A summary of all studies considered for the mammalian toxicology, including the Study ID Matrix is outlined in the following table.

Outline of dataset considered for mammalian toxicology assessment:

Type of toxicity	Study type	Study ID Matrix
Repeated dose toxicity studies in mammals	Repeated dose 90-day oral (feeding) toxicity study in rat	1
	Repeated dose 90-day oral (feeding) toxicity study in rat	2
	Combined chronic toxicity/carcinogenicity oral (feeding) study in rat	3
	One-year interim sacrifice	*3a*
	Final sacrifice	*3b*
	Repeated dose 90-day oral (feeding) toxicity study in mouse	4
	Carcinogenicity oral (feeding) study in mouse	5
	Final sacrifice	*5a*
	Mechanistic part of the study	*5b*
	Repeated dose 90-day oral (feeding) toxicity study in dog	6
	Repeated dose 1-year oral (feeding) toxicity study in dog	7
	Two-generation reproduction oral (feeding) toxicity test in rat	8
	Prenatal developmental toxicity oral (gavage) study in rat	9
	Prenatal developmental toxicity oral (gavage) study in rabbit	10
	Repeated dose 90-day oral (feeding) neurotoxicity study in rat	23
	28-day oral (gavage) study in male rat (mechanistic study - HLR 6-96)	11
	14-day oral (gavage and feeding) TK study in intact male rat (mechanistic study - Report No 49393)	12
	15-day oral (gavage) study in intact male rat (mechanistic study - Report No 50232)	20
	Without hCG challenge	*20a*
	With hCG challenge	*20b*
	2-week oral (gavage) study in male rat (mechanistic study - HLR 575-93)	21
	Without hCG challenge	*21a*
	With hCG challenge	*21b*
	Hormone analysis in the serum of male rats treated for 1-y (carcinogenicity study)	*21e*
***In vitro* mechanistic**	*In vitro* stably transfected human androgen receptor transcriptional activation assays (Report No 50112)	13
	In vitro stably transfected human estrogen receptor-α transcriptional activation assays (Report No 49230)	14
	In vitro dopamine D2 receptor binding assay (DuPont-49680 Rev 1)	15
	In vitro steroidogenesis assay (DuPont-49227)	16
	In vitro hepatic microsome aromatase assay (DuPont-12095)	17
	In vitro aromatase activity assays (Report No 47677)	18
	Rat hepatic microsomes	*18a*
	Rat ovary homogenate	*18b*
	In vitro aromatase inhibition using human recombinant microsomes (DuPont-48651)	19
	15-day oral (gavage) study in intact male rat (mechanistic study - Report No 50232)	20
	Aromatase activity in microsomes prepared from treated rats	*20c*
	In vitro metabolism of testosterone in microsomes prepared from treated rats	*20d*
	2-week oral (gavage) study in male rat (mechanistic study - HLR 575-93)	21
	Aromatase activity in microsomes prepared from treated rats	*21c*
	In vitro hepatic aromatase activity in C8-induced microsomes	*21d*
	In vitro hormonal synthesis in isolated and cultures Leydig cells from treated males	*21f*
	Combination effects of (tri)azole fungicides on hormone production and xenobiotic metabolism in a human placental cell line (Rieke, S *et al.*, 2014)	22
	In vitro ToxCast Androgen (ATG_AR_TRANS_up, AR agonistic activity)	29
	In vitro ToxCast Estrogen (ATG_ERE_CIS_up, ERa agonistic activity)	30
	In vitro ToxCast Estrogen (ATG_ERa_TRANS_up, ERa agonistic activity)	31
	In vitro ToxCast Thyroid (ATG_THRa1_TRANS_up, TRa transactivation)	32
	In vitro aromatase activity in rat ovary homogenate (Report FMC-52754)	33

In silico analysis:

QSAR analysis indicates the active substance can interact with the sulfonylurea receptors

ED assessment for humans

2.1 - ED assessment for T-modality

2.1.1- Have T-mediated parameters been sufficiently investigated?

	Answer
T-mediated parameters	List of available studies in which thyroid adversity (histopathology and/or weight) is addressed: OECD TG 408 - ID: 1#, 2#, 4# OECD TG 409 - ID: 6 OECD TG 452 - ID: 7 OECD TG 453 - ID: 3# OECD TG 451 - ID: 5#

Thyroid weight not measured.

