DATA EVALUATION RECORD

BAS 510 F

STUDY TYPE: CARCINOGENICITY FEEDING STUDY - RAT
[OPPTS 870.4200a (§83-2a); OECD 451]
MRID 45404828

7/24/2002

Prepared for

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

Prepared by

Toxicology and Hazard Assessment Group
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. 02-06

Primary Reviewer: S. Milanez, Ph.D., D.A.B.T.	Signature: Date:	JAN 24 2002
Secondary Reviewers: H.T. Borges, Ph.D., MT(ASCP), D.A.B.T.	Signature: Date:	JAN 24 2002
Robert H. Ross, Group Leader	Signature: Date:	JAN 24 2002
Quality Assurance: S.S. Chang, M.S.	Signature: Date:	JAN 24 2002

Disclaimer

This review may have been altered subsequent to the contractor's signature above.

Oak Ridge National Laboratory, managed and operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

(1)

EPA Reviewer: A. C. Levy, Ph.D.
Registration Action Branch 2, Health Effects Division (7509C)
EPA Work Assignment Manager: G. Dannan, Ph.D.
Registration Action Branch 3, Health Effects Division (7509C)

Signature: Alan C. Levy
Date 7-24-2002
Signature: _____
Date _____

DATA EVALUATION RECORD
TXR#: 0050193

STUDY TYPE: Carcinogenicity feeding- rat; OPPTS 870.4200a [§83-2a]; OECD 451.

PC CODE: 128008

DP BARCODE: D278384
SUBMISSION NO.: S604279

TEST MATERIAL (PURITY): BAS 510 F

SYNONYMS: 2-chloro-N-(4'-chlorobiphenyl-2-yl)nicotinamide (IUPAC name)

CITATION: W. Mellert, K. Deckardt, W. Kaufmann, et al. (2001) Carcinogenicity study in Wistar rats; Administration in the diet for 24 months. Experimental Toxicology and Ecology BASF Aktiengesellschaft, D-67056 Ludwigshafen/Rhein, Germany. Laboratory Project No. 82C0179/97090, BASF Registration Document Number 2001/1000115, February 28, 2001. MRID 45404828. Unpublished.

SPONSOR: BASF Corporation Agricultural Products Division, Research Triangle Park, NC 27709.

EXECUTIVE SUMMARY: In a carcinogenicity study (MRID 45404828) BAS 510 F (94.4% a.i., lot no. N37, Tox-batch III) was administered to 50 Wistar rats/sex/dose in the diet at concentrations of 0, 100, 500, 2500, or 15,000 ppm (equivalent to 0, 4.6, 23.0, 116.1, and 768.8 mg/kg bw/day for males and 0, 6.0, 29.7, 155.6, and 1024.4 mg/kg bw/day for females) for 24 months. The investigators determined that 15,000 ppm was causing excessive weight loss in females and mortality in males and these groups were sacrificed after approximately 17 months and not further analyzed.

No treatment-related effects were seen on clinical observations, survival rates, food consumption, food efficiency, differential blood count or morphology, or gross pathology. Body weights and body weight gains differed consistently from the controls in 2500 ppm females, starting on day 315 and persisting throughout the study (body weights, 84.1-95.3% of controls; overall weight gain, 75.8% of controls). The incidence of liver centrilobular hypertrophy was elevated at 2500 ppm in both sexes ($p \leq 0.01$). The incidence of liver eosinophilic foci was increased slightly but not statistically significantly in 500 and 2500 ppm males, but was not seen in females. The incidences of thyroid follicular cell diffuse hypertrophy and focal hyperplasia were increased in both sexes at 2500 ppm, although the changes were statistically significant in only males. The thyroid lesions (as well as thyroid adenoma) caused a 17-18% increase in the absolute and relative thyroid weight in 2500 ppm males. The liver and thyroid findings in this study, as well as in two

as in two separate mechanistic studies conducted by the registrant, are consistent with an adaptive response of the liver to a xenobiotic that resulted in a secondary toxic effect on the thyroid. **The LOAEL is 2500 ppm for both sexes of rats (116.1 and 155.6 mg/kg/day for males and females, respectively) under the conditions of this study, based on the significant decrease in body weight gain in females and the increased incidence of thyroid follicular cell hyperplasia and hypertrophy in both sexes. The NOAEL is 500 ppm (23.0 and 29.7 mg/kg/day for males and females, respectively).**

