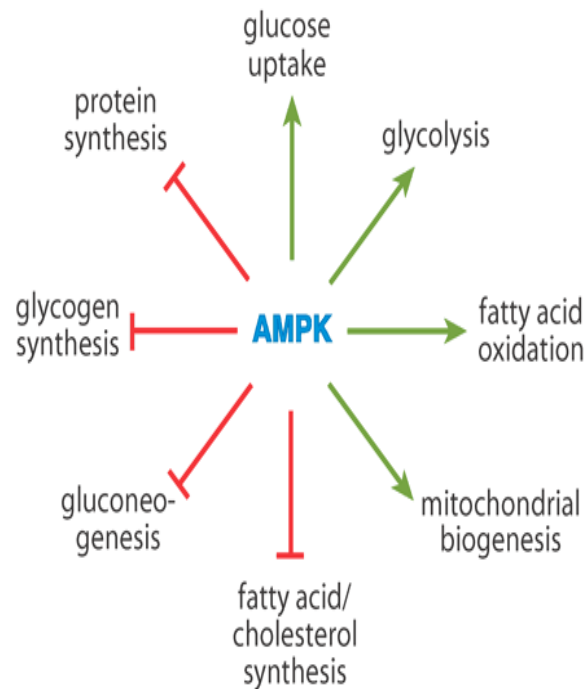
Increased Glomerular Volume in Adipo KO Diabetic Mice



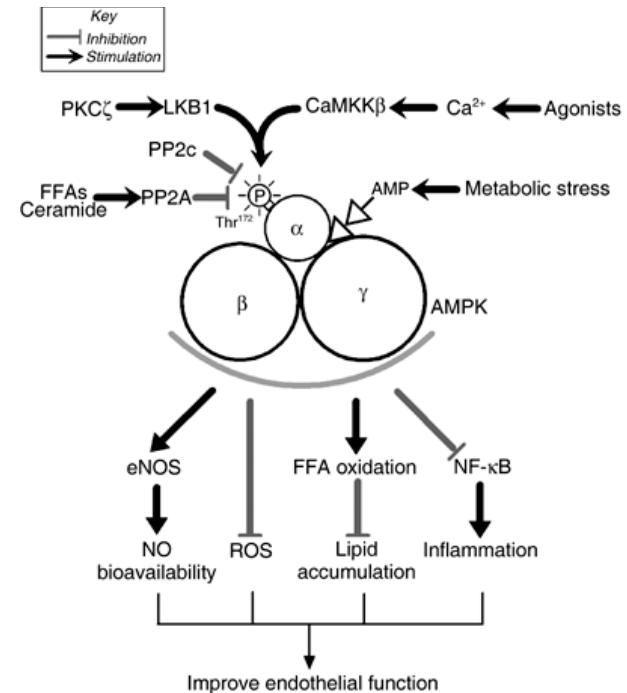
Glomerular Volume in Human Diabetic Nephropathy



Potential Role of Reduced AMPK in Diabetic Kidney Disease

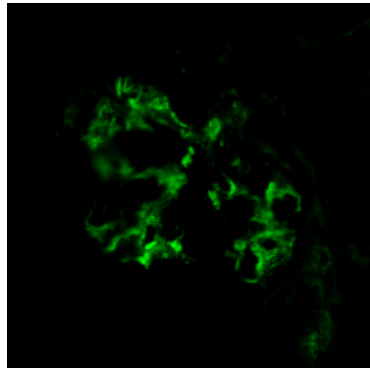


- AMPK stimulated with caloric depletion and inhibited with caloric excess

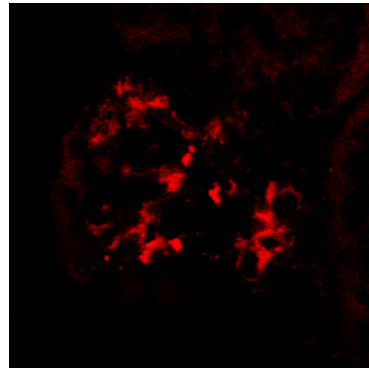


- Reduced AMPK enhances inflammation and inhibits endothelial function

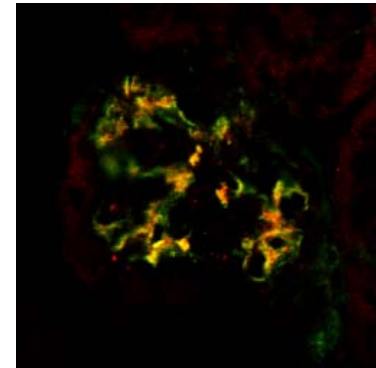
Podocyte AMPK Regulated by Adiponectin in vivo



