(1-L-04)

DATA EVALUATION RECORD

Polyxylenol tetrasulfide (PXTS)
MRID 46062615

Reproductive/developmental oral toxicity screening test of PXTS in rats
OPPTS 870.3550

Prepared for

Antimicrobial Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

ICF Consulting
9300 Lee Highway
Fairfax, VA 22031

Under Subcontract to

Versar
6850 Versar Center
P.O. Box 1549
Springfield, VA 22151

Principal Reviewer	*Katherine Sullivan, M.S.*	Date 1/5/04
Independent Reviewer	*Jennifer Wetham, M.S.*	Date 1/9/04
ICF Program Manager	*Kara Altshuler, Ph.D.*	Date 1/9/04
Versar Program Manager	*Linda Phillips, Ph.D.*	Date 1/12/04

Contract Number: 68-W-01-036
Work Assignment No.: 0248.3000.002.02 TAF 2-2-21
EPA WAM: Killian Swift, Ph.D.

This review may be altered by EPA subsequent to the contractors' signatures above.

1

EPA Reviewer: Tim McMahon, Ph.D.
Senior Toxicologist, AntimicrobialsDivision

Signature
Date 7/11/03

Template version 11/01

DATA EVALUATION RECORD

STUDY TYPE: Reproduction/Developmental Toxicity Screening Test - Rat OPPTS 870.3550 ; OECD 421.

PC CODE: 006929 **DP BARCODE:** D299112

TEST MATERIAL (PURITY): Polyxylenol tetrasulfide (100% a.i.)

SYNONYMS: PXTS

CITATION: Faqi, A.S. (2002) Reproductive/developmental oral toxicity screening test of PXTS in rats. IIT Research Institute (IITRI) Chicago, IL. Laboratory project ID 1636 SN4, November 2002. MRID 46062615. Unpublished.

SPONSOR: Akzo Nobel Functional Chemicals LLC
5 Livingstone Ave.
Dobbs Ferry, NY 10522

EXECUTIVE SUMMARY:

In a reproduction/developmental screening toxicity study (MRID 46062615), polyxylenol tetrasulfide (PXTS) (Lot/batch #1685-26; 100% a.i.) was administered to 11 Sprague-Dawley CD® rats/sex/dose by gavage at dose levels of 0, 125, 250, or 500 mg/kg bw/day. After the quarantine period and group assignment, male and female rats were dosed for a minimum of two weeks prior to mating and during the mating period. Males were dosed for approximately two weeks post-mating up to and including the day before scheduled sacrifice. Females were dosed over the duration of pregnancy and at least four days after delivery.

Treatment-related clinical observations were noted in 500-mg/kg/day parental animals and consisted of salivation, discoloration around the mouth, redness around nose fur, rough coat, discolored paws, coldness to the touch, and hunched posture. Treatment-related observations at necropsy consisted of enlarged spleens, which were observed in 125-mg/kg/day males, 250-mg/kg/day males and females, and 500-mg/kg/day males and females, and pigmentation of the spleen, which was observed in both sexes at all dose levels. **The parental systemic LOAEL is 125 mg/kg bw/day, based on pigmentation of the spleen (which is believed to be an indication of hematotoxicity). A parental systemic NOAEL cannot be determined.**

2

Fetal toxicity was limited to the mid- and high-dose groups. Significantly decreased pup weight was noted on postnatal day 0 and postnatal days 0 and 4 for the 250- and 500-mg/kg/day groups, respectively. At 500 mg/kg/day, a significant increase in the mean number of pup deaths per litter and a significant decrease in pup survival on postnatal day 4 also were observed. **The offspring LOAEL is 250 mg/kg bw/day, based on decreased pup weight. The offspring NOAEL is 125 mg/kg bw/day.**

There was no reproductive toxicity observed at any dose group. **Therefore, the reproductive NOAEL is 500 mg/kg bw/day in males and females. A reproductive LOAEL could not be determined.**

This study is classified as **Acceptable-Guideline** and satisfies the guideline requirement for a reproduction/developmental toxicity screening test (OPPTS 870.3550; OECD 421) in rats.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements are provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: PXTS

Description:	Black solid
Lot/Batch #:	1685-26
Purity:	100% a.i.
Compound Stability:	Stable at room temperature
CAS # of TGAI:	Not provided
	The structure is provided in Appendix B (page 65) of the study report

2. Vehicle and/or positive control: The vehicle was tricaprylin ($C_{27}H_{50}O_6$; Lot # 071K1667 and 022K1667; purity >99% a.i.), a clear, colorless to light yellow liquid.

