Section A6.3.2 **Annex Point IIAV16.3**	**Repeated dose toxicity (dermal)** **Repeated dose toxicity – Dermal route (28 days)**	
	JUSTIFICATION FOR NON-SUBMISSION OF DATA	Official use only
Other existing data [ ] **Limited exposure [ ]**	**Technically not feasible [ ]** **Scientifically unjustified [X]** **Other justification [ ]**	
Detailed justification:	28-day studies are not a mandatory requirement. Therefore no such studies are presented in this submission.	X
Undertaking of intended data submission [ ]	Not applicable	
	Evaluation by Competent Authorities	
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	01/07/2009	
Evaluation of applicant's justification	The 28-days tests are used as a range finding study and are not required when an adequate subchronic toxicity study is available in the rodent, which is the case in this situation. (Dir 98/8, guidance on data requirements for active substances and biocidal products)	
Conclusion	Acceptable A 28-day percutaneous study is required, where the potential dermal exposure is significant which is not the case. (Dir 98/8, guidance on data requirements for active substances and biocidal products)	
Remarks		
	COMMENTS FROM OTHER MEMBER STATE *(specify)*	
Date	*Give date of comments submitted*	
Evaluation of applicant's justification	*Discuss if deviating from view of rapporteur member state*	
Conclusion	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Section A6.3.3 **Annex Point IIA 6.3.3**	**Repeated dose toxicity** **Repeated dose toxicity – Inhalation route (28 days)**	
	JUSTIFICATION FOR NON-SUBMISSION OF DATA	Official use only
Other existing data [ ] **Limited exposure [ ]**	**Technically not feasible [ ]** **Scientifically unjustified [X]** **Other justification [ ]**	
Detailed justification:	28-day studies are not a mandatory requirement. Therefore no such studies are presented in this submission.	X
Undertaking of intended data submission [ ]	Not applicable	
	Evaluation by Competent Authorities	
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	01/07/2009	
Evaluation of applicant's justification	The 28-days tests are used as a range finding study and are not required when an adequate subchronic toxicity study is available in the rodent, which is the case in this situation (Dir 98/8, guidance on data requirements for active substances and biocidal products).	
Conclusion	Acceptable A 28-day inhalation study is required for volatile substances or where the potential inhalation exposure is significant (Dir 98/8, guidance on data requirements for active substances and biocidal products). This is not the case with this active substance.	
Remarks		
	COMMENTS FROM OTHER MEMBER STATE *(specify)*	
Date	*Give date of comments submitted*	
Evaluation of applicant's justification	*Discuss if deviating from view of rapporteur member state*	
Conclusion	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Section A6.4.1/1 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

Official use only

8 Reference

8.1	Reference	[redacted] (2003) Repeated Dose (90-Day) Oral Toxicity Study with Permethrin in Wistar Rats. [redacted] report No.: 3351/02 (unpublished).
		Dates of experimental work: October 8, 2002 – February 4, 2003.
8.2	**Data protection**	Yes
8.2.1	Data owner	Tagros Chemicals India Ltd.
8.2.2	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing a.s for the purpose of its entry into Annex I/IA.

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9 Guidelines and Quality Assurance

9.1	**Guideline study**	Yes, test method was based on OECD guideline 408.
9.2	**GLP**	Yes
9.3	**Deviations**	Yes, with the following deviation: X
		No urinalysis was carried out as is recommended in the guideline.
		This deviation is considered to be minor and does not compromise the scientific validity of the study.

10 MATERIALS AND MethodS

10.1	**Test material**	As given in section 2 (Permethrin 25:75)
10.1.1	Lot/Batch number	143
10.1.2	Specification	As given in section 2 (Permethrin 25:75)

Section A6.4.1/1 Repeated dose toxicity

Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

10.1.2.1	Description	Yellow to pale brown coloured viscous liquid, with a mild characteristic odour, which tends to crystallise partly at room temperature.
10.1.2.2	Purity	92.4%
10.1.2.3	Stability	An in-house stability study (Study No: 3349/02) was carried out on both the 50 and 10000 ppm doses. Results of this showed that the test item was stable in the experimental food up to 30 days in both dietary concentrations when stored at ambient conditions. The percentage reduction from day 0 to day 30 was 6.0% and 5.1% of the nominal concentrations of 50 and 10000 ppm respectively. Results of a homogeneity test indicated that Permethrin distribution in the experimental food was homogeneous at all the tested doses.
10.2	**Test Animals**	
10.2.1	Species	Rat (HsdCpb: WU conventionally bred)
10.2.2	Strain	Wistar
10.2.3	Source	[redacted]
10.2.4	Sex	Male and female
10.2.5	Age/weight at study initiation	7 weeks at start of treatment Males: 187 g – 189 g Females: 141 g – 142 g
10.2.6	Number of animals per group	20 (10 males and 10 females)
10.2.7	Control animals	Yes
10.3	**Administration/ Exposure**	Oral
10.3.1	Duration of treatment	90 days
10.3.2	Frequency of exposure	Daily (in food)
10.3.3	Recovery period	2 additional recovery groups (control and high dose) were maintained on normal food, for a further 28 days after the 90 day period, without being exposed to Permethrin for recovery observations.

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Section A6.4.1/1 Repeated dose toxicity

Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

10.3.3.1	Concentration	Food: 0, 100, 600 and 2000 ppm (equivalent to 0, 8.6, 51.6 and 172 mg/kg bw)
10.3.3.2	Food and water	Available *ad libitum*
10.3.3.3	Vehicle	Acetone
10.3.3.4	Controls	Plain diet
10.4	**Examinations**	
10.4.1.1	Clinical examinations	Yes, once prior to study initiation and weekly thereafter during treatment and recovery periods.
10.4.1.2	Ophthalmological examinations	Yes, on all animals prior to study commencement, at the end of the treatment period and at the end of the recovery period for recovery groups
10.4.1.3	Mortality and general clinical observations	Yes, once a day for general clinical signs and twice daily for morbidity and pre-terminal deaths.
10.4.2	Body weight	Yes, before the start of treatment and weekly thereafter.
10.4.3	Food consumption	Yes, weekly food consumption was recorded.
10.4.4	Water consumption	Not recorded
10.4.5	Haematology	Yes Number of animals: All animals Time points: End of study Parameters: Prothrombin time, Haematocrit (Hct), Haemoglobin (HB), Platelet Count (Plat), Red blood Corpuscles (RBC), White Blood Corpuscles (WBC) The following calculated RBC associated indices were recorded from the haematology analyser: Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC)
10.4.6	Clinical Chemisty	Yes Number of animals: All animals Time points: End of study Parameters: Fasting glucose (Glu) mmol/l, Creatinine (Creat) µmol/l, Total Plasma Protein (Tot. Pro) g/l, Albumin (Alb) g/l, Gamma Glutamyl Transpeptidase (GGT) U/l, Blood Urea Nitrogen (BUN) mmol/l, Urea (mmol/l), Alanine Amino transferase (ALT) U/l, Aspartate Amino transferase (AST) u/l, Total Cholesterol (Chol)

Section A6.4.1/1 **Repeated dose toxicity**

Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

		(mmol/l), Sodium and Potassium
10.4.7	Neurological examinations	Functional observations were carried out during weeks 12 and 13 of the study as follows: Home cage observations: Presence of convulsions or tremors and palpebral closure Handling Observations: ease of removal from cage, ease of handling, lacrimation, chromodacyorrhea, salivation, piloerection, palpebral closure, respiratory observations, degree of eye prominence and muscle tone Open field observations: Mobility, backing (recording no. of backward steps taken in a given time), grooming observations, gait, presence of convulsions or tremors and level of arousal. Sensory observations: Startle (auditory) response, touch response, pupil response, response to nociceptive stimuli and righting reflex Neuromuscular observations: Grip strength, motor activity and hind limb foot splay Physiological observations: Body temperature
10.4.8	Urinalysis	Not conducted
10.5	**Sacrifice and pathology**	
10.5.1	Organ weights	Yes Organs: Liver, kidneys, adrenals, testes, epididymides, uterus, ovaries, thymus, spleen, brain, heart
10.5.2	Gross and histopathology	Yes All dose groups were subjected to gross pathology. Histopatholgical evaluation was carried out on all tissues collected from control and high dose groups and the livers from the low, mid dose and recovery group animals and gross lesions from low, mid and recovery groups. Organs: Brain (including medulla/pons cerebrum and cerebellum), spinal cord at 3 levels – cervical, thoracic and lumbar, pituitary, thyroid, parathyroids, thymus, oesophagus, salivary glands, stomach, duodenum, ileum (with peyer's patches), jejunum, caecum, colon, rectum, liver, pancreas, kidneys, adrenals, spleen, heart, trachea, lungs, aorta, testes, epididymides, uterus, female mammary gland, prostate, urinary bladder, mesenteric lymph nodes, peripheral nerve, sternum with bone marrow, skin, ovaries, seminal vesicles, coagulating glands, sciatic nerves, auxillary lymph nodes and all gross lesions.
10.5.3	Statistics	Functional observations, (body temperature, motor activity, foot splay and grip strength), body weights, net weight gain, food consumption, laboratory investigations (haematology and clinical chemistry), organ weights and organ weight ratio data were analysed using Bartlett's test for homogeneity of intra group variances. Heterogeneous variances underwent appropriate transformation. The data with homogeneous

intra group variances were subjected to one-way analysis of variance (ANOVA – Snedecor and Cochran, 1987). When 'F' was found to be significant, Dunnett's pairwise comparison (Scheffe, 1953) of means of treated and control groups was done individually.

Following a significant difference of a test group with the control group in a minimum of two treatment groups with linear increase or decrease the dose response correlation co-efficient was estimated and subjected to T-test analysis. All analyses and comparisons were evaluated at 5% ($P \leq 0.05$ level).

Mean test item intake (mg/kg body weight) was calculated sexwise and combined sexes, separately for each group.

The following incidences of histopathological changes were statistically compared by Z-test:

Liver – hepatocellular hypertrophy

Lung – lymphocytic infiltration

Lungs – pneumonic foci

Spleen – increased hemosiderosis

11 Results and Discussion

11.1	**Observations**	
11.1.1	Clinical signs	No treatment related clinical signs in any animals
11.1.2	Mortality	No pre-terminal deaths in any animal at any dose level tested
11.2	**Ophthalmological examinations**	No eye abnormalities in any animal at any dose level
11.3	**Body weights and Net body weight gain**	No treatment related changes
11.4	**Food consumption and compound intake**	No treatment related changes
11.5	**Neurological examination**	No treatment related effects
11.6	**Blood analysis**	
11.6.1	Haematology	Males: Statistically significant increase in MCH levels at the mid and high doses Statistically significant increase in MCHC levels at the high dose level at the end of the treatment period. Females: Statistically significant increase in neutrophil counts at the low dose Statistically significant decrease in lymphocyte counts at the low dose.