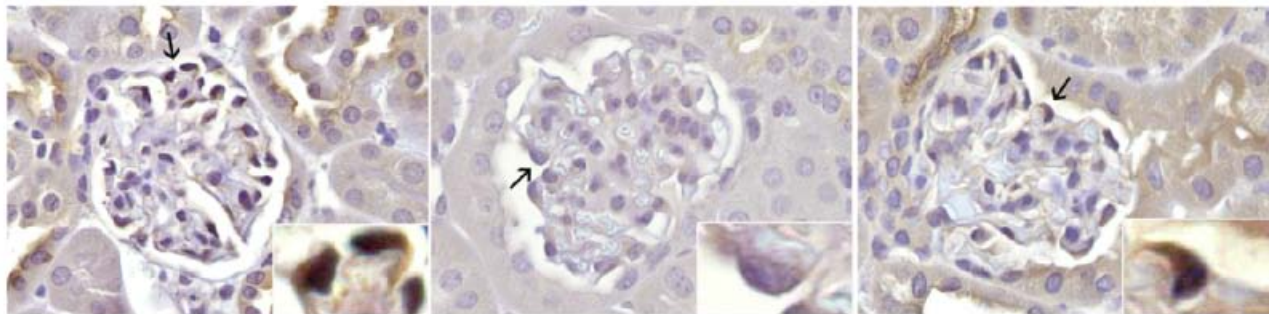
Synpo



P-AMPK



Overlay

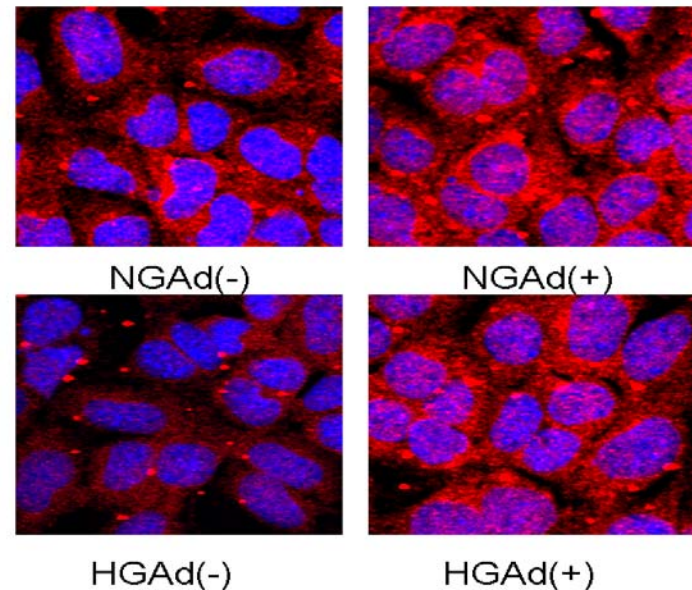
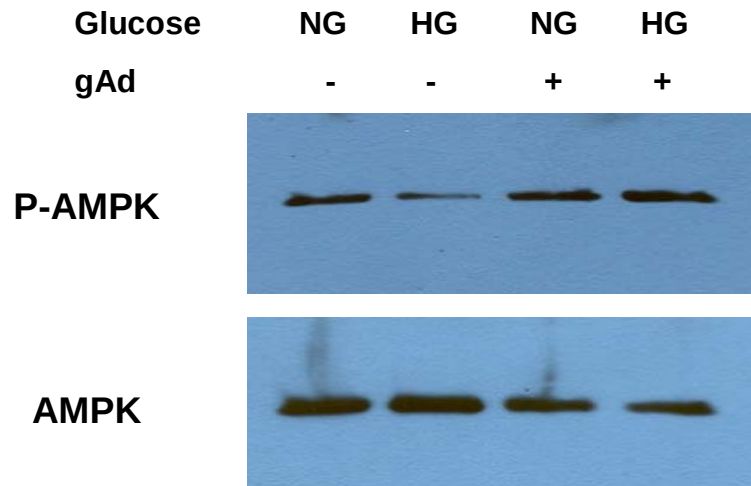


