UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES, AND
TOXIC SUBSTANCES

TXR No.: 0053434

MEMORANDUM

DATE: May 19, 2005

SUBJECT: **BAS 670H:** Report of the Cancer Assessment Review Committee
PC Code: 123009

FROM: Jessica Kidwell, Executive Secretary
Cancer Assessment Review Committee
Health Effects Division (7509C)

TO: Yung Yang, Toxicologist (TB)
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Health Effects Division (7509C)

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Herbicide Branch, Registration Division (7505C)

The Cancer Assessment Review Committee met on March 16, 2005 and April 27, 2005 to evaluate the carcinogenic potential of BAS 670H. Attached please find the Final Cancer Assessment Document.

cc: J. Pletcher
Y. Woo

EXECUTIVE SUMMARY

On March 16, 2005, the Cancer Assessment Review Committee (CARC) of the Health Effects Division of the Office of Pesticide Programs met to evaluate the carcinogenic potential of BAS 670H. This was the first time that this compound was assessed for carcinogenicity by the CARC. On April 27, 2005, the mode of action was re-evaluated in light of the information concerning the registrant's position on the mode of action.

Yung Yang of the Toxicology Branch presented the chronic toxicity/carcinogenicity study in Wistar rats and the carcinogenicity study in C57BL/6J Rj mice. In the rat chronic toxicity/carcinogenicity study, groups of Wistar rats (50/sex/dose) were exposed to BAS 670H (95.2-95.8%, a.i.) in the diet at concentrations of 0, 6, 60, 600, or 6000 ppm nominally (equivalent to 0/0, 0.4/0.5, 3.6/4.7, 36.4/50.8, and 381.5/524.1 mg/kg/day in males/females) for up to 24 months. In the mouse carcinogenicity study, C57BL/6J Rj mice (50/sex/dose) were exposed to BAS 670H (95.2-95.8%, a.i.) in the diet at concentrations of 0, 80, 800, or 8000 ppm nominally (equivalent to 0/0, 19/26, 194/256, and 1903/2467 mg/kg/day in males/females) for up to 78 weeks.

The CARC concluded the following:

Carcinogenicity

Rat

- In male rats, the incidence of thyroid follicular cell adenomas, adenocarcinomas, and combined adenomas and/or adenocarcinomas for the control, 6, 60, 600, and 6000 ppm dose groups was as follows:

Adenomas	8/49 (16%), 12/50 (24%), 13/50 (26%), 18/50 (36%), 23/49 (47%)
Adenocarcinomas	2/47 (4%), 1/46 (2%), 1/46 (2%), 1/46 (2%), 2/42 (5%)
Combined	10/49 (20%), 13/50 (26%), 13/50 (26%), 19/50 (38%), 23/49 (47%)

Male rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 6000 ppm dose group with the controls, for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, all at $p<0.01$. There were significant differences in the pair-wise comparisons of the 600 ppm dose group with the controls for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, both at $p<0.05$. The incidence of thyroid follicular cell adenomas at 600 ppm (36%) and 6000 ppm (47%) exceeded the laboratory historical control mean of 28%. Therefore, the CARC considered the thyroid follicular cell tumors in male rats, driven by adenomas, to be treatment-related.

- In female rats, the incidence of thyroid follicular cell adenomas, adenocarcinomas, and combined adenomas and/or adenocarcinomas for the control, 6, 60, 600, and 6000 ppm

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dose groups was as follows:

Adenomas	2/50 (4%), 4/50 (8%), 7/50 (14%), 8/50 (16%), 13/50 (26%)
Adenocarcinomas	0/50 (0%), 0/50 (0%), 2/50 (4%), 2/50 (4%), 0/50 (0%)
Combined	2/50 (4%), 4/50 (8%), 9/50 (18%), 10/50 (20%), 13/49 (26%)

Female rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 6000 ppm dose group with the controls, for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, all at $p < 0.01$. There were significant differences in the pair-wise comparisons of the 600 ppm dose group with the controls for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, and of the 60 ppm dose group with the controls for thyroid follicular cell adenomas and/or adenocarcinomas combined, all at $p<0.05$. The incidence of thyroid follicular cell adenomas at 600 ppm (16%) and 6000 ppm (26%) exceeded the laboratory historical control mean of 10%. Therefore, the CARC considered the thyroid follicular cell tumors in female rats, driven by adenomas, to be treatment-related.

- In male and female rats, the CARC concluded that dosing at the high dose was considered to be adequate and not excessive based on increased mortality in males, decreased body weight (↓9%/↓8%, males/females) and body weight gains (↓11%/↓10%, males/females), clinical observations (corneal opacity and chronic keratitis) and histopathological findings.

Mouse

- In female mice, the incidence of histiocytic sarcomas was 3/48 (6%), 0/47 (0%), 1/48 (2%), and 7/44 (16%) for the control, 80, 800, and 8000 ppm dose groups, respectively. Female mice had a significant increasing trend in histiocytic sarcomas of the hemolymphoreticular system at $p<0.01$. There were no significant differences in the pair-wise comparisons of the dosed groups with the controls. The incidence of histiocytic sarcomas at 8000 ppm (16%) was within the range of the historical control from the conducting laboratory (15.6%). No preneoplastic lesions were noted. This tumor is known to be common in mice, associated with aging. Therefore, the CARC did not consider the histiocytic sarcomas in female mice to be treatment-related.

- There were no significant treatment-related tumors in male mice.

- The CARC considered dosing at the high dose, which was greater than the limit dose, in both sexes to be adequate and not excessive. In males, this was based on decreased body weight and body weight gain, increased relative liver weight and increased incidence of hepatocellular hypertrophy. In females, this was based on increased mortality, however, no significant decreases in body weight and body weight gains, or histopathological findings were observed.

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Mutagenicity

- There is no mutagenicity concern for BAS 670H based on *in vivo* or *in vitro* assays.

Structure Activity Relationship (SAR)

- The very limited SAR data was not useful in the weight-of-the-evidence analysis.