Section A6.4.1/1 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

		Statistically significant decrease in monocyte count at the mid dose level.	
11.6.2	Clinical chemistry	Males: No statistically significant inter group differences at any dose level, at the end of the treatment or recovery period. Females: No statistically significant inter group differences were noted at any dose level, at the end of the treatment period.	
11.6.3	Urinalysis	Not conducted	
11.7	**Sacrifice and pathology**		
11.7.1	Organ weights	Males: Statistically significant increase in absolute and relative liver weights at the high dose. Statistically significant increase in absolute and relative testes weights, and relative epididymides weights in the high dose recovery animals. Females: Statistically significant increase in absolute liver weights and in relative liver weights in the mid and high dose groups. Statistically significant increase in absolute and relative kidney weights and relative liver and spleen weights in the high dose recovery animals.	X
11.7.2	Gross and histopathology	No treatment-related gross pathological changes. Increased incidence of hepatocellular hypertrophy in the livers of high dose males, and mid and high dose females.	

12 Applicant's Summary and conclusion

12.1	**Materials and methods**	Permethrin was administered for 90 days to groups of CRL: (WI) rats (10 animals/sex/dose) at the following doses 0, 100, 600 and 2000 ppm (equivalent to 0, 8.6, 51.6 and 172 mg/kg bw). Two extra groups of 10 animals/sex/group were treated at the doses of 0 and 2000 ppm (equivalent to 0 and 172 mg/kg bw) for 90 days and were retained for a 28-day post-treatment recovery period. This study in rats was conducted according to OECD 408 and is described under point 3 with the following deviation:

Section A6.4.1/1 Repeated dose toxicity
Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

No urinalysis was carried out.

This deviation is considered to be minor and does not compromise the scientific validity of the study.

12.2 Results and discussion

Results of analyses showed that the analysed concentrations from each batch were within ± 10% of the nominal concentrations of 100, 600, and 2000 ppm.

Veterinary and clinical examinations did not reveal any Permethrin related clinical signs, in any animal, during the course of the study. Some minor incidental signs of hair thinning with subsequent regrowth were observed, however these were not deemed to be of any toxicological significance.

Weekly mean body weights and cumulative net body weight gains showed no statistical inter group difference in any of the doses tested either during treatment or recovery period in any animal. No significant differences in terminal fasting body weights were noted for any animal in any group. Please see Table A6.4.1/1-1 and Table A6.4.1/1-2.

Food intake was unaffected by the presence of Permethrin in males at any of the doses tested either during treatment or recovery periods. However, isolated incidences of significantly lower food intake were seen during week 1 in the high dose main group and during week 5 in the high dose recovery group during the treatment period.

Food intake was unaffected by the presence of Permethrin in females at any of the doses tested either during treatment or recovery periods. However, isolated incidences of significantly lower food intake were seen during weeks 8 to 10 and 13 in the high dose recovery group during the treatment period. Please see Table A6.4.1/1-3.

Functional observations conducted during week 12 and 13 of the treatment period did not reveal any treatment related functional abnormalities in any animal.

In males, haematological investigations revealed significantly higher MCH levels at the mid and high doses in males, and also higher MCHC levels at the high dose level in males at the end of the treatment period. A dose-effect relationship was noted in terms of the higher MCH levels. However, as these changes did not correspond to similar changes in RBC, Hb or Hct parameters, they were considered incidental. At the end of the recovery period, no statistically significant inter group variations were noted for any of the measured parameters.

In females, a significantly higher neutrophil and significantly lower lymphocyte count was noted at the low dose. A lower monocyte count was also noted at the mid dose level. These were the only statistically significant inter group differences noted in any of the tested parameters. Changes noted in the Differential Leukocyte Count, were considered

incidental as no dose effect relationship was observed.

At the end of the recovery period, changes such as significantly lower RBC count, Hb, Hct levels and eosinophil counts, and also significantly higher MCH levels were noted. However, these changes were deemed not to be of toxicological significance, as they were not noted in the high dose main group at the end of the treatment period.

No statistically significant inter group differences were noted at any dose level in males, at the end of the treatment or recovery period.

In females, no statistically significant inter group differences were noted at any dose level, at the end of the treatment period. At the end of the recovery period one female from the high dose recovery group showed significantly higher levels of creatinine. However this finding was not deemed to be of toxicological significance as a similar increase was not observed at the end of the treatment period in the main high dose group.

Absolute and relative liver weights were significantly increased in males at the high dose. A significant increase in absolute and relative testes weights, and relative epididymides weights were observed in the high dose recovery animals. X

A significant increase in absolute liver weights and a significant dose correlated increase in relative liver weights was noted in females in the mid and high dose groups. Absolute and relative kidney weights and relative liver and spleen weights were significantly higher in the high dose recovery animals. The increased liver weights in both males and females were considered to be a treatment related physiological response. Microscopically it was associated with hepatocellular hypertrophy. Some minor changes observed in testes, epididymides, kidneys and spleen in the recovery animals were considered incidental, as they were not observed in the main groups.

There were no treatment-related gross pathological changes. The presence of a uterine polyp in one high dose recovery female was recorded. In one high dose recovery male, an uneven kidney surface and thickened urinary bladder were recorded. These were associated with chronic tubular interstitial nephritis in the kidney and severe epithelial hyperplasia in the urinary bladder. These changes were deemed to be of no toxicological significance.

Frequently observed microscopic findings were as follows: Liver – Hepatocellular hypertrophy (minimal) and necrobiotic foci (minimal); Lungs – Lymphocytic infiltration (minimal and mild); Spleen – Increased hemosiderosis (minimal to moderate) and Pituitary – Dilated Rathke's cleft. An increased incidence of hepatocellular hypertrophy in the livers of high dose males, and mid and high dose females was considered to be the result of treatment with Permethrin. As the incidences of this were greatly reduced in the high dose recovery group, the effect was considered to be reversible. Other microscopic changes, such as lymphocytic infiltration and pneumonic foci in the lungs of high dose females and increased hemosiderosis in spleens of high dose females were not considered to be treatment related.

Section A6.4.1/1	Repeated dose toxicity
Annex Point IIA 6.4	90 day oral toxicity study in the rat

12.3	**Conclusion**	An NOEL of 100 ppm (equivalent to 7.9, 9.3, and 8.6 mg/kg bw day for males, females and combined sex respectively) was established in the study. This was based on findings of increased absolute and relative liver weights which were associated with hepatocellular hypertrophy which were observed in the 600 and 1000 ppm groups.
12.3.1	LO(A)EL	600 ppm (equivalent to 51.6 mg/kg bw day)
12.3.2	NO(A)EL	100 ppm (equivalent to 7.9, 9.3, and 8.6 mg/kg bw day for males, females and combined sex respectively)
12.3.3	Reliability	1
12.3.4	Deficiencies	One deviation was noted and is outlined under points 2.3 and 5.1. However, it does not compromise the scientific validity of the study.

Evaluation by Competent Authorities	
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	Evaluation by Rapporteur Member State
Date	03/07/2009
Materials and Methods	2.3 According to the OECD guideline 408, urinalysis is optional, therefore this is not considered a major deviation, and applicant's version is acceptable.
Results and discussion	Adopt applicant's version 4.7.1 and 5.2. Although there was no statistical significant differences in liver weight in medium dose males, there does appear to be biological significance, strengthening the dose related and treatment related effect.
Conclusion	LO(A)EL: 600 ppm (equivalent to 51.6 mg/kg bw day) NO(A)EL: 100 ppm (equivalent to 7.9, 9.3, and 8.6 mg/kg bw day for males, females and combined sex respectively)
Reliability	1
Acceptability	Acceptable
Remarks	
	Comments from ... *(SPECIFY)*
Date	*Give date of comments submitted*

Section A6.4.1/1 Repeated dose toxicity

Annex Point IIA 6.4 **90 day oral toxicity study in the rat**

Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Table A6.4.1/1-1: Summary of cumulative weekly net body weight gains (g) – Males

G. No. Dose (ppm)	No. of rats	Weeks																
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
G1	10	55	92	126	151	169	181	196	206	216	219	220	222	226	-	-	-	-
0		4.9	7.9	8.3	12.7	14.5	17.9	18.6	21.7	22.4	24.0	24.7	27.5	25.8				
G2	10	53	90	123	150	172	188	206	221	229	230	232	235	240	-	-	-	-
100		7.9	9.8	8.7	10.6	11.9	13.3	18.8	22.7	23.3	21.9	23.9	23.9	25.9				
G3	10	54	93	125	152	170	188	203	219	226	228	233	236	241	-	-	-	-
600		4.1	9.0	12	16.4	20.5	26.1	30.3	31.0	34.6	35.2	36.6	36.4	35.9				
G4	10	51	88	116	143	165	180	195	210	219	222	226	230	234	-	-	-	-
2000		6.1	9.0	12.5	17.4	20.0	22.8	25.3	28.9	28.3	31.0	30	31.5	30.5				
G1R	10	54	90	124	151	170	185	204	217	225	230	233	235	240	251	260	266	275
0		6.0	8.6	9.1	12.7	16.9	18.7	21.8	26.4	27.3	29.2	29.2	28.3	27	(10)	(19)	(26)	(35)
G4R	10	52	91	121	146	162	175	189	204	213	217	218	223	228	238	250	254	263
2000		4.5	7.2	10.7	16.0	21.1	18.9	21.1	20.7	19.3	19.3	20.0	19.6	20.2	(10)	(22)	(26)	(35)

The values in parenthesis indicate the body weight change during the recovery period.

Values: Mean ± SD

Table A 6.4.1/1-2: Summary of cumulative weekly net body weight gains (g) – Females

G. No. Dose (ppm)	No. of rats	Weeks																
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
G1	10	20	34	46	56	64	69	75	81	83	85	88	89	91	-	-	-	-
0		6.2	3.1	4.4	6.2	5.6	5.6	6.5	8.4	7.5	8.6	8.1	9.0	7.6				
G2	10	17	35	46	55	65	73	78	81	85	87	89	90	93	-	-	-	-
100		7.3	5.8	7.0	7.2	8.5	9.0	8.0	10.1	11.6	12.9	12.4	13.0	12.3				
G3	10	19	34	48	58	68	75	80	84	89	91	94	96	97	-	-	-	-
600		5.9	5.3	7.3	7.0	8.8	8.3	11.6	11.1	13.3	12.6	13.6	12.1	12.9				
G4	10	18	32	46	55	63	68	73	80	83	84	87	88	90	-	-	-	-
2000		7.8	7.9	7.5	9.0	9.0	10.1	8.8	11.0	9.7	11.9	12.0	13.5	13.3				
G1R	10	24	38	48	58	67	72	78	84	87	89	91	90	93	97 (3)	100 (7)	102 (9)	108
0		8.3	8.8	10.4	10.8	13.8	15.9	15.3	16.1	16.3	16.4	15.3	14.6	15.6	15	16.5	16.9	(14)
G4R	10	20	35	47	56	65	72	75	80	84	86	87	89	91	98 (7)	100 (9)	101	106
2000		6.3	6.5	7.6	9.5	8.4	9.2	9.8	9.1	9.6	10.1	10.0	10.5	12.3	15.0	12.5	(10)	(15)

The values in parenthesis indicate the body weight change during the recovery period.

