	Conclusions:	dietary administration of Thiabendazole produced no treatment-related mortality. slight increase in the incidence and severity of alopecia was noted along with marked decreases in body weight gain at the two highest dosage levels. no ophthalmic abnormalities were noted. based on body weight changes, doses of 10, 30 and 90 mg/kg/day are recommended for a subsequent carcinogenicity study in this species. based upon all treatment-related changes, the **NOEL** in this study is 10 mg/kg/day.
14	**Statistics**	body weights were analyzed by the following methods: Wilk and Shapiro W. Statistic, Trend Analysis, Rankit Transformaiton, Levene's test, Bartlett's Test, Dunnett's t-Test for Control versus Treatment comparisons **Trend (Dose-Response) Analysis** **Reference**: Tukey, J.W., Ciminera, J.L., and Heyse, J.F., Testing the Statistical Certainty of a Response to Increasing Doses of a Drug, Biometrics, 41: 295-301, 1985. **Multiple Comparisons with a Control** **Reference**: Dunnett, C.W., New Tables for Multiple Comparisons with a Control, Biometrics, 20: 482-491, 1964. **Test for Homogeneity of Variances** **Reference**: Levine, H.: Robust Tests for Equality of Variances, Contributions to Probability and Statistics. Essays in Honor of Harold Hotelling, Stanford University Press, Stanford, CA, 278-292, 1961. **Test for Normality of Data** **Reference**: Shapiro, S.S. and Wilk, M.B., An Analysis of Variance Test for Normality (Complete Samples), Biometrika, 52: 591-611, 1965. **Reference**: Wilk, M.B. and Shapiro, S.S., The Joint Assessment of Normality of Several Independent Samples, Technometrics, 10: 825-839, 1968. **Reference**: Shapiro, S.S. and Wilk, M.B., Approximations for the Null Distribution of the W Statistic, Technometrics, 10: 861-866, 1968. **Rankit Transformation** **Reference**: Harter H.L.: Expected Values of Normal Order Statistics. Biometrika, 48: 151-165, 1961. **Reference**: Tukey, J.W.: The Future of Data Analysis. Annals of Mathematical Statistics, 33: 1-67, 1962. **Bartlett's Test** **Reference**: Bartlett, M.S. (1937). Some examples of statistical methods of research in agriculture and applied biology. J. Royal Statist. Soc. Suppl. IV, pp. 137-170.
15	**References to**	

	publications	none
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	GLP and to US EPA Guidelines.
Results and discussion	[redacted]

[redacted]

Conclusion [redacted]

Reliability [redacted]

Acceptability [redacted]

Remarks [redacted]

98/8 Doc IIIA section No.	**6.5/ 01**	**Chronic oral toxicity test**
91/414 Annex Point addressed	**II 5.3.2 / 02**	Long-term toxicity - oral one year study

1.2	**Title**	Fifty-Three-Week Oral Toxicity Study in Dogs
1.3	**Report No.**	91-068-0
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.3.2/04
1.6	**Authors**	[redacted]
1.7	**Date of report**	20 January 1993
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	13 June 1991 to 11-12 June 1992
3	**Objective**	to determine the toxicity of Thiabendazole in dogs when administered for at least 1 year.
4.1	**Test substance**	Thiabendazole [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	adequate stability of Thiabendazole under the conditions employed within this study has been demonstrated
4.4	**Stability in vehicle**	not applicable
4.5	**Homogeneity in vehicle**	not applicable
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	none used, used as submitted (powder form in gelatin capsules)
6	**Physical form**	off-white powder
7.1	**Test method**	Oral Capsule Dog Toxicity Study
7.2	**Justification**	complied with OECD guidelines according to the 1981 publication
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility**	

	(official or officially recognized)	not applicable
11.3	**Justification**	not applicable
12	**Test system**	
	Animal species:	Dog (*Canis familiaris*) - beagle
	Source:	[redacted]
	Number of animals:	16 males and 16 females, assigned to 4 groups
	Age:	34-37 weeks
	Weight range:	8.9 to 12.3 kg males; 7.8 to 10.7 kg females
	Dosage:	10, 40 or 160 mg/kg/day
	Administration:	oral, by capsule
	Duration:	53 weeks
	General observations:	daily for mortality and clinical signs of drug effect, with less detailed examinations on weekends and holidays
	Ophthalmology:	funduscopic (indirect ophthalmoscopy) examination conducted prior to start of treatment and during drug weeks 27 and 50. Tropicamide (1%) solution was used to dilate the pupils. In drug Week 50, a biomicroscopic (slit lamp) examination was also conducted
	Food consumption:	measured based on an approximate 4-day intake, weekly in Drug Weeks 1-13, and every 4 weeks thereafter
	Body weight:	all animals were weighed pretest and weekly during the study
	Electrocardiograms:	recorded from all dogs, pretest and in approximately Drug Weeks 14, 25 and 50, in lateral recumbency approximately 3-6 hours after dosing
	Hematology:	exams conducted during the pretest period, and in Drug Weeks 4, 12, 26 and 52 on all dogs. In Drug Week 7 blood was also collected from one animal due to physical signs (pale mucous membranes, lethargy). Blood was withdrawn from jugular veins and the following parameters were determined: hematocrit, hemoglobin, platelet count, red blood cell count, white blood cell count (total and differential), prothrombin time, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular Hgb concentration, activated partial thromboplastin time
	Serum Biochemical:	performed at the same time points as hematologic examinations on all dogs. In addition, blood was collected from 2 animals in Drug Week 7 due to the appearance of physical signs. The following parameters were determined: total protein, alkaline phosphatase (AP), albumin, A/G ratio, total bilirubin, chloride, calcium, potassium, glucose, sodium, cholesterol, creatinine, AST, triglycerides, phosphorus, urea nitrogen, ALT, direct bilirubin
	Urinalysis:	were performed pretest and in Drug Weeks 4, 12, 26 and 52 on all dogs; collected overnight and analyzed for: pH, glucose, ketones, urobilinogen, occult blood, volume, specific gravity, protein, bilirubin, microscopy of sediment

13 Findings

Dosages	0 - 10 - 40 - 160 mg/kg/day
Clinical signs	incidence of emesis in group 4 in the first 3 weeks. By altering the feeding regimen the incidence declined in subsequent weeks. No treatment-related increase in emesis in low and mid-dose groups.
Feed intake	comparable to controls in all dose groups.
Mortality	one death during the course of the study, which may have been due to an idiosyncratic reaction to drug treatment or a pre-existing condition in this dog
Body weight development	comparable to controls in all dose groups. Initial weight gain slightly depressed in 2 females of the high dose group. This may be related to emesis, decreased food consumption and possibly other factors (see below)
Changes of uncertain relationship to treatment	2 dogs experienced weight loss over most of the study. One animal was found to have a Campylobacter infection, the other was not tested
Ophthalmoscopy	no ophthalmic changes related to treatment
Electrocardiograms	no changes related to treatment
Hematology	Erythroid parameters were decreased approximately 10-15% in the high-dose compared to controls throughout the study. 2 dogs had very slight to slight splenic erythropoiesis. Several dogs in the high-dose group had sporadically increased numbers of nucleated red blood cells in Drug Weeks 4, 7, 12, 26, or 52. There was also a higher incidence of polychromasia and hypochromia in the high dose relative to controls in Drug Weeks 4, 12 and 26. Mean corpuscular volume was increased in 2 dogs in Drug Week 4. These changes are most likely related to the decrease in erythroid parameters. The changes in erythron parameters were insufficient to affect the overall health of the high dose group animals. The hematology parameters for the mid- and low-dose groups were comparable to controls. Activated PTT was increased about 10% in the high-dose group throughout the study. Platelet number was also increased in the high-dose group, about 60% relative to controls in Drug Weeks 4 through 52. Most dogs in the group had increased values. The increase in platelet number was probably secondary to increased activated PTT. The activated PTT and platelet values for the mid- and low-dose groups were comparable to controls.

Serum Biochemical	no changes related to treatment
Urinalysis	no changes related to treatment

Histologic changes - Findings above control level			
FINDING	TBZ (mg/kg/day)		
	10	40	160
Gallbladder			
Mucosal discoloration (gross)	+	+	+
Cytoplasmic vacuolation	+	+	+
Inspissated secretion	+	+	+
Liver			
Increased liver weight		+	+
Bile duct vacuolation		+	+
Thyroid			
Increased thyroid weight			+
Follicle or follicular cell enlargement			+
Kidney			
Distal tubular vacuolation		+	+
Urinary bladder – epithelial			
Cytoplasmic inclusions		+	+
Spleen			
Hemosiderosis		+	+
Increased erythropoiesis		+	+
Bone marrow			
Increased erythropoiesis			+

	Conclusions:	Thiabendazole was generally well tolerated by dogs receiving doses for one year. Body weights and food consumption in all dose groups were comparable to controls. No serum biochemical, ophthalmic, urinalysis, or electrophysiological changes related to treatment were seen. Increased liver weight, erythropoiesis and hemosiderosis in the spleen, and lipid vacuolation in the urinary bladder, kidney, hepatic bile ducts and gallbladder were seen postmortem and considered treatment-related. With the exception of the gallbladder, the NOEL for all changes is 10 mg/kg/day. The gallbladder epithelial vacuolation was identified as lipid vacuoles by positive staining for oil-red-O and was found to a very slight or slight degree in most animals in the tested groups and in 1 of 8 concurrent controls. However, this change has been seen in historical controls in up to 50% of a given control group (3 of 6). Since the gallbladder change was minimal and similar to that found spontaneously in controls, it is not considered of toxicological significance. Therefore, the no-adverse-effect level (NOAEL) is 10 mg/kg/day.
14	**Statistics**	Statistical evaluation of the absolute and relative weights of liver, kidney, adrenal and thyroid were analyzed by the trend analysis. **Trend (Dose-Response) Analysis** **Reference:** Tukey, J.W., Ciminera, J.L., and Heyse, J.F.: Testing the Statistical Certainty of a Response to Increasing Doses of a Drug. Biometrics, 41: 295-301, 1985.
15	**References to publications**	Smith, P.F., Grossman, S.J., Gerson, R.J. et al: Studies on the Mechanism of Simvastin-induced Thyroid Hypertrophy and Follicular Cell Adenoma in the Rat. Tox. Path. 19: 197-205 (1991).
16	**Unpublished data**	R.N. Hill et al, Fund. and Appl. Toxicol. 12, 629-697, 1989.