Mode of Action

- The Registrant provided data showing effects of BAS 670H on thyroid hormones (T4 and TSH), thyroid weights and thyroid histopathology (hypertrophy and hyperplasia). Overall, there is evidence of concordance between the effects on thyroid homeostasis and thyroid tumor formation. The CARC concluded that the mode of action for thyroid follicular cell tumors in rats has been established. Evidence for the demonstration of antithyroid activity as required by the Agency's Policy Document (EPA/630/R-97/002) is described in detail in Appendix A.

In accordance with the EPA Final Guidelines for Carcinogen Risk Assessment (March 29, 2005), the CARC classified BAS 670H as **"Not likely to be carcinogenic to humans at doses that do not alter rat thyroid hormone homeostasis"**. This decision was based on the following considerations:

(i) No treatment-related tumors were seen in male or female mice when tested at doses (greater than the limit dose) that were adequate to assess carcinogenicity;

(ii) Treatment-related thyroid follicular cell tumors were seen in both male and female rats at 600 and 6000 ppm, which were considered to be adequate, and not excessive, to assess carcinogenicity;

(iii) There is no mutagenicity concern from *in vivo* or *in vitro* assays;

(iv) The non-neoplastic toxicological evidence (i.e., thyroid growth, thyroid follicular cell hypertrophy/hyperplasia, thyroid hormonal changes) indicated that BAS 670H was inducing a disruption in the thyroid-pituitary hormonal status.

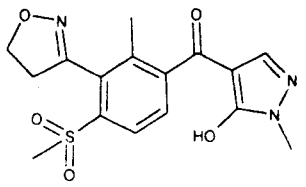
The mechanistic data for thyroid follicular cell tumor formation meet the criteria established by the Agency for the use of a margin of exposure approach for human cancer risk assessment. However, the CARC determined that quantification of human cancer risk is not required since the NOAEL (0.4 mg/kg/day) for non-cancer risk assessment is not expected to alter thyroid hormone homeostasis nor result in thyroid tumor formation.

I. INTRODUCTION

On March 16, 2005, the Cancer Assessment Review Committee (CARC) of the Health Effects Division of the Office of Pesticide Programs met to evaluate the carcinogenic potential of BAS 670H. This was the first time that this compound is assessed for carcinogenicity by the CARC. On April 27, 2005, the mode of action was re-evaluated in light of the information concerning the registrant's position on the mode of action.

II. BACKGROUND INFORMATION

Chemical Name:	BAS 670H; [3-(4-5-Dihydro-isoxazol-3-yl)-4-methanesulfonyl-2-methyl-phenyl]-(5-hydroxy-1-methyl-1H-pyrazol-4-yl)-methanone
Empirical Formula:	$C_{16}H_{17}N_3O_5S$
Molecular Weight:	363.4
CAS Registry No.:	210631-61-8
PC Code:	123009
Structure:	



BAS 670H (common name Topramezone) belongs to the triketone class of chemicals. It inhibits the 4-hydroxyphenylpyruvate dioxygenase (HPPD) enzyme, thereby impairing carotenoid biosynthesis in the chlorophyll pathway, ultimately leading to the breakdown of chloroplasts. In mammals, the inhibition of the HPPD enzyme resulted in elevated tyrosine levels (tyrosinemia).

BAS 670H is a broad-spectrum, post-emergence herbicide used to control grassy and broadleaf weeds in corn. The herbicide is absorbed by the leaves, roots and shoots, then translocated to the growing points of the sensitive weeds. The maximum seasonal use is 0.022 lb/ai/acre. There is a maximum of 2 applications (at least 1 week apart) per season. The single application broadcast rate can be 0.011 lb/ai/acre to the full seasonal maximum use rate (0.011 to 0.22 lb ai/acre). The product controls both broadleaf and grass weeds in all corn types (field, pop, seed, sweet) with a 45 day preharvest application interval.

III. EVALUATION OF CARCINOGENICITY STUDIES

1. Combined Chronic Toxicity/Carcinogenicity Study in Rats

References: Kaspers, U., K. Deckardt, K. Küttler, *et al.* (2003) BAS 670H: Carcinogenicity study in Wistar rats: administration in the diet for 24 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: 82S0124/98099, January 8, 2003. MRID 45902222. Unpublished.

A. Experimental Design

Groups of Wistar rats (50/sex/dose) were exposed to BAS 670H (95.2-95.8%, a.i.) in the diet at concentrations of 0, 6, 60, 600, or 6000 ppm nominally (equivalent to 0/0, 0.4/0.5, 3.6/4.7, 36.4/50.8, and 381.5/524.1 mg/kg/day in males/females) for up to 24 months.

B. Discussion of Mortality and Tumor Data

Survival Analysis

The statistical evaluation of mortality indicated a significant increasing trend with increasing doses of BAS 670H in male rats, as well as a significant difference in the pair-wise comparison of the 6000 ppm dose group with the controls, both at $p<0.05$ (Table 1). There were no statistically significant incremental changes in mortality with increasing doses of BAS 670H in female rats (Table 2) (Memo, L. Brunsman, 2/25/2005, TXR No. 0053149). The statistical evaluation of mortality was based upon the Thomas, Breslow and Gart computer program.

Table 1. BAS 670H - Wistar Rat Study (MRID 45902222)
Male Mortality Rates[+]
Cox or Generalized K/W Test Results

Dose (ppm)	Weeks				
	1-26	27-52	53-78	79-105[f]	Total
0	0/50	1/50	0/49	2/49	3/50 (6)*
6	0/50	0/50	0/50	6/50	6/50 (12)
60	0/50	0/50	0/50	8/50	8/50 (16)
600	0/50	0/50	0/50	8/50	8/50 (16)
6000	0/50	0/50	1/50	11/49	12/50 (24)*

[+]Number of animals that died during interval/Number of animals alive at the beginning of the interval.
[f]Final sacrifice at week 104.
( )Percent.