Values: Mean ± SD

Table A 6.4.1/1-3: Summary of food and test item intake

G. No. Dose (ppm)	G1 0	G2 100	G3 600	G4 2000	G1R 0	G4R 2000
FOOD INTAKE	**MALES**					
g/animal/90 days	2485.4	2554.6	2555.2	2456.9	2521.9	2472.6
g/animal/day	27.6	28.4	28.4	27.3	28.0	27.5
g/kg Bwt/90 days	7022.1	7112.8	7160.0	6972.2	7019.1	7064.1
g/kg Bwt/90 days	78.0	79.0	79.0	77.5	78.0	78.5
TEST ITEM INTAKE						
mg/kg Bwt/90 days	0.0	711.3	4263.6	13944.5	0.0	14128.3
Mg/kg Bwt/day	0.0	7.9	47.4	154.9	0.0	157.0
FOOD INTAKE	**FEMALES**					
g/animal/90 days	1815.6	1742.0	1790.7	1683.6	1833.2	1732.2
g/animal/day	20.2	19.4	19.9	18.7	20.4	19.2
g/kg Bwt/90 days	8709.5	8375.7	8477.7	8128.0	8719.2	8337.9
g/kg Bwt/90 days	96.8	93.1	94.2	90.3	96.9	92.6
TEST ITEM INTAKE						
mg/kg Bwt/90 days	0.0	837.6	5086.6	16255.9	0.0	16675.8
Mg/kg Bwt/day	0.0	9.3	56.5	180.6	0.0	185.3
FOOD INTAKE	**COMBINED SEX**					
g/animal/90 days	2150.5	2148.3	2172.9	2070.2	2177.6	2102.4
g/animal/day	23.9	23.9	24.1	23.0	24.2	23.4
g/kg Bwt/90 days	7865.8	7744.3	7791.9	7550.1	7869.2	7701.0
g/kg Bwt/90 days	87.4	86.0	86.6	83.9	87.4	85.6
TEST ITEM INTAKE						
mg/kg Bwt/90 days	0.0	774.4	4675.1	15100.2	0.0	15402.0
Mg/kg Bwt/day	0.0	8.6	51.9	167.8	0.0	171.1

Section A6.4.1/2 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

Official use only

13 Reference

13.1	Reference	[redacted] (2006), Subacute Oral Toxicity Study with Permethrin technical in Swiss Albino Mice. [redacted] unpublished report no.: 05045.
		Dates of experimental work: October 3, 2005 – February 1, 2006.
13.2	**Data protection**	Yes
13.2.1	Data owner	Tagros Chemicals India Ltd.
13.2.2	Companies with letter of access	Not applicable
13.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing a.s for the purpose of its entry into Annex I/IA.

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14 Guidelines and Quality Assurance

14.1	**Guideline study**	Yes, the test method was based on OECD Guideline 408.
14.2	**GLP**	Yes (certified by the Bundesinstitut für Risikobewertung/Federal Institute for Risk Assessment, Germany)
14.3	**Deviations**	None

15 MATERIALS AND MethodS

15.1	**Test material**	As given in section 2 (Permethrin 25:75)
15.1.1	Lot/Batch number	P-41 and P-26
15.1.2	Specification	As given in section 2 (Permethrin 25:75)

Section A6.4.1/2 Repeated dose toxicity

Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

15.1.2.1	Description	Light yellow to brown liquid
15.1.2.2	Purity	94.04 and 93.61%
15.1.2.3	Stability	Test substance was prepared by mixing with vegetable oil freshly everyday before dosing, hence, stability data of test substance in vegetable oil was not collected.
15.2	**Test Animals**	
15.2.1	Species	Mouse (*Mus musculus*)
15.2.2	Strain	Swiss albino mice
15.2.3	Source	[redacted]
15.2.4	Sex	Male and female
15.2.5	Age/weight at study initiation	7 - 9 weeks Males: 26 – 33 g Females: 25 – 31 g
15.2.6	Number of animals per group	Range finding study: Four groups of 5 animals/sex/group Main study Six groups of 10 animals/sex/group
15.2.7	Control animals	Yes
15.3	**Administration/ Exposure**	Oral
15.3.1	Duration of treatment	90 days
15.3.2	Frequency of exposure	Daily
15.3.3	Post exposure period	Two additional recovery groups (control and high dose) were maintained for a further 28 days after the 90-day period, without being exposed to Permethrin for recovery observations.
15.3.4	**Oral**	

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Section A6.4.1/2 Annex Point IIA 6.4	Repeated dose toxicity 90 day oral toxicity study in the mouse	
15.3.4.1 Type	It is stated that the test substance was administered orally by mixing with vegetable oil.	
15.3.4.2 Concentration	Range finding study: 0, 100, 200 and 400 mg/kg bw/day Main study: 0, 20, 40 and 80 mg/kg bw/day	
15.3.4.3 Vehicle	Vegetable oil	
15.3.4.4 Concentration in vehicle	Not documented	
15.3.4.5 Total volume applied	Not documented	
15.3.4.6 Controls	Vegetable oil	
15.4 Examinations		
15.4.1 Observations		
15.4.1.1 Clinical examinations	Yes, once daily on all animals for general health conditions (changes in skin, fur, eyes, changes in gait, posture and response to handling as well as the presence of clonic or tonic movements, stereotypes or bizarre behaviour), behavioural abnormalities and signs of ill health or reactions to treatment.	
15.4.1.2 Mortality	Yes, once daily on all animals for mortality and morbidity	X
15.4.2 Body weight	Yes, before the start of treatment and weekly thereafter.	
15.4.3 Food consumption	Yes, weekly	
15.4.4 Water consumption	Not recorded	
15.4.5 Ophtalmoscopic examination	Yes, on all animals prior to treatment and at study termination.	
15.4.6 Haematology	Yes Number of animals: All animals Time points: on day 91 for animals in the 0, 20, 40, 80 mg/kg/day and satellite groups (control and high dose) and on day 119 for animals in the satellite groups (control and high dose) Parameters: Haematocrit (HCT), Haemoglobin (HB), Platelet Count (PLT), Erythrocyte count (RBC), Total leukocyte count (WBC)	

Section A6.4.1/2 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

		Differential leukocyte count (Neutrophil (N), Lymphocyte (L), Eosinophil (E), Monocyte (M), Basophil (B)), Clotting time.
15.4.7	Clinical Chemistry	Yes Number of animals: All animals Time points: on day 91 for animals in the 0, 20, 40, 80 mg/kg/day and satellite groups (control and high dose) and on day 119 for animals in the satellite groups (control and high dose) Parameters: Total protein, Albumin, Glucose, Aspartate Amino transferase (AST), Alanine Amino transferase (ALT), Blood Urea Nitrogen (BUN), Creatinine, Calcium, Total Bilirubin, Sodium, Potassium.
15.4.8	Urinalysis	Urinalysis was not conducted.
15.5	**Sacrifice and pathology**	
15.5.1	Organ weights	Yes Organs: brain, heart, liver, spleen, adrenals, kidneys, testes, epididymides, ovaries, uterus and thymus.
15.5.2	Gross and histopathology	Yes All dose groups were subjected to gross pathology. Histopathological evaluation was carried on normal and treated skin and on organs and tissues of all animals in the control and high dose groups. Organs: Spinal cord, brain, pituitary, thyroid, parathyroid, thymus, oesophagus, salivary glands, stomach, small intestine, large intestine, liver, pancreas, kidneys, adrenals, spleen, heart, trachea, lungs, aorta, gonads, uterus, accessory sex organs, female mammary gland, prostate, urinary bladder, gall bladder, lymph nodes, peripheral nerve, bone marrow, skin and all gross lesions.
15.5.3	Other examinations	None
15.5.4	Statistics	The changes in bodyweight, feed consumption, haematology, biochemistry and organ weights in the treatment groups were compared with the control group using Student`s t-test (NCSS, 2000)
15.6	**Further remarks**	None

Section A6.4.1/2 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

16 Results and Discussion

16.1	**Observations**		
16.1.1	Clinical signs	Toxicity signs such as respiratory distress, hyperactivity and tremor were observed in both sexes In the 80 mg/kg/day dose group and in the high dose satellite group from Day 50 until Day 92. No treatment related effects were observed in any other groups.	
16.1.2	Mortality	No treatment related mortality	
16.2	**Body weights**	No treatment related changes	X
16.3	**Food consumption and compound intake**	No treatment related changes	X
16.4	**Ophtalmoscopic examination**	No abnormal ocular lesions were observed in any animal at any dose level.	
16.5	**Blood analysis**		
16.5.1	Haematology	No treatment related changes	
16.5.2	Clinical chemistry	No treatment related changes	
16.5.3	Urinalysis	Not required	
16.6	**Sacrifice and pathology**		
16.6.1	Organ weights	No treatment related changes	
16.6.2	Gross and histopathology	No treatment related finding	
16.7	**Other**	None	

17 Applicant's Summary and conclusion

17.1	**Materials and methods**	Based on the results obtained in the range finding study, four groups of 10 animals/sex were exposed to Permethrin at concentrations of 0, 20, 40 and 80 mg/kg/day during 90 days. Two additional groups of 10 animals/sex used as satellite at 0 and 80 mg/kg/day were treated during 90 days and were then observed during 28 days for delayed occurrence

Section A6.4.1/2 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

toxic effects.

17.2 Results and discussion

Toxicity signs such as respiratory distress, hyperactivity and tremor were observed in both sexes in the 80 mg/kg/day dose group from Day 50 until Day 91 and in males and females in the high dose satellite group from Day 52 until Day 92 and from Day 51 until Day 91, respectively. These toxicity signs disappeared from animals in the high dose satellite group during the post-exposure period.

None of the other treated groups showed any clinical signs of toxicity.

Four males died during the dosing period (two in the 80 mg/kg/day dose group and two in the high dose satellite group). Two females died in the high dose satellite group after the blood collection on Day 92. None of the observed mortalities was considered to be treatment related.

Body weight gain of the 80 mg/kg bw/day dose group and the high dose satellite group animals (both sexes) showed a statistically significant decrease from week 8 onwards when compared to control groups. Other treated groups showed normal body weight gain for the entire study period. Please refer to Table A6.4.1/2-1 and Table A6.4.1/2-2.

A statistically significant decrease was observed in food consumption in the 80 mg/kg bw/day dose group and in the high dose satellite group (both sexes) from week 7 and 8 onwards when compared with control groups. These changes were not considered to be treatment related. Please refer to Table A6.4.1/2-3 and Table A6.4.1/2-4.

Haematological and biochemical values revealed no relevant changes after Permethrin treatment. Some parameters showed statistically significant changes in the treated groups when compared with the control groups, but these variations were within the biological ranges of this species. X

Absolute and relative male right gonad weights were significantly decreased at the 40 mg/kg bw/day treatment dose group. The relative brain weight of males was significantly increased in the 80 mg/kg bw/day dose group. The relative liver, spleen and right gonad weight in the 80 mg/kg bw/day dose group and the relative brain and uterus weight in the high dose satellite group were statistically increased in females. However none of these changes were considered to be treatment related. Please refer to Table A6.4.1/2-5 to Table A6.4.1/2-7.

At necropsy, there was no treatment related findings. All gross observations were incidental and spontaneous in nature and bore no relation to treatment with Permethrin technical.

