

Guidelines/Criteria	
Reference:	Jacobsen PR, Christiansen S, Boberg J, Nellemann C, Hass U. 2010. Combined exposure to endocrine disrupting pesticides impairs parturition, causes pup mortality and affects sexual differentiation in rats. Int J Androl 33:434-442.
In vivo Study Type Route of Administration Species & age of animals	oral gavage pregnant female Wistar rats
Study Duration	Range finding studies: Perinatal study design (GD 7-PND13) Main study:
Type of Mixture Binary >2 components Similar acting or dissimilar What Mode of Action was investigated?	no yes similar action assumed endocrine disruption
Parameters/End points Measured Target organs/Critical effects Pharmacological changes or adverse effects	maternal body weight, gestational lengths, pup mortality, pup weights, male reproduction organ weights, uterus weights, pup liver weights; Anogenital index, nipple/areola retention, malformations of male external genitalia, progesterone, testosterone and estradiol levels in pups, progesterone in dams Adverse
Individual Components Characterisation of individual compounds Name, exact chemical name, CAS no. Were dose responses established for individual components? Were no effect levels established? Were doses below the NO(A)ELs investigated?	No Pesticide mix (Pmix): 25%, 50%, 75%, 100%, 125% Doses in mg/kg bw: Epoxiconazole (3.75, 7.50, 11.25, 15.00, 18.75), Mancozeb (6.25, 12.50, 18.75, 25.00, 31.25), Prochloraz (8.75, 17.50, 26.25, 35.00, 43.75), Tebuconazole (12.5, 25.00, 37.50, 50.00, 62.50), Procymidone ((12.5, 25.00, 37.50, 50.00, 62.50) No, only mixtures at different concentrations tested No, Doses were chosen according to the doses of each individual pesticide that caused no major effects on pregnancy length and pup survival in earlier studies (conducted in the same lab) For some endpoints presumably yes
Mixtures Investigated Number of dose levels How does the mixture make-up compare to individual components? (e.g. low dose) equivalents used?) No. of technical replicates per exposure condition (<i>in vitro</i>) No. of animals per dose group (<i>in vivo</i>)	Range finding study 1: 75, 100, 125% of Pmix; Range finding study 2: 25, 50% of Pmix The lowest doses in the pesticide mixture are considered to be individual NOAELs of the compounds based on gestation lengths and pup mortality not applicable 4-14 animals/group
Observations/Findings	Study 2: Pmix25% and Pmix50%: increased gestation lengths, decreased prostate and epididymides weights; increased anogenital index in female pups, decreased anogenital index in male pups; increased nipple retention in male pups; increased numbers of dysgenesis of the external male genitalia (mild or moderate at Pmix25% and severe effects at Pmix50%) Pmix50% increased perinatal pup mortality, decreased birth weights of female pups, increased liver weights of male and female pups. Pmix75% and Pmix100% could not be evaluated due to pup mortality No effects on hormone levels (however large standard variation)
Overall opinion (e.g. sufficient numbers of groups investigated, group sizes adequate, observations reproducible, low dose levels used investigated)	Inhomogenous animal numbers investigated: Only 4 animals in Pmix 25% dose group tested. The NOAELs (as a basis for actual dose settings) are based on gestation lengths and on pup mortality, not on effects investigated in this study.