

Guidelines/Criteria	
	Reference: Birkhøj M, Nellemann C, Jarfelt K, Jacobsen H, Andersen HR, Dalgaard M, Vinggaard AM. 2004. The combined antiandrogenic effects of five commonly used pesticides. Toxicol Appl Pharmacol 201(1):10-20.
In vivo Study Type Route of Administration Species & age of animals	Oral gavage Castrated male Wistar rats, 6 weeks old
Study Duration	7 days
Type of Mixture Binary >2 components Similar acting or dissimilar What Mode of Action was investigated?	No 6 components Dissimilar Antiandrogenicity
Parameters/End points Measured Target organs/Critical effects Pharmacological changes or adverse effects	Antiandrogenicity Hershberger assay, hormone levels (LH, testosterone, T4, Prolactin) + body weights and adrenals, liver, kidney and thyroid weights
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?	Yes Prochloraz Simazine Tribenuron-methyl Deltamethrin Methiocarb No Each one dose level was investigated, which was for some endpoints a NOAEL No
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>)	Single doses: Deltamethrin: 2.5 mg/kg bw Methiocarb: 2.5 mg/kg bw Prochloraz: 25 mg/kg bw Simazine: 25 mg/kg bw Tribenuron-methyl: 25 mg/kg bw Combination experiment at these doses Not applicable 6 animals/dose group
Observations/Findings	The adrenal gland weights were statistically significantly increased in the combination experiments, whereas no changes were seen for the individual testings. Kidney and thyroid weights were unaffected. For all other parameters the individual chemicals were not included in the mixture at their individual NOAELs. Decreased levator ani/bulbocavernosus muscle weights were observed in the combination experiment. For hormone levels Methiocarb was included in the combination at an effect level.
Overall opinion (e.g. sufficient numbers of groups investigated, group sizes adequate, observations reproducible, low dose levels used investigated)	Limited evidence, as only limited endpoints were investigated. Hershberger assay is only borderline positive for the combination, as only one organ weight was decreased and others were unaffected.