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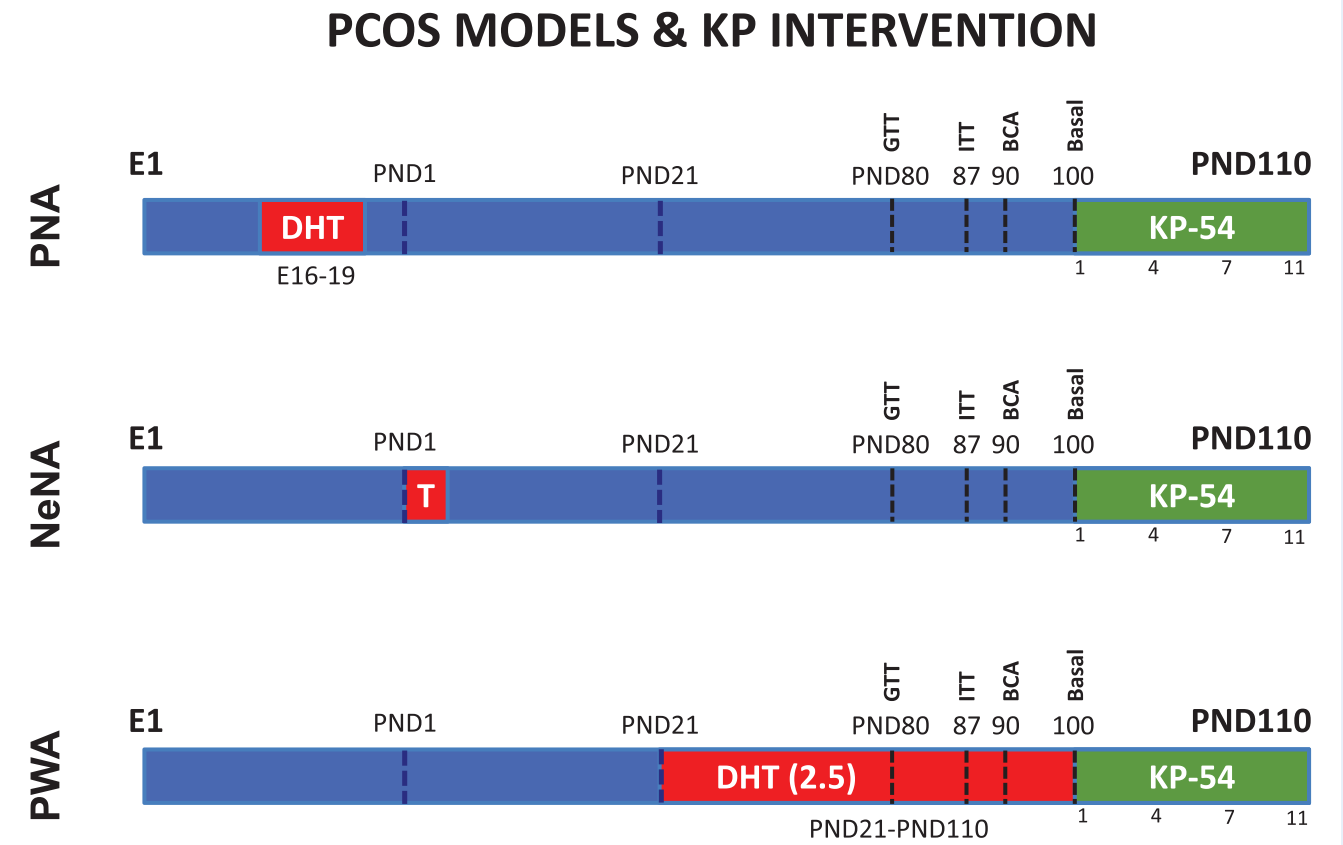
SUPPLEMENTARY DATA



Supplementary Figure S1 Experimental design of preclinical studies: PCOS models and pharmacological intervention. Three preclinical models of polycystic ovary syndrome (PCOS), generated by androgen exposure during specific age windows, were used: PNA (for prenatal androgenization), generated by daily sc injections of dihydrotestosterone (DHT) to pregnant rats between gestational days 16 and 19; NeNA (for neonatal androgenization), produced by a single sc injection of testosterone propionate (T) to newborn female rats; and PWA (for post-weaning androgenization), generated by implanting females rats at the day of weaning (postnatal day 21; PND21) with silastic capsules filled with DHT. The animals ($N = 20$ per model) were followed up to adulthood, when they were subjected to protocols of daily s.c. injections of 100 µg/kg of kisspeptin-54 (KP-54) or vehicle for a period of 11 days. Specific sampling and analyses were conducted at Days 1, 4, 7 and 11 after initiation of KP-54 treatments, as described in Materials and Methods. Animals injected with vehicle served as reference controls. Before initiation of KP-54 administration (at PND100), the animals were subjected to phenotypic characterization, including implementation of glucose and insulin tolerance tests (GTT & ITT, respectively), body composition analyses (BCA) and blood sampling for determination of basal gonadotrophin levels (Basal); the timeline for these analyses is depicted also in the scheme. Group sizes were $N = 10$ per model (PNA, NeNA, PWA) and pharmacological (KP-54, vehicle) treatment.