Evaluation by Competent Authorities	
EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	April 2005
Materials and Methods	GLP and to US EPA Guidelines.
Results and discussion	[redacted]

[redacted]

Conclusion [redacted]

Reliability [redacted]

Acceptability [redacted]

Remarks

98/8 Doc IIIA section No.	**6.3.2 / 01**	**Subchronic dermal toxicity test**
91/414 Annex II Point addressed	**5.3.3 / 01**	Short-term toxicity - other routes

1.2	**Title**	Thiabendazole. Twenty-Three Day Dermal Toxicity Study in Rabbits.
1.3	**Report No.**	89-9011
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.3.3.1/01
1.6	**Authors**	[redacted]
1.7	**Date of report**	20 September 1989
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	8 March 1989 to 30 March 1989
3	**Objective**	to evaluate the dermal toxicity of the test material, Thiabendazole, in rabbits when administered daily via topical application to the unabraded skin for either 21 or 22 consecutive days.
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	satisfactory
4.4	**Stability in vehicle**	not applicable
4.5	**Homogeneity in vehicle**	not applicable
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	the drug was administered by placing the appropriate amount on a gauze pad moistened with about 1 ml of saline
6	**Physical form**	off-white powder
7.1	**Test method**	twenty-three day dermal toxicity study in rabbits
7.2	**Justification**	conducted in accordance with the recommended OECD guidelines
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection

10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable

12 Test system

Animal species:	Hra: (NZW)SPF rabbits
Source:	[redacted]
Number of animals:	40 animals, 20 males and 20 females
Age:	approx. 10 weeks upon receipt, approx. 12 weeks at start of study
Dosage:	50, 200 or 1000 mg/kg/day
Administration:	trunk of all animals was shaved dorsally, ventrally and laterally, care being taken not to abrade the skin. Application under gauze, with use of collars throughout the test
Duration:	6 hours exposure par day for 21 or 22 days
General observations:	twice daily for mortality and moribundity, additional cageside clinical signs were recorded at least once daily. Detailed physical examinations were performed on days 0, 7, 14 and 21.
Food consumption:	estimated daily throughout the study
Body weight:	recorded once prior to treatment (Day 0) and on Days 7, 14 and 21
Dermal irritation:	scored twice daily, immediately prior to the daily 6-hour application period and immediately following removal of the control and test materials, according to the method of Draize (1965)
Hematology:	conducted pretest and in Drug Week 3 for all animals. Prior to sample collection, the animals were food-fasted overnight with water available. Blood samples were collected via the medial ear artery. hematocrit, hemoglobin, platelet count, red blood cell count, white blood cell count (total, corrected and differential), mean cell volume, mean cell hemoglobin, mean cell hemoglobin concentration, cell morphology
Histopathology:	samples of liver, both kidneys, treated skin (application site) and untreated skin, and all gross lesions were analyzed
Blood chemistry:	conducted pretest and in Drug Week 3 for all animals. Prior to sample collection, the animals were food-fasted overnight with water available. Blood samples were collected via the medial ear artery. Blood urea nitrogen, total protein, albumin, A/G ratio (calculated), total bilirubin, chloride, calcium, potassium,

glucose, sodium, inorganic phosphate, globulin, creatinine, aspartate aminotransferase, alanine aminotransferase

Gross pathology: all animals were food-fasted overnight, weighed, anesthetized intravenously with sodium pentobarbital, and exsanguinated. The necropsy included gross examination of the following: external surface (treated and untreated areas), all orifices, cranial cavity, carcass, external surfaces of the brain and spinal cord and the cut surface of the spinal cord, nasal cavity and paranasal sinuses, thoracic, abdominal, and pelvic cavities and their viscera, cervical tissues and organs

Organ weights: the following organs from all animals at the scheduled terminal sacrifice were weighed after careful dissection and trimming of fat and other contiguous tissues: liver with gallbladder (undrained), kidneys (both), testes with epididymides

13 Findings

Summary Incidence of Clinical Observations[a]

	Males				Females			
Group	1	2	3	4	1	2	3	4
Dose level (mg/kg/day)	0[b]	50	200	1000	0	50	200	1000
Number observed	5	5	5	5	5	5	5	5
No. that appeared normal	5	3	5	4	5	5	4	4
Number found dead	0	0	0	0	0	0	0	0
Observation[c]								
Thin	0	1	0	1	0	0	1	0
Pale	0	1	0	0	0	0	0	0
Lacrimation - right eye	0	1	0	0	0	0	0	1
Slight erythema, slight edema and eschar formation surrounded the right eye	0	1	0	0	0	0	0	0

Summary Incidence of Dermal Irritation Scores

	Males				Females			
Group	1	2	3	4	1	2	3	4

[a] includes detailed weekly clinical observations (physical examinations)
[b] control animals received 1.0 ml of saline
[c] the numerals represent the number of animals with the designated finding at least once during the study

Dose level (mg/kg/day)	0[a]	50	200	1000	0	50	200	1000
Number scored	5	5	5	5	5	5	5	5
No. that appeared normal	5	4	5	4	5	4	5	3
Number found dead	0	0	0	0	0	0	0	0
Observation score[b]								
Very slight edema (1)[c] at predose observation	0	0	0	0	0	0	0	1
Very slight erythema (1) at postdose observation	0	1	0	0	0	1	0	1
Very slight edema (1) at postdose observation	0	0	0	1	0	0	0	2
Slight edema (2) at postdose observation	0	0	0	0	0	0	0	1

Conclusion: The dermal application of Thiabendazole did not result in any evidence of systemic toxicity or dermal irritation. Therefore, on the basis of this study, the dermal no-effect level (**NOEL**) for Thiabendazole in rabbits is >1000 mg/kg/day

14 Statistics

Numerical data obtained from this study were subjected to calculations of group mean values and standard deviations of the mean (when appropriate)

Bartlett's Test. Bartlett, M.S. (1937). Some examples of statistical methods of research in agriculture and applied biology. J. Royal Statist. Soc. Suppl. IV, 137-170.

15 References to publications

Draize, J.H. (1959). Dermal toxicity. In Appraisal of the Safety of Chemicals in Foods, Drugs and Cosmetics. The Editorial Committee of the Association of Food and Drug Officals of the United States, Austin, TX, pp. 46-49.

16 Unpublished data

not applicable

Evaluation by Competent Authorities

EVALUATION BY RAPPORTEUR MEMBER STATE

Date

April 2005

Materials and Methods

GLP and to US EPA Guidelines

[redacted]

[a] control animals received 1.0 ml of saline
[b] the numerals represent the number of animals with the designated finding at least once during the study
[c] the numbers in parentheses correspond to the appropriate Draize score

Results and discussion

[redacted]

Conclusion

[redacted]

Reliability

[redacted]

Acceptability

[redacted]

Remarks

98/8 Doc IIIA section No.	**6.6.1 / 01**	**In-vitro gene mutation study in bacteria**
91/414 Annex Point addressed	**II 5.4.1 / 01**	Genotoxicity Studies - *In vitro* testing

1.2	**Title**	Mutagenicity Testing on Thiabendazole in Microbial Systems, Host-mediated Assay.
1.3	**Report No.**	76-9814
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.4/01
1.6	**Authors**	[redacted]
1.7	**Date of report**	1 September 1976
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	1976
3	**Objective**	to investigate the microbial mutagenic potentials of Thiabendazole, a fungicide, in the host-mediated assay using *S. typhimurium* G46 mice
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	within acceptable limits
4.4	**Stability in vehicle**	within acceptable limits
4.5	**Homogeneity in vehicle**	homogenous suspension
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	suspended in water and gum arabic (5%)
6	**Physical form**	off-white powder
7.1	**Test method**	Host-mediated Assay using *S. typhimurium* G46 in Mice.
7.2	**Justification**	although no OECD guideline for this assay is available, this assay for mutation frequency has been extensively validated and is acceptable to Regulatory agencies worldwide.
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	as 10.3 - 10.4
10.2	**Certifying authority**	as 10.3 - 10.4
10.3	**GLP**	no

10.4	**Justification**	the host-mediated assay was conducted prior to the issuance of the Good Laboratory Practices regulations
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable
12	**Test system**	
	Animal species:	mouse (ICR strain)
	Dosage:	300 or 1000 mg/kg/day
	Duration:	5 days, then a single intraperitoneal injection with 2 ml (8.4 x 108 cells/ml) of *S. typhimurium* G46
	Positive control:	Dimethylnitrosamine
13	**Findings**	treated mice showed no increase in the frequency of mutations compared to the control group. Dimethylnitrosamine given as a single oral dose at 50 mg/kg and used as a positive control caused a significant increase in the mutation frequency.
14	**Statistics**	not applicable
15	**References to publications**	Ames, B.N., Durston, W.E., Yamasaki, E. and Lee, F.D. Proc. Natl. Acad. Sci. (U.S.A.) 70: 2281-2285, 1973.
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	[redacted]

[redacted]

Results and discussion [redacted]

Conclusion [redacted]

Reliability [redacted]

Acceptability [redacted]

Remarks

98/8 Doc IIIA section No.	**6.6.2 6/ 01**	**In ~~vitro~~ cytogenicity study in mammalian cells**
91/414 Annex Point addressed	**II 5.4.1 / 04**	Genotoxicity Studies - *In vivo* testing

1.2	**Title**	Dominant Lethal Study with Thiabendazole in Mice.
1.3	**Report No.**	76-9817
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.4/02
1.6	**Authors**	[redacted]
1.7	**Date of report**	1 September 1976
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	1976
3	**Objective**	to investigate the mutagenic potentials of Thiabendazole, a fungicide, in mice
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	within acceptable limits
4.4	**Stability in vehicle**	within acceptable limits
4.5	**Homogeneity in vehicle**	homogenous suspension
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	suspended in water and gum arabic (5%)
6	**Physical form**	off-white powder
7.1	**Test method**	Dominant Lethal Study in Mice.
7.2	**Justification**	study generally in compliance with OECD guidelines published in 1981.
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	as 10.3 - 10.4
10.2	**Certifying authority**	as 10.3 - 10.4
10.3	**GLP**	no
10.4	**Justification**	the host-mediated assay was conducted prior to the issuance of the Good Laboratory Practices regulations
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable
12	**Test system**	

Animal species:	mouse (C3H/HeCr strain)
Dosage:	200 or 600 mg/kg/day
Administration:	orally
Duration:	5 days, then mated with virgin females for 6 successive weeks
Positive control:	Ethylmethansulfonate

13	**Findings**	treated mice showed no increase in the frequency of induced dominant lethal mutations at any stage of spermatogenesis. Ethylmethansulfonate given as a single intraperitoneal dose at 300 mg/kg and used as a positive control did induce lethal mutations at the first and second week of testing. Therefore, Thiabendazole is considered negative in this *in vivo* mutagenicity assay.
14	**Statistics**	not applicable
15	**References to publications**	Röhrborn, G., in F. Vogel and G. Röhrborn (eds.), Chemical Mutagenesis in Mammals and Man: Springer-Verlag, 1970, pp. 148-155.
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	[redacted]

98/8 Doc IIIA section No.	**6.6.3 / 01**	**In-vitro gene mutation assay in mammalian cells**
91/414 Annex Point addressed	**II 5.4.1 / 02**	Genotoxicity Studies - *In vitro* testing