Note: Time intervals were selected for display purposes only.
Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Table 2. BAS 670H - Wistar Rat Study (MRID 45902222)
Female Mortality Rates[+]
Cox or Generalized K/W Test Results

Dose (ppm)	Weeks				
	1-26	27-52	53-78	79-106[f]	Total
0	0/50	0/50	2/50	9/48	11/50 (22)
6	0/50	0/50	6/50	11/44	17/50 (34)
60	0/50	0/50	1/50	9/49	10/50 (20)
600	0/50	0/50	0/50	7/50	7/50 (14)
6000	0/50	0/50	3/50	5/47	8/50 (16)

[+]Number of animals that died during interval/Number of animals alive at the beginning of the interval.
[f]Final sacrifice at week 104.
( )Percent.

Note: Time intervals were selected for display purposes only.
Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Tumor Analysis

Male rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 6000 ppm dose group with the controls, for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, all at $p < 0.01$. There were significant differences in the pair-wise comparisons of the 600 ppm dose group with the controls for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, both at $p<0.05$. The statistical analyses of the male rats were based upon Peto's Prevalence Test (Table 3).

Female rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 6000 ppm dose group with the controls, for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, all at $p < 0.01$. There were significant differences in

the pair-wise comparisons of the 600 ppm dose group with the controls for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, and of the 60 ppm dose group with the controls for thyroid follicular cell adenomas and/or adenocarcinomas combined, all at $p<0.05$. The statistical analyses of the female rats were based upon Fisher's Exact test for pair-wise comparisons and the Exact Test for trend (Table 4).

Table 3. BAS 670H - Wistar Rat Study (MRID 45902222)
Male Thyroid Follicular Cell Tumor Rates+
Peto's Prevalence Test Results

	Dose (ppm)				
	0	6	60	600	6000
Adenomas (%)	8/49 (16)	12/50 (24)	13/50 (26)	18[a]/50 (36)	23/49 (47)
p =	0.00090**	0.14212	0.12539	0.01469*	0.00048**
Adenocarcinomas (%)	2/47 (4)	1/46 (2)	1/46 (2)	1/46 (2)	2[b]/42 (5)
p =	0.32032	0.91559	0.68664	0.68664	0.65578
Combined (%)	10/49 (20)	13/50 (26)	13[c]/50 (26)	19/50 (38)	23[d]/49 (47)
p =	0.00271**	0.28477	0.25741	0.02786*	0.00220**

+Number of tumor bearing animals/Number of animals examined, excluding those that died before observation of the first tumor.

[a]First adenoma observed at week 80, dose 600 ppm.
[b]First carcinoma observed at week 98, dose 6000 ppm.
[c]One animal in the 60 ppm dose group had both an adenoma and a carcinoma.
[d]Two animals in the 6000 ppm dose group had both an adenoma and a carcinoma.

Note: Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

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Table 4. BAS 670H - Wistar Rat Study (MRID 45902222)
Female Thyroid Follicular Cell Tumor Rates+
Fisher's Exact Test and Exact Trend Test Results

	Dose (ppm)				
	0	6	60	600	6000
Adenomas (%)	2/50 (4)	4/50 (8)	7/50 (14)	8/50 (16)	13[a]/50 (26)
p =	0.00208**	0.33887	0.07975	0.04582*	0.00191**
Adenocarcinomas (%)	0/50 (0)	0/50 (0)	2[b]/50 (4)	2/50 (4)	0/50 (0)
p =	0.3872	1.00000	0.24747	0.24747	1.00000
Combined (%)	2/50 (4)	4/50 (8)	9/50 (18)	10/50 (20)	13/50 (26)
p =	0.00694**	0.33887	0.02556*	0.01387*	0.00191**

+Number of tumor bearing animals/Number of animals examined, excluding those that died before week 53.

[a]First adenoma observed at week 79, dose 6000 ppm.
[b]First carcinoma observed at week 100, dose 60 ppm.

Note: Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Historical Control Data

Historical control data from one in-house 24 months study conducted in 2002 showed that rates for thyroid follicular adenoma in Wistar rats are 28% for males and 10% for females, and the historical control rates for thyroid follicular adenocarcinomas are 4% for males and 0% for females.

C. Non-neoplastic Findings

The incidence of selected non-neoplastic lesions is shown in the Table 5 below extracted from the DER. Non-neoplastic microscopic pathology revealed: (i) focal follicular cell hyperplasia in the thyroid gland in all treated male groups (46-64% treated vs 36% controls) and female groups (26-56% treated vs 16% controls); (ii) minimal to slight diffuse follicular cell hypertrophy in the

thyroid gland in all treated male groups (42-76% treated vs 32% controls) and female groups (28-58% treated vs 22% controls); (iii) minimal to severe loss of sperm in the epididymides of all treated male groups (10-18% treated vs 4% controls); (iv) minimal to marked chronic keratitis in the ≥60 ppm males (16-82% treated vs 0% controls) and females (14-88% treated vs 0% controls); (v) minimal to severe hematopoiesis in the spleen in ≥60 ppm males (38-54% treated vs 32% controls); and (vi) minimal to marked diffuse degeneration in the pancreas in ≥600 ppm males (46-66% treated vs 0% controls) and ≥60 ppm females (10-38% treated vs 0% controls).

Table 5. Incidence of selected microscopic lesions in rats treated with BAS 670H for up to 728 days. [a]