WT

Ad-/-

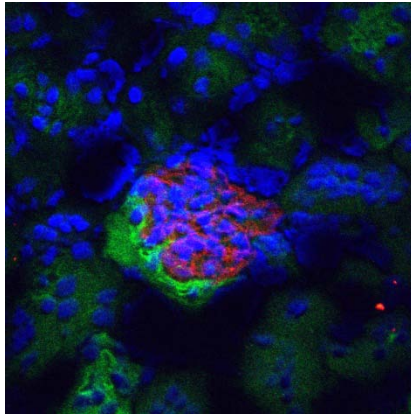
Ad-/-
gAd

High Glucose Inhibits AMPK in Podocytes: Reversal with Adiponectin

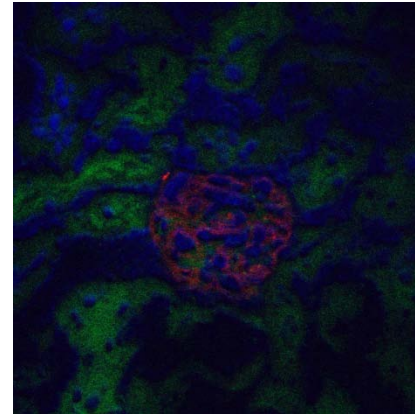


pAMPK in kidney tissues

Adipo ^{+/+}

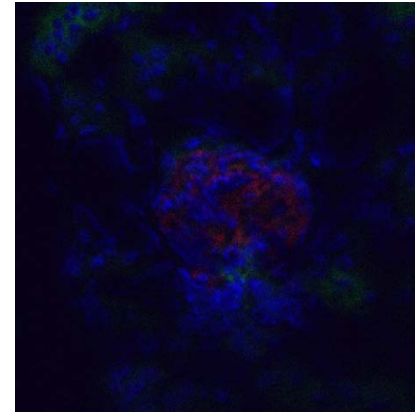
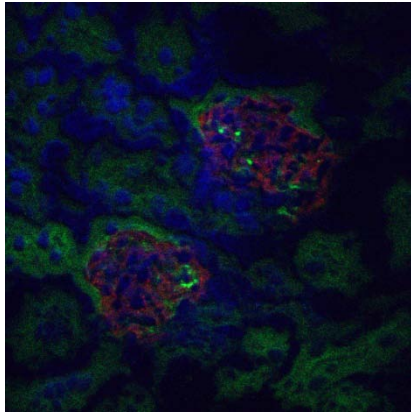


