Mitochondrial adaptations and dynamics in the human placenta

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The placenta performs many key functions, transporting nutrients, gases and wastes between maternal and fetal systems, and modifying maternal systems to maintain the pregnancy. During gestation the placenta undergoes significant growth and structural changes; initially sustaining the fetus in a low-oxygen environment before the onset of maternal blood flow, then in the third trimester delivering oxygen rich blood to support the exponential rise in energy demands of the growing fetus. Preeclampsia is an important pregnancy disorder caused by placental dysfunction. Placenta blood flow and oxygen exposure are thought to be altered in preeclampsia, when blood is delivered in a pulsatile manner leading to placental oxidative stress and subsequent maternal hypertension and endothelial dysfunction. Mitochondria have an array of roles linked to oxygen consumption, including providing the majority of cellular energy through oxidative phosphorylation. Placental mitochondria must adapt to changes in the energetic demands of the placenta throughout gestation, and may also be critically affected in preeclampsia. Given the importance of placental mitochondrial function over gestation and in preeclampsia, we investigated mitochondria at important times during gestation, and in healthy and preeclamptic pregnancies.

Placental tissue was obtained from first trimester pregnancies, and healthy and preeclamptic term pregnancies following delivery. Mitochondrial content was determined by qPCR for mitochondrial and nuclear genes. Respiratory states were assessed by respirometry. Expression levels of mitochondrial complexes were determined by western blot. Western blot and qPCR were used to determine levels of markers of mitochondrial biogenesis, mitochondrial fission and fusion, autophagy, apoptosis, and cellular replication. Protein carbonyls were measured by enzyme linked immunosorbent assay. Reactive oxygen species (ROS) were measured by resorufin fluorescences. Total antioxidant activity was measured *via* the ABTS (2,2′-Azino-di-[3-ethylbenzthiazoline sulphonate]) radical cation. In addition, healthy term tissue was subjected to 20 min of hypoxia followed by reoxygenation *in vitro*, and respiration and ROS monitored.

In first trimester placentae, mitochondrial content was increased at 12–13 relative to 7–10 weeks. Respiration decreased at 11 weeks compared to earlier gestations, and increased from 12 weeks. In term placentae, mitochondrial content was increased and total capacity of the respiratory system was higher compared to first trimester. *In vitro* hypoxia/reoxygenation led to a decrease in respiration and no change in ROS. In preeclamptic placentae, protein carbonyls were not different; ROS, antioxidant activity, and the level of mitochondrial complexes II, III, and V were increased relative to control. Preeclamptic mitochondrial content and respiration were increased, and reserve respiratory capacity was reduced relative to control. No change was found in markers of mitochondrial biogenesis, mitochondrial fusion, autophagy, apoptosis, and cellular replication. A marker of mitochondrial fission was decreased in preeclamptic placentae relative to control.

Placental mitochondria adapt over gestation and in preeclampsia. Changes over gestation may relate to the onset of maternal blood flow to the placenta. *In vitro* hypoxia/reoxygenation increased respiration, providing a potential mechanism for mitochondrial adaptation in the placenta. Increased antioxidant activity in preeclampsia may be a partially successful response to increased ROS, which therefore leads to no overt tissue damage (as measured *via* protein carbonyls). However, placental mitochondria are affected in preeclampsia, with decreased respiratory reserve capacity and increased expression of some mitochondrial complexes that may account for this decrease mitochondrial efficiency. Increased mitochondrial content and respiration in preeclampsia may represent adaptation to mitochondrial damage to maintain appropriate energy levels, and this appears to be regulated through decreased mitochondrial fission.