3. Test animals:

Species:	Rat	
Strain:	Sprague-Dawley CD® [CRL:CD(SD)IGSBR]	
Age at study initiation:	Females: 8 weeks old upon receipt Males: 9 weeks old upon receipt	
Wt. at study initiation:	Females: random sample (5 rats) weighed 184-205 g the day after receipt Males: random sample (5 rats) weighed 272-303 g the day after receipt	
Source:	Charles River Laboratories, Raleigh, NC	
Housing:	Individually-housed (except prior to quarantine and during mating) in suspended polycarbonate cages equipped with automatic watering systems and hardwood bedding (male and non-mated female cages) or corncob bedding and shredded paper towel (mated female rat cages). Animals were housed 2 per cage upon receipt.	
Diet:	Certified Rodent Diet #5002 (PMI Nutrition International, Inc., Brentwood, MO) provided *ad libitum*. The diet reportedly did not contain levels of contaminants that would interfere with the study.	
Water:	Tap water (City of Chicago) provided *ad libitum*. The water reportedly did not contain levels of contaminants that would interfere with the study.	
Environmental conditions:	**Temperature:**	20-25°C
	Humidity:	47-70 %
	Air changes:	Not provided
	Photoperiod:	12 hrs dark/ 12 hrs light
Acclimation period:	9 days	

B. PROCEDURES AND STUDY DESIGN

1. Mating procedure: During the mating period, one male was caged with one female from the same test group until sperm cells were observed in vaginal smears. The day on which a sperm-positive smear was collected was considered gestation day (gd) 0. Females were allowed to undergo natural parturition.

2. Study schedule: After the quarantine period and group assignment, male rats were dosed for a minimum of four weeks (minimum of two weeks prior to mating, during the mating period, and approximately two weeks post-mating). Female rats were dosed throughout the study (two weeks

prior to mating, variable time to conception, the duration of pregnancy, and at least four days after delivery).

3. **Animal assignment:** Three days prior to dosing, animals were assigned to test groups using a computerized randomization procedure based on body weights as seen in Table 1. Each rat was identified by a number on its tail.

TABLE 1: Animal Assignment

Test Group	Dose mg/kg	Animals/group	
		Males	Females
Control	0	11	11
Low (LDT)	125	11	11
Mid (MDT)	250	11	11
High (HDT)	500	11	11

4. **Dose selection rationale:** According to the study protocol, the doses selected were chosen on the basis of a range-finding study. No further details are provided.

5. **Dosage preparation and analysis:** Dosing formulations at concentrations of 41.7, 83.3, and 166.7 mg/mL (to deliver 125, 250, and 500 mg/kg, respectively) were prepared weekly by mixing appropriate amounts of heated test substance (approximately 90°C) with tricaprylin. Dilution to final volume was performed after the dosing formulations were cooled and stirred; dosing formulations were then placed in amber glass jars and stored at room temperature until use. Homogeneity and stability of the dose formulations were determined during a previous study (ITRI Project No. 1636 Study No. 1). Two samples (~2 mL) were collected from each dose level on 5/2/2002 and 5/9/2002 to evaluate achieved concentration; samples were analyzed the day following collection.

Results

Homogeneity Analysis: Results from homogeneity analyses performed for a previous study are not provided. Although no further details are provided in the study report, review of a developmental toxicity study in rabbits (MRID 46062616) indicates that formulations of PXTS in tricaprylin were determined to be homogeneous at doses up to 62.5 mg/kg. .

Stability Analysis: Results from stability analyses performed for a previous study are not provided. Although no further details are provided in the study report, review of a developmental toxicity study in rabbits (MRID 46062616) indicates that formulations of PXTS in tricaprylin up to 62.5 mg/kg dose level were determined to be stable for up to 14 days.