1.2	**Title**	Thiabendazole In Vitro DNA Alkaline Elution/Rat Hepatocyte Assay.
1.3	**Report No.**	89-8312
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.4/03
1.6	**Authors**	[redacted]
1.7	**Date of report**	12 May 1989
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	6 February 1989 - 10 February 1989
3	**Objective**	to determine whether Thiabendazole induces DNA strand breaks without concomitant induction of cytotoxicity in primary rat hepatocytes dosed *in vitro*.
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	within acceptable limits
4.4	**Stability in vehicle**	within acceptable limits
4.5	**Homogeneity in vehicle**	within acceptable limits
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	Dimethylsulfoxide (DMSO)
6	**Physical form**	off-white powder
7.1	**Test method**	In Vitro Alkaline Elution/Rat Hepatocyte Assay.
7.2	**Justification**	although no OECD guideline for this assay is available, this assay for genetic damage has been extensively validated and is acceptable to regulatory agencies worldwide.
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially**	

	recognized)	not applicable
11.3	**Justification**	not applicable
12	**Test system**	
	Cell type:	primary rat hepatocytes
	Cell source:	[redacted]: CD®(SD)BR, Sprague-Dawley, male
	Dosage:	0.3 to 1.3 mM (the maximum soluble concentration)
	Positive control:	Aflotoxin B at 1 mM
13	**Findings**	the relative survival after treatment with Thiabendazole ranged from 102 to 97% over the dose range. Alkaline elution results show that Thiabendazole did not produce any three-fold or greater increases in the elution slopes relative to the concurrent negative control slope at any non-toxic concentration tested, whereas the positive control, aflotoxin B, at a final concentration of 1 mM gave a 21.57-fold increase in elution slope. Based on these findings, Thiabendazole does not induce DNA strand breaks in isolated rat hepatocytes and thus is not likely to be a mammalian mutagen or carcinogen.
14	**Statistics**	none used
15	**References to publications**	none
16	**Unpublished data**	Range-finding Cytotoxicity in Rat Hepatocytes TT #88-8850

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	The study complied with GLP and the laboratory is subject to US EPA inspection. [redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	6.6.2 / 01	In-vitro cytogenicity assay in mammalian cells

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Reference	[redacted]
Comment	[redacted]
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	
Reliability	
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	**6.6.~~3~~ ~~4~~/ 01**	**In ~~vitro~~ gene mutation assay in mammalian cells**
91/414 Annex Point addressed	**II 5.4.1 / 02**	Genotoxicity Studies - *In ~~vitro~~ vivo* testing

1.2	**Title**	Thiabendazole: Assay for Chromosomal Aberrations in Mouse Bone Marrow
1.3	**Report No.**	94-8603
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.4/04
1.6	**Authors**	[redacted]
1.7	**Date of report**	11 July 1994
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	8 February 1994 to 4 May 1994
3	**Objective**	to determine the potential of Thiabendazole to induce chromosomal aberrations in bone marrow cells of male Crl: CD-1®(ICR)BR mice
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	tested and found to be within acceptable limits
4.4	**Stability in vehicle**	tested and found to be within acceptable limits
4.5	**Homogeneity in vehicle**	samples from the top, middle and bottom levels of the suspension of thiabendazole were assayed for drug concentration and uniformity and found to be within acceptable limits
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	0.5% methylcellulose, Fisher Lot #712680
6	**Physical form**	off-white powder
7.1	**Test method**	Assay for Chromosomal Aberrations in Mouse Bone Marrow
7.2	**Justification**	study in compliance with OECD guidelines published in 1981.
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable

11.1 GEP not applicable

11.2 Type of facility (official or officially recognized)
not applicable

11.3 Justification not applicable

12 Test system

Animal species: mouse [Crl: CD-1®(ICR)BR]

Source: [redacted]

Age: weanling, about 4 weeks old

Weight: males: 20.3 to 28.7 g (24.5 g average weight)

Dosage: 200, 667, 2000 mg/kg of compound at 0.1 ml/10 g body weight.
Single dose at time 0 for all groups

Number of animals, treatment and sacrifice schedule:

		No. of animals sacrificed		
		Sacrifice time		
Group	Treatment	6 HR	24 HR	48 HR
1	Negative control	12	12	12
2	TBZ: 200 mg/kg	8	8	8
3	TBZ: 667 mg/kg	8	8	8
4	TBZ: 2000 mg/kg	10[a]	10[a]	10[a]
5	Positive control high dose	-	4	-
6	Positive control low dose	-	8	-

a = according to our SOP, 10 animals are treated with the high dose to allow for the possibility of treatment-related deaths. Slides are analyzed from only 8 animals at each sacrifice time.

Administration: compound and negative control: oral gavage
positive control: intraperitoneal injection

Duration: positive control animals sacrificed at 24 hours.
compound and negative control animals sacrificed at 6, 24 and 48 hours.

Positive control: Mitomycin C 3.5 and 1.0 mg/kg, Sigma Lot #83H-2510

Negative control: vehicle, Fisher Lot #712680

Clinical observations: animals were examined prior to dosing, at selected times after drug administration, and before each sacrifice time

Sacrifice of animals and harvest of bone marrow cells:
intraperitoneal injection of 2 mg colchicine/kg body weight 3 hours prior to sacrifice.Animals were sacrificed by cervical dislocation. Both femurs of each animal were quickly removed and crushed, and bone marrow was harvested.

Slide analysis: generally, 50 cells were scored per mouse, except for high dose positive controls, where fewer cells were scored

Data calculation: for each mouse: percentage mitotic cells, total number of aberrant cells, percentage aberrant cells, total number of aberrations, frequency of aberrations/100 cells;for each group: mean percentage mitotic index, total number of aberrant cells, mean percentage of aberrant cells, total number of aberrations, mean frequency of aberrations/100 cells, total number of animals with aberrations.

13 Findings

Dosages	200, 667, 2000 mg/kg of compound at 0.1 ml/10 g body weight
Clinical signs	decreased activity and ptosis seen in the mid- and high-dose levels within 6 hours post-dosing. High-dose: bradypnea also seen. All mice appeared normal at 24 hours.
Mortality	no deaths

Conclusions: The positive control, Mitomycin C, induced highly significant increases in aberrations in this study.

There were no statistically significant increases in percentage of cells with chromosome aberrations in male mice treated with Thiabendazole when compared with the combined control mean.

A positive response is when 2 groups show significant increases in aberrations. Since there are no such increases in the present study, Thiabendazole was negative in the chromosome aberration test in mouse bone marrow under the conditions of this assay.

14 Statistics for each sacrifice time, the data were analyzed using a rankit transformation that was computed across the four TBZ dose groups. Trend was assessed for each time period separately using a simple linear regression analysis on three candidate (arithmetic, ordinal, arithmetic-logarithmic) dosage scalings.

15 References to publications

Tukey, J.W., Ciminera; J.L., and Heyse, J.F., (1985), "Testing the Statistical Certainty of a Response to Increasing Doses of a Drug", Biometrics, 41

16 Unpublished data Acute Toxicity Study. TT #94-2536

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	[redacted]
Results and discussion	[redacted]

	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	6.6.4/ 02	In–gene mutation assay in mammalian cells

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Reference	[redacted]
Comment	[redacted]
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	
Reliability	
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	**6.6.3 / 02**	**In-vitro gene mutation assay in mammalian cells**
91/414 Annex Point addressed	**II 5.4.1 / 03**	Genotoxicity Studies - *In vitro* testing

1.2	**Title**	Microbial Mutagenicity Assay in (*Salmonella typhimurium* and *Escherichia coli*)
1.3	**Report No.**	92-8074 and 92-8079
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.4.1.1/01; 5.4.1.2/01
1.6	**Authors**	[redacted]
1.7	**Date of report**	28 April 1993
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	21 October 1992 to 05 November 1992
3	**Objective**	to determine the potential of the test material to induce mutations in *Salmonella typhimurium* and *Escherichia coli*
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	within acceptable limits
4.4	**Stability in vehicle**	within acceptable limits
4.5	**Homogeneity in vehicle**	within acceptable limits
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	DMSO, 0.1 ml per plate
6	**Physical form**	solution
7.1	**Test method**	as 7.2
7.2	**Justification**	conducted in compliance with OECD and internationally accepted guidelines
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable

11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable
12	**Test system**	
	Salmonella typhimurium	strains TA1535, TA97a, TA98 and TA100,
	Escherichia coli	strains WP2, WP2 uvrA, and WP2 uvrApKM101,
		with and without activation by rat liver microsomal enzymes.
	Concentration range:	3 to 6000 μg/plate (the highest soluble concentration)
	Positive Control compounds:	2-aminoanthracene and hydrazine sulfate
13	**Findings**	Thiabendazole did not produce any 2-fold or greater increases in revertants relative to the solvent control and is thus considered negative in this assay. The positive control compounds, 2-aminoanthracene and hydrazine sulfate, produced the expected increase in revertants.
14	**Statistics**	none
15	**References to publications**	Ames, B.N., J. McCann, and E. Yamasaki. Methods for detecting carcinogens and mutagens with the *Salmonella*/mammalian microsome mutagenicity test. Mutation Research 31: 347-364, 1975.
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	[redacted]
Results and discussion	Precipitate was seen on the plates at test concentrations as low as 1000 g/plates. [redacted]

	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	**6.7 / 01**	**Carcinogenicity study**
91/414 Annex II Point addressed	**II 5.5.1 / 01**	Long-term Toxicity and Carcinogenicity

1.2	**Title**	Thiabendazole: Lifetime Carcinogenic Study in Mice
1.3	**Report No.**	77-014-0
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.5/01
1.6	**Authors**	[redacted]
1.7	**Date of report**	12 December 1979
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	29 March 1977 to 28 March 1979
3	**Objective**	to determine the carcinogenic potential of thiabendazole in mice
4.1	**Test substance**	Thiabendazole, active substance as manufactured, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	material remained chemically unchanged during the study
4.4	**Stability in vehicle**	confirmed
4.5	**Homogeneity in vehicle**	concentration and uniformity confirmed
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	Purina Certified Rodent Chow
6	**Physical form**	white powder
7.1	**Test method**	Lifetime Carcinogenic Study in Mice
7.2	**Justification**	OECD and US EPA guidelines
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable

11.3 Justification not applicable

12 Test system

Animal species: Charles River CD-1 (HaM/ICR) mice

Source: [redacted]

No. of animals: 50 male and 50 female in each dosage group: 300 of each sex in all

Age: approximately 4 weeks old

Dosage (a.s.): 0.022, 0.066 and 0.2% for males,

0.066, 0.2 and 0.533% for females: different cc used for males and females after range-finding study

Starting the 7th week, lowest concentrations for males and females reduced to 0.006%.

