Guidelines/Criteria	
Reference:	Kolle SN, Melching-Kollmuss S, Krennrich G, Landsiedel R, van Ravenzwaay B. Assessment of combinations of antiandrogenic compounds vinclozolin and flutamide in a yeast based reporter assay. Regul Toxicol Pharmacol 60:373-380.
***In vitro* Study Type** Route of Administration Species & age of animals	YAS assay (Human androgen receptor mediated androgenic and antiandrogenic activity): Yeast stably transformed with the human androgen receptor (hAR) gene and expression plasmids carrying androgen-responsive sequences. Test substance is determined to be an androgen agonist if the ß-galactosidase activity is increased concentration dependently by more than 20% and to be an androgen antagonist if the ß-galactosidase activity was decreased concentration dependently to levels of androgen antagonistic control (dihydrotestosterone)
Study Duration	not applicable
Type of Mixture Binary >2 components Similar acting or dissimilar What Mode of Action was investigated?	 yes no Similar acting or dissimilar Antiandrogenicity
Parameters/End points Measured Target organs/Critical effects Pharmacological changes or adverse effects *In vitro*	 Endocrine disruption at androgen receptor level
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 Yes. Full dose-response curves were determined Yes 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 (*in vitro*) No. of animals per dose group (*in vivo*)	 6 concentrations of the individual components and the mixtures were measured (10-4 - 10-9 mol/L) Quadruplicates
Observations/Findings	Vinclozolin and Flutamide were antiandrogenic with similar potency. Receptor saturation was observed at around 10-4 mol/L. Both vinclozolin and flutamide were antiandrogens of similar potency in the YAS assay. In the concentration range tested the two antiandrogens vinclozolin and flutamide did not act synergistically. Concentration additivity was observed in the linear, non-receptor-saturated concentration range. At high concentrations of one of the two substances tested the contribution of the second at lower concentration levels was less than additive. The combined response of both compounds at high concentration levels was also less than additive (saturation effect). At concentration levels which did not elicit a response of the individual compounds, the combination of these compounds also did not elicit a response.
Overall opinion (e.g. sufficient numbers of groups investigated, group sizes adequate, observations reproducible, low dose levels used investigated)	 Reliable study; adequate study design and reporting. Quadruplicate testing.