The registrant should provide justification for stability and homogeneity of PXTS in the dosing solutions at the dose levels used in this study.

Concentration Analysis: The measured concentration of dose formulations ranged from 101% to 106% of the nominal concentration.

The analytical data indicates that the variance between nominal and actual dosage to the study animals is acceptable.

6. **Dosage administration:** All doses were administered once daily by gavage at a volume of 3.0 mL/kg body weight (bw)/day. Dosing was based on the most recent body weight determination.

C. OBSERVATIONS

1. **Parental animals:** The parental rats were observed at least once daily during the quarantine period for moribundity and mortality. Complete physical examinations were performed prior to treatment initiation and at termination. During treatment, the rats were observed twice daily, at least 4 hours apart, for moribundity and mortality. Clinical signs were monitored daily 3-6 hours after dosing. Body weights were measured the day after receipt, at randomization, on the first day of dosing (day 1), at least weekly thereafter, and at termination. Females were not weighed during mating, but female body weights were recorded on gd 0, 7, 14, and 20, as well as on postnatal days 0 and 4. During the treatment period, food consumption was recorded on the same days that body weights were measured.

2. **Litter observations:** Litter observations were made on postnatal days 0 and 4 as shown in Table 2. In addition, the number of runts was determined as soon as possible after delivery.

TABLE 2: Litter Observations [a]

Observation	Time of observation (lactation day)	
	Day 0	Day 4
Number of live pups	X	X
Pup weight	X	X
Litter weight	X	X
Gross abnormalities/lesions	X	X
Number of dead pups	X	X
Sex of each pup (M/F)	X	X

a Data obtained from pages 13 and 45-49 in the study report.

3. **Postmortem observations**:

a) Parental animals: All surviving parental males were sacrificed approximately two weeks after mating. Parental females were sacrificed on postnatal day 4. These animals were subjected to a limited necropsy.

The following tissues (X) were prepared for microscopic examination and weighed (XX).

XX	Ovaries	XX	Testes
	Uterus	XX	Epididymides
	Vagina/cervix		Prostate
X	Lesions		Seminal vesicles

Tissues were fixed in 10% neutral buffered formalin (ovaries and lesions) or Bouin's fixative (testes and epididymides). A detailed histopathology examination (with special emphasis placed on the stages of spermatogenesis and histopathology of interstitial cell structure) was performed on the ovaries, testes, and epididymides of the control and high-dose rats.

b) Offspring: On day 4 postpartum, all surviving pups were sacrificed; necropsy evaluations were not performed.

D. DATA ANALYSIS

1. Statistical analyses: Statistical analysis was performed using Systat software. For all comparisons, the minimum significance level was $p \leq 0.05$. Analysis of variance (ANOVA) followed, where appropriate, by the *post-hoc* Dunnett's test, was used to assess parental body weight, parental body weight gain, food consumption, fertility index, reproductive organ weights, pup body weight, and litter survival. Mating and pregnancy indices were evaluated using the Chi-Square test. The analyses used are considered appropriate.

2. **Indices**:

Reproductive indices: The following reproductive indices were calculated and/or defined from breeding and parturition records of animals in the study:

- Mating index: (number of females made sperm-positive/ number of males mated) X 100
- Pregnancy index: (number of males that made females pregnant/ number of males that made females sperm-positive) X 100
- Fertility index: number of days elapsed until the male had fertilized its female partner

Offspring viability indices: No offspring viability indices were calculated.

3. **Historical control data**: Historical control data are not provided.

II. RESULTS

A. PARENTAL ANIMALS

1. **Mortality and clinical signs:** There were no treatment-related mortalities. One control female was found dead on gd 23. No other females or males died prior to scheduled sacrifice. Treatment-related clinical signs were observed only in high-dose males and/or females. These included salivation (3/11 females and 2/11 males), discoloration around the mouth (4/11 females and 2/11 males), redness around nose fur (2/11 males), rough coat (1/11 males), discolored paws (1/11 males), coldness to the touch (1/11 males), and hunched posture (1/11 males). Discoloration around the mouth and paws was attributed to the dark color of the test article. Other isolated clinical signs observed in the vehicle control, 125-, and/or 250-mg/kg/day dose groups are considered incidental and not related to treatment. Treatment-related clinical signs are summarized in Table 3.