Administration: oral by feeding

Duration: up to 105 weeks

General observations: daily for physical signs, although less detailed on weekends and on holidays. All animals were palpated for masses generally once a week

Food consumption: determined once a week

Body weight: determined pretest and once a week

Ocular examinations: on all surviving mice in the 82 or 83 and 101 weeks of the study

Haematology, Clinical chemistry, Urinalysis, Enzyme induction assay

were not analyzed as the rat study is the definitive chronic toxicity study, whereas in the mouse, only tumours are analysed

Gross pathology: conducted on all animals

Histopathology: conducted on all animals

13 Findings

Dosages	0.022, (0.006% DW 7-termination) 0.066 and 0.2% for males 0.066, (0.006% DW 7-termination) 0.2 and 0.533% for females
Clinical signs	
Feed intake	decreased feed intake for males and females given highest and middle concentrations
Mortality	greater mortality seen in males and females given middle and highest concentrations
Body weight development	decreased weight gains for males and females given highest and middle concentrations
Gross pathology	increased incidence of atrial thrombosis in males and females of the highest concentration, and females of the middle concentration

Organ weights	liver weights increased in highest concentration, and females on middle concentrations kidney weights in middle concentrations and females on highest concentration were lower than controls

Conclusions: The 0.006% concentration is equal to 60 parts per million and is considerably above the Acceptable Daily Intake (ADI) value of 0.1 parts per million (mg/kg) and, therefore, thiabendazole is safe for use on agricultural commodities.

14 Statistics

All tumor data were evaluated using a modified Mantel-Haenszel procedure taking into account time to tumor using a life-table analysis.

Statistical analyses were performed on terminal body weights and organ weights. Methods employed were tests for homogeneity of variance (Levene's test), tests for normality of data (Wilk and Shapiro), and analysis of variance.

15 References to publications

Reference: Walker, A.I.T., Thorpe, E. and Stevenson, D.E. (1972). The Toxicology of Dieldrin (HEOD).
1. Long-term toxicity studies in mice, Fd Cosmet, Toxicol. II: 415-432.

Leven's Test:

Reference: Draper, N.R., and Hunter, W.G. (1969). Transformations: some examples revisited. Technometrics, 11: pp. 23-40.

Reference: Levene, H. (1960). Robust tests for equality of variances. In Contributions to Probability and Statistics (I. Olkin, ed.). Stanford University Press, Palo Alto, pp. 278-292.

Test for Normality or Data

Reference: Shapiro, S.S. and Wilk, M.B., An Analysis of Variance Test for normality (Complete Samples), Biometrika, 52: 591-611, 1965.

Reference: Wilk, M.B. and Shapiro, S.S., The Joint Assessment of Normality of Several Independent Samples, Technometrics, 10: 825-839, 1968.

Reference: Shapiro, S.S. and Wilk, M.B., Approximations for the Null Distribution of the W Statistic, Technometrics, 10: 861-866, 1968.

16 Unpublished data

not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	**6.7 / 02**	**Carcinogenicity study**
91/414 Annex Point addressed	**II 5.5.2 / 01**	Long-term Toxicity and Carcinogenicity

1.2	**Title**	Thiabendazole: One-Hundered-Six-Week Dietary Toxicity/Carcinogenicity Study in Rats
1.3	**Report No.**	90-9009
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.5/02
1.6	**Authors**	[redacted]
1.7	**Date of report**	27 September 1993
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	23 August 1990 to 28 August 1992
3	**Objective**	to evaluate the chronic toxicity and carcinogenic potential of thiabendazole when fed to rats for at least 104 weeks
4.1	**Test substance**	Thiabendazole, [redacted] (based on thin layer chromatography $HClO_4$ titration)
4.2	**Specification**	[redacted]
4.3	**Storage stability**	was conducted and found to be within acceptable limits prescribed by GLP (documented in data)
4.4	**Stability in vehicle**	thiabendazole is stable in rodent feed at room temperature for at least 3 weeks at the concentration range used in this study
4.5	**Homogeneity in vehicle**	results of analyses indicated that the diet mixtures were homogenous
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	Purina Certified Rodent Chow #5002
6	**Physical form**	white powder
7.1	**Test method**	106-week dietary toxicity/carcinogenicity in rats
7.2	**Justification**	in compliance with the OECD guidelines according to the 1981 publication
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	Log of Protocol and/or GLP Deviations in Appendix II of final report. These deviations were considered minor and did not affect the conclusion of the study

10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable

12 Test system

Animal species:	rats [strain: Crl:CD®(SD)BR]
Source:	[redacted]
No. of animals:	500 animals (250 male and 250 female)
Age:	approximately 6 weeks old on dosing
Weights:	216 to 280 g for the males, and 130 to 194 g for the females
Dosage (a.s.):	10, 30, 90 mg/kg/day
Administration:	oral by feeding
Duration:	at least 104 weeks
General observations:	observations for mortality and moribundity twice daily. Cageside observations for obvious indications of toxic effects were performed daily. Physical examinations were conducted at each weighing interval
Food consumption:	recorded weekly
Body weight:	recorded at randomization, prior to treatment, and weekly thereafter
Ophthalmoscopic examinations:	indirect ophthalmoscopic examination on each animal prior to treatment, during week 52 and at termination week (week 104) using 1% Mydriacyl® as the mydriatic agent
Haematology:	samples obtained via orbital sinus venipuncture of the right eye of animals anethetized via CO_2/O_2 inhalation. Samples were also collected for haematology on animals sacrificed in a moribund condition (when possible) Following parameters were determined: activated partial thromboplastin time (APTT), cell morphology, corrected leukocyte count (COR WBC), erythrocyte count (RBC), mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), hematocryt (HCT), hemoglobin (HGB), leukocyte count (WBC), leukocyte differential mean cell volume (MCV), Platelet, prothrombin time (PT)
Clinical chemistry:	Following parameters were determined: alanine aminotransferase (ALT), albumin, albumin/globulin ratio (A/G), alkaline phosphatase (ALK P), aspartate aminotransferase (AST), blood urea nitrogen (BUN), calcium, chloride, creatine kinase (CK), creatinine (CREAT), globulin,

	glucose, inorganic phosphorus (IN PHOS), potassium, sodium, total bilirubin (T BILI), total cholesterol (T CHOL), total protein (T PROT)
Urinalysis:	Following parameters were determined: appearance, bilirubin, glucose, ketones, microscopic examination of sediment, occult blood, protein, specific gravity, urine volume
Gross necroscopy:	all animals which were found dead or sacrificed in extremis during the study were subjected to a gross postmortem examination. All surviving animals were fasted overnight, weighed on the day of scheduled necroscopy, anesthetized by CO_2 inhalation, and exsanguinated. Necropsies included examination of the following: all orifices, carcass, cervical tissues and organs, cranial cavity, external surface of the body, oral cavity and tongue, external surface of the brain and spinal cord, nasal cavity and paranasal sinuses, thoracic, abdominal and pelvic cavities and their viscera
Organ weights:	at terminal sacrifice, the following organs from the first 10 surviving animals/sex/group (except only 9 group 4 females) were weighed after careful dissection and trimming of fat and other contiguous tissue: adrenals (postfixation), brain (including brainstem), epididymides, heart, kidneys, liver, ovaries (postfixation), prostate (ventral), pituitary (postfixation), testes, thyroid/parathyroids (postfixation), uterus
Histopathology:	conducted on all animals

13 Findings

Dosages	0, 0, (2 control groups) 10, 30, 90 mg/kg/day
Clinical signs	no apparent compound-related differences noted between control and test groups
Feed intake	decreased feed intake for males and females correlated with decreases in body weight gain. No effect on food consumption was found for the low-dose males and females when compared to controls
Mortality	at the completion of the 104 weeks of treatment, survival of the high-dose males (74%) and females (51%) was higher than either respective control group (62 and 70% for males and 36 and 46% for females). No significant decrease in survival for any of the treated groups as compared to controls.
Body weight development	mean body weight generally were lower for the. mid- and high-dose males and high-dose females throughout the study. No effects on body weight gain was found for low-dose males and females
Haematology	statistical evaluation of the erythrocyte, hemoglobin and hemocrit values showed significant diffrences: see table 1 below. These changes may be treatment-related, but the magnitude of the changes was low (about 6% or less as compared to controls) and they occured inconsistently over time. The remaining haematology data were comparable for treated and control groups.

Clinical chemistry	mild, treatment-related increases in total cholesterol were observed in high-dose animals. No other treatment-related findings were noted in the remaining clinical chemistry data
Urinalysis	treated and control groups were comparable throughout the study
Ophthalmology	no compound-related ocular abnormalities
Gross pathology	the incidence of findings was comparable for treated and control groups for the scheduled and unscheduled deaths
Organ weights	statistical analysis of the liver and thyroid/parathyroid data showed significantly higher liver-to-body-weight ratio for the high-dose males and thyroid/parathyroid-to-body-weight ratio for the high-dose females as compared to the values for the respective control groups.
Histopathology	Also an increased incidence of thyroid follicular cell hypertrophy and hyperplasia and benign adenomas were seen. See Table 2 below.

Key: ↓ = significantly decreased as compared to combined control groups 1 and 2 value, $p \leq 0.05$

Table 1: Clinical Haematology data

	Males			Females		
Parameter Group	3	4	5	3	4	5
Erythrocyte count						
Week 14			↓			
Week 53			↓			
Hemoglobin						
Week 14			↓			
Week 53			↓		↓	↓
Hematocrit						
Week 14			↓			
Week 53			↓		↓	↓
Week 105		↓				

Table 2: The Incidence of Various Thyroid Lesions

	Control 1		Control 2		Group 3		Group 4		Group 5	
Thyroids	F	M	F	M	F	M	F	M	F	M
Number examined	50	50	50	50	50	50	50	50	50	50
Diffuse Follicular Cell Hypertrophy	0	0	0	0	0	0	0	1	2	4

Focal Cystic Foll. Cell Hyperplasia	2	0	1	4	0	2	3	1	6	3
Follicular Cell Adenoma	1	0	2	0	0	1	1^{NS}	5^{NS}	5^{S}	6^{S}
Follicular Cell Carcinoma	1	0	0	1	0	0	0	0	0	1

S = statistically different compared to combined control (P≤0.05)
NS = not statistically different compared to combined control (P>0.05)

Conclusions: based on an analysis of all antemortem data, the no-observed-effect level (**NOEL**) for thyroid adenomas in this study is 10 mg/kg/day and for all other tumour sites is > 90 mg/kg/day.

due to increased liver weight and the hepatic metabolism of thiabendazole, the increased incidence of thyroid tumours is likely to be related to increased clearance of thyroxine and increased thyroid-stimulating hormone levels. This mechanism is well documented in rodents but does not occur in humans (Hill, et al.). Therefore, this increase in benign thyroid adenomas is not considered a risk for human exposure.

14 Statistics

Cumulative survival data were analyzed. Analysis of body weights were performed. The incidence of various tumor types were analyzed for statistically significant trend (P ≤ 0.05) with adjustments made for variations such as mortality.

Analysis of Variance
Reference: Winer, B.J. (1971). *Statistical Principles in Experimental Design*, McGraw-Hill, New York, 2nd Edition, pp. 149-220.

Dunnett's t-Test for Control vs. Treatment Comparisons
Reference: Dunnett, C.W. (1955). A multiple comparison procedure for comparing several treatments with a control. *J.Am.Stat.Assoc.* **50**, pp. 1096-1121.