2.1.2 - Lines of evidence for adverse effects and endocrine activity related to T-modality

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
Thyroid receptor	Human liver cell line HepG2	24	Hours	Uptake from the medium (in vitro)			No effect	Negative for TR agonist (ATG_THRa1_TRANS_up)		
Thyroid histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Thyroid weight	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
Adrenals histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Adrenals weight	rat	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Brain histopathology examination	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Brain weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative brain weight		
	rat	90	Days	Oral	2000	ppm	Increase	Mean relative brain weight		
	rat	90	Days	Oral	2000	ppm	Decrease	Mean absolute brain weight		
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	90	Days	Oral	1500	ppm	Increase	Mean relative brain weight		
	rat	12	Months	Oral	1500	ppm	Increase	Mean relative brain weight		
	rat	22	Months	Oral	1500	ppm	Increase	Mean relative brain weight		
	mouse	18	Months	Oral		ppm	No effect			
Fertility (mammals)	rat	131	Days	Oral		ppm	No effect			
Litter size	rat	131	Days	Oral		ppm	No effect			
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral		mg/kg bw/day	No effect			
Litter viability	rat	131	Days	Oral		ppm	No effect			
Litter/pup weight	rat	131	Days	Oral	750	ppm	Decrease	Pup weight (<10%)		
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral	270	mg/kg bw/day	Decrease	Decreased fetal weight (no clear relationship but low nb of litters in the HD group)		
Number of implantations, corpora lutea	rat	131	Days	Oral		ppm	No effect			
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral		mg/kg bw/day	No effect			
Number of live births	rat	131	Days	Oral		ppm	No effect			

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
Numbers of embryonic or foetal deaths and viable foetuses	rat	131	Days	Oral		ppm	No effect			
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral		mg/kg bw/day	No effect			
Pituitary histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	131	Days	Oral		ppm	No effect			
	Rat	15	Days	Oral		mg/kg bw/day	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Pituitary weight	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the absorption)		
	Rat	15	Days	Oral		mg/kg bw/day	No effect			
Post implantation loss	rat	131	Days	Oral		ppm	No effect			
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral	270	mg/kg bw/day	Increase	Abortions: 8/16 at 270 mkd, 12/20 at 800 mkd		
Pre implantation loss	rat	131	Days	Oral		ppm	No effect			
Presence of anomalies (external, visceral, skeletal	rat	22	Days	Oral	350	mg/kg bw/day	Increase	Variations - unossified skulls (350); slight retardation renal dev (1000); partially ossified vertebra (1000); Malf - incidence of external/skeletal, 4 fetuses from 4 litters, no specific pattern (1000)		

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
	rabbit	13	Days	Oral		mg/kg bw/day	No effect	Insufficient number of litters in the 2 highest dose groups according to OECD guideline - may compromise assessment of teratogenicity		
Pup survival index	rat	131	Days	Oral		ppm	No effect			
Sex ratio	rat	131	Days	Oral		ppm	No effect			
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral		mg/kg bw/day	No effect			
Time to mating	rat	131	Days	Oral		ppm	No effect			
Heart weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative heart weight		
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Kidney histopathology	rat	90	Days	Oral	10000	ppm	Increase	Hemosiderosis: pigment in prox tubules		
	rat	90	Days	Oral	2000	ppm	Increase	Hemosiderosis: pigment in prox tubules		
	rat	90	Days	Oral	15000	ppm	Increase	Tubular epithelial cell atrophy		
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Kidney weight	rat	90	Days	Oral	10000	ppm	Decrease	Mean abs weight		

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
	rat	90	Days	Oral	10000	ppm	Increase	Mean rel weight		
	rat	90	Days	Oral	2000	ppm	Decrease	Mean absolute kidney weight		
	rat	90	Days	Oral	10000	ppm	Increase	Mean relative kidney weight		
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral	1500	ppm	Decrease	Mean absolute kidney weight		
	rat	22	Months	Oral	1500	ppm	Decrease	Mean absolute kidney weight		
	mouse	18	Months	Oral		ppm	No effect			
Liver histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral	750	ppm	Increase	Hepatocelluar hypertrophy		
	dog	90	Days	Oral	4000	ppm	Increase	Bile stasis (f); Pigmented sinusoidal macrophages (m&f)		
	dog	1	Years	Oral	3500	ppm	Increase	Hepatocelluar hypertrophy		
	Rat	15	Days	Oral		mg/kg bw/day	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral	750	ppm	Decrease	Dec. incidence of periportal fatty change (m&f), eosinophilic & total foci of cellular alt (m), basophilic & total foci of cellular alt (f)		
	mouse	18	Months	Oral	2500	ppm	Increase	Hepatic foci of cellular alteration, presence of intrahepatocellular erythrocytes, pigment accumulation in Kupffer cells and individual hepatocellular necrosis. Hepatocellular		