TABLE 3. Mortality and Clinical Signs [a]

Observation	Dose Group(mg/kg/day)			
	0	125	250	500
Males				
Animals found dead	0	0	0	0
Salivation	0	0	0	2
Discoloration around the mouth	0	0	0	2
Redness around the nose	0	0	0	2
Rough coat	0	0	0	1
Discolored paws	0	0	0	1
Cold to the touch	0	0	0	1
Hunched posture	0	0	0	1
Females				
Animals found dead	1	0	0	0
Salivation	0	0	0	3
Discoloration around the mouth	0	0	0	4
Redness around the nose	0	0	0	0
Rough coat	0	0	0	0
Discolored paws	0	0	0	0

Observation	Dose Group(mg/kg/day)			
	0	125	250	500
Cold to the touch	0	0	0	0
Hunched posture	0	0	0	0

a Data obtained from pages 21, 25, 30, and 34 in the study report.

2. **Body weight and food consumption:** Body weight was significantly decreased in high-dose males on study days 8 and 15. Mean body weight gain during study days 1-8 and mean total body weight gain also were significantly reduced at this dose level when compared to the corresponding control group. There were no significant changes in mean body weight or mean body weight gain for the females in any of the treatment groups during the pre-mating, gestation, or lactation periods.

Food consumption was significantly decreased for high-dose male and female rats during study days 1-8 compared to the control. There were no changes in group food consumption for any of the female treatment groups throughout the gestation and lactation periods.

Reported body weight and food consumption results are summarized in Tables 4a, 4b, 4c, and 4d.

TABLE 4a. Pre-mating Male Mean (±SD) Body Weight and Food Consumption [a]

Observations/study day	Dose Group (mg/kg/day)			
	0	125	250	500
Mean body weight (g)				
Day 1	362±14.9	360±12.1	363±13.7	362±12.8
Day 8	384±22.3	383±16.1	375±18.6	360*±19.7
Day 15	410±31.5	405±18.9	395±23.6	379*±24.8
Day 22	423±34.7	416±25.9	412±18.8	393±30.5
Day 29	453±44.4	444±24.0	439±26.6	422±31.1
Day 36	472±52.5	458±26.3	457±23.5	436±28.8
Mean weight gain (g)				
Days 1-8	22±10.3	23±6.0	12±10.9	-1**±11.8
Days 8-15	26±11.2	22±6.6	20±9.3	19±15.9
Days 15-22	13±7.5	10±13.8	18±8.8	13±19.2
Days 22-29	30±13.0	29±8.1	27±14.7	30±12.4
Days 29-36	20±9.9	13±7.3	18±9.5	14±6.3
Total gain (Days 1-36)	110±42.2	98±18.3	95±15.1	75**±20.0

Observations/study day	Dose Group (mg/kg/day)			
	0	125	250	500
Mean food consumption (g/animal/day)				
Days 1-8	136±14.4	154±19.2	143±13.5	108**±24.7
Days 8-15	157±16.4	151±10.8	150±14.5	147±18.4
Days 15-29	164±16.5	157±13.4	160±21.1	161±18.4
Days 29-36	167±21.2	168±28.9	158±12.6	150±16.2

a Data obtained from pages 35-37 in the study report.
* Statistically different from control, $p<0.05$.
** Statistically different from control, $p<0.01$.

TABLE 4b. Pre-mating Female Mean (±SD) Body Weight and Food Consumption [a]

Observations/study day	Dose Group (mg/kg/day)			
	0	125	250	500
Mean body weight (g)				
Day 1	227±8.5	227±10.3	228±11.1	226±9.7
Day 8	233±13.4	234±10.9	230±16.4	227±11.8
Day 15	245±13.5	244±9.6	237±14.7	241±13.2
Mean weight gain (g)				
Days 1-8	7±7.5	7±7.2	2±9.8	1±5.4
Days 8-15	12±3.7	10±9.9	8±6.6	14±4.4
Total gain (Days 1-15)	18±8.6	17±10.4	9±8.2	15±7.8
Mean food consumption (g/animal/day)				
Days 1-8	106±11.5	113±16.3	111±25.8	88*±13.5
Days 8-15	114±8.4	117±15.6	117±15.9	114±23.4

a Data obtained from pages 22-24 in the study report.
* Statistically different from control, $p<0.05$.