Reference: Dunnett, C.W. (1964). New tables for multiple comparisons with a control. *Biometrics* **20**, pp. 482-491.

Levene's Test:
Reference: Draper, N.R., and Hunter, W.G. (1969). Transformations: some examples revisited. *Technometrics* **11**, pp. 23-40.

Reference: Levene, H. (1960). Robust tests for equality of variances. In *Contributions to Probability and Statistics* (I. Olkin, ed.). Stanford University Press, Palo Alto, pp. 278-292.

Life Table/Time-to-Tumor Analysis (The NCI Package)
Reference: Thomas, D.G., Breslow, N., and Gart, J.J. (1977). Trend and homogeneity analysis of proportions and life table data. *Comput. Biomed. Res.* **10**, pp. 373-381.

Testing for Trend/Assessment of Mortality
Reference: Mantel, N. (1957). Chi-square Tests with One Degree of Freedom; Extensions of the Mantel-Haenszel Procedure. *Journal of the American Statistical Association* **58**, 1963, pp. 690-700.

Pairwise t-Test

Winer, B.J. (1971). Statistical Principles in Experimental Design, McGraw-Hill, New York, 2nd Edition, pp. 26-44.

Bartlett's Test

Bartlett, M.S. (1937). Some examples of statistical methods of research in agriculture and applied biology. J. Royal Statist. Soc. Suppl. IV, pp. 137-170.

15	**References to publications**	R.N. Hill et al., Fundamental and Applied Toxicology, 12, 629-697, 1989. Fujii, T., Mikuriya, H., Sasaki, M., Fd. Chem. Toxic., 29, 771-775, 1991.
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	April 2005
Materials and Methods	[redacted]
Results and discussion	[redacted]

[redacted]

Conclusion [redacted]

Reliability [redacted]

Acceptability [redacted]

Remarks

98/8 Doc IIIA section No.	**6.8.1 / 01**	**Teratogenicity test**
Annex Point addressed	**II 5.6.2 / 01**	Developmental toxicity studies

1.2	**Title**	Thiabendazole: Oral Developmental Toxicity Study in Rabbits
1.3	**Report No.**	89-9005
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.6.2/01
1.6	**Authors**	[redacted]
1.7	**Date of report**	27 October 1989
1.8	**Published**	Published in Food and Chemical Toxicology, 31 (1993), pp. 199-207.
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	8 February 1989 to 31 March 1989
3	**Objective**	To assess the developmental toxicity of Thiabendazole when administered orally on Days 6 through 18 of gestation in rabbits
4.1	**Test substance**	Thiabendazole [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	within acceptable limits
4.4	**Stability in vehicle**	suspensions of thiabendazole in the concentration range used have been documented to be stable for at least 24 hours.
4.5	**Homogeneity in vehicle**	homogeneity of the dosing suspensions was within acceptable limits.
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	0.5% methylcellulose
6	**Physical form**	off-white powder
7.1	**Test method**	1981 OECD guidelines
7.2	**Justification**	not applicable
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	rabbits dosed on days 6-18 of gestation instead of 7-19 as suggested in the guidelines (does not significantly affect the validity of the study)
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes

10.4 Justification not applicable

11.1 GEP not applicable

11.2 Type of facility (official or officially recognized) not applicable

11.3 Justification not applicable

12 Test system

Animal species: rabbits (Hazleton Research Products [Hra: (NZW)SPF]

Source: [redacted]

Pennsylvania 17517, USA

Number of animals: 72, all female

Age at artificial insemination: approx. 23 weeks

Dosage: 600 mg/kg/day: high dose120 mg/kg/day: mid-dose24 mg/kg/day: low dose

Administration: orally by gavage

Duration: once daily, days 6 through 18 of presumed gestation

General observations: daily check during acclimation period and predosage periodat least twice daily check during dosage (Days 6-18)daily check of general health and/or signs of abortionpostdosage (Days 19-29)

Food consumption: daily check from day 0 to 29

Body weight: at least twice prior to study assignment, on Day 0 and daily during dosage (Days 6-18) and postdosage (Days 19-29)

Sacrifice and reproductive status: on day 29 of presumed gestation, the animals were sacrificed by intravenous injection of T-61® Euthanasia Solution.

The uterus of each rabbit was examined to determine the reproductive status. The rabbits were also examined for gross lesions

Necropsy: gross evaluation of skin, fur, thoracic and abdominal viscera (including reproductive status)

Examination of fetuses: uterine implantation sites were counted and classified as alive or dead fetuses, or as early or late resorptions.All live fetuses were weighed and examined for external lterations. All fetuses were sexed and examined for visceral alterations by fresh visceral dissection, including examination of the brain. Skeletal examination.

Histopathology: tissues with gross lesions were saved in neutral buffered 10% formalin for possible histological evaluation

13 Findings

Dosages	0, 24 (low), 120 (mid), 600 (high) mg/kg/day
Clinical signs	one high-dose death was preceded by clonic convulsions and vocalization

Feed intake	decreased for mid to high dosage groups. no effect for low group.
Mortality	in high group, one treatment-related death on Day 14 of gestation and 4 treatment-related abortions, 2 on Day 20, and 1 on Days 21 and 26
Body weight development	0.23 kg average body weight loss for high group, decreased body weight gain (86% of control) for mid group. no effect for low group.
Embryo-fetal loss	treatment-related increases at mid and high doses. Mid dose: resorption of litters in 4 does. High dose: 4 abortions and 17.8% resorption rate, compared to 4.7% in controls.
Fetus alterations	2 in the high group, one in the mid group showed hydrocephaly and related alterations.

Conclusion: the no observed effect level (**NOEL**) for both maternal and developmental toxicity is 24 mg/kg/day.

14 Statistics NOSTASOT (sequential trend) analysis.

1. Tukey, J.W., Ciminera, J.L., and Heyse, J.F., "Testing the Statistical Certainty of a Response to Increasing Doses of a Drug", Biometrics, Vol.41, March, 1985, pp. 295-301.
2. Haseman, J.K. and Hogan, M.D., "Selection of the Experimental Unit in Teratology Studies", Teratology, Vol. 12, 1976, pp. 165-171.
3. Edgington, E.S., Randomization Tests, New York: Marcel Dekker, Inc., 1980.
4. Snedecor, G.W. and Cochran, W.G. (1967). Analysis of Variance. Statistical Methods, 6th Edition, Iowa State University Press, Ames, Iowa, pp. 259-275.
5. Sokol, R.R. and Rohlf, F.J. (1969). Bartlett's test of homogeneity of variances. Biometry, W.H. Freeman and Co., San Francisco, pp. 370-371.
6. Dunn, O.J. (1964). Multiple comparisons using rank sums. Technometrics, 6(3): 241-252.
7. Dunnett, C.W. (1955). A multiple comparison procedure for comparing several treatments with a control. J. Amer. Stat. Assoc., 50: 1096-1129.
8. Sokol, R.R. and Rohlf, F.J. (1969). Kruskal-Wallis Test. Biometry, W.H. Freeman and Co., San Francisco, pp. 388-389.

15 References to publications

see point 1.8 of this document, reference included at the end of the K document

Hamilton, W.J., Boyd, J.D. and Mossman, H.W. (1972). Human Embryology, 4th Edition, Williams and Wilkins Company, Baltimore, MD

16 Unpublished data not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	May 2005
Materials and Methods	The study complied with GLP and the laboratory is subject to US EPA inspection [redacted]
Results and discussion	[redacted]
Conclusion	Maternal toxicity was apparent at 600 mg/kg/day as 1 death, 4 abortions, and [redacted]
Reliability	[redacted]

Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	**6.8.1 / 02**	**Teratogenicity test**
Annex Point addressed	**II 5.6.2 / 02**	Developmental toxicity studies

1.2	**Title**	Thiabendazole: Oral Developmental Toxicity Study in Rats
1.3	**Report No.**	90-713-0
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.6.2/02
1.6	**Authors**	[redacted]
1.7	**Date of report**	28 November 1990
1.8	**Published**	Published in Food and Chemical Toxicology, 31 (1993), pp. 199-207.
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	2 July 1990 to 26 July 1990
3	**Objective**	To evaluate the potential developmental toxicity of Thiabendazole (TBZ) when administered to pregnant rats from gestational days (GD) 6 to 17.
4.1	**Test substance**	Thiabendazole [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	not applicable
4.4	**Stability in vehicle**	the compound has been shown to be stable in this vehicle under the conditions of this study
4.5	**Homogeneity in vehicle**	all assay results were within acceptable limits
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	0.5% (w/v) methylcellulose in deionized water
6	**Physical form**	off-white powder
7.1	**Test method**	Oral Developmental Toxicity Study - Rats
7.2	**Justification**	in compliance with the recommended OECD guidelines according to the 1981 publication
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	analysis of the test substance was conducted under GMPs and not under GLPs
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes

10.4 Justification not applicable

11.1 GEP not applicable

11.2 Type of facility (official or officially recognized) not applicable

11.3 Justification not applicable

12 Test system

Animal species: rat (Sprague-Dawley) Crl:CD(SD)BR

Source: [redacted]

Number of animals: 100, all female

Age: approx. 10 weeks at initiation

Weight: 210-310 g

Dosage: 80 mg/kg/day: high dose
40 mg/kg/day: mid-dose
10 mg/kg/day: low dose

Administration: orally by gavage with metal catheter

Duration: once daily, days 6 through 17 of gestation

Mating: each female was housed with 1 untreated male of the same strain. Females were selected for the study when daily examination revealed the presence of copulatory plugs. Females with only one plug below the cage floor were lavaged to check for the presence of sperm in the vagina. The day of finding the plugs or sperm was considered GD 0

Physical signs: once daily check on GD 0 and 6-20, with an additional observation at 1-5 hours after dosing during the treatment period.

Food consumption: was measured for all animals on GD 3-5, 6-8, 9-11, 12-14, 15-17, 18-20 (2-day periods)

Body weight: recorded on GD 0, 6, 8, 10, 12, 14, 16, 18, and 20

Sacrifice and pregnancy status: all females were euthanized on GD 20 by CO_2 asphyxiation. The uterus of each female was examined to determine pregnancy status. The number of corpora lutea were counted

Necropsy: gross evaluation of thoracic and abdominal viscera was done on all F0 females

Examination of fetuses: implants were counted and classified as alive or dead fetuses, or resorption. All fetuses were weighed and examined externally. Approximately one-half of the fetuses in each litter were given a visceral examination by dissection.

