Hypoxia-induced growth restriction leads to sexually dimorphic changes to the renal and cardiovascular systems in adult mouse offspring

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Adverse *in utero* conditions are associated with intrauterine growth restriction and subsequent low birth weight. Growth restriction increases the risk of perinatal morbidity and mortality, but may also impact on health outcomes in later life. Reduced oxygen supply to the fetus (hypoxia) is a common gestational perturbation linked to cardiovascular dysfunction in adulthood. However little attention has been placed on the effects on the developing kidney, which has been highlighted as particularly susceptible to *in utero* perturbations. Currently, we are examining the impact of maternal hypoxia on the structure and function of the kidney. Of particular interest are the relationship between low glomerular number, a strong risk factor for hypertension, and the compensatory capacity of the renal tubule system.

To assess adult outcomes of hypoxia-induced growth restriction, pregnant CD1 mice were housed in a hypoxic chamber (12.0% O_2; N=11, HYP) or control (21% O_2; N=11, CON) environment from embryonic day (E) 14.5 to birth (E19.5). A subset of offspring (N=8-11 per sex per treatment) was culled at postnatal day (P) 21 for assessment of nephron number and renal tubule lengths using unbiased stereology. Remaining offspring were aged to 12 months to assess renal function and blood pressure. Offspring (N=8-10 per sex per treatment) were housed in metabolic cages to collect a 24h urine sample under basal conditions and in response to 24h water deprivation. 24 h urinary sodium and albumin excretion were measured using a COBAS Integra Plus and a murine ELISA kit respectively. Systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR) were assessed in the offspring (N=5-6 per sex per treatment) *via* radiotelemetry recording over three days.

At P1, pooled male and female HYP offspring body weight was reduced by approximately 7% (P=0.001). This level of growth restriction was maintained at P21, where body weight was reduced by approximately 7-8% in both HYP male and female offspring ($P_{TREATMENT}$ = 0.01). A reduction in left kidney weight of approximately 7-8% in male and female HYP offspring was also observed ($P_{TREATMENT}$ = 0.03). Heart weight and brain weight were unaffected by maternal hypoxia. Maternal hypoxia led to a nephron deficit of 24% in male HYP offspring (CON: 12,886±515, HYP: 9,782±517; P=0.0006) thereby reducing total glomerular density (CON: 347 n/mm^3, HYP: 261 n/mm^3 P=0.04). This was accompanied by substantial elongation in total proximal tubule length in male HYP offspring (control: 104±8m, hypoxia: 159±17m; P=0.007). Interestingly, female HYP offspring had normal glomerular number and renal tubule lengths. Aged male and female offspring maintained urine flow and sodium excretion, however male HYP offspring exhibited mild albuminuria (P=0.009). In response to 24h water deprivation, male HYP offspring did not reduce urine flow (P=0.04). Male and female HYP offspring had significantly higher MAP throughout both the light and dark cycle compared to control counterparts (males: $P_{TREATMENT}$=0.01; females: $P_{TREATMENT}$<0.0001). In male HYP offspring, this was driven by an increase in DBP ($P_{TREATMENT}$<0.0001) with no change in SBP. In contrast, female HYP offspring had an increase in both SBP ($P_{TREATMENT}$=0.0001) and DBP ($P_{TREATMENT}$<0.0001). No change in HR was observed between groups in male or female offspring.

In summary, hypoxia-induced growth restriction led to reduced nephron endowment, compensatory proximal tubule elongation and impaired response to a water deprivation challenge in male offspring only. However, both male and female offspring developed hypertension. This suggests that female offspring are afforded some form of renoprotection *in utero* or during early postnatal life, however this protection does not extend to the cardiovascular system. We are currently investigating the molecular mechanisms that lead to the sexually dimorphic alterations seen in the fetal renal and cardiovascular systems.