TABLE 4c. Female Mean (±SD) Body Weight and Food Consumption during Gestation [a]

Observations/study day	Dose Group (mg/kg/day)			
	0	125	250	500
Mean body weight (g)				
Day 0	244±12.2	248±12.1	243±18.2	247±17.5
Day 7	279±16.0	281±10.1	277±17.7	277±19.3

Observations/study day	Dose Group (mg/kg/day)			
	0	125	250	500
Day 14	310±21.7	315±11.3	308± 22.0	306±27.3
Day 20	386±31.8	390±17.5	373±30.9	372±31.8
Mean weight gain (g)				
Days 0-7	35±6.3	33±5.8	35±6.8	30±8.6
Days 7-14	32±7.2	34±8.8	31±9.6	29±11.9
Days 14-20	75±11.2	75±9.2	64±13.1	66±8.9
Total gain (Days 0-20)	142±21.3	142±17.1	130±18.6	125±18.3
Mean food consumption (g/animal/day)				
Days 0-7	129±12.2	133±9.0	133±14.9	121±14.4
Days 7-14	140±15.2	144±13.1	138±18.0	129±17.5
Days 14-20	131±16.4	133±10.6	127±14.7	125±10.8

a Data obtained from pages 26-28 in the study report.

TABLE 4d. Female Mean (±SD) Body Weight and Food Consumption during Lactation [a]

Observations/study day	Dose Group (mg/kg/day)			
	0	125	250	500
Mean body weight (g)				
Day 0	291±25.5	296±16.1	283±25.0	285±20.7
Day 4	309±20.7	314±14.9	295±22.3	303±24.5
Mean weight gain (g)				
Days 0-4	18±14.7	19±11.8	11±19.6	18±13.5
Mean food consumption (g/animal/day)				
Days 0-4	125±21.4	133±18.7	115±20.5	127±10.5

a Data obtained from pages 31-33 in the study report.

3. **Reproductive performance**: There were no treatment-related effects on mating, fertility, or reproduction. All mated dams found to be sperm-positive became pregnant, except for one sperm-positive female in the 500-mg/kg/day group. Results for the parental animals are summarized from the report in Table 5.

TABLE 5. Reproductive Performance [a]

Observation	Dose Group (mg/kg/day)			
	0	125	250	500
Mean (±SD) precoital interval (days)	2±1.1	2±1.2	2±1.3	2±1.7
MALES				
Number mated	11	11	11	11
Number fertile	11	11	11	11
Fertility not determined	0	0	0	0
Intercurrent deaths	0	0	0	0
FEMALES				
Number mated	11	11	11	11
Number fertile	11	11	11	10
Fertility not determined	0	0	0	1
Mating index	100	100	100	100
Pregnancy index	100	100	100	91
Intercurrent deaths	1	0	0	0
Mean gestation interval (days)	22	22	22	22

a Data obtained from pages 29 and 38 in the study report.

4. Parental postmortem results:

a) Organ weights: There were no treatment-related effects on the reproductive organ weights.

b) Pathology

1) Macroscopic examination: A summary of macroscopic findings is presented below in Table 6. Treatment-related gross necropsy observations were limited to the spleen. The spleens of 9 male and 4 female low-dose rats, 11 male and 11 female mid-dose rats, and 10 male and 11 female high-dose rats were darkly pigmented. In the mid- and high-dose groups, all of the darkly pigmented spleens also were enlarged. Only 3/9 low-dose males that exhibited pigmentation also had enlarged spleens; there were no incidences of enlarged spleens in low-dose females. The spleen of 1 high-dose male also had a rough surface, but this finding is considered to be incidental.