13 Findings

Dosages	0, 10 (low), 40 (mid), 80 (high) mg/kg/day

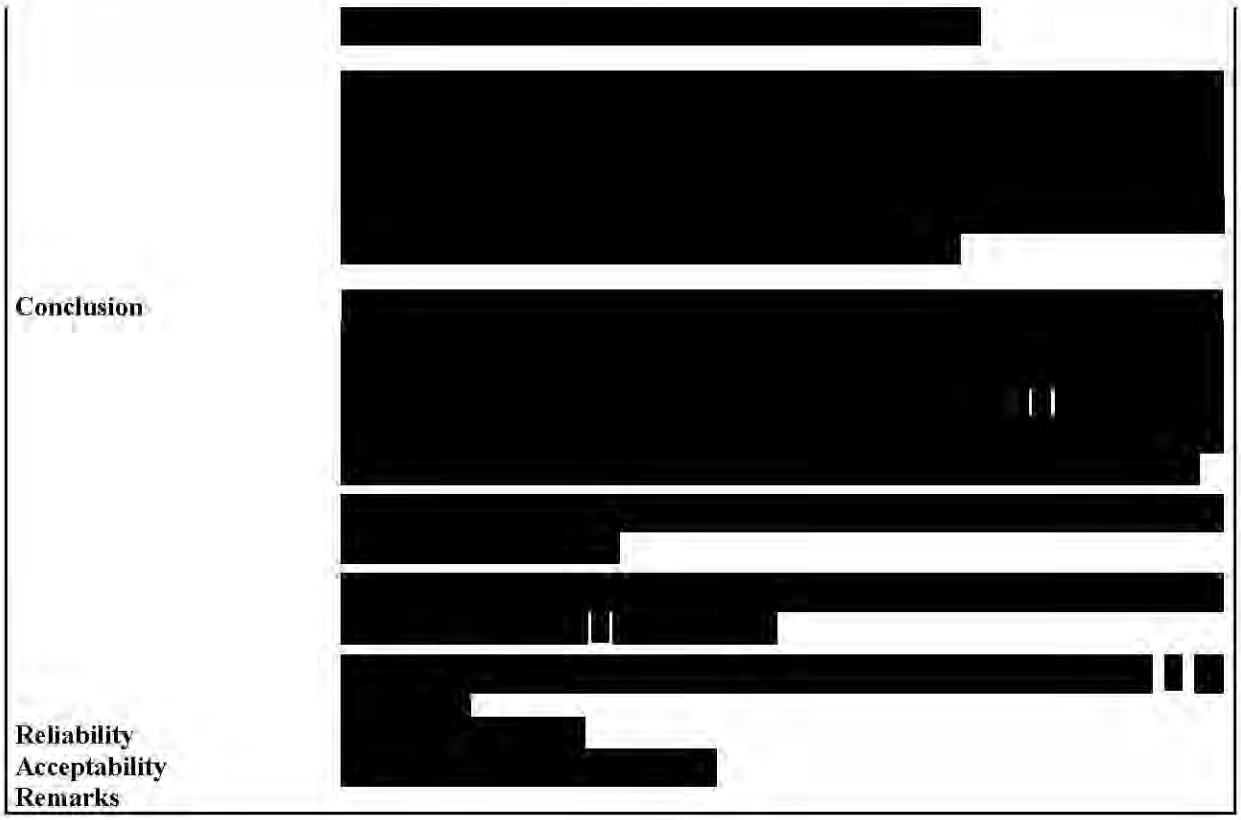
Physical signs	treatment-related physical signs were observed only in the high-dose group and consisted of ptosis in 4 females on GD 6 and regurgitation of some dosage suspension in 3 females on 1 or 2 days Alopecia observed in 1 to 3 females of all groups and considered incidental
Food consumption	There were dose- and treatment-related decreases in average maternal food consumption in the mid- and high-dose groups during the dosing period. There were no effects on average maternal food consumption in the low-dose group
Mortality	no deaths or abortions during the study.
Maternal body weight	a significant ($P \leq 0.05$) dose and treatment-related decreases in average maternal weight gain in the mid- and high-dose groups during the dosing period (GD 6 to 18). These effects were mainly due to a significant ($P \leq 0.05$) decrease in weight gain in the mid-dose group and a weight loss in the high-dose group between GD 6 and 8. There were no effects on average maternal weight gain in the low-dose group.
Embryo survival	there were no treatment-related effects based on pre-implantation loss, the percent resorptions plus dead fetuses/implants, implants/pregnant female, and live fetuses/pregnant female.
Live fetal weight	slight but dose- and treatment-related decreases in mean live fetal weight in the mid- and high-dose group. These decreases, except for the mid-dose males, were statistically significant ($P \leq 0.05$). No significant ($P > 0.05$) or treatment-related effects in the low-dose group. Mean gravid uterine weights were comparable in all groups.
Fetal examinations	no external anomalies were observed in any group. Visceral examinations revealed no treatment-related effects. Single fetuses in the mid- and high-dose groups had visceral variations, but due to their isolated occurrence these findings were not considered to be treatment-related. Skeletal examinations revealed no treatment-related effects.
Maternal necroscopy	no treatment-related gross lesions

Conclusion: The only evidence of developmental toxicity was slight but statistically significant ($P \leq 0.05$) decreases in the mean live fetal weight in the mid- and high-dose groups, but there was no concomitant evidence of alteration in external, visceral, or skeletal morphology.

Based on these results, the **NOEL** of TBZ in rats for maternal and developmental toxicity is 10 mg/kg/day.

14	**Statistics**	Statistical analyses were based on a trend test in which it was determined if there was a significant ($P \leq 0.05$) trend with increasing dosage including all treatment groups. Nonparametric data were normalized by a rankit method when appropriate. **Estimation of Linear and Quadratic Coefficients (Body Weights)** **Reference**: Robson, D.S.: A Simplified Method for Constructing Orthogonal Polynomials When Independent Variable is Unequally Spaced. Biometrics 15: 187-191, 1959. **Trend (Dose Response) Analysis** **Reference**: Tukey, J.W., Ciminera, J.L., and Heyse, J.F., "Testing the Statistical Certainty of a Response to Increasing Doses of a Drug", Biometrics, Vol. 41, March, 1985, pp. 295-301. **Rankit Transformation** **Reference**: Harter, H.L.: Expected Values of Normal Order Statistics. Biometrika 48: 151-165, 1961. **Reference**: Tukey, J.W.: The Future of Data analysis. Annals of Mathematical Statistics 33: 1-67, 1962.
15	**References to publications**	see point 1.8 of this document, reference included at the end of the K document in report 5.6.2/01.
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	May 2005
Materials and Methods	The study complied with GLP and the laboratory is subject to US EPA inspection [redacted]
Results and discussion	[redacted]


Conclusion
Reliability
Acceptability
Remarks

98/8 Doc IIIA section No.	6.8.1 / 03	Teratogenicity test
Annex Point addressed	II 5.6.2 / 02	Developmental toxicity studies

1.2	Title	Thiabendazole: Oral Developmental Toxicity Study in Rabbits
1.3	Report No.	90-734-0
1.4	Lab. report No.	not applicable
1.5	Cross reference	5.6.2/03
1.6	Authors	[redacted]
1.7	Date of report	10 June 1991
1.8	Published	Published in Food and Chemical Toxicology, 31 (1993), pp. 199-207.
2.1	Testing facility	[redacted]
2.2	Dates of experimental work	28 November 1990 to 28 December 1990
3	Objective	To examine the potential for development toxicity in rabbits following oral administration of Thiabendazole (TBZ) on Gestational Days (GD) 6 through 18. In addition, this study was designed to help clarify the findings of a previous Oral Developmental Toxicity Study in Rabbits (TT #89-9005).
4.1	Test substance	Thiabendazole [redacted]
4.2	Specification	[redacted]
4.3	Storage stability	not applicable
4.4	Stability in vehicle	the compound has been shown to be stable in this vehicle under the conditions of this study
4.5	Homogeneity in vehicle	all assay results were within acceptable limits
4.6	Validity	not applicable
5	Vehicle/solvent	0.5% methylcellulose
6	Physical form	off-white powder
7.1	Test method	Oral Developmental Toxicity Study - Rabbits
7.2	Justification	in compliance with the recommended OECD guidelines according to the 1981 publication
7.3	Copy of method	not applicable
8	Choice of method	not applicable
9	Deviations	analysis of the test substance was conducted under GMPs and not under GLPs
10.1	Certified laboratory	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	Certifying authority	the study complied with GLP and the laboratory is subject to US EPA inspection

10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable

12 Test system

Animal species:	rabbits (New Zealand White)
Source:	[redacted]
Number of animals:	72, all female
Age at artificial insemination:	25 to 26 weeks
Weight at initiation:	2897 to 3811 g
Dosage:	600 mg/kg/day: high dose 150 mg/kg/day: mid-dose 50 mg/kg/day: low dose
Administration:	orally by rubber catheter
Duration:	once daily, days 6 through 18 of presumed gestation
Artificial insemination:	following administration of 25 USP units of HCG (human chorionic ganodotropin) intravenously on Day 0, virgin females were inseminated with at least 0.25 ml of a diluted pooled semen sample which was collected from 3 untreated males and contained motile sperm
Physical signs:	once daily check except during the dosage period when females were observed at dosing and 1 to 5 hours after the dosing. In addition, a postdose examination for mydriasis and/or slowed pupillary reflex was performed on all females on GD 12.
Food consumption:	was measured during 24-hour intervals from GD 0-1, 3-4, 6-7, 9-10, 12-13, 15-16, 18-19, 21-22, 24-25, and 27-28.
Body weight:	recorded on GD 0, 6, 8, 10, 12, 14, 16, 18, 19, 22 and 28.
Sacrifice and pregnancy status:	all females were euthanized by intravenous injection of sodium pentobarbital on GD 28 and pregnancy status was determined. Gravid uterine weights were recorded and total corpora lutea were counted. The rabbits were also examined for gross lesions.
Examination of fetuses:	implants were counted and classified as alive or dead fetuses, or resorption. All fetuses were weighed, examined externally, and after euthanasia by intravenous injection of sodium pentobarbital, given a visceral examination by dissection.
Necropsy:	a gross examination of thoracic and abdominal viscera was performed on all animals.

13 Findings

Dosages	0, 50 (low), 150 (mid), 600 (high) mg/kg/day
Clinical signs	no changes related to treatment, incidental physical signs appeared also in controls (alopecia, pulling fur, blood in pan, diarrhea, and soft or mucoid feces)
Feed intake	a significant ($P \leq 0.05$) treatment-related decrease in average maternal food consumption in the high-dose group. No treatment-related effects in other groups.
Mortality	no deaths during the study. 2 control females aborted on GD 21 and 1 female in the low-dose group. This was not considered treatment-related.
Body weight development	a significant ($P \leq 0.05$) treatment-related decrease in average maternal weight gain between GD 6 and 19 in the high-dose group. After the treatment period (GD 19 to 28) there was a significant ($P \leq 0.05$) increase in average weight gain in this group. No treatment-related effects in other groups.
Embryo survival	a slight but significant ($P \leq 0.05$) treatment-related increase in the percent resorptions per implant in the high-dose group. This increase was due to 8 of 16 litters which had from 1 to 4 resorptions compared to 5 of 12 control litters with 1 or 2 resorptions each. There were no whole-litter resorptions and no dead fetuses in the study. There were no treatment-related effects on the remaining embryo survival parameters (i.e. implants/pregnant female, % preimplantation loss, and live fetuses/pregnant female). No significant ($P > 0.05$) or treatment-related effects in other groups.
Live fetal weight	a significant ($P \leq 0.05$) treatment-related decrease in average fetal weight in the high-dose group. No significant ($P > 0.05$) or treatment-related effects in other groups.
Fetal examinations	variation in lung lobation and incompletely ossified metacarpal, 2 relatively common minor anomalies, were increased in the high-dose group. This was considered to be treatment-related since they were outside the range of historical controls. The latter anomaly may be related to the overall decreased weight of the fetuses in this group.
Maternal necroscopy	no treatment-related changes

Result: the no-effect level for both maternal and developmental toxicity is 150 mg/kg/day.