There was no Permethrin related histological findings in this study. All microscopic findings were either agonal such as lymphangiectasis, haemangiectasis, related to infection such as bronchitis and bronchiolitis or of the type routinely observed in Swiss albino mice of this age such

Section A6.4.1/2 **Repeated dose toxicity**
Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

	as arteritis in the heart and microgranuloma in the liver. No changes in any of the sexual organs were considered to be treatment related. Therefore based on these results, Permethrin cannot be considered as an endocrine disruptor.
17.3 Conclusion	A LO(A)EL of 80 mg/kg bw/day for males and females was established in the study. This was based on findings such as decreases in food consumption and body weight, variations in the haematological and biochemical values and changes in organ weights. However none of these changes was considered to be treatment related at 80 mg/kg bw/day. A NO(A)EL of 40 mg/kg bw/day day for males and females was established in the study. This was based on findings such as decreases in food consumption and body weight, variations in the haematological and biochemical values and changes in organ weights. However none of these changes was considered to be treatment related at 80 mg/kg bw/day.
17.3.1 LO(A)EL	Male: 80 mg/kg bw/day Female: 80 mg/kg bw/day
17.3.2 NO(A)EL	Male: 40 mg/kg bw/day Female: 40 mg/kg bw/day
17.3.3 Reliability	1
17.3.4 Deficiencies	No

Evaluation by Competent Authorities	
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	Evaluation by Rapporteur Member State
Date	03/07/2009
Materials and Methods	applicants version is acceptable
Results and discussion	Adopt applicant's version 5.2. There are significant differences and biological variations in biochemical and haematological parameters, but do not appear to be treatment related.

Section A6.4.1/2 Repeated dose toxicity

Annex Point IIA 6.4 **90 day oral toxicity study in the mouse**

Conclusion	LO(A)EL: Male: 80 mg/kg bw/day Female: 80 mg/kg bw/day NO(A)EL: Male: 40 mg/kg bw/day Female: 40 mg/kg bw/day
Reliability	1
Acceptability	Acceptable
Remarks	

Comments from ...

Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Table A6.4.1/2-1: Summary of male body weights (g)

Assessment time	Males (g)					
	Control	20 mg/kg bw	40 mg/kg bw	80 mg/kg bw	Satellite control	Satellite 80 mg/kg bw
	Mean ± SD					
Week 0	29.63 ± 1.94	28.47 ± 1.73	30.25 ± 1.70	30.64± 1.95	30.09 ± 2.35	29.70 ± 2.63
Week 1	30.38 ± 1.67	29.03 ± 1.68	31.01 ± 1.37	30.85 ± 2.39	31.31 ± 2.16	30.75 ± 1.81
Week 2	31.12 ± 1.64	30.17 ± 1.60	30.87 ± 1.84	30.66 ± 2.64	31.96 ± 2.07	31.03 ± 1.64
Week 3	31.55 ± 2.12	30.67 ± 1.53	31.59 ± 1.33	31.16 ± 2.25	32.64 ± 2.17	31.68 ± 1.83
Week 4	32.11 ± 1.73	30.75 ± 1.36	32.63 ± 1.10	32.12 ± 2.39	33.38 ± 2.38	32.13 ± 1.48
Week 5	32.82 ± 2.55	32.31 ± 1.14	33.41 ± 1.84	32.43 ± 2.23	34.07 ± 1.84	32.60 ± 1.44
Week 6	33.18 ± 2.41	32.76 ± 1.46	34.39 ± 1.87	32.53 ± 2.28	34.87 ± 1.73	33.53 ± 1.16
Week 7	33.83 ± 2.05	33.66 ± 1.67	35.89 ± 2.03	32.55 ± 2.29	35.83 ± 1.72	34.52 ± 1.18
Week 8	35.52 ± 1.89	34.73 ± 1.54	36.30 ± 2.56	33.08 ± 2.16*	36.93 ± 2.42	34.51 ± 1.61*
Week 9	36.90 ± 2.02	35.53 ± 1.79	37.30 ± 2.34	34.13 ± 2.05*	37.61 ± 2.37	35.16 ± 1.77*
Week 10	37.64 ± 2.23	36.32 ± 2.05	37.86 ± 2.36	34.78 ± 2.33*	38.53 ± 2.26	36.41 ± 2.19*
Week 11	37.80 ± 2.23	37.67 ± 2.18	39.25 ± 2.56	35.54 ± 2.50*	39.71 ± 2.02	36.30 ± 1.91*
Week 12	39.64 ± 1.98	38.33 ± 2.37	39.67 ± 2.58	36.00 ± 2.64*	40.50 ± 2.25	36.99 ± 1.66*
Week 13	40.46 ± 1.93	39.13 ± 2.37	40.19 ± 2.50	36.63 ± 2.54*	41.39 ± 2.12	37.56 ± 1.69*
Week 14	-	-	-	-	41.74 ± 1.89	37.94 ± 1.79*
Week 15	-	-	-	-	41.85 ± 1.72	38.37 ± 1.93*
Week 16	-	-	-	-	42.23 ± 1.66	38.80 ± 1.88*
Week 17	-	-	-	-	42.60 ± 1.49	39.77 ± 1.85*

* Significantly different from the control group; $p < 0.05$

Table A6.4.1/2-2: Summary of female body weights (g)

Assessment time	Females (g)					
	Control	20 mg/kg bw	40 mg/kg bw	80 mg/kg bw	Satellite Control	Satellite 80 mg/kg bw
	Mean ± SD					
Week 0	26.62 ± 1.48	26.44 ± 1.33	27.20 ± 1.73	26.56 ± 1.85	26.90 ± 1.47	26.75 ± 1.48
Week 1	27.31 ± 2.00	27.25 ± 1.46	27.85 ± 1.94	26.98 ± 1.82	27.39 ± 1.52	26.93 ± 2.22
Week 2	27.73 ± 1.88	27.76 ± 1.57	28.29 ± 1.94	27.36 ± 1.99	27.96 ± 1.57	27.17 ± 1.95
Week 3	28.58 ± 1.97	28.37 ± 1.62	29.01 ± 1.85	27.49 ± 2.06	28.35 ± 1.62	27.25 ± 1.85
Week 4	28.94 ± 2.03	28.84 ± 1.39	29.55 ± 1.89	27.77 ± 1.79	29.34 ± 1.68	27.72 ± 1.87
Week 5	29.61 ± 2.44	29.39 ± 1.61	30.24 ± 1.67	27.78 ± 2.12	29.96 ± 1.77	28.58 ± 1.68
Week 6	30.18 ± 2.37	30.12 ± 1.51	30.99 ± 1.99	28.39 ± 2.02	30.49 ± 1.51	29.11 ± 1.78
Week 7	30.73 ± 2.22	30.59 ± 1.75	31.53 ± 1.74	28.93 ± 1.92	31.08 ± 1.63	29.86 ± 1.82
Week 8	31.15 ± 2.31	31.12 ± 1.69	32.24 ± 1.61	29.00 ± 1.73*	31.82 ± 1.86	29.38 ± 2.08*
Week 9	32.29 ± 1.79	31.91 ± 1.60	33.02 ± 1.66	29.37 ± 1.58*	32.36 ± 1.64	29.53 ± 2.27*
Week 10	33.33 ± 2.05	32.73 ± 1.70	33.76 ±1.69	29.74 ± 1.85*	32.97 ± 1.60	29.87 ± 2.57*
Week 11	33.75 ± 1.42	33.16 ± 1.56	34.26 ± 1.69	30.52 ± 1.83*	33.88 ± 1.62	30.48 ± 2.36*
Week 12	34.67 ± 1.84	34.05 ± 1.27	34.80 ± 1.58	31.13 ± 1.71*	34.99 ± 1.64	31.44 ± 2.25*
Week 13	35.57 ± 1.92	34.99 ± 1.16	35.82 ± 1.51	32.03 ± 1.77*	35.86 ± 1.86	32.27 ± 2.33*
Week 14	-	-	-	-	36.51 ± 1.91	33.07 ± 2.37*
Week 15	-	-	-	-	36.90 ± 1.86	33.48 ± 2.25*
Week 16	-	-	-	-	37.45 ± 2.04	33.09 ± 2.36*
Week 17	-	-	-	-	37.83 ± 1.92	34.65 ± 2.51*

* Significantly different from the control group; $p < 0.05$

Table A6.4.1/2-3: Summary of male feed consumption (g)

Assessment time	Males (g)					
	Control	20 mg/kg bw	40 mg/kg bw	80 mg/kg bw	Satellite Control	Satellite 80 mg/kg bw
	Mean ± SD					
Week 1	14.60 ± 0.65	14.44 ± 0.58	14.25 ± 0.22	14.47 ± 0.63	14.41 ± 0.29	14.23 ± 0.30
Week 2	15.07 ± 0.46	14.69 ± 0.50	14.65 ± 0.21	14.86 ± 0.60	14.74 ± 0.34	14.68 ± 0.50
Week 3	15.94 ± 0.20	15.51 ± 0.56	15.30 ± 0.69	15.41 ± 0.56	15.30 ± 0.27	15.11 ± 0.42
Week 4	16.67 ± 0.57	16.37 ± 0.22	15.64 ± 0.69	15.98 ± 0.12	16.13 ± 0.30	15.71 ± 0.51
Week 5	16.88 ± 0.48	16.97 ± 0.31	16.32 ± 0.92	16.28 ± 0.15	16.60 ± 0.45	16.18 ± 0.42
Week 6	17.47 ± 0.48	17.62 ± 0.03	16.69 ± 0.93	16.63 ± 0.23	16.85 ± 0.55	16.47 ± 0.52
Week 7	17.93 ± 0.53	18.26 ± 0.11	16.99 ± 0.97	16.33 ± 0.63*	17.37 ± 0.51	16.13 ± 0.22*
Week 8	18.03 ± 0.38	18.22 ± 0.15	17.51 ± 0.58	16.58 ± 0.59*	17.75 ± 0.37	16.36 ± 0.36*
Week 9	18.21 ± 0.44	17.56 ± 0.07	17.45 ± 0.98	16.73 ± 0.45*	18.05 ± 0.42	16.71 ± 0.49*
Week 10	18.32 ± 0.46	17.78 ± 0.21	17.84 ± 0.59	16.96 ± 0.24*	18.32 ± 0.52	16.95 ± 0.60*
Week 11	18.31 ± 0.56	17.70 ± 0.41	17.58 ± 1.06	17.01 ± 0.10*	18.00 ± 0.34	16.70 ± 0.44*
Week 12	18.41 ± 0.64	17.70 ± 0.24	17.39 ± 0.62	16.89 ± 0.33*	18.16 ± 0.32	17.28 ± 0.31*
Week 13	18.97 ± 0.18	18.17 ± 0.07	17.90 ± 0.91	17.38 ± 0.15*	18.01 ± 0.09	17.27 ± 0.18*
Week 14	-	-	-	-	18.23 ± 0.10	17.35 ± 0.16*
Week 15	-	-	-	-	18.25 ± 0.03	17.41 ± 0.12*
Week 16	-	-	-	-	18.22 ± 0.23	17.52 ± 0.02*
Week 17	-	-	-	-	18.39 ± 0.20	17.48 ± 0.20*

* Significantly different from the control group; $p < 0.05$

Table A6.4.1/2-4: Summary of female feed consumption (g)

