Prenatal corticosterone exposure induces dysregulation of the renal renin angiotensin system in male offspring

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Introduction: Maternal exposure to the stress hormone corticosterone, can impair fetal development and program long term disease. Fetal exposure to corticosterone is regulated by the placental glucocorticoid metabolizing enzyme, Hydroxysteroid (11β) dehydrogenase 2 (HSD11B2) which is elevated to a greater extent in response to maternal corticosterone in placentas of female fetuses. Thus, male fetuses are exposed to greater levels of corticosterone than females. Fetal exposure to the synthetic glucocorticoid, dexamethasone has been shown to reduce nephron endowment and program long term cardiovascular outcomes. While a reduction in nephron number may result in impaired renal function and hypertension, additional renal factors have been shown to be affected in other models of programmed disease physiology/pathology. The renal renin angiotensin system (RAS) in particular has been shown to be affected by prenatal perturbations and is an important regulator of cardiovascular function. However, the effects of maternal corticosterone exposure *in utero* on the long term renal RAS in offspring is poorly understood. Therefore, this study aimed to identify the long term renal RAS outcomes in mice prenatally exposed to maternal corticosterone.

Methods: Pregnant C57/BL/6 mice were anesthetised (induced with 5% isoflurane in air and maintained at 2.5% isoflurane in air) at E12.5 for the surgical implantation of osmotic minipumps primed to release corticosterone (33μg/kg/h for 60h beginning) or were left untreated. A subset of dams was killed during corticosterone exposure at E14.5, the fetuses sexed by genotyping and the kidneys removed and snap frozen for subsequent analysis. mRNA levels of markers of glucocorticoid exposure, the glucocorticoid receptor (Nr3c1) and the mineralocorticoid receptor (Nr3c2), were assessed using QPCR. Additional dams were allowed to litter down naturally and male and female offspring were killed at 6 months of age and kidneys collected. Renal mRNA levels of Nr3c1, Nr3c2, renin (Ren1), Angiotensin converting enzyme (Ace), angiotensin II receptor 1 a (Agtr1a) and the Mas proto-oncogene (Mas1) were assessed by QPCR. Renal tissue levels of Renin, Angiotensin II and Angiotensin 1-7 were assessed by radioimmuoassay.

Results: Maternal corticosterone exposure increased renal mRNA levels of Nr3c1 and Nr3c2 in male but not female E14.5 fetuses. Similarly, maternal corticosterone exposure increased the mRNA levels of Nr3c1 and Nr3c2 in the kidneys of 6 month old male but not female offspring. The mRNA levels of Ren1 and Mas1 were increased in the kidneys of male offspring of pregnant mice exposed to corticosterone compared to controls while Ace and Agtr1a mRNA levels were not affected. In contrast, female kidney mRNA levels of Ren1, Ace and Mas1 were not affected by maternal corticosterone exposure and Agtr1a was increased compared to controls. Renal tissue levels of Renin were significantly increased in male but not female offspring while renal tissue levels of Angiotensin II and Angiotensin 1-7 were not affected.

Conclusion: This study demonstrates that kidneys of male and female fetuses have different renal responses to a maternal stress challenge. Male fetuses appear to be directly affected by maternal glucocorticoid exposure, resulting in direct and long term upregulation of the renal glucocorticoid and mineralocorticoid receptors. Additionally, increased renal exposure to glucocorticoids in male fetuses has resulted in the long term disruption of the renal RAS. The increase in renal renin levels may play a significant role in disrupting overall physiology in the male offspring. In contrast, the kidneys of female fetuses appear to be largely protected from maternal corticosterone and have no long term effects on renal glucocorticoid receptors and minimal effects on the renal RAS. These sexually dimorphic effects on the renal RAS which have their origins *in utero* may contribute towards the more severe programmed cardiovascular phenotypes often seen in males.