Microscopic lesion	Dose (ppm)				
	0	6	60	600	6000
Males (n=50, except as noted)					
Thyroid gland,					
focal follicular cell hyperplasia	18 (36%)	23 (46%)	25 (50%)	32 (64%)	27 (54%)
Diffuse follicular cell hypertrophy	16 (32%)	21 (42%)	31 (62%)	30 (60%)	38 (76%)
Genital organs,					
Epididymides, loss of sperm	2 (4%)	5 (10%)	6 (12%)	7 (14%)	9 (18%)
Testes, degeneration, diffuse	3 (6%)	4 (8%)	6 (12%)	5 (10%)	9 (18%)
Seminal vesicles, Atrophy	2 (4%)	3 (6%)	6 (12%)	6 (12%)	12 (24%)
Prostate, Atrophy	0	0	2 (4%)	1 (2%)	7 (14%)
Skin, ulcerative inflammation	16(33%)	21 (81%)[b]	24(77%)[b]	27 (87%)[b]	28 (56%)
Spleen, hematopoiesis (total)	16 (32%)	13 (26%)	19 (38%)	25 (50%)	27 (54%)
Pancreas, diffuse degeneration (total)	0	0	1 (2%)	23 (46%)	33 (66%)
Liver, Focal necrosis	4 (8%)	5 (10%)	8 (16%)	7 (14%)	10 (20%)
Centrilobular hypertrophy	0	0	0	0	14 (28%)
Bone marrow femur activation,	7 (14%)	9 (18%)	13 (26%)	9 (18%)	23 (46%)
Bone marrow Sternum activation,	4 (8%)	9 (18%)	10 (20%)	9 (18%)	14 (28%)
Sciatic nerves, focal degeneration	10 (20%)	2 (33%)[b]	2 (25%)[b]	0(0%)[b]	19 (38%)
Pituitary, pars distalis hyperplasia	8 (16%)	1(13%)[b]	2 (11%)[b]	1(11%)[b]	17 (34%)
Adrenal cortices, accessory adrenal tissue	6 (12%)	2 (18%)[b]	3 (30%)[b]	4 (33%)[b]	15 (30%)
Adrenal cortices,Focal fatty change	14 (28%)	5 (45%)[b]	3 (30%)[b]	7 (58%)[b]	21 (42%)
Eyes, chronic keratitis (total)	0	0	8 (16%)	32 (64%)	41 (82%)
Females (n=50, except as noted)					
Thyroid gland, focal follicular cell hyperplasia	8 (16%)	13 (26%)	22 (44%)	28 (56%)	26 (52%)
Diffuse follicular cell hypertrophy	11 (22%)	14 (28%)	17 (34%)	28 (56%)	29 (58%)

(Table continues next page)

Microscopic lesion	Dose (ppm)				
	0	6	60	600	6000
Eyes, chronic keratitis (total)	0	0	7 (14%)	36 (72%)	44 (88%)
Pancreas, diffuse degeneration (total)	0	0	5 (10%)	10 (20%)	19 (38%)
Liver, Focal necrosis	4 (8%)	7 (14%)	6 (12%)	8 (16%)	10 (20%)
Pigment storage	4 (8%)	3 (6%)	5 (10%)	4 (8%)	10 (20%)
Pituitary, pars distalis hyperplasia	5 (10%)	8 (21%)[b]	8 (27%)[b]	11 (28%)[b]	13 (26%)
Bone marrow activation, femur	5(10%)[d]	7 (41%)[b]	4 (40%)[b]	2 (29%)[b]	11 (22%)
Adrenal cortices, accessory adrenal tissue	4 (8%)	3 (14%)[b]	3 (18%)[b]	3 (23%)[b]	9 (18%)
Sciatic nerves, focal degeneration	7 (14%)	2 (12%)[b]	1(10%)[b]	2 (25%)[b]	16 (32%)

a Data (n=50, except as noted) were obtained from pages 48-51 and 185-206 of MRID 45902222. Severity data is reported as grades 1-5 in the individual data and minimal to marked in the results section. Although a code explanation was not provided, it was assumed by the reviewers that grades 1-4 corresponded to minimal to marked, respectively. Grade 5 was referred to as *severe* in this table. For clear presentation, severity data is not presented in this table for treatment-related effects that occurred only at 6000 ppm.

b Reported as # affected/# examined; the percentage affected may not be reflective of the entire group.

c Reported as # affected

d n=49

D. Adequacy of Dosing for Assessment of Carcinogenicity

In male rats, dosing was considered adequate and not excessive based on increased mortality at the high dose, clinical observations and histopathological findings. Increased mortality was observed at the high dose (6%, 12%, 16%, 16%, or 24% for doses 0, 6, 60, 600, or 6000 ppm, respectively); however, the mortality did not exceed the guideline requirement. Decreased body weight (↓6-9%) and body weight gains (↓7-11%) were observed at doses ≥60 ppm throughout the study. Dose-dependent increases of corneal opacity were observed at doses ≥60 ppm. Histopathological findings showed dose-dependent increases of incidence in thyroid (follicular cell hyperplasia), pancreas (diffuse degeneration), liver (focal necrosis), and eyes (chronic keratitis) at doses ≥60 ppm.

In female rats, dosing was considered adequate and not excessive based on slightly decreased body weight, body weight gains, clinical observation, and histopathological findings. Slight decreases of body weights (↓3-8%) and body weight gains (↓7-10%) were observed at doses ≥600 ppm throughout the study. Dose-dependent increases of corneal opacity were observed at doses ≥60 ppm. Histopathological findings showed dose-dependent increases of incidence in thyroid (follicular cell hyperplasia), pancreas (diffuse degeneration), liver (focal necrosis), and eyes (chronic keratitis) at doses ≥60 ppm.

2. Carcinogenicity Study in Mice

Reference: Mellert, W., K. Deckardt, K. Küttler, et al. (2002) BAS 670H: carcinogenicity study in C57BL mice: administration in the diet for 18 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: 76C0124/98112, December 6, 2002. MRID 45902221. Unpublished.

A. Experimental Design

C57BL/6J Rj mice (50/sex/dose) were exposed to BAS 670H (95.2-95.8%, a.i.) in the diet at concentrations of 0, 80, 800, or 8000 ppm nominally (equivalent to 0/0, 19/26, 194/256, and 1903/2467 mg/kg/day in males/females) for up to 78 weeks.

B. Discussion of Tumor Data

Survival Analysis

There were no statistically significant incremental changes in mortality with increasing doses of BAS 670H in male mice (Table 6). Female mice showed a significant increasing trend in mortality at $p < 0.01$ with increasing doses of BAS 670H, as well as a significant difference in the pair-wise comparison of the 8000 ppm dose group with the controls at $p < 0.05$ (Table 7). The statistical evaluation of mortality was based upon the Thomas, Breslow and Gart computer program.

Table 6. BAS 670H - C57BL/6JRj Mouse Study (MRID 45902221)
Male Mortality Rates[+]
Cox or Generalized K/W Test Results

Dose (ppm)	Weeks			
	1-26	27-52	53-79	Total
0	1/50	1/49	1/48	3/50 (6)
80	0/50	1/50	2/49	3/50 (6)
800	1/50	1/49	0/48	2/50 (4)
8000	1/50	1/49	1/48	3/50 (6)

[+]Number of animals that died during interval/Number of animals alive at the beginning of the interval.
[f]Final sacrifice at week 79.
( )Percent.