TABLE 6. Gross Necropsy Observations [a]

Observation	Dose Group (mg/kg/day)			
	0	125	250	500
Males				
No gross lesions	11	2	0	0
Spleen, parenchyma				
-Pigmentation	0	9	11	10
-Enlarged	0	3	11	10
- Rough surface	0	0	0	1
Females				
No gross lesions	11	7	0	0
Spleen, parenchyma				
-Pigmentation	0	4	11	11
-Enlarged	0	0	11	11
- Rough surface	0	0	0	0

a Data obtained from page 43 in the study report.

2) **Microscopic examination:** There were no treatment-related microscopic findings. Findings that were considered by the pathologist to be common or incidental included follicular cysts (found in 1 control female), multifocal tubules with retained step 19 spermatids with an intensity scale of minimal (found in the testes of 2 high-dose males), and lymphatic infiltrates in the interstitial tissue of the epididymis (observed in 3 control rats and 5 high-dose rats). All ovaries of female rats were considered by the pathologist to have been within normal limits.

B. OFFSPRING

1. **Viability and clinical signs:** There was no significant difference in the mean number of pups per litter in any of the treatment groups compared to the control group. Compared to the control, the mean number of pup deaths per litter on lactation day 4 was significantly increased and the mean percent survival on lactation day 4 was significantly decreased in the high-dose group. These differences are indicative of test substance-induced fetal toxicity. Litter parameters from pups during lactation is summarized from the report in Table 7.

TABLE 7. Litter parameters [a]

Observation		Dose Group (mg/kg)			
		0	125	250	500
Number born live		140	164	150	142
Number born dead		0	6	4	3
Sex Ratio Day 0 (% ♂) [b]		45	51	55	52
# Deaths Days 0-4		3	10	7	21
Mean litter size	Day 0	14±1.4	15±2.3	14±2.2	14±1.7
	Day 4	14±1.6	15±2.2	13±2.2	12±3.1
Mean # pups dead/litter					
Day 0		0±0.0	1±0.5	0±0.7	0±0.5
	Day 4	0±0.5	0±0.9	0±0.5	2*±2.4
Mean Percent Survival on Day 0		100±0.0	96±3.7	98±4.5	98±3.3
Mean Percent Survival on Day 4		98±3.7	98±5.8	98±3.2	87*±18.1

a Data obtained from pages 47-49, 145-163, and 166 in the study report.
b Calculated by ICF reviewers.
* Statistically different from control, $p<0.05$
** Statistically different from control, $p<0.01$

2. Body weight: At the high-dose level, mean pup body weight was significantly decreased compared to the respective control on postnatal days 0 and 4 in males, on postnatal day 4 in females, and postnatal days 0 and 4 in both sexes combined. Postnatal day 0 body weight was decreased in high-dose female pups compared to the control, but the difference did not reach statistical significance. Mean pup body weight also was significantly decreased compared to the respective control in mid-dose males, females, and both sexes combined on postnatal day 4 (but not postnatal day 0). Selected mean pup body weight data are presented in Table 8.

TABLE 8. Mean (±SD) Litter and Pup Weights (g) [a]

Lactation Day	Dose Group (mg/kg)			
	0	125	250	500
Mean Litter Weight [b]				
Day 0	89.40±7.07	90.53±12.59	83.61±11.63	82.109±9.14
Day 4	141.13±12.49	140.07±19.49	122.09±12.70	108.40±28.90
Mean Individual Pup Weight-Males				
Day 0	6.58±0.403	6.24±0.458	6.33±0.567	5.92*±0.510
Day 4	10.64±0.657	9.91±0.877	9.54*±1.46	8.79*±0.732
Mean Individual Pup Weight-Females				
Day 0	6.23±0.261	5.94±0.396	5.96±0.580	5.75±0.392
Day 4	10.06±0.585	9.418±0.793	8.95*±1.470	8.70*±0.827
Mean Individual Pup Weight-Both Sexes				
Day 0	6.40±0.320	6.10±0.414	6.18±0.565	5.80*±0.386
Day 4	10.35±0.626	9.67±0.791	9.31*±1.478	8.71*±0.699

a Data obtained from pages 44-46 and 165-166 in the study report.
b Calculated by ICF reviewers.
* Statistically different from control, $p<0.05$
** Statistically different from control, $p<0.01$