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
								adenomas.		
Liver weight	rat	90	Days	Oral	2000	ppm	Increase	Mean relative liver weight (NSS, 11%; >20% and in M at higher doses)		
	rat	90	Days	Oral	2000	ppm	Increase	Mean relative liver weight		
	mouse	90	Days	Oral	750	ppm	Increase	Mean absolute & relative liver weight		
	dog	90	Days	Oral	4000	ppm	Increase	Absolute and relative liver weight		
	dog	1	Years	Oral	3500	ppm	Increase	Mean absolute and relative liver weight		
	Rat	28	Days	Oral		mg/kg bw/day	No effect			
	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the absorption)		
	Rat	15	Days	Oral		mg/kg bw/day	No effect			
	Rat	15	Days	Oral	1500	mg/kg bw/day	Increase	Mean relative liver weight		
	Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean absolute liver weight		
	rat	12	Months	Oral	1500	ppm	Increase	Mean relative liver weight		
	mouse	18	Months	Oral	2500	ppm	Increase	Absolute & relative liver weight		
Lung histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
	mouse	18	Months	Oral		ppm	No effect			
Pancreas histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral		ppm	No effect			
	dog	1	Years	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
	mouse	18	Months	Oral		ppm	No effect			
Peripheral nerve histopathology	rat	90	Days	Oral		ppm	No effect			
	rat	22	Months	Oral	1500	ppm	Increase	Incidence and severity of myelin/axon degeneration of the sciatic nerve		
Spinal cord histopathology	rat	90	Days	Oral		ppm	No effect			
Spleen histopathology	rat	90	Days	Oral	2000	ppm	Increase	Extramedullary hematopoiesis (in M and F at higher doses)		
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral		ppm	No effect			
Spleen weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative spleen weight		
	rat	90	Days	Oral	2000	ppm	Increase	Mean relative spleen weight		
	mouse	90	Days	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect			
	rat	22	Months	Oral	1500	ppm	Decrease	Mean absolute spleen weight		
	mouse	18	Months	Oral		ppm	No effect			
Body weight	rat	90	Days	Oral	2000	ppm	Decrease	Mean body weight (7% in M, 3% in F) & body weight gain (11% in M, 7% in F)		
	rat	90	Days	Oral	2000	ppm	Decrease	Mean body weight (9% in M, 16% in F) & body weight gain (16% in M,		

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
								40% in F)		
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral	8000	ppm	Decrease	Mean body weight (25% in M, 15% in F) & body weight gain (83% in M, 57% in F)		
	dog	1	Years	Oral	3500	ppm	Decrease	Mean body weight gain (18% in M, 20% in F)		
	rat	131	Days	Oral	750	ppm	Decrease	Mean body weight (5% in F0 M) & body weight gain (13% in F0 M, 14% in F0 F) during premating		
	rat	131	Days	Oral	750	ppm	Decrease	Mean body weight in F0 females during lactation (7%) and gestation (6%).		
	rat	131	Days	Oral	750	ppm	Decrease	Mean body weight in F1 females during pre-mating, lact., gest. (7%) and in M at 1500 ppm		
	rat	22	Days	Oral	350	mg/kg bw/day	Decrease	Body weight loss GD7-9, body weight gain (GD7-17: 19% compared to controls/30% at 1000 mkd)		
	rabbit	13	Days	Oral	90	mg/kg bw/day	Decrease	Body weight loss GD7-10, body weight gain (32% compared to controls GD7-20)		
	Rat	28	Days	Oral		mg/kg bw/day	No effect			
	rat	90	Days	Oral	750	ppm	Decrease	Mean body weight (10% in F) & body weight gain (22% in F) - at		

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
								3000 ppm in M		
	Rat	15	Days	Oral	500	mg/kg bw/day	Decrease	Mean body weight (7-15% on D15) & body weight gain (64-115% D1-8, 24-68% D1-15)		
	Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean body weight (8% on D15) & body weight gain (68% D1-8, 42% D1-15)		
	Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease	Mean body weight (10% on D15) & body weight gain (74% D1-15)		
	rat	12	Months	Oral	750	ppm	Decrease	Mean body weight (7% in M, 7% in F) & body weight gain (11% in M, 13% in F)		
	rat	22	Months	Oral	1500	ppm	Decrease	Mean body weight (14% in M, 15% in F) & body weight gain (20% in M, 23% in F)		
	mouse	18	Months	Oral	7000	ppm	Decrease	Mean body weight gain (11% in M, 16% in F)		
Clinical chemistry and haematology	rat	90	Days	Oral	2000	ppm	Decrease	RBC count, Hb, Ht		
	rat	90	Days	Oral	10000	ppm	Increase	MCV, Reticulocytes		
	rat	90	Days	Oral	10000	ppm	Increase	Lymphocytosis		
	rat	90	Days	Oral	2000	ppm	Decrease	RBC count, Hb, Ht		
	rat	90	Days	Oral	2000	ppm	Increase	MCV, Reticulocytes		
	dog	90	Days	Oral	8000	ppm	Decrease	RBC count, Hb, Ht		
	dog	90	Days	Oral	8000	ppm	Increase	MCV, Reticulocytes; Hypercellularity of sternal, femoral BM		

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
								(regenarative anemia)		
	dog	90	Days	Oral	4000	ppm	Increase	ALAT, ASAT (and ALP at 8000 ppm)		
	dog	1	Years	Oral	3500	ppm	Decrease	RBC count, Hb, Ht		
	dog	1	Years	Oral	3500	ppm	Increase	ALP		
	rat	12	Months	Oral	750	ppm	Decrease	RBC count, Hb, Ht		
Food consumption	rat	90	Days	Oral	2000	ppm	Decrease	Food efficiency		
	rat	90	Days	Oral	2000	ppm	Decrease	Food efficiency & food consumption		
	mouse	90	Days	Oral		ppm	No effect			
	rat	131	Days	Oral	750	ppm	Decrease	Food consumption and/or food efficiency		
	rat	131	Days	Oral	750	ppm	Decrease	Food consumption during premating		
	rat	22	Days	Oral	350	mg/kg bw/day	Decrease	Food consumption - days 7-9G, 9-11G; increased days 17-22G		
	rabbit	13	Days	Oral	270	mg/kg bw/day	Decrease			
	Rat	28	Days	Oral		mg/kg bw/day	No effect			
	rat	90	Days	Oral	750	ppm	Decrease			
	Rat	15	Days	Oral	500	mg/kg bw/day	Decrease	Food consumption and/or food efficiency		
	Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Food consumption and/or food efficiency		
	Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease			
	mouse	18	Months	Oral		ppm	No effect	Food consumption & efficiency		
Mortality	rat	90	Days	Oral		ppm	No effect			
	rat	90	Days	Oral		ppm	No effect			

Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the line of evidence
	mouse	90	Days	Oral		ppm	No effect			
	dog	90	Days	Oral	8000	ppm	Increase	2/4 females		
	dog	1	Years	Oral	3500	ppm	Increase	1/5 male and 1/5 female		
	rat	22	Days	Oral		mg/kg bw/day	No effect			
	rabbit	13	Days	Oral	270	mg/kg bw/day	Increase	Dose-related deaths: 2/20 at 270 mkd, 9/20 at 800 mkd		
	Rat	28	Days	Oral		mg/kg bw/day	No effect			
	rat	90	Days	Oral		ppm	No effect			
	rat	12	Months	Oral		ppm	No effect	No effect		
	rat	22	Months	Oral		ppm	No effect	Poor survival in all groups typical for strain, not a compound-related effect. Study terminated at 22-m.		
	mouse	18	Months	Oral		ppm	No effect			

2.2 - ED assessment for EAS-modalities

2.2.1 - Have EAS-mediated parameters been sufficiently investigated?

	answer
EAS-mediated parameters	lack of the following studies: OECD TG 443 OECD TG 416, test protocol according to latest version of January 2001*

* Note: the two-generation reproduction study was conducted in 1990-1991 according to a former version of the OECD TG 416. Several EAS-mediated parameters were not investigated:

EAS-mediated parameters not investigated

sperm parameters,

oestrus cycle length,

vaginal opening,

preputial separation,

anogenital distance,

uterus weight,

ovary weight,

epididymis weight,

prostate weight,

adrenals weight

pituitary weight

coagulating gland weight

seminal vesicles weight

2.2.2 - Lines of evidence for adverse effects and endocrine activity related to EAS-modalities

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
	In vitro mechanistic	Androgen receptor	Human cells	23	Hours	Uptake from the medium		µM	No effect	Negative for AR agonist/antagonist in the stably Transfected Human AR Transactivation Assay (AR STTA, OECD 458)			EAS
			Human liver cell line HepG2	24	Hours	Uptake from the medium (in vitro)			No effect	Negative for AR agonist (ATG_AR_TRANS_up)			
		Estrogen receptor	Human cells	20-22	Hours	Uptake from the medium		µM	No effect	Negative for ER agonist/antagonist in the stably Transfected Human ERα Transcriptional Activation Assay (ER STTA, OECD 455)			
			Human liver cell line HepG2	24	Hours	Uptake from the medium (in vitro)			No effect	Negative for ER agonist (ATG_ER_CIS_up)			
			Human liver cell line HepG2	24	Hours	Uptake from the medium (in vitro)			No effect	Negative for ER agonist (ATG_ERa_TRANS_up)			
		CYP19	Rat microsomes	3	Hours	Uptake from the medium	500	µM	Decrease	Aromatase inhibition - no IC50 calculated (approx. 25% inhibition at 500 µM) [Release of 3H2O from 3H-androstenedione]			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			Human recombinant microsomes	15	Minutes	Uptake from the medium		µM	No effect	[Release of 3H2O from 3H-androstenedione] (Aromatase inhibition assay using human recombinant microsomes, OPPTS 890.1200)			
			Human placental cells	48	Hours	Uptake from the medium		µM	No effect	No change in CYP19 gene expression			
			Rat liver microsomes	3	Hours	Uptake from the medium	800	µM	Decrease	Aromatase inhibition - no IC50 calculated (approx. 50% inhibition at 800 µM) [Release of 3H2O from 3H-androstenedione]			
			Rat ovary homogenate	20	Minutes	Uptake from the medium	250	µM	Decrease	Equivocal aromatase inhibition / No firm conclusion, limitations [Measure of the conversion of testosterone to estradiol]			
			Rat microsomes from treated animals	15	Days	Oral		mg/kg bw/day	No effect	No aromatase inhibition in microsomes prepared from liver of rats treated for 15 days [Release of 3H2O from 3H-androstenedione]			
			Rat microsomes from treated animals	15	Days	Oral	1000	mg/kg bw/day	No effect	Hepatic aromatase activity not altered in microsomes prepared from liver of rats treated for 15 days (but some control animals had higher activity than treated animals)			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			Rat C8-induced liver microsomes			Uptake from the medium	173.6	µM	Decrease	Dose-dependent aromatase inhibition, SS at all concentrations [Release of 3H2O from 3H-androstenedione]			
			Rat ovary homogenate	20	Minutes	Uptake from the medium	100	µM	Decrease	Aromatase inhibition - no IC50 calculated (22% inhibition at 100 µM, 30% inhibition at 500 µM) [Measure of the conversion of testosterone to estradiol]			
		Estradiol synthesis	Human H295R cells	48	Hours	Uptake from the medium		µM	Increase	Equivocal induction of E biosynthesis (highest concentration only) or not interpretable (Steroidogenesis assay, OECD 456)			
			Human placental cells	48	Hours	Uptake from the medium	40	µM	Decrease	Decreased estradiol concentration (70% of control, p≤0.01)			
			Rat Leydig cells	5	Hours	Uptake from the medium	100	µM	Decrease	With and without hCG			
		Testosterone synthesis	Human H295R cells	48	Hours	Uptake from the medium		µM	Decrease	Equivocal inhibition of T biosynthesis (1 or 2 highest concentrations) (Steroidogenesis assay, OECD 456)			
			Rat Leydig cells	5	Hours	Uptake from the medium	100	µM	Increase	Without hCG			