Assessment time	Females (g)					
	Control	20 mg/kg bw	40 mg/kg bw	80 mg/kg bw	Satellite control	Satellite 80 mg/kg bw
	Mean ± SD					
Week 1	11.61 ± 0.78	12.20 ± 0.62	11.68 ± 0.33	11.80 ± 0.40	12.03 ± 0.43	11.74 ± 0.25
Week 2	12.39 ± 0.53	12.52 ± 0.20	12.12 ± 0.24	12.56 ± 0.58	12.39 ± 0.17	12.23 ± 0.20
Week 3	12.80 ± 0.49	13.11 ± 0.34	12.70 ± 0.51	13.16 ± 0.37	13.13 ± 0.24	12.91 ± 0.05
Week 4	13.23 ± 0.45	13.76 ± 0.35	13.05 ± 0.39	13.50 ± 0.22	13.57 ± 0.36	13.31 ± 0.03
Week 5	13.71 ± 0.46	14.49 ± 0.20	13.86 ± 0.96	13.55 ± 0.10	13.89 ± 0.23	13.71 ± 0.16
Week 6	14.06 ± 0.62	14.87 ± 0.52	14.08 ± 1.06	13.45 ± 0.26	14.46 ± 0.25	14.02 ± 0.17
Week 7	14.42 ± 0.61	15.07 ± 0.15	14.27 ± 1.01	13.16 ± 0.32	14.79 ± 0.24	14.01 ± 0.24
Week 8	14.62 ± 0.44	15.33 ± 0.06	14.54 ± 0.81	13.54 ± 0.07*	15.00 ± 0.29	14.44 ± 0.05*
Week 9	14.79 ± 0.48	14.95 ± 0.25	14.86 ± 0.92	13.41 ± 0.22*	15.45 ± 0.30	14.72 ± 0.06*
Week 10	15.02 ± 0.41	15.02 ± 0.18	14.75 ± 0.23	13.61 ± 0.53*	15.58 ± 0.28	15.00 ± 0.05*
Week 11	15.01 ± 0.24	15.01 ± 0.17	14.49 ± 0.30	13.65 ± 0.39*	15.98 ± 0.33	15.17 ± 0.13*
Week 12	15.15 ± 0.36	14.83 ± 0.55	14.35 ± 0.73	13.70 ± 0.40*	16.05 ± 0.19	15.18 ± 0.33*
Week 13	15.18 ± 0.23	15.13 ± 0.49	14.53 ± 0.88	13.63 ± 0.10	16.25 ± 0.13	15.18 ± 0.52*
Week 14					16.40 ± 0.26	15.44 ± 0.45*
Week 15					16.45 ± 0.26	15.45 ± 0.41*
Week 16					16.76 ± 0.16	15.64 ± 0.26*
Week 17					16.90 ± 0.16	15.80 ± 0.17*

* Significantly different from the control group; $p < 0.05$

Table A6.4.1/2-5: Summary of absolute organ weight in male mice(g)

Group/dose	Male	
	Gonad left	Gonad right
	Mean ± SD	
Control	0.142 ± 0.040	0.144 ± 0.039
20 mg/kg bw	0.117 ± 0.014	0.117 ± 0.019
40 mg/kg bw	0.116± 0.021	0.114 ± 0.021*
80 mg/kg bw	0.129 ± 0.023	0.126 ± 0.022
Satellite control	0.115 ± 0.025	0.118 ± 0.023
Satellite 80 mg/kg bw	0.112 ± 0.008	0.118 ± 0.010

* Significantly different from the control group; p< 0.05

Table A6.4.1/2-6: Summary of relative organ weight in male mice (%)

Group/dose	Male		
	Brain	Gonad left	Gonad right
	Mean ± SD		
Control	1.136 ± 0.120	0.350 ± 0.092	0.355 ± 0.092
20 mg/kg bw	1.214 ± 0.094	0.296 ± 0.037	0.295 ± 0.041
40 mg/kg bw	1.137 ± 0.087	0.289 ± 0.058	0.283 ± 0.054*
80 mg/kg bw	1.265 ± 0.110*	0.353 ± 0.066	0.344 ± 0.061
Satellite control	1.107 ± 0.068	0.270 ± 0.061	0.276 ± 0.058
Satellite 80 mg/kg bw	1.212 ± 0.131	0.282 ± 0.024	0.298 ± 0.026

* Significantly different from the control group; p< 0.05

Table A6.4.1/2-7: Summary of relative organ weight in female mice (%)

Group/dose	Female					
	Brain	Liver	Spleen	Gonad left	Gonad right	Uterus
	Mean ± SD					
Control	1.416 ± 0.114	4.056 ± 0.720	0.379 ± 0.093	0.059 ± 0.018	0.053 ± 0.016	0.370 ± 0.120
20 mg/kg bw	1.328 ± 0.145	4.653 ± 1.119	0.503 ± 0.236	0.057 ± 0.027	0.056 ± 0.017	0.454 ± 0.085
40 mg/kg bw	1.361 ± 0.184	3.859 ± 1.298	0.654 ± 0.727	0.057 ± 0.010	0.058 ± 0.007	0.346 ± 0.176
80 mg/kg bw	1.487 ± 0.088	5.031 ± 0.663*	0.584 ± 0.190*	0.066 ± 0.014	0.066 ± 0.011*	0.343 ± 0.119
Satellite control	1.179 ± 0.111	3.509 ± 1.098	0.365 ± 0.150	0.050± 0.008	0.054 ± 0.011	0.231 ± 0.013
Satellite 80 mg/kg bw	1.342 ± 0.118*	3.787 ± 0.889	0.486 ± 0.243	0.052± 0.007	0.053 ± 0.005	0.266 ± 0.013*

* Significantly different from the control group; $p < 0.05$

Official use only

18 Reference

1.1	Reference	[redacted] (2006), Subacute Dermal Toxicity Study with Permethrin technical in Wistar Rats, [redacted] unpublished report no.: 14996 Dates of experimental work: May 17, 2005 – September 5, 2005
1.2	**Data protection**	Yes
1.2.1	Data owner	Tagros Chemicals India Ltd.
1.2.2	Companies with letter of access	Not applicable
1.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing a.s. for the purpose of its entry into Annex I/IA.

Comment [T21]: Confidential
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2 Guidelines and Quality Assurance

2.1	**Guideline study**	Yes, the test method was based on OECD Guideline 411.
2.2	**GLP**	Yes (certified by Bundesinstitut für Risikobewertung, Germany)
2.3	**Deviations**	None

3 MATERIALS AND MethodS

3.1	**Test material**	As given in section 2 (Permethrin 25:75)
3.1.1	Lot/Batch number	P-41, P-11
3.1.2	Specification	As given in section 2 (Permethrin 25:75)

Section A6.4.2 Repeated dose toxicity

Annex Point IIA6.4 **90-day dermal toxicity study in the rat**

3.1.2.1	Description	Light yellow to brown liquid	
3.1.2.2	Purity	94.04, 92.86% m/m	
3.1.2.3	Stability	The test material is considered to be stable throughout the study.	
3.2	**Test Animals**		
3.2.1	Species	*Rattus norvegicus* - Rat	
3.2.2	Strain	Wistar	
3.2.3	Source	[redacted]	
3.2.4	Sex	Male and female	
3.2.5	Age/weight at study initiation	Age not documented Male: 204 – 336 g Females: 178 – 228 g	X
3.2.6	Number of animals per group	6 groups of 10 animals/sex/group	
3.2.7	Control animals	Yes	
3.3	**Administration/ Exposure**	Dermal	X
3.3.1	Duration of treatment	13 weeks	
3.3.2	Frequency of exposure	5 days per week	X
3.3.3	Postexposure period	28 days for a satellite control group and high dose level satellite group	
3.3.4	**Dermal**		

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3.3.4.1	Area covered	4 x 5 cm^2 of the dorsal area of each rat
3.3.4.2	Occlusion	Occlusive
3.3.4.3	Vehicle	Undiluted (as received)
3.3.4.4	Concentration in vehicle	Not applicable
3.3.4.5	Total volume applied	Not documented
3.3.4.6	Duration of exposure	6 hours
3.3.4.7	Removal of test substance	The skin was washed with lukewarm water and sponged with gauze
3.3.4.8	Controls	Yes
3.4	**Examinations**	
3.4.1	Observations	
3.4.1.1	Clinical signs	General health condition, behavioural abnormalities and signs of ill health or reactions to treatment were recorded every day in individual rats of all groups.
3.4.1.2	Mortality	Yes, daily
3.4.2	Body weight	Yes, on day 0 and weekly thereafter
3.4.3	Food consumption	Yes, weekly
3.4.4	Water consumption	Not documented
3.4.5	Ophthalmoscopic examination	Yes, prior to exposure and at termination of the study in high and control groups
3.4.6	Haematology	Yes, Number of animals: all animals at 0, 500, 1000 and 2000 mg/kg bw/day Time points: on day 91 Parameters: Haematocrit, haemoglobin concentration, erythrocyte count, total and differential leukocyte count, platelet count, clotting time and prothrombin time Number of animals: all animals in the control satellite group and in the 2000 mg/kg bw/day satellite group Time points: on days 92 and 119 Parameters: Haematocrit, haemoglobin concentration, erythrocyte count, total and differential leukocyte count, platelet count, clotting

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		time and prothrombin time
3.4.7	Clinical Chemisty	Yes, Number of animals: all animals at 0, 500, 1000 and 2000 mg/kg bw/day Time points: on day 91 Parameters: calcium, phosphorus, chloride, sodium, potassium, glucose, alanine aminotransferase, aspartate aminotransferase, gamma glutamyl transpeptidase, blood urea nitrogen, albumin, total protein, total bilirubin and creatinine Number of animals: all animals in the control satellite group and in the 2000 mg/kg bw/day satellite group Time points: on days 92 and 119 Parameters: calcium, phosphorus, chloride, sodium, potassium, glucose, alanine aminotransferase, aspartate aminotransferase, gamma glutamyl transpeptidase, blood urea nitrogen, albumin, total protein, total bilirubin and creatinine
3.4.8	Urinalysis	Not required
3.5	**Sacrifice and pathology**	
3.5.1	Organ Weights	Yes All animals that were found dead or moribund and at the terminal sacrifice (scheduled on Days 91 and 119). Organs: liver, kidneys, adrenals, testes and ovaries
3.5.2	Gross and histopathology	Yes, All animals were subjected to a gross necropsy The following organs and tissues were preserved: All gross lesions, brain (including sections of medulla/pons), cerebellar cortex and cerebral cortex, pituitary, thyroid/parathyroid, thymus, lungs, heart, aorta, salivary glands, liver, spleen, kidneys, adrenals, pancreas, gonads, accessory genital organs, oesophagus, stomach, duodenum, jejunum, ileum, caecum, colon, rectum, urinary bladder, representative lymph nose, peripheral nerve. All gross lesions were examined. Full histopathology was carried out on normal and treated skin and on organs and tissues of all animals in the control and high dose groups. Histopathology was performed in satellite groups on tissues and organs identified as showing effects in other treated groups.
3.5.3	Other examinations	None
3.5.4	Statistics	The data were tested for normality. Normal data were subjected to one-way ANOVA and Post ANOVA comparison was carried out using the Newman-Keuls test. Non normal data was subjected to Kruskal-Wallis one way ANOVA on Ranks and post ANOVA comparison was carried

		out using Kruskal-Wallis Z test.
3.6	**Further remarks**	**None**

4 Results and Discussion

4.1	**Observations**	
4.1.1	Clinical signs	Signs of tremor and piloerection were noted at 2000 mg/kg bw between days 39 and 91.
4.1.2	Mortality	No mortality was observed in any group
4.2	**Body weight gain**	Decrease in body weight gain from week 3 to week 13 in males at 2000 mg/kg bw. Decrease in body weight gain from week 3 to week 17 in males at 2000 mg/kg bw in the satellite group
4.3	**Food consumption and compound intake**	Decrease in food consumption from week 4 to week 13 in males at 2000 mg/kg bw Decrease in food consumption from week 4 to week 13 in males at 2000 mg/kg bw satellite group
4.4	**Ophtalmoscopic examination**	No abnormal ocular lesions were observed in any of the treated groups.
4.5	**Blood analysis**	
4.5.1	Haematology	No treatment related effects were recorded.
4.5.2	Clinical chemistry	Decrease in phosphorus on day 91 in males and females at 1000 and 2000 mg/kg bw
4.5.3	Urinalysis	Not applicable
4.6	**Sacrifice and pathology**	
4.6.1	Organ weights	Male Increase in mean relative liver weight at 500, 1000 and 2000 mg/kg bw Increase in mean relative left and right kidney weight at 2000 mg/kg bw Female Increase in mean relative liver weight at 1000 and 2000 mg/kg bw Increase in mean absolute liver weights at 1000 and 2000 mg/kg bw Increase in mean absolute left kidney weight at 1000 and 2000 mg/kg bw Decrease in mean relative right gonad weight at 1000 and 2000 mg/kg bw

