Sex differences in glomerular protein expression and effects of soy-based diet on podocyte signaling. Afreeda Mahesaniya et al. 2022. Canadian Journal of Kidney Health and Disease.

Supplemental Materials

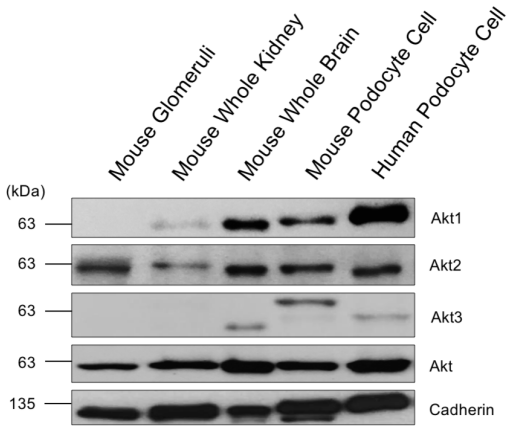
Supplemental Figure 1: Baseline protein expression of Akt1-3 paralogs in total kidney, isolated glomerular and cultured podocytes.

Supplemental Figure 2: Baseline expression profile of estrogen receptor β (ERβ) in male and female glomeruli.

Supplemental Figure 3: Urinary albumin assessment following consumption of variable amounts of isolated soy protein (ISP).

Supplemental Figure 4: Total and phosphorylated p38 expression in female human podocyte cells (HPCs).

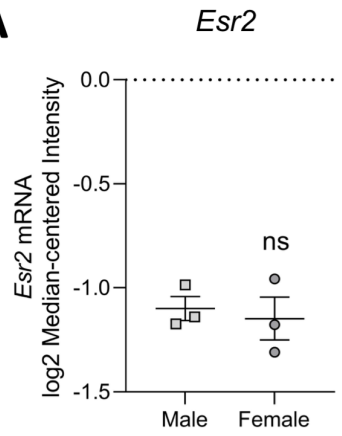
Supplemental Table 1: Profiling of short tandem repeat (STR) sites for male and female human podocyte cells (HPCs).



Supplemental Figure 1

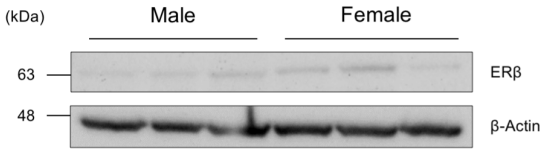
Supplemental Figure 1: Baseline protein expression of Akt1-3 paralogs in total kidney, isolated glomerular and cultured podocytes. Immunoblots of lysates prepared from mouse kidney, glomeruli and brain (used as a positive control), as well as from mouse and human podocyte cell lines. Use of paralog-specific antibodies confirmed that Akt2 is the primary variant in mouse glomeruli, while both Akt1 and Akt2 are found in whole kidney. Podocytes grown in culture express all three paralogs. Cadherin was used as a loading control.