III. DISCUSSION and CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS: The study author concluded that oral administration of polyxylenol tetrasulfide (PXTS) and vehicle tricaprylin resulted in parental systemic toxicity at all dose levels in males and at the mid- and high-dose levels in females. Treatment-related clinical observations included salivation, discoloration around the mouth, redness around nose fur, rough coat, discolored paws, coldness to the touch, and hunched posture in males and/or females at 500 mg/kg/day. Systemic toxicity also was manifested as enlarged spleens, which were observed at necropsy in males at all dose levels and females at 250 and 500 mg/kg/day. Although no specific reasoning or supportive information is provided, the study author does not appear to have considered the dark pigmentation of the spleen in males and females at all dose groups an indication of systemic toxicity. Furthermore, NOAEL/LOAEL values for parental systemic toxicity were not established.

The study author concluded that there were no treatment-related effects on mating, fertility, or reproduction, nor on the testes, epididymides, or ovaries. As a result, the study author stated that the reproductive NOAEL is >500 mg/kg/day.

The study author also concluded that fetal toxicity was limited to the mid- and high-dose groups. There was a significant decrease in pup weight on postnatal days 0 (500 mg/kg/day) and 4 (250 and 500 mg/kg/day), as well as a significant reduction in pup survival on postnatal day 4 (500 mg/kg/day). The study author further stated that fetal toxicity in the high-dose group may have

been the result of maternal toxicity. Based on these results, the study author established a NOAEL for offspring of 125 mg/kg/day and a LOAEL of 250 mg/kg/day.

B. REVIEWER COMMENTS: Treatment-related clinical observations were noted in 500-mg/kg/day parental animals and consisted of salivation, discoloration around the mouth, redness around nose fur, rough coat, discolored paws, coldness to the touch, and hunched posture. Treatment-related observations at necropsy consisted of enlarged spleens, which were observed in 125-mg/kg/day males, 250-mg/kg/day males and females, and 500-mg/kg/day males and females, and pigmentation of the spleen, which was observed in both sexes at all dose levels. In a 90-day oral rat study (MRID 46062614), PXTS also produced effects in the spleen at a dose of 200 mg/kg/day which included increased spleen weight, passive congestion of the red pulp, and hemosiderin deposition. Hematologic toxicity was also evident in the 90 day study. Thus, the effects on the spleen observed in the present study are likely related to test article administration. Thus, the parental systemic LOAEL is 125 mg/kg bw/day, based on pigmentation of the spleen (which is believed to be an indication of hematotoxicity). A parental systemic NOAEL cannot be determined. This lack of a NOAEL is consistent with effects observed at 50 mg/kg/day in the 90-day oral toxicity study.

Because there were no effects on reproduction measures or reproductive organs, the reproductive NOAEL is $\geq$ 500 mg/kg/day, and the reproductive LOAEL is > 500 mg/kg/day. .

Fetal toxicity in the high-dose group (500 mg/kg/day) consisted of significantly decreased pup body weight on postnatal days 0 and 4, significantly increased number of mean pup deaths per litter on postnatal day 4, and significantly decreased pup survival on postnatal day 4. Fetal toxicity in the mid-dose group (250 mg/kg/day) was limited to significantly decreased pup body weight on postnatal day 4. Therefore, that the NOAEL for offspring is 125 mg/kg/day, and the LOAEL is 250 mg/kg/day.

C. STUDY DEFICIENCIES: There are no major study design deficiencies. Some minor study design and/or reporting deficiencies are:

- The number of air changes in the study room is not provided.
- The homogeneity and stability of the test material in vehicle is not provided. Although information was available from another study (MRID 46062616), the data in that study only reported on dosing solutions up to a dose of 62.5 mg/kg. The registrant should provide justification for homogeneity and stability of the dosing solutions used in this study on the basis of data from the previous study.
- The number of corpora lutea was not determined. Pre- and post-implantation loss also were not determined.

D. STUDY CLASSIFICATION: This study is classified as **Acceptable-Guideline** and satisfies the guideline requirements for a reproduction/developmental toxicity screening test (OPPTS 870.3550; OECD 421) in rats.