		Decrease in absolute right gonad weight at 1000 and 2000 mg/kg bw Other changes were noted but not considered to be treatment related.
4.6.2	Gross and histopathology	No treatment related changes or histological findings were observed.
4.7	**Other**	None

5 Applicant's Summary and conclusion

5.1	**Materials and methods**	Permethrin was applied to the clipped skin of Wistar rats (10 animals/sex/group) at the following concentrations 0, 500, 1000 and 2000 mg/kg bw for 5 consecutive days in a week during thirteen weeks. Two additional groups, a control satellite group and a 2000 mg/kg bw satellite group, were studied for another four weeks after the treatment period. This study was conducted according to OECD Guideline 411 and is described under point 3 with no deviations
5.2	**Results and discussion**	No mortality was recorded in any group throughout the study. Clinical signs such as tremor and piloerection were noted at 2000 mg/kg bw on day 39 to 90. These clinical signs of toxicity were deemed to be treatment related. However these signs disappeared during the post exposure period. No other clinical signs were recorded in any control and treated groups. No abnormal ocular lesions were reported in any control and treated groups. At 2000 mg/kg bw, males exhibited a statistically significant decrease in body weight gain from week 3 until study termination. This reduction was also noted in males in the 2000 mg/kg bw satellite group. Similarly the decrease in food consumption was statistically significant in these groups at the same time. No changes in body weight gain or in food consumption were observed in the other treated groups. Please refer to Tables A6.4.2-1 to A6.4.2-4. No statistically significant changes in haematological parameters were noted in any of the treated groups during the treatment period and post exposure treatment. Both males and females exhibited a statistically significant reduction in urine levels of phosphorus at 1000 and 2000 mg/kg bw on day 91. However these changes were not observed in the 2000 mg/kg bw satellite group on day 92. Therefore, this decrease may not be due to

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the application of Permethrin. Please refer to Tables A6.4.2-5 and A6.4.2-6.

Male rats showed an increase in mean relative liver weight at 500, 1000 and 2000 mg/kg bw. This was deemed to be treatment related. A similar trend in females was also reported and this was correlated with the increase in mean absolute liver weight in females. However changes were not statistically significant. Furthermore at the end of the recovery, absolute and relative liver weights in the 2000 mg/kg bw satellite group were comparable to or below those of control rats. No histological correlation could be established.

Males revealed an increase in mean relative left and right kidney weights at 2000 mg/kg bw. Females exhibited an increase in mean absolute left kidney weight, a decrease in mean relative right gonad weight and a decrease in mean absolute right gonad weight at 1000 and 2000 mg/kg bw. These changes in both males and females were deemed to be treatment related. However, at the end of the post exposure period, absolute and relative kidney weights were comparable to or below those of the control satellite. Therefore these changes in kidney weights were not considered to be adverse since only the left kidney in females was affected and no histological correlation could be established. A similar conclusion was drawn for the changes in gonads. Please refer to Tables A6.4.2-7 to A6.4.2-10.

No treatment related gross or histological lesions were noted in any of the treated groups.

5.3 Conclusion

Based on clinical signs of toxicity such as tremors and piloerections and liver weight changes, the NOAEL in rats was established to be 1000 mg/kg bw by dermal route under the conditions described in the study.

5.3.1	LO(A)EL	2000 mg/kg bw/day
5.3.2	NO(A)EL	1000 mg/kg bw/day
5.3.3	Other	None
5.3.4	Reliability	1
5.3.5	Deficiencies	No

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	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	Evaluation by Rapporteur Member State
Date	*26 June 2009*
Materials and Methods	2.3 The study was performed using technical rather than high purity material. 3.2.5 Female rats weights were less than 200g at the start of the study. 3.3 Animal room temperature is not described in the study report. 3.3 The top dose of 2000 mg/kg bw/d is in excess of the limit dose (1000 mg/kg bw/d) 3.3.2 The substance was applied for 5 rather than 7 days a week.
Results and discussion	*Adopt applicant's version.*
Conclusion	**LO(A)EL: 2000 mg/kg bw/d** **NO(A)EL: 1000 mg/kg bw/d** **Other conclusions:** Adopt applicants version.
Reliability	1
Acceptability	Acceptable
Remarks	The NOAEL is based on clinical signs (piloerection and tremor) at the top dose. The liver weight increases (greater than 10% at all doses in males) have not been considered as adverse because the only other liver effects seen are centrilobular hypertrophy at the top dose. The liver weight and hypertrophy have been regarded as adaptive responses. Aside from the minor materials and methods deviations the study is of good quality.
	Comments from ... *(SPECIFY)*
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Table A6.4.2-1: Summary of body weight values in grams (males)

Week of study	Control	500 mg/kg bw/day	1000 mg/kg bw/day	2000 mg/kg bw/day	Satellite control	Satellite 2000 mg/kg bw/day
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
0	269.60 ± 12.81	271.20 ± 47.42	286.90 ± 26.68	261.80 ± 28.82	255.50 ± 21.36	259.90± 31.68
1	310.60 ± 32.95	300.90 ± 50.28	313.40 ± 27.69	282.50 ± 30.93	281.10 ± 24.96	287.90 ± 36.71
2	330.80 ± 14.93	324.70 ± 50.69	337.50 ± 27.04	308.50 27.83	305.80 ± 26.28	314.30 ± 35.37
3	335.60 ± 15.01	332.40 ± 45.91	331.90 ± 35.18	308.30 ± 33.97[a]	325.60 ± 27.09	321.70 ± 35.62[a]
4	348.70 ± 13.16	350.30 ± 58.22	347.00 ± 35.97	318.60 ± 30.52[a]	355.80 ± 24.40	332.80 ± 31.39[a]
5	357.20 ± 15.68	363.00 ± 56.97	358.30 ± 39.30	332.10 ± 32.05[a]	376.60 ± 21.54	341.00 ± 31.41[a]
6	364.90 ± 16.52	374.10 ± 56.76	360.80 ± 43.72	337.40 ± 38.82[a]	383.10 ± 22.64	349.20 ± 31.46[a]
7	369.00 ± 11.47	382.60 ± 56.68	360.60 ± 50.27	348.10 ± 42.64[a]	394.00 ± 30.77	357.60 ± 31.51[a]
8	371.80 ± 15.31	390.80 ± 60.60	373.10 ± 43.61	353.40 ± 34.90[a]	410.00 ± 33.24	365.70 ± 31.95[a]
9	383.50 ± 11.52	402.60 ± 61.87	376.50 ± 41.11	360.10 ± 34.53[a]	423.10 ± 34.57	374.10 ± 31.89[a]
10	393.70 ± 15.64	418.00 ± 56.63	388.70 ± 42.77	366.60 ± 34.45[a]	438.30 ± 33.64	382.50 ± 32.10[a]
11	402.60 ± 19.95	423.10 ± 57.44	399.90 ± 40.31	373.00 ± 34.28[a]	453.60 ± 35.05	390.80 ± 32.22[a]
12	401.00 ± 15.79	426.50 ± 58.19	409.90 ± 29.22	379.60 ± 34.33[a]	456.90 ± 35.36	399.20 ± 32.44[a]
13	414.20 ± 12.54	426.00 ± 60.64	414.20 ± 33.02	386.10 ± 34.38[a]	453.80 ± 34.25	407.60 ± 32.79[a]
14	-	-	-	-	463.60 ± 33.22	415.90 ± 33.17[a]
15	-	-	-	-	471.20 ± 35.66	424.10 ± 33.46[a]
16	-	-	-	-	477.70 ± 32.26	432.30 ± 33.63[a]
17	-	-	-	-	487.40 ± 33.41	440.20 ± 33.64[a]

values are expressed as mean ± SD (n=10)
[a] statistically different ($p<0.05$)

Table A6.4.2-2: Summary of body weight values in grams (females)

Week of study	Control	500 mg/kg bw/day	1000 mg/kg bw/day	2000 mg/kg bw/day	Satellite control	Satellite 2000 mg/kg bw/day
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
0	203.90 ± 7.39	194.50 ± 11.20	210.00 ± 9.17	204.70 ± 14.78	197.40 ± 9.73	199.40 ± 15.30
1	213.40 ± 15.55	203.60 ± 13.62	224.40 ± 10.42	219.3 ± 16.19	212.10 ± 10.26	214.10 ± 17.51
2	222.80 ± 16.46	215.90 ± 10.28	235.60 ± 13.64	231.40 ± 18.39	226.90 10.10	225.40 ± 19.65
3	226.30 ± 23.39	220.20 ± 13.05	235.30 ± 18.84	220.00 ± 24.89	229.60 16.37	223.30 ± 19.18
4	236.40 ± 23.14	223.10 ± 13.67	241.40 ± 15.71	227.80 ± 29.22	238.30 ± 13.22	232.80 ± 19.00
5	240.60 ± 23.99	225.00 ± 16.54	237.80 ± 18.25	236.60 ± 24.97	242.00 ± 11.47	235.60 ± 20.28
6	245.20 ± 24.77	231.40 ± 10.13	241.40 ± 16.61	241.60 ± 23.82	241.90 ± 10.75	234.80 ± 22.51
7	248.50 ± 25.49	235.20 ± 13.68	234.10 ± 22.45	242.60 ± 25.36	240.50 ± 15.53	239.00 ± 27.39
8	247.20 ± 25.54	239.40 ± 14.09	242.30 ± 16.76	246.20 ± 29.68	245.50 ± 16.08	244.40 ± 24.86
9	254.20 ± 26.45	245.10 ± 14.87	247.20 ± 14.25	253.80 ± 27.15	250.30 ± 13.93	247.10 ± 24.15
10	258.00 ± 26.42	249.80 ± 15.82	242.70 ± 14.24	250.60 ± 23.92	254.00 ± 13.00	245.40 ± 28.34
11	258.20 ± 28.22	253.00 ± 14.28	244.40 ± 15.42	253.50 ± 30.05	257.80 ± 17.44	246.70 ± 26.82
12	258.50 ± 28.62	250.90 ± 16.88	249.60 ± 16.04	255.00 ± 23.98	258.50 ± 18.53	244.00 ± 26.40
13	250.40 ± 25.58	249.70 ± 11.13	246.90 ± 18.81	254.20 ± 23.79	259.30 ± 19.43	243.10 ± 30.06
14	-	-	-	-	258.20 ± 12.87	244.80 ± 31.09
15	-	-	-	-	260.90 ± 17.95	244.10 ± 31.63
16	-	-	-	-	261.30 ± 19.49	246.30 ± 36.46
17	-	-	-	-	263.00 ± 11.16	244.30 ± 44.37

values are expressed as mean ± SD (n=10)
[a] statistically different ($p<0.05$)