A

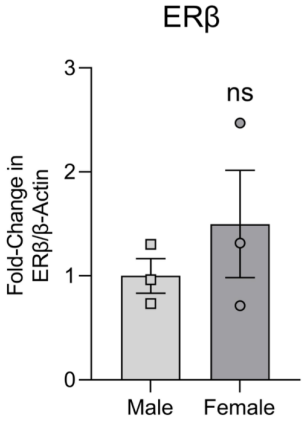


Human glomerular dataset sourced from
Lindenmeyer et al., 2010 using Nephroseq

B

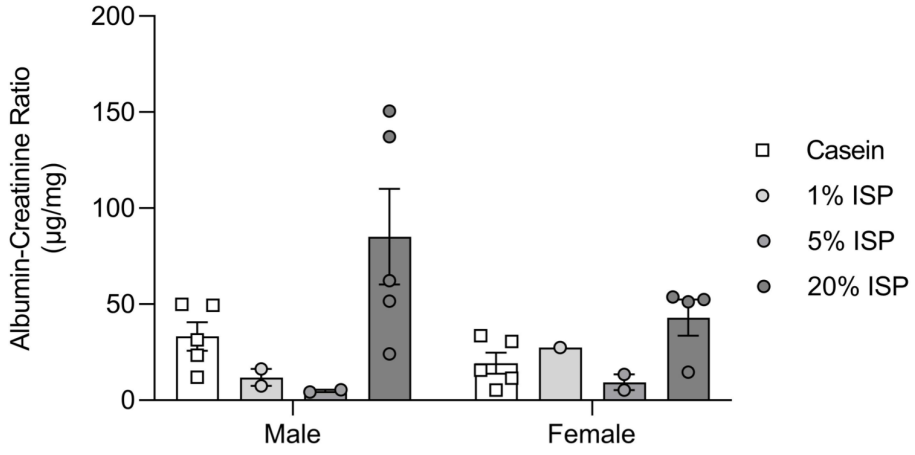


C



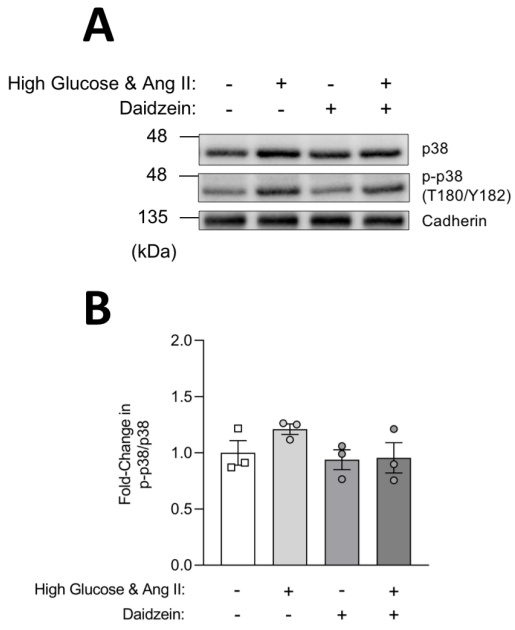
Supplemental Figure 2

Supplemental Figure 2: Baseline expression profile of estrogen receptor β (ERβ) in male and female glomeruli. (A) mRNA expression of ERβ in male and female human glomeruli. Data were acquired using the Nephroseq database using microarray datasets published by Lindenmeyer et al., 2010.[47] (B) Immunoblots of glomerular whole cell lysate from male and female control mice. β-actin was used as a loading control. (C) Densitometry of immunoblot in (B), normalized to β-actin and male mice. Data are presented as mean +/- SEM. Statistical significance compared to male mice was determined using Student's *t*-test. "ns" denotes no significance.



Supplemental Figure 3

Supplemental Figure 3: Urinary albumin assessment following consumption of variable amounts of isolated soy protein (ISP). Albumin-Creatinine Ratio (ACR) of spot urine collections from mice fed casein with or without 1%, 5%, or 20% ISP.



Supplemental Figure 4

Supplemental Figure 4: Total and phosphorylated p38 expression in female human podocyte cells (HPCs). (A & B) Differentiated female HPCs were treated with a combination of high glucose (25mM)/1µM angiotensin II (Ang II), daidzein (10µM), or high glucose/Ang II and daidzein for 24 hours. Differentiated HPCs with 19.5mM mannitol and 0.1% DMSO were used as controls. (A) Immunoblots of total and phospho-p38 from female HPC lysates with cadherin shown as a loading control. (B) Densitometry from p38 immunoblots (A) from three separate biological replicates (n=3) normalized to control cells. All data are presented as mean +/- SEM. Statistical significance compared to control cells was determined using one-way ANOVA with post-hoc Sidak's test.

Supplemental Table 1: Profiling of short tandem repeat (STR) sites for male and female human podocyte cells (HPCs)

Male HPCs (AB line – Saleem et al., 2002, *J Am Soc Nephrol*)		
STR Marker	Allele 1	Allele 2
Amelogenin	X	Y
CSF1PO	12	12
D13S317	8	11
D16S539	11	13
D21S11	29	30
D5S818	11	12
D7S820	10	11
TH01	7	7
TPOX	8	9
vWA	16	18
Female HPCs (LY line – Haley et al., 2018, *Sci Adv*)		
STR Marker	Allele 1	Allele 2
Amelogenin	X	X
CSF1PO	10	12
D13S317	11	11
D16S539	11	11
D21S11	29	30
D5S818	11	12
D7S820	11	12
TH01	7	8
TPOX	9	11
vWA	16	17
D10S1248	12	13
D12S391	17	21
D18S51	12	18
D19S433	15	15
D1S1656	15.3	17.3
D22S1045	12	15
D2S1338	20	26
D2S441	11.3	14
D3S1358	15	16
D8S1179	14	14
FGA	21	22.2
Penta D	11	12
Penta E	7	12