At the doses tested, there was a small but not statistically significant increase in thyroid follicular cell adenoma in 2500 ppm males and females when compared to controls. The incidence at 0, 100, 500, and 2500 ppm in males was 0/50, 0/50, 1/50, and 4/50, respectively and in females was 0/50, 1/50, 0/50, and 3/50, respectively. The thyroid adenomas were supported by correlating thyroid histological changes, and appeared to be a secondary effect of the chronically induced liver metabolism. No dose-related increase was seen for the number of animals with tumors (benign or malignant) or the total number of primary neoplasms/group. Dosing was considered adequate based on the body weight decreases in females and thyroid lesions in both sexes at 2500 ppm.

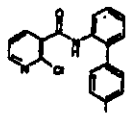
This carcinogenicity study in the rat is **Acceptable/Guideline**, and satisfies the guideline requirement for a carcinogenicity study [OPPTS 870.4200; OECD 451] in rats.

COMPLIANCE: Signed and dated Flagging Criteria, GLP, Quality Assurance, and Data Confidentiality statements were provided. The GLP statement indicated that the study meets the requirements of 40 CFR Part 160, but was conducted in accordance with GLP provisions of FR Germany and the OECD. This study met or exceeded the criteria for flagging studies.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test material: BAS 510 F
   - Description: white solid
   - Lot/Batch #: N37 (Tox-batch III)
   - Purity: 94.4% a.i.
   - Compound Stability: Stable at room temperature during conduct of study
   - CAS for TGAI#: 188425-85-6
   - Structure:



2. Vehicle and/or positive control: The test material was administered continuously in the diet; no positive control was used.

3. **Test animals:**

Species:	Rat
Strain:	Wistar Chbb:THOM (SPF)
Age/weight at study initiation:	42 days old males, 163.5-211.5 g; 43 day old females, 128.3-159.9 g
Source:	Boehringer Ingelheim Pharma KG, Biberach/Riss, Germany
Housing:	Singly in type DK III stainless steel wire mesh cages
Diet:	Ground Kliba maintenance diet rat/mouse/hamster, meal, supplied by Provimi Kliba SA, Kaiseraugst, Switzerland: ad libitum
Water:	Drinking water from water bottles: ad libitum
Environmental conditions:	**Temperature:** 20-24°C **Humidity:** 30-70% **Air changes:** not specified, but animal room was fully air-conditioned **Photoperiod:** 12 hours dark / 12 hours light
Acclimation period:	9 days for males, 10 days for females

B. STUDY DESIGN:

1. **In life dates:** Start: February 4, 1998 for males, February 19, 1998 for females; End (Groups 0-3): February 3-25, 2000 for males, February 18-March 3, 2000 for females; End (Group 4): June 30-July 1, 1999 for males, July 2-5, 1999 for females.

2. **Animal assignment/dose levels:** Animals were assigned by weight (using computer randomization) to the test groups noted in Table 1.

TABLE 1: Study Design

Test Group	Conc. in Diet (ppm)	Dose to animal (mg/kg/day)		Number of animals	
		Male	Female	Male	Female
0 (Control)	0	0	0	50	50
1 (Low dose)	100	4.6	6	50	50
2 (Mid dose)	500	23	29.7	50	50
3 (Mid dose)	2500	116.1	155.6	50	50
4 (High dose)[1]	15000	768.8	1024.4	50	50

Data from pp. 23 and 37, MRID 45404828.
[1]Surviving Group 4 animals were all sacrificed after 17 months due to excessive toxicity; other groups were sacrificed after 24 months.

3. **Dose selection:** The high test concentration of 15,000 ppm was expected to have a test substance intake of >1000 mg/kg body weight/day, which is a "limit test" concentration for testing toxic substances. The other dietary concentrations were 2,500 and 500 ppm as "mid concentrations" and 100 ppm as the "low concentration." No previous toxicity study results were mentioned for BAS 510 F.