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		Other hormones (in vitro)	Rat microsomes from treated animals	15	Days	Oral		mg/kg bw/day	No effect	No change in in vitro metabolism of testosterone in microsomes prepared from treated animals incubated for 30 min with testosterone			
		Progesterone (in vitro)	Human placental cells	48	Hours	Uptake from the medium		µM	No effect				
			Rat Leydig cells	5	Hours	Uptake from the medium	100	µM	No effect	With and without hCG			
		Binding to the Dopamine D2 receptor	Human HEK 293 cells	2	Hours	Uptake from the medium		µM	No effect				
	In vivo mechanistic	Estradiol level	Rat	28	Days	Oral	5	mg/kg bw/day	Decrease	Lower mean rate of increase in serum estradiol from pre-study to wk4 at 5 mkd, as well as NSS decreased serum estradiol at wk4 in all tested groups compared to control			EAS
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
										questionable			
			Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease	Decreased serum estradiol, no effect interstitial fluid estradiol			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Decreased serum estradiol, no effect interstitial fluid estradiol			
			Rat	12	Months	Oral	750	ppm	Decrease	Decreased serum estradiol in 1-y interim sacrifice rats of the 2-y rat study			
		Testosterone level	Rat	28	Days	Oral		mg/kg bw/day	No effect				
			Rat	14	Days	Oral		mg/kg bw/day	No effect	No significant differences in the plasma AUC0-6 or CL0-6 of testosterone from controls or treated rats, but high variability in a same group			
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral	1000	mg/kg bw/day	No effect	No effect serum and interstitial fluid testosterone			