Note: Time intervals were selected for display purposes only.
Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Table 7. BAS 670H - C57BL/6JRj Mouse Study (MRID 45902221)
Female Mortality Rates[+]
Cox or Generalized K/W Test Results

Dose (ppm)	Weeks			
	1-26	27-52	53-79	Total
0	1/50	0/49	3/49	4/50 (8)**
80	2/50	0/48	2/48	4/50 (8)
800	1/50	1/49	1/48	3/50 (6)
8000	5/50	0/45	6/45	11/50 (22)*

[-]Number of animals that died during interval/Number of animals alive at the beginning of the interval.
[f]Final sacrifice at week 79.
( )Percent.

Note: Time intervals were selected for display purposes only.
Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Tumor Analysis

There were no significant compound-related tumors in male mice though tumor tables are provided for lymphomas and histiocytic sarcomas of the hemolymphoreticular system for comparison with the females. The statistical analyses of the male mice were based upon Fisher's Exact test for pair-wise comparisons and the Exact Test for trend (Table 8).

Female mice had a significant increasing trend in histiocytic sarcomas of the hemolymphoreticular system at $p < 0.01$. There were no significant differences in the pair-wise comparisons of the dosed groups with the controls. The statistical analyses of the female mice were based upon Peto's Prevalence Test (Table 9).

Table 8. BAS 670H - C57BL/6J Rj Mouse Study (MRID 45902221)
Male Hemolymphoreticular System Tumor Rates+
Fisher's Exact Test and Exact Trend Test Results

	Dose (ppm)			
	0	80	800	8000
Lymphomas (%)	2[a]/48 (4)	4[a]/49 (8)	4[a]/48 (8)	4[a]/48 (8)
p =	0.3128	0.34863	0.33865	0.33865
Histiocytic Sarcomas (%)	0[b]/48 (0)	1[b]/49 (2)	2[b]/48 (4)	3[b]/48 (6)
p =	0.05345	0.50515	0.24737	0.12105

+Number of tumor bearing animals/Number of animals examined, excluding those that died before week 53.
[a]First lymphoma observed at week 79, simultaneously in final sacrifice animals, at all dose groups.
[b]First histiocytic sarcoma observed at week 79, simultaneously in final sacrifice animals, at all dose groups.

Note: Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Table 9. BAS 670H - C57BL/6J Rj Mouse Study (MRID 45902221)
Female Hemolymphoreticular System Tumor Rates+
Peto's Prevalence Test Results

	Dose (ppm)			
	0	80	800	8000
Histiocytic Sarcomas (%)	3/48 (6)	0/47 (0)	1/48 (2)	7[a]/44 (16)
p =	0.00276**	0.96006	0.85051	0.12236

+Number of tumor bearing animals/Number of animals examined, excluding those that died before week observation of the first tumor..
[a]First histiocytic sarcoma observed at week 62, dose 8000 ppm.

Note: Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

Historical Control Data

BASF provided historical control data for the mouse oncogenicity study with BAS 670 H for the finding "lymphoma, malignant" and "histiocytic sarcoma" in both sexes of C57BL mice. From historical control data at the conducting laboratory (BASF), nine 18-month carcinogenicity studies are available that show data from the years 1991 up to 2001. The mean (and range) for malignant lymphomas was 12.2% (0-26%) for males and 21.5% (4-54%) for females. For histiocytic sarcomas, the mean (and range) was 9.6% (2-32%) for males and 15.6% (3-42%) for females.

C. Non-Neoplastic Liver Lesions

An increased incidence of central hepatocellular hypertrophy was observed in the males as follows: controls (6%), and 80 (2%), 800 (22%), and 8000 (38%) ppm. Other differences in incidence of histological lesions were minor.

D. Adequacy of Dosing for Assessment of carcinogenicity

In males, dosing was considered adequate and not excessive even the high dose was exceed the limit dose based on decreased body weight and body weight gain, increased relative liver weight and increased incidence of hepatocellular hypertrophy in males.

In females, dosing was considered adequate and not excessive although the high dose was above 2x the limit dose. Increased mortality was observed at the high dose; however, no significant decreases in body weight and body weight gains, or histopathological findings were observed.

IV. TOXICOLOGY

1. Metabolism

In a rat metabolism study (MRIDs 45902305 and 45902306), [^{14}C]-BAS 670H in 0.5% aqueous Tylose or Cremophor EL/CMC was administered to Wistar rats by gavage. In an initial plasma kinetics studies, a single oral dose of [pyrazole-4-^{14}C]-BAS 670H (Batch # 706-1013; radiochemical purity of >= 98%) was administered to 4 Wistar rats/sex/dose at nominal doses of 10, 100, 200, 400, or 500 mg/kg. In the main mass balance/excretion/metabolism studies, 4 rats/sex/dose were given [pyrazole-4-^{14}C]-BAS 670H as a single oral dose of 10 or 300 mg/kg, a repeated oral dose of 300 mg/kg (14 days unlabeled + 1 day radiolabeled), or 300 mg/kg [phenyl-U-^{14}C]-BAS 670H (Batch #714-1026; radiochemical purity of >= 98%). Tissue distribution time course and biliary excretion studies were also performed. Metabolites were identified and quantified in the urine, feces, bile, kidney, and liver in the main studies at 10 and 300 mg/kg. In an additional study, metabolite profiles were determined in urine and feces of rats given a single oral dose of 500 mg/kg.
Absorption of [^{14}C]-BAS 670H following a single oral dose was rapid but limited, with the highest plasma concentrations observed at 1 hour (first time point measured). In the 10 mg/kg group, a second smaller peak in plasma concentration occurred at 8 hours post-dose. Plasma concentrations declined bi-phasically at 10 and 100 mg/kg and tri-phasically at >= 200 mg/kg. The change in AUC was proportional to dose in both sexes at <= 200 mg/kg and overproportional with dose at 400 and 500 mg/kg.