4. **Diet preparation and analysis:** Diets were prepared weekly (usually) by mixing appropriate amounts of test substance with ground Kliba maintenance diet rat/mouse/hamster meal and were stored at ambient temperature. The stability of the test material in the diet was determined prior to study initiation using 100 ppm samples. Three or four samples were assayed at times 0, 12, and 32 days after preparation and storage at ambient temperature. Homogeneity and concentration were tested at the beginning of the study as well as after 3

months of treatment; concentration only was tested after 6, 9, 12, 15, and 18 months of treatment (except 15,000 ppm diets were not assessed after 18 months). Three samples of the highest and lowest concentration were assayed for homogeneity and the dietary concentration was assayed for all doses (duplicate analyses were conducted for each sample). No dietary analyses were performed after 18 months due to protocol deviation, but the study investigators assumed that these concentrations were correct based on the results of the analyses up to 18 months.

Results:

Homogeneity analysis: The mean and standard deviation of the measured concentration of 100 ppm samples (duplicate analyses of three samples) was 100.1 ± 3.8% of nominal at the beginning of the study and 102.8 ± 1.8% after 3 months. Analogous analyses for the 15,000 ppm samples yielded 96.2 ± 1.7% of nominal at study initiation and 93.2 ± 1.5% at 3 months. The low standard deviations demonstrated the homogeneity of the feed.

Stability analysis: Concentrations of samples taken after 13 and 32 days ranged from 95.4-99.6% and 94.5-99.6% of nominal values (mean of 97.5 and 97.9% of time 0 concentration).

Concentration analysis: Through 18 months of the study, analytical concentrations (mean of duplicates) as a percent of nominal concentrations ranged from 89.9-102.9% at 100 ppm, 91.6-98.4% at 500 ppm, 90.5-104.5% at 2500 ppm, and 92.4-98.9% at 15,000 ppm (latter samples only taken through 15 months).

The analytical data indicated that the mixing procedure was adequate and that the variance between nominal and actual dosage to the animals was acceptable.

5. Statistics: Food consumption, body weight, body weight change, and food efficiency data were analyzed by parametric one-way analysis using the F-test and 2-sided ANOVA. If the resulting p-value was ≤ 0.05, Dunnett's 2-sided test was used to compare each test group with the control group; significance was reported as $p \leq 0.05$ or 0.01. Organ weights were evaluated with the two-sided Kruskal-Wallis test (non-parametric one-way analysis). If the resulting p-value was ≤ 0.05, each test group was compared with the control group using the Wilcoxon test (significance reported as $p \leq 0.05$ or 0.01). The reviewer considers the analyses used to be appropriate, and also analyzed intergroup differences for histology and selected gross necropsy lesions using Fisher's exact probability test.

C. METHODS:

1. Observations:

1a. **Cageside observations:** Animals were inspected for signs of toxicity and for mortality twice a day Monday - Friday and once a day on Saturday and Sunday and on holidays.

1b. **Clinical examinations:** Thorough clinical examinations, including palpation, were conducted weekly.

2. **Body weight:** Animals were each weighed prior to dosing, on study day 0, and weekly through week 13. Thereafter, rats were weighed every 4 weeks and prior to necropsy.

3. **Food consumption and compound intake:** Food consumption was determined once a week over a period of 7 days through the first 13 dosing weeks. Thereafter, food consumption was determined one week in 4 and prior to necropsy. The mean daily diet consumption was calculated as g food/animal/day. Food efficiency (body weight gain in g/food consumption in g per unit time X 100) and compound intake (mg/kg bw/day) values were calculated as time-weighted averages from the food consumption and body weight gain data.

4. **Ophthalmoscopic examination:** Not conducted; not a guideline requirement.

5. **Hematology & Clinical chemistry:** Blood was collected by decapitation from all rats surviving to the study termination to prepare differential blood smears. The rats were fasted overnight prior to the collection. Only the control and 2500 ppm groups were evaluated, which is the minimum required for carcinogenicity studies based on Guideline 870.4200 & OECD 451. Clinical chemistry studies were not conducted and are not required for carcinogenicity studies based on Guideline 870.4200 & OECD 451.

6. **Urinalysis:** Urinalysis was not conducted and is not required for carcinogenicity studies based on Guideline 870.4200 & OECD 451.