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			Rat	15	Days	Oral	2000	mg/kg bw/day	Increase	Increased serum testosterone, no effect interstitial fluid testosterone			
			Rat	12	Months	Oral	750	ppm	Increase	Increased serum testosterone in 1-y interim sacrifice rats of the 2-y rat study			
		Follicle Stimulating Hormone (FSH) level	Rat	28	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral	1000	mg/kg bw/day	Increase	Slight increased serum FSH (NSS, high variability)			
			Rat	12	Months	Oral	1500	ppm	Increase	Increased serum FSH in 1-y interim sacrifice rats of the 2-y rat study			
		Luteinizing Hormone (LH) level	Rat	28	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral	1000	mg/kg bw/day	Increase	Slight increased serum LH (NSS, high variability)			
			Rat	12	Months	Oral	1500	ppm	Increase	Increased serum LH in 1-y interim sacrifice rats of the 2-y rat study			
		Other hormones	Rat	15	Days	Oral		mg/kg bw/day	No effect	Dihydrotestosterone level. No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral		mg/kg bw/day	No effect	Dihydrotestosterone level. No consistent change, high variability, reliability of the hormone analysis questionable			
		Prolactin	Rat	28	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent change, high variability, reliability of the hormone analysis questionable			
			Rat	15	Days	Oral	1000	mg/kg bw/day	Increase	Slight increased serum prolactin (NSS, high variability)			
			Rat	12	Months	Oral		ppm	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
	EATS-mediated	Coagulating gland weight	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the absorption)			EAS
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease	Mean absolute and relative acc sex glands weight -without dose response			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean absolute and relative acc sex glands weight			
		Epididymis histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral	15000	ppm	Increase	Oligospermia, atrophy - associated with decreased BW (40%) and BWG (70%)			
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral	4000	ppm	Increase	Aspermatogenesis, oligospermia - associated with decreased BW (25%) and BWG (83%) at 8000 ppm but no effect on BW(G) at 4000 ppm. Nevertheless, effect not observed in the 1-y dog study, may be due to immaturity of dogs in the 90-d study			
			dog	1	Years	Oral		ppm	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			rat	131	Days	Oral		ppm	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Epididymis weight	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the absorption)			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
		Prostate histopathology (with seminal vesicles and coagulating glands)	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Prostate weight	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the absorption)			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease	Mean absolute and relative acc sex glands weight -without dose response			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean absolute and relative acc sex glands weight			
		Seminal vesicles histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Seminal vesicles weight	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the absorption)			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease	Mean absolute and relative acc sex glands weight -without dose response			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean absolute and relative acc sex glands weight			
		Testis	rat	90	Days	Oral		ppm	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
		histopathology	rat	90	Days	Oral	15000	ppm	Increase	Atrophy, degeneration, bilateral Leydig cell hyperplasia - associated with decreased BW (40%) and BWG (70%)			
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral	4000	ppm	Increase	Bilateral tubular atrophy, decrease thickness of the seminiferous tubules, cytoplasmic vacuolation of germinal epithelium (at 8000 ppm) - associated with decreased BW (25%) and BWG (83%) at 8000 ppm but no effect on BW(G) at 4000 ppm. Nevertheless, effect not observed in the 1-y dog study, may be due to immaturity of dogs in the 90-d study			
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral		ppm	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral	750	ppm	Increase	Leydig cell hyperplasia, adenoma (Carc 2 H351, RAC 2013)			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			mouse	18	Months	Oral		ppm	No effect				
		Testis weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative testes weight			
			rat	90	Days	Oral	15000	ppm	Decrease	Mean absolute testes weight - associated with important decreased BW (40%) and BWG (70%)			
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral	4000	ppm	Decrease	Absolute and relative testes weight			
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral	750	ppm	Decrease	Mean relative testes weight			
			rat	131	Days	Oral	1500	ppm	Decrease	Mean relative testes weight			
			Rat	28	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect	Trend towards increased rel testes weight at 1500 and 2000 mkd. No consistent effect (no dose relationship but saturation of the absorption)			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral	1000	mg/kg bw/day	Increase	Mean relative testes weight - without dose response			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Increase	Mean relative testes weight			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			rat	12	Months	Oral	750	ppm	Increase	Mean relative testes weight (and also absolute at 1500 ppm)			
			rat	22	Months	Oral	1500	ppm	Increase	Mean relative testes weight			
			mouse	18	Months	Oral		ppm	No effect				
		Mammary gland histopathology (female)	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral	750	ppm	Decrease	Dec. incidence of mammary masses (fibroadenomas). Only controls, high dose and decedents examined.			
			mouse	18	Months	Oral		ppm	No effect				
		Ovary histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			mouse	18	Months	Oral		ppm	No effect				
		Uterus histopathology (with cervix)	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Vagina histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
	Sensitive to, but not diagnostic of, EATS	Adrenals histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				

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			dog	1	Years	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Adrenals weight	rat	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Brain histopathology examination	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Brain weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative brain weight			
			rat	90	Days	Oral	2000	ppm	Increase	Mean relative brain weight			
			rat	90	Days	Oral	2000	ppm	Decrease	Mean absolute brain weight			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	90	Days	Oral	1500	ppm	Increase	Mean relative brain weight			
			rat	12	Months	Oral	1500	ppm	Increase	Mean relative brain weight			
			rat	22	Months	Oral	1500	ppm	Increase	Mean relative brain weight			
			mouse	18	Months	Oral		ppm	No effect				
		Fertility (mammals)	rat	131	Days	Oral		ppm	No effect				
		Litter size	rat	131	Days	Oral		ppm	No effect				
			rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral		mg/kg bw/day	No effect				
		Litter viability	rat	131	Days	Oral		ppm	No effect				
		Litter/pup weight	rat	131	Days	Oral	750	ppm	Decrease	Pup weight (<10%)			
			rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral	270	mg/kg bw/day	Decrease	Decreased fetal weight (no clear relationship but low nb of litters in the HD group)			
		Number of	rat	131	Days	Oral		ppm	No effect				

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		implantations, corpora lutea	rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral		mg/kg bw/day	No effect				
		Number of live births	rat	131	Days	Oral		ppm	No effect				
		Numbers of embryonic or foetal deaths and viable foetuses	rat	131	Days	Oral		ppm	No effect				
			rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral		mg/kg bw/day	No effect				
		Pituitary histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	131	Days	Oral		ppm	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Pituitary weight	Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the			

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
										absorption)			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
		Post implantation loss	rat	131	Days	Oral		ppm	No effect				
			rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral	270	mg/kg bw/day	Increase	Abortions: 8/16 at 270 mkd, 12/20 at 800 mkd			
		Pre implantation loss	rat	131	Days	Oral		ppm	No effect				
		Presence of anomalies (external, visceral, skeletal	rat	22	Days	Oral	350	mg/kg bw/day	Increase	Variations - unossified skulls (350); slight retardation renal dev (1000); partially ossified vertebra (1000); Malf - incidence of external/skeletal, 4 fetuses from 4 litters, no specific pattern (1000)			
			rabbit	13	Days	Oral		mg/kg bw/day	No effect	Insufficient number of litters in the 2 highest dose groups according to OECD guideline - may compromise assessment of teratogenicity			
		Pup survival index	rat	131	Days	Oral		ppm	No effect				
		Sex ratio	rat	131	Days	Oral		ppm	No effect				