In the main mass balance/excretion studies, 94-103% of the dose was recovered after 168 hours, with <= 0.12% dose remaining in the tissues and <0.1% of the dose in exhaled air. The majority of the dose was recovered within 48 hours in the feces (73-91% dose) and urine (8-29% dose). In a separate experiment, bile was collected for up to 48 hours from rats and accounted for 19-32% dose at 10 mg/kg and 7-9% dose at 300 mg/kg. The pattern of excretion was similar between the sexes and dose groups, although urinary excretion was higher in the low dose groups than in the high dose groups. Urinary excretion was also higher in females, while biliary excretion was higher in males.

At 168 hours post-dose, concentrations of radioactivity remaining in the tissues were generally similar between the sexes and across the dose groups. Concentrations were highest in liver and kidneys of all groups, and in the thyroid of rats from the single 300 mg/kg [^{14}C-pyrazole] dose group. Concentrations in the thyroid were also generally above levels in the blood for the other 300 mg/kg dose groups, but not the 10 mg/kg group. For the two groups dosed at 300 mg/kg

with different ^{14}C-labels, concentrations in the adrenal glands, ovaries, uterus, bone marrow, and pancreas of the [^{14}C-phenyl] group were higher than in the [^{14}C-pyrazole] group, suggesting some differential distribution of metabolites. Repeated dosing at 300 mg/kg had no effect on the concentration of radioactivity in the tissues.

Similar findings in the relative distribution of radioactivity in tissues were observed in the time course study. Concentrations in the kidneys and liver were higher than in blood in both sexes at all time points in both the 10 and 300 mg/kg groups. Compared to levels in the blood, radioactivity was also higher in the ovaries and uterus beginning at 1 hour in the 10 mg/kg rats and beginning at 8 hours in the 300 mg/kg rats. In the thyroid, increases over blood levels were observed transiently in the low dose at 8 hours and consistently in the high dose beginning at 2 hours.

Radio-HPLC identified and quantified parent and up to four metabolites (M670H01, M670H02, M670H05, and M670H13) in the urine, feces, bile, liver, and kidney, and the identity of each compound was confirmed by LC/MS, LC/MS/MS, and /or NMR analysis. In the main study groups, parent and identified metabolites in excreta accounted for 91.8-104.5% dose, while unidentified compounds accounted for <1% dose.

In all groups, parent was the predominant compound identified in both urine (4.0-21.3% dose) and feces (66.3-91.7% dose). The primary metabolites in urine were M670H02 (1.0-5.3% dose) and M670H01 (0.2-1.2% dose), along with minor amounts (<0.5% dose) of M670H05 and M670H13. Metabolites identified in feces included M670H02 (1.3-3.1% dose) and M670H01 (0.6-6.7% dose). In the bile, parent was again the predominant compound identified, accounting for 3.4-13.7% of the dose, along with minor amounts of M670H02 (2.2-12.1% dose), M670H01 (0.2-1.3% dose), and M670H13 (<0.5% dose). In liver and kidneys sampled at T_{max} (1 hour) from both 10 and 300 mg/kg dose groups, the major compound identified was parent, accounting for 48.3-82.5% of the total radioactive residues (TRR), along with M670H02 (14.5-34.6% TRR) and M670H01 (0.9-14.0% TRR).

Regardless of sex, dose level, and the position of the ^{14}C-label, the overall metabolism of [^{14}C]BAS 670H in rats was similar. The ^{14}C-dose was excreted primarily as unchanged parent (82.7-98.3% dose), which was recovered primarily in the feces and to a lesser extent in the urine. Biotransformation of ^{14}C-BAS 670H was limited and primarily involved oxygenation of the isoxazole ring to form M670H02 (1.0-5.3% dose) and subsequent ring opening and loss of the acetic acid moiety to yield M670H01 (0.4-7.9% dose). A minor fraction of parent (<1% dose) was also hydrolyzed at methanone bridge to yield M670H13 and M670H05. The proposed pathway for biotransformation of [^{14}C]BAS 670H in rats is presented in Figure 1.

This metabolism study in the rat is classified **acceptable/guideline** and satisfies the guideline requirement for a Tier 1 metabolism study [OPPTS 870.7485, OPP 85-1] in rats.

2. Mutagenicity

BAS 670 H was not mutagenic in *Salmonella typhimurium* or *Escherichia coli* in a series of reverse mutation assays using the standard plate incorporation and preincubation protocols. Negative results were obtained with three different batches of the test material, and purity ranged from 97.7 to 99.3% (MRID 45902225 through 45902227). By contrast, a single batch (N 26) of the test material induced a reproducible and concentration-dependent mutagenic response in the nonactivated phase of plate and preincubation tests with *S. typhimurium* TA98 (MRID 45902228). This finding should be viewed with caution for the following reasons: 1) the response was seen at high concentrations including and exceeding the limit dose (2500-7000 µg/plate); 2) the batch of BAS 670 H used in this study had the lowest percentage active ingredient (95.8% vs. 97.7-99.3 % ai for the other batches) and 3) microbial tests performed with the three batches mentioned above were negative up to the limit dose. Therefore, the mutagenic effect was likely due to impurities in the test article. BAS 670 H was also not mutagenic in independently performed gene mutation assays in Chinese hamster ovary (CHO) cells up to insoluble and cytotoxic concentrations (MRID 45902230).

There was, however, evidence of concentration-related and reproducible clastogenicity in two trials of an initial *in vitro* chromosome aberration assay with Chinese hamster lung (V79) (MRID 45902232) and in a confirmatory assay performed 3 years later (MRID 45902233). Purity for both batches (Batches N 14 and 30786/22) used in these assays was 97.7 and 99.3%, respectively. In all cases, BAS 670 H was positive up to S9-activated levels that were insoluble but not cytotoxic; the most frequently observed aberration was exchanges. In contrast to the positive findings from the *in vitro* assays for chromosome damage, BAS 670 H (Batch N 14, 97.7%) was neither clastogenic nor aneugenic when tested in excess of the limit dose in the bone marrow of NMRI mice (MRID No. 45902234). Although the Registrant claimed that the results of a pharmacokinetic study indicate that the test material reaches the bone marrow, it was noted that the pharmacokinetic study in question was conducted in rats not in mice. Therefore, no link can be made between the pharmacokinetic data and the micronucleus results. Nevertheless, BAS 670H tested negative in excess of the limited dose. BAS 670 H was also negative for induction of unscheduled DNA synthesis in primary rat hepatocytes (MRID 4590302).