7. **Sacrifice and pathology:** All animals that died and those sacrificed on schedule (decapitation under CO_2 anesthesia) were subjected to gross pathological examination and the CHECKED (X) tissues were collected. Histological examination was conducted on all control and 2500 ppm animals, but the thyroid glands, thymus, lungs, liver, kidneys, and urinary bladder and gross lesions were also examined in the 100 and 500 ppm groups. The animals were fasted overnight prior to necropsy. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
-	Tongue	x	Aorta, thoracic*	xx	Brain (multiple sections)*+
x	Salivary glands*	x	Heart*+	x	Periph.nerve*
x	Esophagus*	x	Bone marrow*	x	Spinal cord (3 levels)*
x	Stomach*	x	Lymph nodes*	x	Pituitary*
x	Duodenum*	x	Spleen*+	x	Eyes (retina, optic nerve)*
x	Jejunum*	x	Thymus		GLANDULAR
x	Ileum*			xx	Adrenal gland*+
x	Cecum*		UROGENITAL	x	Lacrimal gland
x	Colon*	xx	Kidneys*+	x	Parathyroids*
x	Rectum*	x	Urinary bladder*	xx	Thyroids*
xx	Liver*+	xx	Testes*+		OTHER
-	Gall bladder* (not rat)	x	Epididymides*+	x	Bone (sternum and/or femur)
-	Bile duct (rat)	x	Prostate*	x	Skeletal muscle
x	Pancreas*	x	Seminal vesicle*	x	Skin*
	RESPIRATORY	xx	Ovaries*+	x	All gross lesions and masses*
x	Trachea*	x	Uterus*+		
x	Lung*	x	Mammary gland* (female only)		
-	Nose*	x	Oviducts		
-	Pharynx*	x	Vagina		
-	Larynx*				

* Required for carcinogenicity studies based on Guideline 870.4200.
+Organ weight required in carcinogenicity studies. - Not examined

II RESULTS

A. OBSERVATIONS:

1. **Clinical signs of toxicity:** The incidence of abnormal clinical observations was comparable in control and treated animals.

2. **Mortality:** The mortality rates at study termination (day 728) for rats administered 0, 100, 500, and 2500 ppm BAS 510 F were, respectively, 14, 24, 22, and 32% for males and 28, 16, 22 and 20% for females. The elevated mortality in the 2500 ppm males appeared to be incidental because many deaths in the 2500 ppm males occurred shortly before day 728, and mortality was comparable among the control and/or three dose groups on days 700 and 751 (day 700 mortality:10, 20, 16, 18%; day 751 mortality: 18, 26, 26, 32% at 0, 100, 500, and 2500 ppm, respectively). The mortality rate of the 15,000 ppm males was higher than for the other groups: the first animal died on day 196 compared to day 420-504 in the control and three lower dose groups, and at day 504, the mortality rate was 6% for the 15,000 ppm males and 2% for all the other groups. The 15,000 ppm females did not have an elevated mortality rate compared to the other study groups.

B. BODY WEIGHT: Body weights of control and all groups of treated males were comparable throughout the study (97.9-104.4% of controls), including the 15,000 ppm group through day 483 [day 483 was the last weighing before the 15,000 ppm males and females were sacrificed; all other groups were weighed through day 728]. The body weight gains of the males were also similar to that of the controls (97.2-107.1% of controls); the statistically significant increases ($p \leq 0.05$ or 0.01) seen for some groups through day 42 were not dose-

related. Females had significantly reduced body weights ($p \leq 0.05$ or 0.01) starting on day 147 at 15,000 ppm and on day 315 at 2500 ppm, and persisting through the last weighing (15,000 ppm, 86.8-95.8% of controls; 2500 ppm, 84.1-95.3% of controls). Body weight gains of females parallelled their lowered body weights, significant decreases ($p \leq 0.05$ or 0.01) occurring consistently starting on day 147 at 15,000 ppm and on day 287 at 2500 ppm. The final weight gain of the 15,000 ppm females was 79.1% of controls (day 483) and of the 2500 ppm females was 75.8% of controls (day 728). The body weights and body weight gains of the females are shown in Table 2.