14	**Statistics**	Statistical analyses were based on a trend test in which it was determined if there was a significant ($P \leq 0.05$) trend with increasing dosage including all treatment groups. (If there was a significant ($P \leq 0.05$) trend, data from the high-dose group were excluded and the trend test was repeated. This process was repeated through the low-dose level or until the trend test was not statistically significant ($P > 0.05$). The highest dose level with no significant ($P > 0.05$) trend was designated the NOSTASOT (NO Statistical Significance Of Trend) dose. **Trend (Dose Response) Analysis** **Reference**: Tukey, J.W., Ciminera, J.L., and Heyse, J.F., "Testing the Statistical Certainty of a Response to Increasing Doses of a Drug", Biometrics, Vol. 41, March, 1985, pp. 295-301. **Estimation of Linear and Quadratic Coefficients (Body Weights)** **Reference**: Robson, D.S.: A Simplified Method for Constructing Orthogonal Polynomials When Independent Variable is Unequally Spaced. Biometrics 15: 187-191, 1959. **Rankit Transformation** **Reference**: Harter, H.L.: Expected Values of Normal Order Statistics. Biometrika, 48: 151-165, 1961. **Reference**: Tukey, J.W.: The Future of Data Analysis. Annals of Mathematical Statistics, 33: 1-67, 1962.
15	**References to publications**	see point 1.8 of this document, reference included at the end of the K document in report 5.6.2/01.
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	May 2005
Materials and Methods	The study complied with GLP and the laboratory is subject to US EPA inspection [redacted] [redacted]

[redacted]

Results and discussion

[redacted]

Conclusion

[redacted]

	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	

98/8 Doc IIIA section No.	**None**	
91/414 Annex Point addressed	**II 5.8.2 / 01**	Toxicity of metabolites

Not Applicable

98/8 Doc IIIA section No.	6.8.2 / 01	**Two generations reproduction study**
Annex Point addressed	**II 5.6.1 / 01**	Multigeneration studies

1.2	**Title**	Thiabendazole: Two-Generation Dietary Reproduction Study in Rats.
1.3	**Report No.**	90-733-0
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.6.1/01
1.6	**Authors**	[redacted]
1.7	**Date of report**	21 May 1992
1.8	**Published**	yes, in Food and Chemical Toxicology, 32: 239-246, 1994.
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	Initiated: 6 November 1990 Terminated: 11 March 1991 (date of last F0 necroscopsy) 5 August 1991 (date of last F1 necroscopsy) 5 August 1991 (date of last F2 sacrifice)
3	**Objective**	to assess the effects of thiabendazole on the growth and reproductive performance during two consecutive generations in the rat
4.1	**Test substance**	Thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	within acceptable limits
4.4	**Stability in vehicle**	was conducted and found to be within acceptable limits
4.5	**Homogeneity in vehicle**	was conducted and found to be within acceptable limits
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	milled rodent chow (Purina Certified Rodent Chow #5002M)
6	**Physical form**	white powder
7.1	**Test method**	2-Generation Dietary Reproduction Study in Rats
7.2	**Justification**	study complied with OECD guidelines according to the 1981 publication
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable
9	**Deviations**	not applicable

10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable

12 Test system

Animal species: rat (Sprague-Dawley [Crl:CD®(SD)BR])

Source: [redacted]

Number of animals: F0 Males/Females: 132/132, F1 Males/Females: ± 560/560, F2 Males/Females: ± 370/370

Age: approx. 8 weeks at initiation

Weight: 303-403 g (F0 Males)
185-266 g (F0 Females)

Dosage: 10, 30 or 90 mg/kg/day; continuous dosing.

Administration: orally in diet

Duration: F0 animals were given the various diets beginning at approx. 8 weeks of age and continuing until sacrifice (after weaning the F1 generation). F1 animals were given the various diets beginning at 3 weeks of age (weaning) and continuing until sacrifice.

Physical signs: daily during the study

Body weights:

Males: recorded one day prior to the start of dosing and at least once weekly thereafter.

Females:

Premating period: one day prior to the start of dosing and once weekly thereafter;

Gestation period: GD 0, 4, 8, 12, 16, 20 and 24;

Lactation Period: LD 0, 4, 8, 12, 16, 20, and 21

In addition, during cohabitation weights were recorded weekly, and females without any live pups were weighed weekly until sacrifice.

Food consumption:

Males: measured over a 6-day interval every week, except during Drug Weeks 7, 8, and 9 when 3- to 7-day intervals were measured. No consumption values were recorded during cohabitation.

Females:

Premating period:	measured over 6-day interval every week (except 4-day interval in Drug Week 9).
Cohabitation:	not measured
Gestation period:	GD 0 to 4, 4 to 8, 8 to 12, 12 to 16 and 16 to 20.
Lactation period:	LD 0 to 4, 4 to 8, 8 to 12, 12 to 16 and 16 to 20.
Postcohabitation:	Females which did not breed or deliver pups were weighed weekly until termination.

Mating:

in the 9th week of treatment, F0 females were housed with males in the same dose group in a 1:1 ratio for a maximum of 21 nights. A check was made each morning for seminal plugs in the pan and/or vagina and a vaginal lavage was examined for the presence of sperm. The day of finding plug and/or sperm was considered GD0.

Observation of Parturition and length of gestation:

from GD 21 until the completion of delivery, each presumed-pregnant female was observed on 4 occasions on each workday (~7:30am, 10:30am, 1:30pm and 4:30pm) and on 2 occasions on each weekend day (~7:30am and 10:30am). Whether delivery had begun or had been completed was noted as well as any signs of difficulty in parturition.

Sacrifice and necroscopsy:

All F0 males were sacrificed by CO2 asphyxiation after all the pregnant females initiated delivery (Drug Week 14). All males were examined grossly and the testes, epididymides, prostate, and seminal vesicles were examined in control and high dose groups. All gross lesions were examined for histomorphological changes.

Females which delivered were euthanized by CO2 asphyxiation between LD 22 to 27 and the uterus of each female was examined to count metrial glands. Mated females that did not deliver pups were euthanized between presumed GD 30 to 48. Females that did not mate were euthanized 28 days after the end of the mating period. The females were examined grossly and the ovaries, uterus, and vagina were examined in control and high dose groups. All gross lesions were examined for histomorphological changes.

F1 generation

1. Preweaning:

Physical signs:	daily observation for mortality and physical signs
Body weights:	recorded on PND 0, 4, 7, 14 and 21
External examination:	examined for malformations on PND 0, 4 and 21. The external sex of each pup was recorded on PND 0, and confirmed on PND 4, 7, 14 and 21 in remaining pups
Dead pups:	examined for visceral abnormalities including an examination for bedding material in the trachea and esophagus
2. Postweaning:	(PND 21 to termination)
Physical signs:	daily observation for mortality and physical signs

Body weights:

Males: once a week from weaning till termination.

Females: once a week from weaning until breeding or termination.

average age of animals at time of first postweaning weight was 25 days

Food consumption:

Males: measured over a 6-day interval every week. No consumption values were recorded during cohabitation

Females:

Premating period: measured over a 6-day interval every week

Cohabitation: not measured; Gestation period: GD 0 to 4, 4 to 8, 8 to 12, 12 to 16 and 16 to 20

Postcohabitation: females which did not mate or deliver pups were weighed weekly until termination

Lactation period: LD 0 to 4, 4 to 8, 8 to 12, 12 to 16 and 16 to 20

Mating:

During Postnatal Weeks 17, one female and one male (non-siblings) per litter were caged together for mating. The mating period was limited to 21 days. The day on which spermatozoa were detected in the daily vaginal lavage was considered GD 0 and the mated females were removed and individually caged

Observation of Parturition and length of gestation:

on GD 16, the females were transferred to plastic boxes containing dry bedding in preparation for delivery of F2 pups. Observation of parturition and determination of length of gestation were the same as for F0 dams.

Sacrifice and necroscopsy:

all F1 males used for mating were euthanized by CO2 asphyxiation after all of the pregnant females initiated delivery (Drug Week 20). The males were examined grossly and testes, epididymides, prostate, and seminal vesicles were examined in the control and high-dose groups. All gross lesions were examined for histomorphological changes.

F1 females which delivered were euthanized by CO2 asphyxiation between LD 21 to 27 and the uterus of each female was examined to count metrial glands. Mated females that did not deliver pups were euthanized on the presumed GD 25-44, substitute females were also euthanized at this time.

All F1 females were examined grossly and the ovaries, vagina, and uterus from the control and high-dose group were examined. All gross lesions were examined for histomorphological changes

F2 generation

Physical signs: daily observation for mortality and physical signs

Body weights: recorded on PND 0, 4, 7, 14 and 21

External examination: examined for malformations on PND 0, and culled pups on PND 4 and 21. The external sex of each pup was recorded on PND 0, and confirmed on PND 4, 7, 14 and 21 in remaining pups

Dead pups: examined for visceral abnormalities including an examination for bedding material in the trachea and esophagus

Sacrifice of F2 pups: on PND 21 all remaining pups were euthanized and discarded without further examination

13 Findings

F0 Generation

Dosages	0, 10 (low), 30 (mid), 90 (high) mg/kg/day
Physical signs	Females: no treatment-related physical signs Males: no treatment-related physical signs
Food consumption	significant ($P \leq 0.05$) and/or treatment-related effects on average food consumption are outlined in **Table 1**. No significant ($P > 0.05$) or treatment-related effects in the low-dose group
Mortality	no treatment-related deaths. 1 female in each of the low- and high-dose groups was sacrificed or died, respectively, during parturition. Multiple dead, intra-uterine pups were found in both cases. These female mortalities were considered incidental due to their isolated nature, and to the occasional occurrence of maternal deaths during parturition in control rats
Reproductive performance	no treatment-related or statistically significant ($P > 0.05$) adverse effects on reproductive performance
Body weights	treatment-related effects on average body weights are outlined in **Table 2**. There were no significant ($P > 0.05$) or treatment-related effects in the low-dose group.
Necrospy	no treatment-related effects on the gross or histomorphological appearance of the reproductive system of F0 animals

Table 1: Food Consumption: F0 generation

	Dose	% change v. controls	
Period	Level	Females	Males
Premating	90	down 12%*	down 13%*
	30		down 4%*
After cohabitation	90	NA**	down 11%*
	30		down 3%*
Gestation	90	down 4-16%	NA
Lactation	90	None	NA