non DM



DM 4m

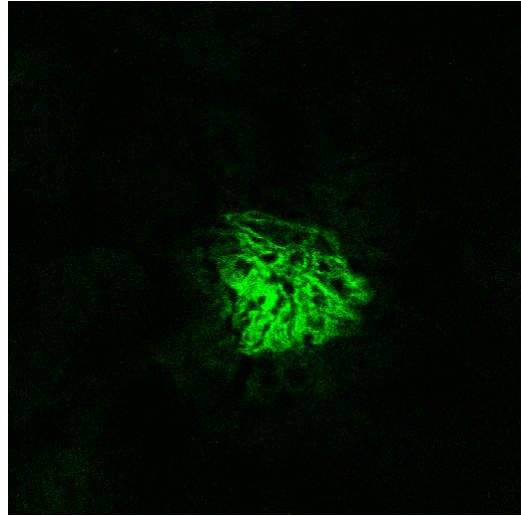
Adipo ^{-/-}



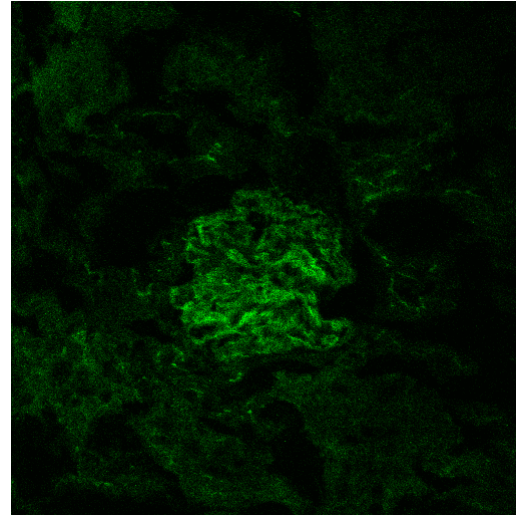
Green: pAMPK, Red: podocin

ZO-1

Adipo ^{+/+}

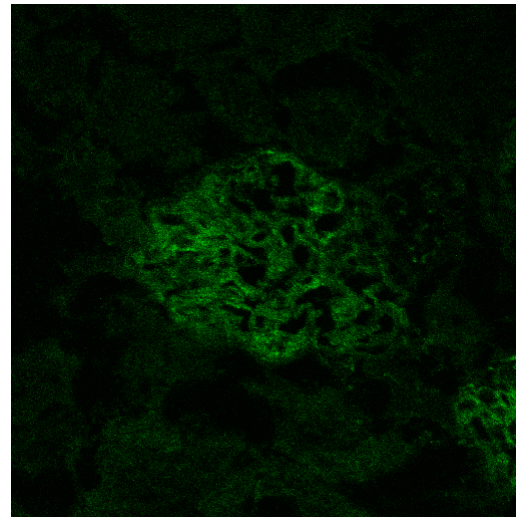
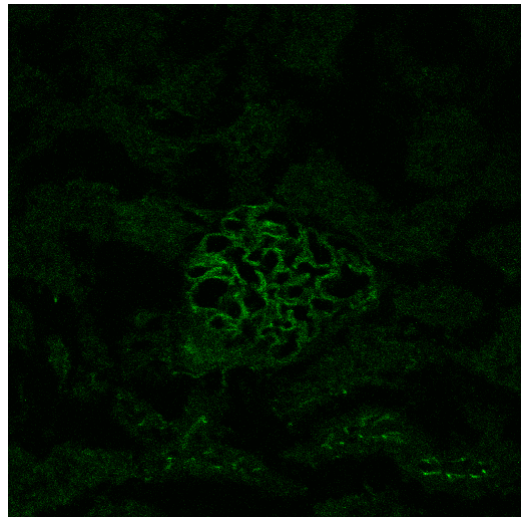


non DM



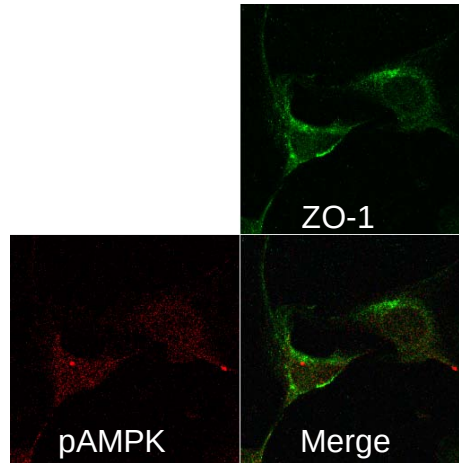
DM 4m

Adipo ^{-/-}

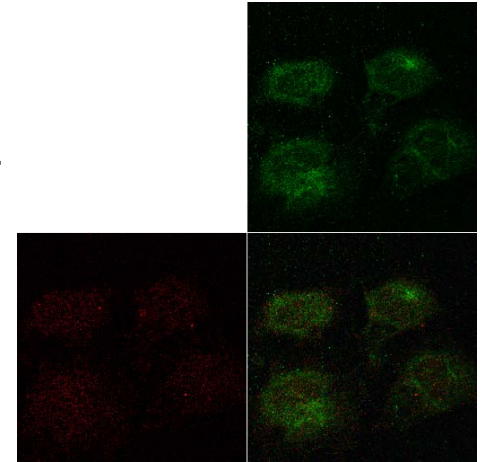


Podocyte (ZO-1/pAMPK)