TABLE 2: Mean body weights and body weight gains (± standard deviation) for female rats fed BAS 510 F for up to 2 years.[1]

Study day	Dietary concentration (ppm)				
	0	100	500	2500	15000
Mean body weights (g) – females					
0	144.0 ± 7.2	145.9 ± 7.7	145.5 ± 7.4	145.2 ± 8.0	143.1 ± 7.3
91	284.8 ± 22.1	276.9 ± 18.9	279.5 ± 20.5	276.6 ± 24.2	276.0 ± 20.2
175	311.4 ± 25.3	309.1 ± 22.2	310.3 ± 24.4	306.0 ± 27.1	297.8 ± 22.2* (95.6)
371	351.8 ± 44.6	339.0 ± 31.2	339.5 ± 32.4	331.8 ± 33.6* (94.3)	319.5 ± 29.4** (90.8)
483	381.9 ± 48.2	362.4 ± 40.2	364.9 ± 42.1	347.4 ± 38.6** (91.0)	331.4 ± 34.0** (86.8)
728	425.8 ± 75.7	410.0 ± 63.9	397.0 ± 70.0	358.0 ± 56.2** (84.1)	N/A
Mean body weight gains (g) – females					
0-91	140.8 ± 17.7	130.9 ± 15.2* (93.0)	134.0 ± 17.0	131.4 ± 19.3* (93.3)	132.9 ± 15.4
0-175	167.4 ± 21.7	163.2 ± 19.2	164.8 ± 21.4	160.8 ± 21.9	154.7 ± 18.0** (92.4)
0-371	207.8 ± 42.6	193.0 ± 28.1	194.0 ± 31.4	186.6 ± 29.2** (89.8)	176.4 ± 26.0** (84.9)
0-483	238.0 ± 46.6	216.5 ± 37.0* (91.0)	219.4 ± 41.7	202.3 ± 34.9** (85.0)	188.3 ± 30.9** (79.1)
0-728	281.1 ± 74.8	263.9 ± 62.1	252.6 ± 70.2	213.1 ± 55.1** (75.8)	N/A
175-371[2]	40.4	29.9	29.2	25.8	21.7
371-483[2]	30.1	23.4	25.4	15.6	11.9

Data taken from pp. 87-99, MRID 45404828.
N/A = not available (animals were sacrificed prior to this time point).
[1]Numbers in parentheses are the percent of control.
[2]Calculated by the reviewer and not statistically analyzed.
*$p \leq 0.05$, **$p \leq 0.01$, significantly different than the control group.

C. FOOD CONSUMPTION AND COMPOUND INTAKE:

1. **Food consumption:** Food consumption was not affected by treatment. Males given 100, 500, and/or 2500 ppm had slightly greater consumption than the controls (3.2-7.6%, $p \leq 0.05$ or 0.01) primarily between days 28 and 84 of the study. Females administered 100, 500, and/or 2500 ppm had slightly decreased consumption intermittently during the first treatment year (3.9-9.1%, $p \leq 0.05$ or 0.01). A dose-response was not seen for either sex.

2. **Compound consumption:** (time-weighted average): The time-weighted-average doses for each treatment group are presented in Table 1.

3. **Food efficiency:** No consistent differences were seen among the control and treated groups of either sex; both increased and decreased values ($p \leq 0.05$ or 0.01) were seen sporadically during the study.

D. **OPHTHALMOSCOPIC EXAMINATION:** Not conducted.

E. **BLOOD ANALYSES:**

1. **Hematology:** No treatment-related changes were seen in the leukocyte differential count or in the morphology of leukocytes or erythrocytes.

2. **Clinical chemistry:** Not performed.

F. **URINALYSIS:** Not performed.

G. **SACRIFICE AND PATHOLOGY:** Rats treated with 15,000 ppm BAS 510 F were sacrificed after approximately 17 months of treatment, but necropsy and histopathology were not performed on these groups.