Table A6.4.2-3: Summary of weekly feed consumption in grams (males)

Week of study	Control	500 mg/kg bw/day	1000 mg/kg bw/day	2000 mg/kg bw/day	Satellite control	Satellite 2000 mg/kg bw/day
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
1	14.50 ± 1.45	15.2 ± 0.5	16.36 ± 1.23	16.1 ± 0.6	16.0 ± 0.2	16.5 ± 0.3
2	15.04 ± 1.09	15.0 ± 0.2	15.37 ± 0.64	15.2 ± 0.3	14.6 ± 0.1	16.6 ± 1.2
3	17.40 ± 0.94	20.4 ± 0.3	17.91 ± 0.96	15.5 ± 0.3	17.4 ± 0.8	16.1 ± 0.7
4	20.90 ± 3.22	19.2 ± 3.7	20.54 ± 1.51	17.1 ± 0.9[a]	23.9 ± 2.7	16.5 ± 0.3[a]
5	20.29 ± 1.56	17.5 ± 0.9	21.17 ± 2.75	16.6 ± 2.2[a]	22.9 ± 0.8	16.2 ± 0.1[a]
6	19.00 ± 2.38	17.3 ± 3.4	19.23 ± 3.48	14.3 ± 0.2[a]	20.3 ± 0.6	15.9 ± 0.5[a]
7	20.84 ± 2.86	18.9 ± 0.6	22.69 ± 3.62	18.2 ± 0.7[a]	22.5 ± 0.9	15.4 ± 0.6[a]
8	19.16 ± 3.48	20.2 ± 1.7	20.89 ± 1.68	17.3 ± 1.4[a]	21.5 ± 0.7	15.5 ± 0.5[a]
9	21.90 ± 1.92	22.2 ± 2.3	21.77 ± 1.93	19.1 ± 2.1[a]	23.1 ± 0.5	15.1 ± 0.1[a]
10	20.51 ± 3.16	21.9 ± 2.0	20.17 ± 3.53	17.4 ± 0.9[a]	23.4 ± 0.7	17.1 ± 0.1[a]
11	21.83 ± 2.45	19.1 ± 1.5	20.51 ± 1.81	18.9 ± 0.8[a]	24.2 ± 0.3	15.6 ± 0.5[a]
12	21.93 ± 2.15	19.1 ± 0.2	21.34 ± 2.22	19.1 ± 0.5[a]	23.6 ± 0.6	15.6 ± 0.2[a]
13	20.20 ± 2.32	19.8 ± 0.7	21.34 ± 1.62	18.6 ± 0.2[a]	21.4 ± 1.1	18.2 ± 4.7[a]
14	-	-	-	-	25.6 ± 2.8	24.0 ± 0.2
15	-	-	-	-	21.8 ± 3.1	21.4 ± 3.7
16	-	-	-	-	21.3 ± 3.6	20.5 ± 3.5
17	-	-	-	-	22.2 ± 1.9	20.7 ± 4.5

values are expressed as mean ± SD (n=10)
[a] statistically different ($p<0.05$)

Table A6.4.2-4: Summary of weekly feed consumption in grams (females)

Week of study	Control	500 mg/kg bw/day	1000 mg/kg bw/day	2000 mg/kg bw/day	Satellite control	Satellite 2000 mg/kg bw/day
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
1	14.83 ± 1.49	14.21 ± 0.30	16.19 ± 1.22	14.50 ± 0.71	17.36 ± 0.51	14.50 ± 0.30
2	14.77 ± 0.56	14.00 ± 0.20	14.23 ± 1.16	13.93 ± 0.91	14.29 ± 0.40	15.29 ± 0.61
3	19.46 ± 0.38	15.79 ± 0.30	19.00 ± 0.54	16.71 ± 0.20	15.79 ± 0.30	13.43 ± 1.41
4	16.53 ± 3.67	16.36 ± 0.30	16.41 ± 1.34	18.50 ± 0.71	16.64 ± 1.72	15.21 ± 0.71
5	15.03 ± 1.40	14.36 ± 2.53	16.33 ± 1.60	16.07 ± 1.52	16.64 ± 0.30	15.29 ± 2.02
6	15.03 ± 1.30	17.07 ± 1.72	17.06 ± 1.90	20.43 ± 1.01	16.86 ± 0.40	16.36 ± 1.11
7	15.92 ± 1.72	18.93 ± 0.51	18.67 ± 1.26	16.86 ± 2.83	18.07 ± 2.32	16.29 ± 1.62
8	15.07 ± 1.07	17.00 ± 1.82	17.27 ± 1.68	17.86 ± 0.61	18.79 ± 1.52	16.71 ± 0.20
9	17.63 ± 1.41	18.43 ± 2.02	18.19 ± 0.92	19.07 ± 0.10	20.14 ± 0.81	18.14 ± 0.20
10	15.23 ± 1.31	20.57 ± 2.83	15.99 ± 3.06	16.57 ± 1.62	17.71 ± 1.82	14.71 ± 0.81
11	15.02 ± 1.45	15.93 ± 1.92	15.80 ± 2.21	16.57 ± 0.00	15.57 ± 1.41	15.07 ± 1.72
12	15.13 ± 1.09	16.43 ± 2.02	17.33 ± 2.36	20.86 ± 2.22	18.36 ± 0.91	14.79 ± 0.91
13	14.90 ± 1.10	19.00 ± 0.47	17.29 ± 1.37	18.14 ± 0.61	16.50 ± 1.31	17.14 ± 4.04
14	-	-	-	-	13.50 ± 2.12	12.86 ± 1.41
15	-	-	-	-	16.43 ± 2.02	13.50 ± 1.31
16	-	-	-	-	14.36 ± 0.51	13.07 ± 0.30
17	-	-	-	-	13.50 ± 2.12	14.75 ± 1.77

values are expressed as mean ± SD (n=10)

Table A6.4.2-5: Summary of urine phosphorus measurements (in mg/dl) – Male rats

Week of study	Control	500 mg/kg bw/day	1000 mg/kg bw/day	2000 mg/kg bw/day	Satellite control	Satellite 2000 mg/kg bw/day
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
Phosphorus on day 91	6.4 ± 0.9	6.1 ± 0.5	4.2 ± 0.7[a]	3.7 ± 0.4[a]	-	-
Phosphorus on day 92	-	-	-	-	4.6 ± 1.5	5.4 ± 0.7
Phosphorus on day 119	-	-	-	-	5.5 ± 0.9	4.6 ± 0.7

values are expressed as mean ± SD
[a]statistically different ($p<0.05$)

Table A6.4.2-6: Summary of urine phosphorus measurements (in mg/dl) - Female rats

Week of study	Control	500 mg/kg bw/day	1000 mg/kg bw/day	2000 mg/kg bw/day	Satellite control	Satellite 2000 mg/kg bw/day
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
Phosphorus on day 91	6.2 ± 0.5	5.9 ± 0.4	4.1 ± 0.6[a]	3.9 ± 0.7[a]	-	-
Phosphorus on day 92	-	-	-	-	4.7 ± 0.8	5.2 ± 0.9
Phosphorus on day 119	-	-	-	-	4.2 ± 0.8	4.2 ± 1.0

values are expressed as mean ± SD
[a] statistically different ($p<0.05$)

Table A6.4.2-7: Summary of absolute organ weight (g) – Male rats

Group/Dose	Liver	Kidney left	Kidney right	Gonad left	Gonad right
Control	15.303 ± 0.557	1.600 ± 0.049	1.617 ± 0.054	1.662 ± 0.035	1.673 ± 0.032
500 mg/kg bw	17.851 ± 1.157	1.683 ± 0.091	1.594 ± 0.080	1.615 ± 0.051	1.665 ± 0.089
1000 mg/kg bw	17.448 ± 0.533	1.609 ± 0.050	1.652 ± 0.050	1.598 ± 0.057	1.595 ± 0.043
2000 mg/kg bw	17.892 ± 0.673	1.702 ± 0.087	1.704 ± 0.070	1.601 ± 0.049	1.601 ± 0.057
Satellite control	17.174 ± 0.713	1.852 ± 0.065	1.860 ± 0.058	1.649 ± 0.104	1.499 ± 0.170
Satellite group 0.4363 mg/L	14.829 ± 0.588[a]	1.699 ± 0.073	1.627 ± 0.061[a]	1.640 ± 0.037	1.614 ± 0.069

[a] Statistically different from control ($p < 0.05$)

Table A6.4.2-8: Summary of absolute organ weight (g) – Female rats

Group/Dose	Liver	Kidney left	Kidney right	Gonad left	Gonad right
Control	8.986 ± 0.359	0.981 ± 0.031	1.034 ± 0.036	0.073 ± 0.002	0.075 ± 0.003
500 mg/kg bw	9.846 ± 0.194	0.976 ± 0.035	0.979 ± 0.025	0.068 ± 0.003	0.071 ± 0.002
1000 mg/kg bw	11.379 ± 0.337[a]	1.092 ± 0.031[a]	1.115 ± 0.043	0.061 ± 0.002	0.065 ± 0.002[a]
2000 mg/kg bw	12.862 ± 0.514[a]	1.105 ± 0.038[a]	1.108 ± 0.042	0.063 ± 0.002	0.065 ± 0.001[a]
Satellite control	8.655 ± 0.262	1.111 ± 0.035	1.130 ± 0.042	0.086 ± 0.002	0.085 ± 0.002
Satellite group 0.4363 mg/L	8.467 ± 0.291	1.095 ± 0.026	1.099 ± 0.011	0.080 ± 0.002	0.076 ± 0.003[a]

[a] Statistically different from control ($p < 0.05$)

Table A6.4.2-9: Summary of relative organ weight (%) – Male rats

Group/Dose	Liver	Kidney left	Kidney right	Gonad left	Gonad right
Control	3.701 ± 0.150	0.387 ± 0.013	0.391 ± 0.015	0.403 ± 0.011	0.405 ± 0.011
500 mg/kg bw	4.169 ± 0.131[a]	0.394 ± 0.008	0.375 ± 0.013	0.385 ± 0.018	0.393 ± 0.017
1000 mg/kg bw	4.247 ± 0.192[a]	0.390 ± 0.012	0.400 ± 0.010	0.389 ± 0.019	0.387 ± 0.013
2000 mg/kg bw	4.630 ± 0.094[a]	0.440 ± 0.016[a]	0.441 ± 0.011[a]	0.416 ± 0.011	0.416 ± 0.014
Satellite control	3.526 ± 0.133	0.380 ± 0.012	0.382 ± 0.012	0.338 ± 0.021	0.308 ± 0.035
Satellite group 0.4363 mg/L	3.442 ± 0.174	0.392 ± 0.020	0.374 ± 0.017	0.370 ± 0.010	0.364± 0.016