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			rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral		mg/kg bw/day	No effect				
		Time to mating	rat	131	Days	Oral		ppm	No effect				
	Target organ toxicity	Heart weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative heart weight			
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Kidney histopathology	rat	90	Days	Oral	10000	ppm	Increase	Hemosiderosis: pigment in prox tubules			
			rat	90	Days	Oral	2000	ppm	Increase	Hemosiderosis: pigment in prox tubules			
			rat	90	Days	Oral	15000	ppm	Increase	Tubular epithelial cell atrophy			
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
			mouse	18	Months	Oral		ppm	No effect				
		Kidney weight	rat	90	Days	Oral	10000	ppm	Decrease	Mean abs weight			
			rat	90	Days	Oral	10000	ppm	Increase	Mean rel weight			
			rat	90	Days	Oral	2000	ppm	Decrease	Mean absolute kidney weight			
			rat	90	Days	Oral	10000	ppm	Increase	Mean relative kidney weight			
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				
			rat	12	Months	Oral	1500	ppm	Decrease	Mean absolute kidney weight			
			rat	22	Months	Oral	1500	ppm	Decrease	Mean absolute kidney weight			
			mouse	18	Months	Oral		ppm	No effect				
		Liver histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral	750	ppm	Increase	Hepatocelluar hypertrophy			
			dog	90	Days	Oral	4000	ppm	Increase	Bile stasis (f); Pigmented sinusoidal macrophages (m&f)			
			dog	1	Years	Oral	3500	ppm	Increase	Hepatocelluar hypertrophy			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			rat	12	Months	Oral		ppm	No effect				

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			rat	22	Months	Oral	750	ppm	Decrease	Dec. incidence of periportal fatty change (m&f), eosinophilic & total foci of cellular alt (m), basophilic & total foci of cellular alt (f)			
			mouse	18	Months	Oral	2500	ppm	Increase	Hepatic foci of cellular alteration, presence of intrahepatocellular erythrocytes, pigment accumulation in Kupffer cells and individual hepatocellular necrosis. Hepatocellular adenomas.			
		Liver weight	rat	90	Days	Oral	2000	ppm	Increase	Mean relative liver weight (NSS, 11%; >20% and in M at higher doses)			
			rat	90	Days	Oral	2000	ppm	Increase	Mean relative liver weight			
			mouse	90	Days	Oral	750	ppm	Increase	Mean absolute & relative liver weight			
			dog	90	Days	Oral	4000	ppm	Increase	Absolute and relative liver weight			
			dog	1	Years	Oral	3500	ppm	Increase	Mean absolute and relative liver weight			
			Rat	28	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral		mg/kg bw/day	No effect	No consistent effect (no dose relationship but saturation of the			

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										absorption)			
			Rat	15	Days	Oral		mg/kg bw/day	No effect				
			Rat	15	Days	Oral	1500	mg/kg bw/day	Increase	Mean relative liver weight			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean absolute liver weight			
			rat	12	Months	Oral	1500	ppm	Increase	Mean relative liver weight			
			mouse	18	Months	Oral	2500	ppm	Increase	Absolute & relative liver weight			
		Lung histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Pancreas histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral		ppm	No effect				
			dog	1	Years	Oral		ppm	No effect				

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			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
			mouse	18	Months	Oral		ppm	No effect				
		Peripheral nerve histopathology	rat	90	Days	Oral		ppm	No effect				
			rat	22	Months	Oral	1500	ppm	Increase	Incidence and severity of myelin/axon degeneration of the sciatic nerve			
		Spinal cord histopathology	rat	90	Days	Oral		ppm	No effect				
		Spleen histopathology	rat	90	Days	Oral	2000	ppm	Increase	Extramedullary hematopoiesis (in M and F at higher doses)			
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral		ppm	No effect				
		Spleen weight	rat	90	Days	Oral	10000	ppm	Increase	Mean relative spleen weight			
			rat	90	Days	Oral	2000	ppm	Increase	Mean relative spleen weight			
			mouse	90	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect				
			rat	22	Months	Oral	1500	ppm	Decrease	Mean absolute spleen weight			
			mouse	18	Months	Oral		ppm	No effect				

dy ID trix	Effect classification	Effect target	Species	Duration of exposure	Duration unit	Route of administration	Lowest Effect dose	Dose unit	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modali
	Systemic toxicity	Body weight	rat	90	Days	Oral	2000	ppm	Decrease	Mean body weight (7% in M, 3% in F) & body weight gain (11% in M, 7% in F)			
			rat	90	Days	Oral	2000	ppm	Decrease	Mean body weight (9% in M, 16% in F) & body weight gain (16% in M, 40% in F)			
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral	8000	ppm	Decrease	Mean body weight (25% in M, 15% in F) & body weight gain (83% in M, 57% in F)			
			dog	1	Years	Oral	3500	ppm	Decrease	Mean body weight gain (18% in M, 20% in F)			
			rat	131	Days	Oral	750	ppm	Decrease	Mean body weight (5% in F0 M) & body weight gain (13% in F0 M, 14% in F0 F) during premating			
			rat	131	Days	Oral	750	ppm	Decrease	Mean body weight in F0 females during lactation (7%) and gestation (6%).			
			rat	131	Days	Oral	750	ppm	Decrease	Mean body weight in F1 females during pre-mating, lact., gest. (7%) and in M at 1500 ppm			
			rat	22	Days	Oral	350	mg/kg bw/day	Decrease	Body weight loss GD7-9, body weight gain (GD7-17: 19% compared to controls/30% at 1000 mkd)			