1. **Organ weight:** Statistically significant changes ($p \leq 0.05$ or 0.01) in males consisted of increased absolute and relative thyroid weight at 2500 ppm (117-118% of controls), and increased absolute testes weight and decreased relative adrenal weight at 500 ppm (120% and 86% of controls, respectively). Females had decreased absolute kidney weight at 500 and 2500 ppm (93-95% of controls). The changes in adrenal, testes, and kidney weights appeared to be incidental to treatment because there was no clear dose-response. Additionally, the relative testes and kidney weights did not differ significantly from controls. The increased thyroid weight in 2500 ppm males was correlated with increased incidences of thyroid follicular cell hypertrophy and hyperplasia as well as follicular cell adenoma. The terminal body weight of the high-dose females was significantly lower (17%, $p \leq 0.01$) than of the control group. The thyroid and body weights of males are shown in Table 3.

TABLE 3. Thyroid weights (± standard deviation) in male rats fed BAS 510 F for 2 years[1].

Organ	Dietary concentration (ppm)			
	0	100	500	2500
Body wt, (g) [n=34-41]	659.722 ± 92.923	661.059 ± 95.945	682.724 ± 102.428	668.541 ± 79.653
Thyroid glands (mg)	38.872 ± 6.879	37.583 ± 7.71	41.703 ± 9.055	45.818 ± 9.156** (118)
% body weight [n=33-39]	0.006 ± 0.001	0.006 ± 0.001	0.006 ± 0.002	0.007 ± 0.002** (117)

Data taken from pp. 126 and 128, MRID 45404828.
**$p \leq 0.01$, significantly different from the control group.
[1]Numbers in parentheses are the percent of control, calculated by the reviewer.

2. **Gross pathology:** The only lesion with a significantly increased incidence ($p \leq 0.05$) was "mass" in the thymus of 2500 ppm females (0/50, 3/50, 3/50, 6/50 at 0, 100, 500, and 2500 ppm, respectively). The incidence of this lesion was not increased in males (1/50, 2/50, 3/50, 1/50 at 0, 100, 500, and 2500 ppm, respectively) and a correlating microscopic lesion with an increased incidence was not found.

3. Microscopic pathology:

a) Non-neoplastic: The liver and thyroid were target organs in both sexes. The incidence of liver centrilobular hypertrophy was increased significantly at 2500 ppm ($p \leq 0.01$) in males and females. The incidence of liver eosinophilic foci was increased slightly but not statistically in 500 and 2500 ppm males, but was not seen in females. The hypertrophy was characterized by an increase in the hepatocyte size and organelle content, and a ground-glass appearance sometimes containing eosinophilic droplets. The eosinophilic foci in 2500 ppm males had cytoplasmic inclusions.

The incidences of thyroid follicular cell diffuse hypertrophy and of focal hyperplasia were also increased in both sexes of 2500 ppm rats, but the change was significant ($p \leq 0.01$) only in the males. The thyroid lesions were correlated with an increase in the weight of this organ in males. The decreased incidence ($p \leq 0.05$) of focal degeneration of the adrenal cortex in 2500 ppm females was likely incidental to treatment, and the small (non-significant) increase in the incidence of urinary bladder diffuse papillary hyperplasia in males was of unknown etiology. The liver and thyroid microscopic lesions are summarized in Table 4.

TABLE 4: Incidence of non-neoplastic microscopic pathology findings in rats fed BAS 510 F for 2 years.

Organ: lesion	Dietary concentration (ppm)							
	0	100	500	2500	0	100	500	2500
	Males				Females			
Liver: Centrilob. hypertrophy	0/50	0/50	2/50	27/50**	0/50	0/50	0/50	11/50**
Eosinophilic foci	3/50	4/50	8/50	9/50	0/50	0/50	0/50	0/50
Thyroid glands:								
Hypertr. Follic. Cell. diffuse	2/50	5/50	6/50	22/50**	2/50	0/50	0/50	4/50
Hyperpl. Folicl. Cell. focal	1/50	1/50	1/50	9/50**	2/50	2/50	1/50	7/50

Data from pp. 41-42 and 145-208. MRID 45404828.
*$p \leq 0.05$. **$p \leq 0.01$: Significantly different from controls. determined by reviewer using Fisher exact test.

b) Neoplastic: There were no statistically significant differences in the incidence of any type of tumor in either sex. A slight but not statistically significant increase in thyroid follicular cell adenoma was seen in 2500 ppm males and females (4/50 males and 3/50 females vs. 0/50 each control). This finding was correlated with enlargement of the thyroid glands in one male given 500 ppm and one given 2500 ppm. The adenomas contained well-defined areas of hyperplastic follicles with hypertrophic epithelial cells with compressed adjacent parenchyma. Historical control tumor data provided by BASF showed that the mean rate of adenoma was 0.8% from 41 studies (37 feeding studies) that included 1710 animals, the range was 0-10%, and ≥6% adenoma occurred in only 2 of the 41 studies.