It was concluded that BAS 670 H induced a clastogenic response in cultured mammalian cells but this activity is not expressed in whole animals. The lack of a clastogenic effect in vivo casts doubts on the relevance of the in vitro response to a possible mutagenic mode of action for BAS 670H. Consequently, there is no concern for mutagenicity. The submitted assays satisfy the FIFRA guidelines for mutagenicity testing. No additional testing is required at this time.

Within the mutagenicity package, two studies were found on test material Reg. No. 388 010 which has a different CAS number than BAS 670 H technical. Results for these studies were negative for gene mutations in Chinese hamster lung (V79) cells up to the limit concentration

(MRID 45902231) and negative for micronucleus induction in the bone marrow of NMRI mice up to an intraperitoneal dose that represents 80% of the lethal dose (MRID 45902301).

MUTAGENICITY STUDIES WITH BAS 670 H TECHNICAL		
GENE MUTATIONS–BACTERIA		
Test System	MRID (year) Purity Batch No.	Result
870.5100 Bacterial Gene Mutation Assay(plate or preincubation) *Salmonella typhimurium, Escherichia coli*	45902225 (1999) 97.7% N 14 Acceptable/guideline 20-5000 µg/plate - /+ S9 or 4-2500 µg/plate - /+ S9	**Negative** up to the highest dose tested (5000 µg/plate -/+ S9-**plate** or 2500 µg/plate - / + S9-**preincubation**) which was also cytotoxic.
870.5100 Bacterial Gene Mutation Assay(plate or preincubation) *S. typhimurium, E. coli*	45902226 (2002) 99.3% 30786/22 Acceptable/guideline 20-5000 µg/plate - /+ S9 or 4-2500 µg/plate - /+ S9	**Negative** up to the highest dose tested (5000 µg/plate- /+ S9-**plate** or 2500 µg/plate - / + S9-**preincubation**) which was also cytotoxic.
870.5100 Bacterial Gene Mutation Assay(plate or preincubation) *S. typhimurium, E. coli*	45902227 (2002) 98.8% N 33 Acceptable/guideline 20-5000 µg/plate - /+ S9 or 4-2500 µg/plate - /+ S9	**Negative** up to the highest dose tested (5000 µg/plate -/+ S9-**plate** or 2500 µg/plate - / + S9-**preincubation**) which was also cytotoxic.

870.5100 Bacterial Gene Mutation Assay(plate or preincubation) *S. typhimurium, E. coli*	45902228 (2003) 95.8% N 26 Acceptable/guideline 20-5000 µg/plate - /+ S9 (plate) 4-2500 µg/plate - /+ S9 (pre-incubation) 3000-7000 µg/plate - S9 (repeat: plate: 2 trials; pre-incubation: 1 trial	Cytotoxic at 5000 µg/plate - S9; ≥2500 µg/plate + S9 **(plate)** 2500 µg/plate-S9; ≥500 µg/plate + S9 **(preincubation)**. **Positive :** ~1.4- 1.5 x ↑ mutant colonies of TA98 at 2500 & 5000 µg/plate - S9 **(plate)** ~1.7 x ↑ mutant colonies TA98 at 2500 µg/plate - S9 **(preincubation)** ~1.9-5.2 x ↑ mutant colonies TA98 at 3000-7000 µg/plate - S9 **(repeat plate)**. Cytotoxic at ≥4000 µg/plate- S9; ~1.6 x ↑ mutant colonies TA98 at 3000 µg/plate - S9 **(repeat preincubation)**.

MUTAGENICITY STUDIES WITH BAS 670 H TECHNICAL		
GENE MUTATIONS–BACTERIA		
Test System	MRID (year) Purity Batch No.	Result
870.5100 Bacterial Gene Mutation Assay(plate or preincubation) *S. typhimurium, E. coli*	45902229 (2001) 99.3% N 3 **Reg. No. 388 010** Acceptable/guideline 10-5000 µg/plate - /+ S9 or 2-500 µg/plate - /+ S9	**Negative** up to cytotoxic levels (5000 µg/plate - S9; ≥300 µg/plate + S9-**plate** or ≥250 µg/plate - /+ S9-**preincubation**).
GENE MUTATIONS– MAMMALIAN CELLS		
870.5300 *In vitro* mammalian cell gene mutation Chinese hamster ovary (CHO) cells	45902230 (2000) 95.8% N 26 Acceptable/guideline 93.75- 3000 µg/mL -/+S9 (30%) Trial 1 93.75- 3000 µg/mL -S9 78.13- 2500 µg/mL +S9 (10%) Trial 2	**Negative** up to insoluble & cytotoxic levels (≥3000 µg/mL -/+ S9).
870.5300 *In vitro* mammalian cell gene mutations Chinese hamster lung V79 cells	45902231 (2001) 99.3% N 3 **Reg. No. 388 010** Acceptable/guideline 175-2800 µg/mL -/+S9 (10 or 30%)	**Negative** up to the limit concentration (2800 µg/mL, equiv to10 mM).