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			rabbit	13	Days	Oral	90	mg/kg bw/day	Decrease	Body weight loss GD7-10, body weight gain (32% compared to controls GD7-20)			
			Rat	28	Days	Oral		mg/kg bw/day	No effect				
			rat	90	Days	Oral	750	ppm	Decrease	Mean body weight (10% in F) & body weight gain (22% in F) - at 3000 ppm in M			
			Rat	15	Days	Oral	500	mg/kg bw/day	Decrease	Mean body weight (7-15% on D15) & body weight gain (64-115% D1-8, 24-68% D1-15)			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Mean body weight (8% on D15) & body weight gain (68% D1-8, 42% D1-15)			
			Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease	Mean body weight (10% on D15) & body weight gain (74% D1-15)			
			rat	12	Months	Oral	750	ppm	Decrease	Mean body weight (7% in M, 7% in F) & body weight gain (11% in M, 13% in F)			
			rat	22	Months	Oral	1500	ppm	Decrease	Mean body weight (14% in M, 15% in F) & body weight gain (20% in M, 23% in F)			
			mouse	18	Months	Oral	7000	ppm	Decrease	Mean body weight gain (11% in M, 16% in F)			
		Clinical chemistry and	rat	90	Days	Oral	2000	ppm	Decrease	RBC count, Hb, Ht			
			rat	90	Days	Oral	10000	ppm	Increase	MCV, Reticulocytes			

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		haematology	rat	90	Days	Oral	10000	ppm	Increase	Lymphocytosis			
			rat	90	Days	Oral	2000	ppm	Decrease	RBC count, Hb, Ht			
			rat	90	Days	Oral	2000	ppm	Increase	MCV, Reticulocytes			
			dog	90	Days	Oral	8000	ppm	Decrease	RBC count, Hb, Ht			
			dog	90	Days	Oral	8000	ppm	Increase	MCV, Reticulocytes; Hypercellularity of sternal, femoral BM (regenarative anemia)			
			dog	90	Days	Oral	4000	ppm	Increase	ALAT, ASAT (and ALP at 8000 ppm)			
			dog	1	Years	Oral	3500	ppm	Decrease	RBC count, Hb, Ht			
			dog	1	Years	Oral	3500	ppm	Increase	ALP			
			rat	12	Months	Oral	750	ppm	Decrease	RBC count, Hb, Ht			
		Food consumption	rat	90	Days	Oral	2000	ppm	Decrease	Food efficiency			
			rat	90	Days	Oral	2000	ppm	Decrease	Food efficiency & food consumption			
			mouse	90	Days	Oral		ppm	No effect				
			rat	131	Days	Oral	750	ppm	Decrease	Food consumption and/or food efficiency			
			rat	131	Days	Oral	750	ppm	Decrease	Food consumption during premating			
			rat	22	Days	Oral	350	mg/kg bw/day	Decrease	Food consumption - days 7-9G, 9-11G; increased days 17-22G			
			rabbit	13	Days	Oral	270	mg/kg bw/day	Decrease				

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			Rat	28	Days	Oral		mg/kg bw/day	No effect				
			rat	90	Days	Oral	750	ppm	Decrease				
			Rat	15	Days	Oral	500	mg/kg bw/day	Decrease	Food consumption and/or food efficiency			
			Rat	15	Days	Oral	2000	mg/kg bw/day	Decrease	Food consumption and/or food efficiency			
			Rat	15	Days	Oral	1000	mg/kg bw/day	Decrease				
			mouse	18	Months	Oral		ppm	No effect	Food consumption & efficiency			
		Mortality	rat	90	Days	Oral		ppm	No effect				
			rat	90	Days	Oral		ppm	No effect				
			mouse	90	Days	Oral		ppm	No effect				
			dog	90	Days	Oral	8000	ppm	Increase	2/4 females			
			dog	1	Years	Oral	3500	ppm	Increase	1/5 male and 1/5 female			
			rat	22	Days	Oral		mg/kg bw/day	No effect				
			rabbit	13	Days	Oral	270	mg/kg bw/day	Increase	Dose-related deaths: 2/20 at 270 mkd, 9/20 at 800 mkd			
			Rat	28	Days	Oral		mg/kg bw/day	No effect				
			rat	90	Days	Oral		ppm	No effect				
			rat	12	Months	Oral		ppm	No effect	No effect			

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			rat	22	Months	Oral		ppm	No effect	Poor survival in all groups typical for strain, not a compound-related effect. Study terminated at 22-m.			
			mouse	18	Months	Oral		ppm	No effect				