The number of animals with tumors (benign or malignant), with more than one primary neoplasm, and the total number of primary neoplasms/group were similar for the control and treated groups or were not dose-related. The ratio of benign to malignant primary tumors differed somewhat among the dose groups, but there was no consistent pattern. The thyroid and overall tumor incidences of the animals are shown in Table 5.

TABLE 5: Incidence of thyroid neoplasia in rats fed BAS 510 F for 2 years.								
Neoplasm	Dietary concentration (ppm)							
	0	100	500	2500	0	100	500	2500
	Males				Females			
Thyroid follicular cell adenoma	0/50	0/50	1/50	4/50	0/50	1/50	0/50	3/50
No. rats with neoplasms	43/50	48/50	43/50	43/50	45/50	46/50	42/50	42/50
No. rats with benign neoplasms	40/50	44/50	38/50	42/50	41/50	41/50	40/50	38/50
No. rats with malignant neoplasms	6/50	15/50	13/50	4/50	8/50	15/50	9/50	15/50
No. rats with >1 primary neoplasm	19/50	28/50	28/50	28/50	23/50	28/50	21/50	27/50
Total no. primary neoplasms (number benign-malignant)	77 (69-8)	95 (79-16)	93 (79-14)	94 (90-4)	79 (71-8)	87 (70-17)	75 (64-11)	77 (61-16)

Data from pp. 41 and 209-235, MRID 45404828.

III. DISCUSSION and CONCLUSIONS

A. **INVESTIGATORS' CONCLUSIONS:** The investigators determined that the 15,000 ppm treatment was causing excessive weight loss in females and excessive mortality in males, and therefore these groups were sacrificed after approximately 17 months and not further analyzed. A significant decrease in body weight and body weight gain occurred in only the 2500 ppm females during the 2-year study. Treatment did not affect the differential blood count or morphology, and was not responsible for the small but statistically significant decreases in absolute kidney weight (500 and 2500 ppm females) or relative adrenal weight (500 ppm males).

Treatment with BAS 510 F was associated with an increased incidence of hepatocyte centrilobular hypertrophy in 2500 ppm males and females ($p \leq 0.01$) and a slight (non-statistical) increase in 500 ppm males. The 500 and 2500 ppm males also had a small increase in eosinophilic foci. The incidences of thyroid hypertrophy and hyperplasia were elevated in 2500 ppm males ($p \leq 0.01$) and slightly, but not significantly, in 2500 ppm females. The thyroid lesions (and adenoma) resulted in increased thyroid weights in 2500 ppm males ($p \leq 0.01$). The investigators postulated that BAS 510 F induced liver enzymes and liver hypertrophy as an adaptive response, which caused a concomitant increase in the turnover of T3/T4, chronic feedback stimulation of thyroid stimulating hormone, and the resulting thyroid lesions (including adenoma). This assertion was consistent with two mechanistic studies in which Wistar rats were given 15,000 ppm BAS 510 F in the diet: increased microsomal enzyme levels were seen in a 2-week study (MRID 45404902), and decreased T3/T4 thyroid hormone levels and increased thyroid stimulating hormone levels were found in a 4-week study (MRID 45404903). The investigators identified the liver and thyroid as target organs and stated that treatment-related effects occurred in both sexes at 2500 ppm and in males at 500 ppm (centrilobular liver cell hypertrophy in 2/50 males vs. 0/50 controls). Therefore, the no observed effect level for males was 100 ppm and for females was 500 ppm.

