Abnormal Pattern of Ca2+ Oscillations During Fertilization In Vivo Impairs Offspring Growth Trajectory In The Mouse. Virginia Savy, National Institute of Environmental Health Sciences, USA

During mammalian fertilization, the sperm triggers a series of oscillations in the egg’s intracellular Ca2+ concentration ([Ca2+]i), which is the hallmark signal for egg activation. Interestingly, experimental manipulation of [Ca2+]i in vitro during or immediately after fertilization results in alterations in the blastocyst transcriptome, implantation success and offspring health. However, it remains unclear whether the findings reported from in vitro studies recapitulate the complex regulation of in vivo fertilization. Here we tested the hypothesis that, even in the highly specialized environment of the oviduct, appropriate Ca2+ signaling after fertilization is critical for proper preimplantation embryo development and offspring growth. In somatic cells, plasma membrane Ca2+ ATPase (PMCA) pumps are responsible for clearing excess Ca2+ from the cell following Ca2+ release events. We found that in mouse eggs, the most highly expressed PMCA isoform was PMCA1 (encoded by Atb2b1). To generate mouse eggs with abnormally increased [Ca2+]i exposure following fertilization, we conditionally deleted PMCA1 (cKO) in oocytes using the Zp3-cre transgene. As anticipated, in vitro fertilized cKO eggs had a much longer first Ca2+ transient than controls (mean ± SEM: 11.3 ± 1.0 min, N=30 vs 3.6 ± 0.3 min, N=49; p<0.0001); however, the oscillation frequency was similar between groups. Assuming a comparable difference in Ca2+ dynamics between control and cKO eggs during in vivo fertilization, we evaluated the impact of altered Ca2+ signaling following fertilization on offspring growth trajectory. Heterozygous (Atp2b1+/-) offspring from Atb2b1-flox/flox; Cre+ females mated with wild type males were weighed weekly for 8 weeks. Wild type (Atp2b1+/flox) offspring from Atb2b1-flox/flox females mated to wild type males served as controls. Offspring weigh at birth was similar between groups for both females and males; however, growth trajectory was different between groups by the 1st week of age. On average the experimental males were 14.7% smaller than controls at 8 weeks (mean weight ± SEM: 22.9 ± 0.3 g, N=33 vs 20.4 ± 0.3 g, N=34, males from 17 litters), whereas females were 5.9% smaller than controls (18.1 ± 0.2 g, N=29 vs 17.0 ± 0.2 g, N=30, females from 17 litters). Despite the altered postnatal growth rate, cKO-derived mice had normal glucose and insulin tolerance at 3 months of age. Our findings strongly support the idea that appropriate Ca2+ signaling in the first few hours following fertilization is necessary to ensure appropriate embryo “quality” and offspring health. Given the essential role of Ca2+ signaling for egg activation and embryo development, further research is necessary to decode the link between Ca2+ dynamics and long-term effects on offspring health, to ensure safe clinical practices during assisted reproductive procedures.