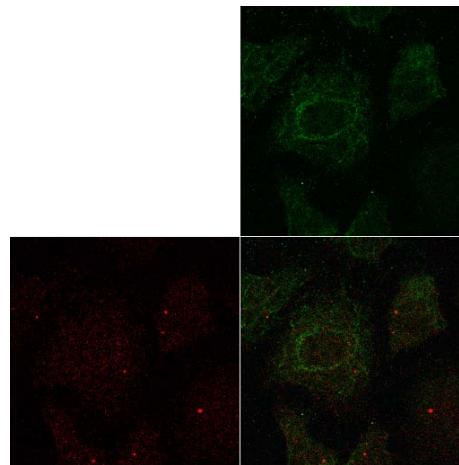
Normal Glucose



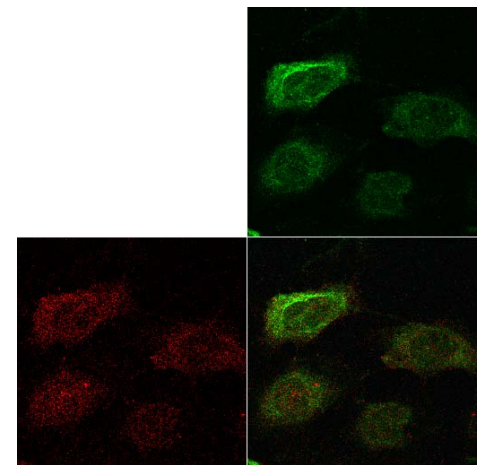
High Glucose 24hr

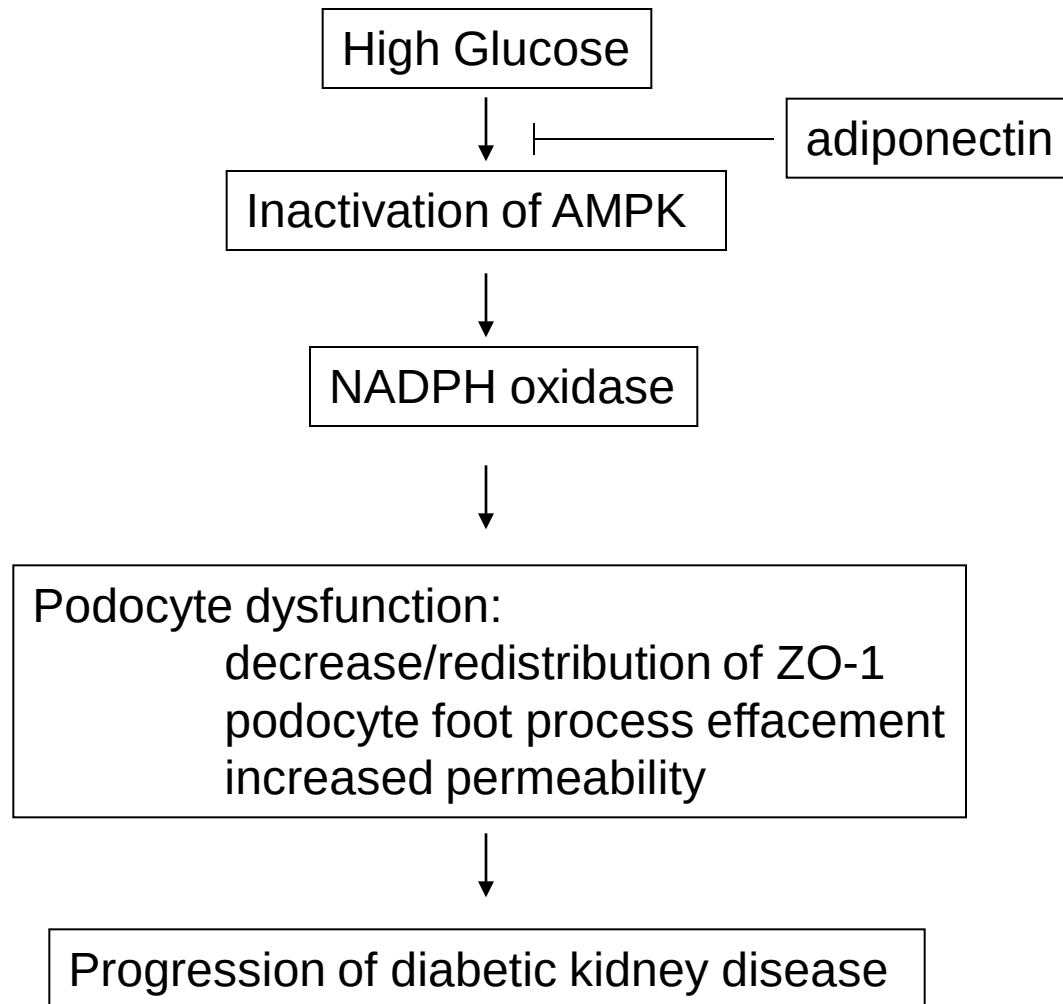


Ad-AMPK-DN



Ad-AMPK-CA

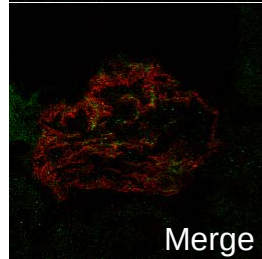
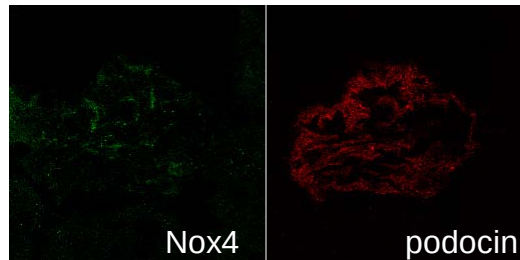