Thyroid follicular cell adenoma occurred in both sexes at 2500 ppm (4/50 males and 3/50 females vs. 0/50 each control, NS) and in one 500 ppm male and one 100 ppm female. The investigators considered the adenoma in the 500 ppm male as treatment-related based on a parallel chronic toxicity study (MRID 45404827) but the adenoma in the 100 ppm females as spontaneous and within historical control levels. The investigators concluded that BAS 510 F

spontaneous and within historical control levels. The investigators concluded that BAS 510 F is not a relevant human oncogen because the increase in thyroid adenoma was not due to an organ-specific carcinogenic effect but a secondary mechanism, a small number of animals were affected, only adenomas were found, and BAS 510 F is not genotoxic.

B. REVIEWER COMMENTS: The reviewer concurs with the investigators' conclusion that BAS 510 F caused centrilobular hypertrophy in both sexes and eosinophilic foci in males at 2500 ppm. The reviewer also agrees that there is sufficient evidence, especially when taking into consideration the two BASF mechanistic studies and the concurrent 2-year chronic toxicity study, to conclude that the thyroid lesions (including adenomas) in both sexes were indirectly treatment-induced and likely due to chronically decreased T3/T4 levels and elevated TSH levels.

The LOAEL is 2500 ppm for both sexes of rats (116.1 and 155.6 mg/kg/day for males and females, respectively) under the conditions of this study, based on the significant decreases in body weight and body weight gains in females and the increased incidence of thyroid follicular cell hyperplasia and hypertrophy in both sexes (statistically significant only in males). The NOAEL is 500 ppm (23.0 and 29.7 mg/kg/day for males and females, respectively). This disagrees with the investigators' conclusion for the males. because the reviewer considers that the occurrence of hepatocellular centrilobular hypertrophy in 2/50 males (vs. 0/50 in controls) at 500 ppm with no associated thyroid toxicity was a minor adaptive response that does not establish 500 ppm as a systemic toxicity effect level.

A small increase in thyroid follicular cell adenoma was seen in 2500 ppm males and females. The incidence at 0, 100, 500, and 2500 ppm in males was 0/50, 0/50, 1/50, and 4/50, respectively and in females was 0/50, 1/50, 0/50, and 3/50, respectively. Although this is within the range of the testing laboratory's historical control values, it is well above the mean of 0.8% and the correlating thyroid histological changes indicate that the adenomas were treatment-related. Contrary to the study author's opinion, the reviewer considers that chemically-induced thyroid adenoma is relevant to human carcinogenicity, and this is also the position of the U.S. EPA (Hill et al. 1998. *Environ. Health Perspect.* 106: 447-457). Dose-related increases were not seen for the number of animals with tumors (benign or malignant) or the total number of primary neoplasms/group. Dosing was considered adequate based on the body weight decreases in females and thyroid lesions in both sexes at 2500 ppm.

This carcinogenicity study in the rat is **Acceptable/Guideline**, and satisfies the guideline requirement for a carcinogenicity study [OPPTS 870.4200; OECD 451] in rats. No major deficiencies were identified.

C. STUDY DEFICIENCIES: No major deficiencies were identified. Minor deficiencies that are unlikely to affect the outcome or interpretation of this study include failure to weigh the spleen and uterus, and to evaluate microscopically the nose, pharynx, and larynx. Although not required, it would have been helpful for the interpretation of the study if an intermediate (12 month) sacrifice and analysis had been conducted and/or if the 15,000 ppm rats sacrificed after 17 months were necropsied and analyzed microscopically.

DATA FOR ENTRY INTO ISIS

Carcinogenicity Study - rats (870.4200a)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range: ppm, (mg/kg/day)[1]	Doses tested: ppm, (mg/kg/day)[1]	NOAEL: ppm, (mg/kg/day)	LOAEL: ppm (mg/kg/day)	Target organ(s)	Comments
128008	45404828	carcinogenicity	rats	24 months	oral	diet	100-2500 (4.6-155.6)	0, 100, 500, 2500 (♂: 0, 4.6, 23.0, 116.1; ♀: 0, 6.0, 29.7, 155.6)	500 (23.0 for ♂, 29.7 for ♀)	2500 (116.1 for ♂, 155.6 for ♀)	Thyroid and liver in both sexes; body weight decrease in females.	

[1]A fourth test concentration, 15, 000 ppm (equivalent to 768.8 mg/kg bw/day for males and 1024.4 mg/kg bw/day for females), was given to males and females but was discontinued after 17 months due to excessive toxicity in both sexes.