* $P \leq 0.05$ by trend analysis

** group sizes too small for valid comparisons

Table 2: Body Weights: F0 generation			
	Dose	% change v. controls	
Period	Level	Females	Males
Premating	90	down 29%*	down 29%*
	30		down 10%*
During and after cohabitation	90	NA**	down 46%*
	30		down 16%*
Gestation	90	down 8%*	NA
Lactation	90	up 3.5x*	NA

* $P \leq 0.05$ by trend analysis
** group sizes too small for valid comparisons

F1 Generation from birth through lactation

Physical signs	no treatment-related physical signs observed in F1 pups during lactation
Mortality	no treatment-related effects on pup survival from birth to the end of lactation (PND 21) in the drug-treated groups
Examinations of F1 pups	no treatment-related external malformations or variations in F1 pups on PND 0, 4, or 21. There were no treatment-related visceral malformations or variations in dead or externally malformed F1 pups
Pup body weights	average pup weight at birth was comparable across all groups. Thereafter on PND 4, 7, 14 and 21 there were slight but significant ($P \leq 0.05$) treatment-related decreases in average weights (both sexes) in the high-dose group (5 to 8% below control). No significant ($P > 0.05$) or treatment-related effects in the low- or mid-dose groups

F1 Generation from weaning until sacrifice

Physical signs	no treatment-related physical signs

Food consumption	treatment-related effects on average food consumption are outlined in **Table 3**. No significant ($P > 0.05$) or treatment-related effects in the low-dose group
Mortality	no treatment-related deaths during the post-weaning phase of the study. One female in the high-dose group died on LD 2 due to spontaneous interstitial moderate nephritis. One female in the low-dose group was sacrificed on LD 5 because of no surviving pups
Reproductive performance	no treatment-related or statistically significant ($P > 0.05$) adverse effects on reproductive performance
Body weights	significant ($P \leq 0.05$) and/or treatment-related effects on average body weight gain are outlined in Table 4. There were no significant ($P > 0.05$) or treatment-related effects in the low-dose group
Histomorphologic Examinations	no treatment-related changes in the gross or histomorphological appearance of the reproductive system of F1 animals

Table 3: Food Consumption: F1 generation

	Dose	% change v. controls	
Period	Level	Females	Males
Premating	90	down 10%	down 9%
	30		down 4%
During and after cohabitation	90	NA*	down 11%
	30		down 5%
Gestation	90	down 4-10%	NA
Lactation	90	None	NA

* group sizes too small for valid comparisons

Table 4: Body Weights: F1 generation

	Dose	% change v. controls	
Period	Level	Females	Males
Premating	90	down 14%*	down 13%*
	30		down 7%*
During and after cohabitation	90	NA**	down 41%*
	30		down 18%*
Gestation	90	no effects	NA

Lactation	90	18g vs. -5 g in control	NA

* $P \leq 0.05$ by trend analysis
** group sizes too small for valid comparisons

F2 Generation from birth through lactation

Physical signs	no treatment-related physical signs observed in F2 pups during lactation
Mortality	no treatment-related or statistically significant ($P > 0.05$) effects
Examinations of F2 pups	no treatment-related effects
Pup body weights	a significant ($P \leq 0.05$) treatment-related decrease in average weight of male and female pups in the high-dose group on PND 14 and 21 (7 to 10% below control). No treatment-related effects in the low- or mid-dose groups

Conclusion:

F0 generation: dose-related decreases in average body weight gain and food consumption in mid- and high-dose groups. Slight treatment-related increase in average lactational weight gain in high dose group. No treatment-related effects on reproductive performance or gross lesions at any dose level or histomorphology of reproductive organs at the high dose. No treatment-related effects of any kind in the low dose group.

F1 generation: dose-related decreases in average weight gain and food consumption in mid- and high-dose groups, and slight increase in average lactational weight gain in the high-dose group. No treatment-related effects on survival, reproductive performance, or gross lesions at any dose level or histomorphology of reproductive organs at the high-dose level. No treatment-related effects of any kind in the low dose group.

F2 generation: no treatment-related effects in the survival, physical signs, or external morphology to PND 21. A slight treatment-related decrease in average PND 14 and 21 pup body weights occurred in the high-dose group only.

The **NOAEL** (No Observed Adverse Effect Level) for all growth, survival, and reproductive performance parameters assessed in this study was 10 mg/kg/day. Treatment of the F0 and F1 generations of rats with Thiabendazole up to 90 mg/kg/day had no effect on the gross or microscopic appearance of the reproductive system of the F0 or F1 animals.

14 Statistics Statistical analyses were done by an analysis of variance or covariance (continuous variables) or by an extended Mantel-Heanszel test (discrete variables). A rankit method was used to normalize nonparametric data. Results were considered to be statistically significant if $P \leq 0.05$. A trend

analysis was used to determine if there was a significant ($P \leq 0.05$) trend with increasing dosage across all treatment groups. If there was a significant ($P \leq 0.05$) trend, data from the high dose group were excluded and the trend test was repeated.

Analysis of Variance and Analysis of Covariance

Reference: Snedecor, G.W. and Cochran, W.G., Statistical Methods, 7th Ed., Iowa State University Press, Ames, Iowa, Chapter 12, 1980.

Reference: Snedecor, G.W. and Cochran, W.G., Statistical Methods, 7th Edition, Iowa State University Press, Ames, Iowa, Chapter 18, 1980.

Trend (Dose Response) Analysis

Reference: Tukey, J.W., Ciminera, J.L., and Heyse, J.F., Testing the Statistical Certainty of a Response to Increasing Doses of a Drug. Biometrics, 41: 295-301, 1985.

Test for Parallelism (Analysis of Covariance)

Reference: Villars, D.S., Statistical Design and Analysis of Experiments for Development Research, W.C. Brown, Co., Dubuque, Iowa, 173-177, 1951.

Rankit Transformation

Reference: Harter, H.L., Expected Values of Normal Order Statistics. Biometrika, 48: 151-165, 1961.

Reference: Tukey, J.W., The Future of Data Analysis, Annals of Mathematical Statistics. 33: 1-67, 1962.

Estimation of Linear and Quadratic Coefficients (Body Weights)

Reference: Robson, D.S., A Simplified Method for Constructing Orthogonal Polynomials When Independent Variable is Unequally Spaced. Biometrics, 15: 187-191, 1959.

Mantel-Haenszel Analysis

Reference: Mantel, N. (1957). Chi-square Tests with One Degree of Freedom. J. Am. Statistical Assoc., 58: 690-700, 1963.

15	**References to publications**	see point 1.8 of this document, reference included at the end of the K document
16	**Unpublished data**	not applicable

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	May 2005
Materials and Methods	The study complied with GLP and the laboratory is subject to US EPA inspection
	[redacted]

F animals were observed for physical signs daily and body weights were

Results and discussion

[redacted]

Conclusion [redacted]

Reliability [redacted]
Acceptability
Remarks

98/8 Doc IIIA section No.	**6.9**	**Neurotoxicity studies**
91/414 Annex Point addressed	**II 5.8.**	Other toxicological studies - Supplementary studies on the active substance

[redacted]

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	February 2005
Materials and Methods	
Results and discussion	
Conclusion	[redacted]
Reliability	
Acceptability	
Remarks	

98/8 Doc IIIA section No.	**6.10**	**Mechanistic study - any studies necessary to clarify effects reported in toxicity studies**
91/414 Annex Point addressed	**II 5.8.2 / 01**	Other toxicological studies - Supplementary studies on the active substance

1.2	**Title**	Thiabendazole: Fourteen-Week Dietary Thyroxine Clearance Study in Rats with a 14-Week Recovery Period
1.3	**Report No.**	94-024-0
1.4	**Lab. report No.**	not applicable
1.5	**Cross reference**	5.5/03
1.6	**Authors**	[redacted]
1.7	**Date of report**	16 February 1995
1.8	**Published**	no
2.1	**Testing facility**	[redacted]
2.2	**Dates of experimental work**	23 March 1994 to 21 September 1994
3	**Objective**	to determine if the fungicide thiabendazole alters thyroxine clearance and affects Thyroid Stimulating Hormone (TSH) or thyroid hormone levels in rats treated for approximately 14 weeks and to determine if the thyroid hyperplasia produced by thiabendazole treatment is reversible.
4.1	**Test substance**	technical grade thiabendazole, [redacted]
4.2	**Specification**	[redacted]
4.3	**Storage stability**	of the bulk drug over the duration of the study was confirmed by assays of a sample obtained in Week 14
4.4	**Stability in vehicle**	thiabendazole is stable in rodent feed at room temperature during the study
4.5	**Homogeneity in vehicle**	diet mixtures were homogenous
4.6	**Validity**	not applicable
5	**Vehicle/solvent**	Certified Purina Rodent Chow
6	**Physical form**	*white powder*
7.1	**Test method**	14-weeks
7.2	**Justification**	in compliance with the OECD guidelines according to the 1981 publication
7.3	**Copy of method**	not applicable
8	**Choice of method**	not applicable

9	**Deviations**	GLP deviation on p17 of final report. This was considered minor and did not affect the conclusion of the study
10.1	**Certified laboratory**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.2	**Certifying authority**	the study complied with GLP and the laboratory is subject to US EPA inspection
10.3	**GLP**	yes
10.4	**Justification**	not applicable
11.1	**GEP**	not applicable
11.2	**Type of facility (official or officially recognized)**	not applicable
11.3	**Justification**	not applicable
12	**Test system**	

Animal species: albino rats [strain: Crl:CD®(SD)BR]

Source: [redacted]

No. of animals: 140 male animals (3 drug-treated groups and one control group, each containing 35 males)

Age: 59 days at initiation of compound administration

Weights: 249 to 366 g

Identification: individual by a Biomedic implant

Dosage (a.s.): 10, 90, 270 mg/kg/day

Administration: oral by feeding

Duration: 91 days for 30 males/group and 94 days for 5 males/group

General observations: daily observations for mortality and clinical signs. Less detailed examinations on weekends and holidays. Animals were weighed pretest and once a week thereafter.

Food consumption: recorded weekly

TSH, T3 and T4 serum levels:

were determined with 2 ml of whole blood/nonfasted rat, pretest (in 30 rats/group), in Weeks 2, 4, 8 and 13 (in 15 rats/group), and in Recovery weeks 6 and 13 (in generally 15 rats/group)

Determination of thyroxine clearance:

in Week 13/14, from 5 non-fasted rat/group, about 1.5 ml of heparinized blood/rat/time point were taken at approximately 8, 22, 34, 48 and 72 hours after intravenous injection of ^{125}I-thyroxine.

Gross examination: in Week 14, fifteen male rats per group were killed by exsanguination after being rendered unconscious with CO_2. Gross examination was limited to the thyroids and liver of all animals and weights of these organs as well as the terminal body weight and brain weight were taken.