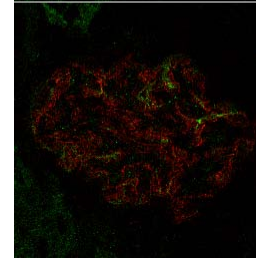
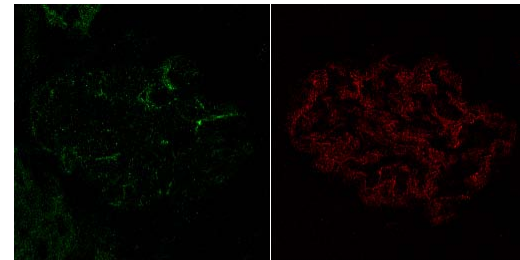
Future Studies

- Study adiponectin KO mice with Akita on B16, DBA background
- Evaluate glomerular hemodynamics in diabetic adiponectin KO mice
- Evaluate role of adiponectin receptors in AMPK regulation (VA Merit)

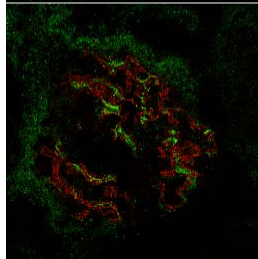
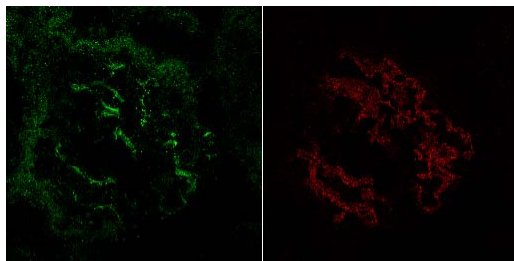
Nox4



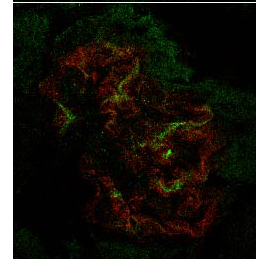
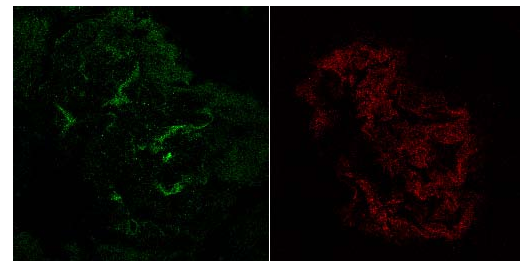
adipo ^{+/+} non DM