[a] Statistically different from control ($p < 0.05$)

Table A6.4.2-10: Summary of relative organ weight (%) – Female rats

Group/Dose	Liver	Kidney left	Kidney right	Gonad left	Gonad right
Control	3.609 ± 0.156	0.396 ± 0.021	0.417 ± 0.022	0.029 ± 0.001	0.030 ± 0.001
500 mg/kg bw	3.946 ± 0.077	0.391 ± 0.014	0.393 ± 0.011	0.027 ± 0.001	0.028 ± 0.001
1000 mg/kg bw	4.619 ± 0.129[a]	0.444 ± 0.016	0.454 ± 0.023	0.025 ± 0.001	0.026 ± 0.001[a]
2000 mg/kg bw	5.082 ± 0.205[a]	0.439 ± 0.022	0.440 ± 0.024	0.025 ± 0.000	0.026 ± 0.001[a]
Satellite control	3.300 ± 0.123	0.422 ± 0.012	0.430 ± 0.014	0.033 ± 0.001	0.032 ± 0.001
Satellite group 0.4363 mg/L	3.545 ± 0.183	0.461 ± 0.027	0.465 ± 0.029	0.034 ± 0.002	0.031 ± 0.001

[a] Statistically different from control ($p < 0.05$)

Section A6.4.3 **Subchronic toxicity**
Annex Point IIA6.4 **90-day inhalation toxicity study in the rat**

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19 Reference

19.1	**Reference**	[redacted] (2006), Subchronic inhalation toxicity study of Permethrin technical in Wistar rats, [redacted] unpublished report no.: 14995
		Dates of experimental work: August 8, 2005 –December 5, 2005
19.2	**Data protection**	Yes
19.2.1	Data owner	Tagros Chemicals India Ltd.
19.2.2	Companies with letter of access	Not applicable
19.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing a.s for the purpose of its entry into Annex I/IA.

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20 Guidelines and Quality Assurance

20.1	**Guideline study**	Yes, the study was carried out according to OECD Guideline 413.
20.2	**GLP**	Yes
20.3	**Deviations**	Yes with the following deviation: X
		Data on haematological and clinical chemistry parameters are not available as recommended in the guideline.
		This deviation is not considered to compromise the scientific validity of the study.

21 MATERIALS AND MethodS

21.1	**Test material**	As given in section 2 (Permethrin 25:75)
21.1.1	Lot/Batch number	P-41, P-13, P-17 and P-26
21.1.2	Specification	As given in section 2 (Permethrin 25:75)

Section A6.4.3 Subchronic toxicity

Annex Point IIA6.4 **90-day inhalation toxicity study in the rat**

21.1.2.1	Description	Light yellow to brown liquid
21.1.2.2	Purity	94.04%, 93.1%, 92.29% and 93.61%
21.1.2.3	Stability	The test material was considered to be stable throughout the study.
21.2	**Test Animals**	
21.2.1	Species	Rat
21.2.2	Strain	Wistar
21.2.3	Source	[redacted]
21.2.4	Sex	Male and female
21.2.5	Age/weight at study initiation	Not documented. Males/Females: 80-90g
21.2.6	Number of animals per group	6 groups of 10 animals/sex/group
21.2.7	Control animals	Yes, distilled water
21.3	**Administration/ Exposure**	Inhalation
21.3.1	Duration of treatment	13 weeks
21.3.2	Frequency of exposure	5 days per week (6 hours per day)
21.3.3	Post exposure period	28 days for a satellite control group and high dose level satellite group
21.3.4	**Inhalation**	

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Section A6.4.3 Subchronic toxicity

Annex Point IIA6.4 **90-day inhalation toxicity study in the rat**

21.3.4.1	Concentrations	Actual concentration: Analytical concentration:	0, 0.1149, 0.2201 and 0.4363 mg/L 13.84-14.11, 27.69-28.21 and 46.15-47.02 mg/L
21.3.4.2	Particle size	Particles were below 10 µm. During exposure period, At 0.1149 mg/L, 75.86 to 99.08% of the particles generated were < 5.8 µm. At 0.2201 mg/L, 48.98 to 97.89% of the particles generated < 5.8 µm. At 0.4363 mg/L, 43.39 to 82.17% of the particles generated < 5.8 µm.	
21.3.4.3	Type or preparation of particles	A nebuliser was used to generate respirable particles. The test substance from the infusion syringe pump or peristaltic pump was delivered through a cannula into the nebuliser. The rates of introduction of Permethrin were set at 0.15, 0.3 and 0.5 mL/min. Filter air was passed through the chamber at a rate of 10L/min.	
21.3.4.4	Type of exposure	Head only inhalation	
21.3.4.5	Vehicle	Not applicable (administered in air)	
21.3.4.6	Concentration in vehicle	Not relevant	
21.3.4.7	Duration of exposure	6 hours per day	
21.3.4.8	Controls	Exposed to distilled water	
21.4	**Examinations**		
21.4.1	Observations		
21.4.1.1	Clinical signs	Yes, once daily	
21.4.1.2	Mortality	Yes, once daily	
21.4.2	Body weight	Yes, prior to study initiation and weekly thereafter until sacrifice	
21.4.3	Food consumption	Yes, weekly during the study	
21.4.4	Water consumption	Not measured	
21.4.5	Ophthalmoscopic examination	Yes, prior to study initiation and termination in the control and high dose groups	
21.4.6	Haematology	Yes Number of animals: all animals at 0, 0.1149, 0.2201 and 0.4363 mg/L	

Section A6.4.3 Subchronic toxicity
Annex Point IIA6.4 **90-day inhalation toxicity study in the rat**

		Time points: at study termination (on days 91 and 92) Parameters: red blood cells (RBC), white blood cells (WBC), haemoglobin (Hb), haematocrit (PCV), platelet count, differential leucocyte count (DC), prothrombin time and clotting time. Number of animals: all animals in the control satellite group and high dose satellite group Time points: at study termination (on day 119) Parameters: red blood cells (RBC), white blood cells (WBC), haemoglobin (Hb), haematocrit (PCV), platelet count, differential leucocyte count (DC), prothrombin time and clotting time.
21.4.7	Clinical chemistry	Yes Number of animals: all animals at 0, 0.1149, 0.2201 and 0.4363 mg/L Time points: at study termination (on days 91 and 92) Parameters: albumin, protein, alanine aminotransferase, aspartate aminotansferase, gamma glutamyl transpeptidase, creatinine, total bilirubin, alanine phosphatase, phosphorus, chloride, glucose, blood urea nitrogen, calcium, sodium and potassium Number of animals: all animals in the control satellite group and high dose satellite group Time points: at study termination (on day 119) Parameters: albumin, protein, alanine aminotransferase, aspartate aminotansferase, gamma glutamyl transpeptidase, creatinine, total bilirubin, alanine phosphatase, phosphorus, chloride, glucose, blood urea nitrogen, calcium, sodium and potassium
21.4.8	Urinalysis	Not required
21.5	**Sacrifice and pathology**	
21.5.1	Organ weights	Yes All animals that were found dead or moribund and at the terminal sacrifice (scheduled on days 91, 92 and 119) Organs: liver, adrenals, kidneys, gonads and lungs
21.5.2	Gross and histopathology	Yes All animals were subjected to a gross necropsy The following organs and tissues were preserved: All gross lesions, lungs, brain (including sections of medulla/pons), cerebellar cortex, liver, spleen, kidneys, adrenals, pancreas, gonads, uterus, accessory genital organs, oesophagus, stomach, duodenum, jejunum, ileum, caecum, colon, rectum, urinary bladder, representative lymph node, peripheral nerve, sternum with bone marrow.

Section A6.4.3 **Subchronic toxicity**
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		All gross lesions and lungs were examined in all animals. Full histopathology was carried out on the trachea and other organs and tissues of all animals in the control and high dose groups.
21.5.3	Other examination	None
21.5.4	Statistics	Body weight, body weight change, feed consumption, haematology and clinical chemistry parameters of rats belonging to the control and experimental groups were tested for normality. Normal data was subjected to one-way ANOVA. Post ANOVA comparison is carried out using Newman-Keul`s Test. Non-Normal data was subjected to Kruskal-Wallis one-way ANOVA on Ranks. Post ANOVA comparison is carried out using Kruskal-Wallis Z Test.

22 Results and Discussion

22.1	**Observations**	
22.1.1	Clinical signs	Clinical signs such as nasal irritation and mild tremor were noted on day 7 at 0.4363 mg/L and in the high dose satellite group.
22.1.2	Mortality	No mortality was observed throughout the study.
22.2	**Body weight gain**	No treatment related changes were observed. The following changes were noted but not deemed to be treatment related: Statistically significant decrease in absolute body weights and body weights gains for both males and females at 0.04363 mg/L and in the high dose satellite group on week 2.
22.3	**Food consumption and compound intake**	No changes were observed in any groups.
22.4	**Ophtalmoscopic examination**	No treatment related ocular effects were observed.
22.5	**Blood analysis**	
22.5.1	Haematology	No treatment related changes in haematological parameters were observed in any groups.
22.5.2	Clinical chemistry	No treatment related changes in clinical chemistry values in any groups.

Section A6.4.3 **Subchronic toxicity**
Annex Point IIA6.4 **90-day inhalation toxicity study in the rat**

22.5.3	Urinalysis	Not carried out
22.6	**Sacrifice and pathology**	
22.6.1	Organ weights	No treatment related organ weight changes were noted The following changes were noted but not deemed to be treatment related: Males: Increase in absolute adrenal weight at 0.4363 mg/L and in the high dose satellite group Increase in relative liver weight at 0.1149 mg/L Increase in relative kidney weight at 0.2201 and 0.4363 mg/L Increase in relative adrenal weight at 0.1149 and 0.2201 mg/L Increase in relative gonad weight at 0.2201 and 0.4363 mg/L Females: Increase in absolute liver weight at 0.1149 and 0.2201 mg/L Increase in absolute kidney weight at 0.2201, 0.4363 mg/L and in the high dose satellite group Increase in absolute adrenal weight at 0.1149, 0.2201 and 0.4363 mg/L and in the high dose satellite group Increase in relative liver weight at 0.1149 and 0.2201 mg/L Increase in relative adrenal weights at 0.1149, 0.2201 and 0.4363 mg/L and in the high dose satellite group Increase in relative kidney weights at 0.1149, 0.2201 and 0.4363 mg/L Increase in relative gonads weights at 0.1149 mg/L Increase in relative lungs weights at 0.1149 and 0.2201 mg/L
22.6.2	Gross pathology and histopathology	No treatment related findings were observed at necropsy. No treatment related histological findings were noted.

23 Applicant's Summary and conclusion

23.1 Materials and methods

Four groups of 10 animals/sex were exposed to Permethrin at aerosol levels of 0, 0.1149, 0.2201 and 0.4363 mg/L, 6 hours per day, 5 days a week during 13 weeks. Two additional groups of 10 animals/sex used as satellite at 0 and 0.4363 mg/L were treated during 13 weeks and were then observed during 28 days for delayed occurence toxic effects.

This study was conducted according to OECD Guideline 413 and is described in section 3 with the following deviation:

Data on haematological and clinical chemistry parameters are