adipo ^{+/+} DM



adipo ^{-/-} non DM

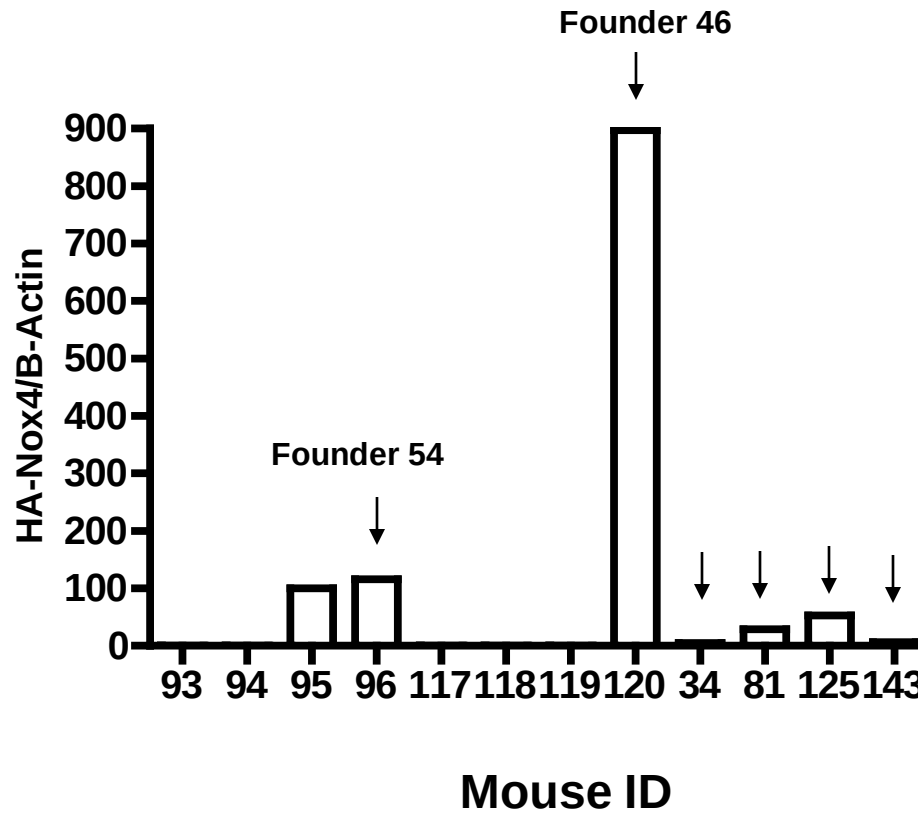


adipo ^{-/-} DM

Development of Inducible Podocyte Nox4 Transgenic

- Creation of HA/HNox4 tg mice, 10 founder lines in FVB background
- Cross with podocin rtTA mice (obtained from Jeff Kopp) in FVB
- Administer doxycycline for 2 weeks
- Screen litters for expression of HA/HNox4

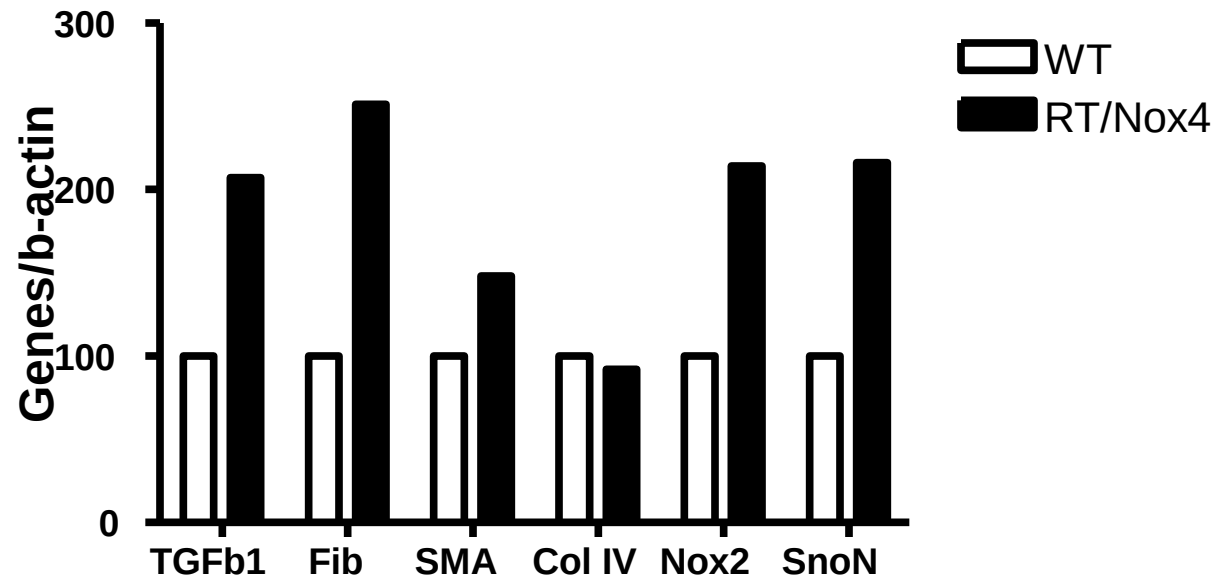
Q-RT-PCR for HA/HNox4



↓ Double TG
RT/HANox4

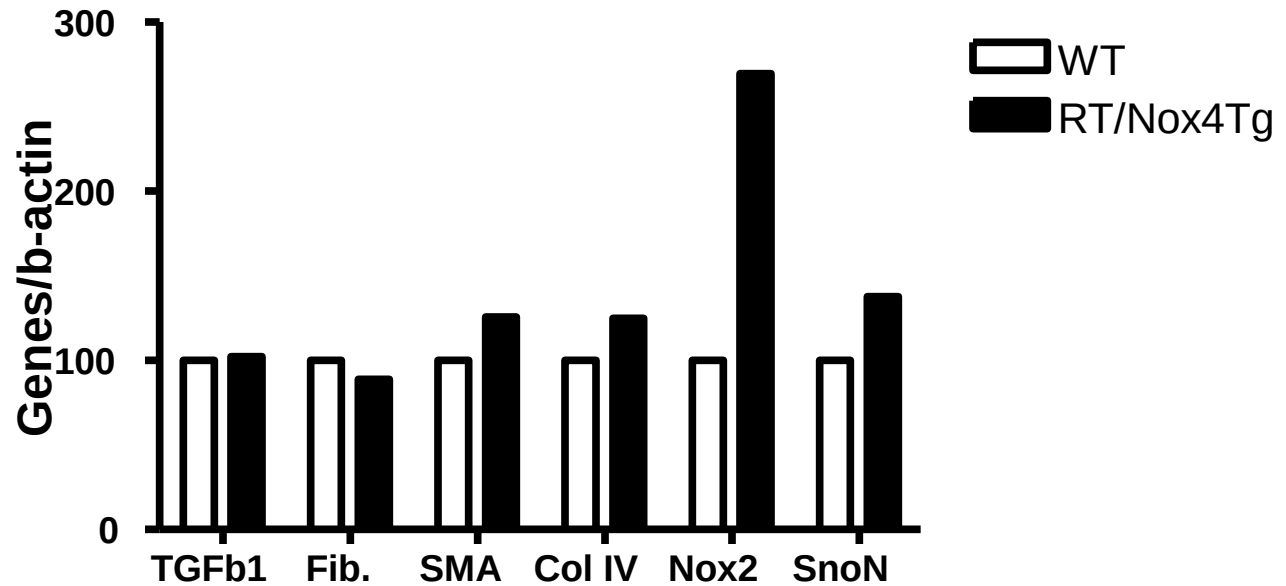
Sep 2009

Q-RT-PCR of podocinrtTA/HAHNox4 tg mouse



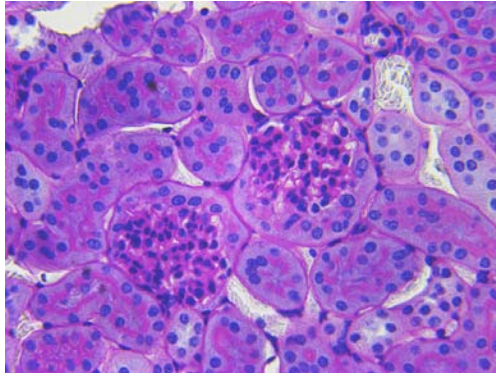
Founder 54

Q-RT-PCR of podocinrtTA/HAHNox4 tg mouse

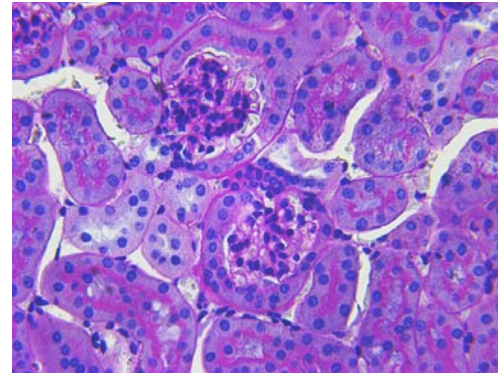


Founder 46

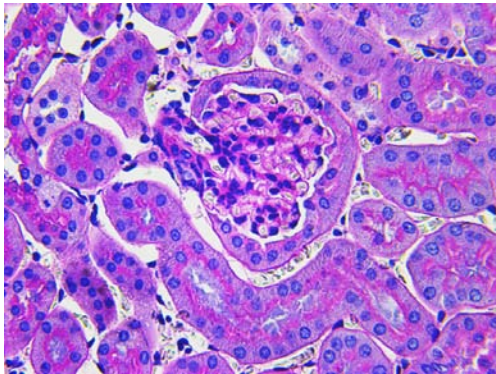
PAS staining of rtTA/HANox4 TG mouse



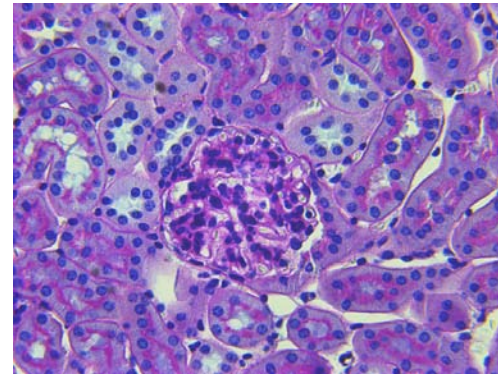
WT



Podocin rtTA



rtTA/HANox4 line 54



rtTA/HANox4 line 46

Future Studies

- Expand lines of rtTA/HHANox4 lines based on expression levels
- Cross with Akita on FVB background to assess impact of diabetes with Nox4 upregulation
- Evaluate on DBA background