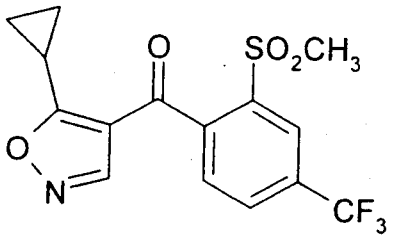
MUTAGENICITY STUDIES WITH BAS 670 H		
CHROMOSOME ABERRATIONS –*IN VITRO*		
870.5375 Cytogenetics *In vitro* mammalian cell chromosomal aberration assay Chinese hamster lung (V79) cells	45902232 (1999) 97.7% N 14 Acceptable/guideline 225-3600 µg/mL -/+S9 Trial 1 1800-3600 µg/mL +S9 Trial 2	**Negative** up to insolubility w/o cytotoxicity at 3600 µg/mL -S9. **Positive** S ↑ in the percent of cells w aberrations and exchanges) at 3600 µg/mL +S9 (Trial 1). S↑ in the percent of cells w aberrations and exchanges at 3600 µg/mL +S9 (Trial 2); NS conc.-related ↑ also seen at 1800 and 2700 µg/mL +S9. Confirmed in a later study (MRID 45902233).
870.5375 Cytogenetics *In vitro* mammalian cell chromosomal aberration assay Chinese hamster lung (V79) cells	45902233 (2002) 99.3% 30786/22 Acceptable/guideline 900-3600 µg/mL -/+S9	**Negative** up to insolubility w/o cytotoxicity at ≥1800 µg/mL -S9. **Positive** S ↑ in the percent of cells w aberrations and exchanges at 3600 µg/mL +S9; NS conc.-related ↑ also seen at 2700 µg/mL +S9. Confirmed in an earlier study (MRID 45902232).
CHROMOSOME ABERRATIONS –*IN VIVO*		
870.5395 Mammalian Erythrocyte Micronucleus Test NMRI Mice	45902234 (1999) 97.7% N 14 Acceptable/guideline 0, 370, 630 1130 mg/kg x 2 days, ip	**Negative** up to a dose (1130 mg/kg x 2) in excess of the limit dose (2000 mg/kg).
870.5395 Mammalian Erythrocyte Micronucleus Test NMRI Mice	45902301 (2001) 99.3% N 3 **Reg. No. 388 010** Acceptable/guideline 0, 200, 400 800 mg/kg, ip	**Negative** up to an acceptable high dose (800 mg/kg) that represents 80% of the lethal dose.

MUTAGENICITY STUDIES WITH BAS 670 H		
OTHER MUTAGENIC MECHANISMS		
870.5550 Other Genotoxicity *In vitro* UDS in Primary Rat Hepatocytes	45902302 (1999) 97.7% N 14 Acceptable/guideline 10-5000 µg/mL (Trial 1) 78.125-2500 µg/mL (Trial 2)	**Negative** up to cytotoxic concentrations (≥3750µg/mL). Compound precipitation was seen at 5000 µg/mL.

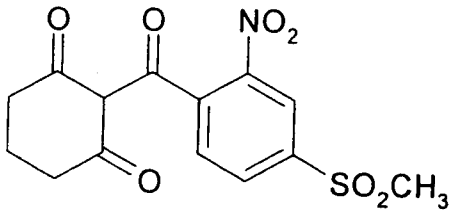
S= Significant ($p<0.01$)
NS= Not significant

3. Structure-Activity Relationship

BAS 670H belongs to a chemical class of triketone (e.g., mesotrione). It is an inhibitor of the enzyme 4-hydroxyphenylpyruvate dioxygenase (HPPD) and it shares this property with mesotrione and isoxaflutole. Literature searches conducted on triketone did not show evidence of carcinogenicity or mutagenicity. Isoxaflutole was characterized as "likely to be a human carcinogen" based on increases in liver tumors in both sexes of mice and rats, and increases in thyroid follicular cell tumor in male rats. The CARC determined that there are too few chemicals and too little structural similarity to draw any firm conclusion toward the structure-activity relationship.



Isoxaflutole



Mesotrione

4. Subchronic Toxicity

a. Subchronic Toxicity in Rats

In a subchronic oral toxicity study (MRIDs 45902204 and 45902203), BAS 670H (99.3% a.i., Batch # 30786/22) was administered to 10 Wistar rats/sex/dose in the diet at dose levels of 0, 15, or 30 ppm (equivalent to 0/0, 1.1/1.3, and 2.1/2.5 mg/kg/day in males/females) for 13 weeks.

No treatment-related effect was observed on mortality, clinical signs, body weight, body weight gain, food consumption, food efficiency, ophthalmology, hematology, urinalysis, organ weights, or gross pathology.

A minimal diffuse degeneration was observed in the pancreas of the 30 ppm males (2/10 treated vs 0/10 controls). The Sponsor stated that the lesions most resembled a chronic interstitial pancreatitis morphologically. This lesion was also observed in a subchronic toxicity and neurotoxicity study (MRID 45902201). Minimal to moderate flaky colloid was observed (vs 0/10 controls) in ≥15 ppm males (9-10/10) and females (1-3/10). There was no other histological thyroid lesion and no evidence of impairment of function; therefore, this effect was not considered adverse.

In the 30 ppm males, cholesterol was increased ($p \leq 0.01$) by 28%; and alkaline phosphatase was decreased ($p \leq 0.01$) by 21%. Corroborating evidence of adverse effect was absent for both parameters.

The LOAEL for males is 30 ppm (equivalent to 2.1 mg/kg/day), based on diffuse degeneration in the pancreas. The NOAEL is 15 ppm (equivalent to 1.1 mg/kg/day). The LOAEL for females was not established, the NOAEL is 30 ppm (2.5 mg/kg/day).

This study is classified as **acceptable/guideline** and satisfies the guideline requirements (OPPTS 870.3100a; OECD 408) for a subchronic oral toxicity study in the rat.

b. Subchronic Toxicity Study in Mice

In a subchronic oral toxicity study (MRID 45902202), BAS 670H (97.7% a.i., Batch/Lot #: N14) was administered to C57BL mice (10/sex/dose) in the diet at doses of 0, 125, 1000, or 8000 ppm (equivalent to 0/0, 37/51, 288/406, or 2289/3010 mg/kg/day [M/F]) for up to 91 days.

There were no adverse treatment-related effects observed on mortality, clinical signs, body weight, body weight gain, food consumption, hematology, clinical chemistry, or gross or microscopic pathology at any dose in either sex. No serum tyrosine level was measured.

Increased relative liver weight was observed in females only at 1000 and 8000 ppm; however, there was no corroborative histopathological evidence for this finding. Decreased testes weights were observed in males at 125 and 1000 ppm. There was no dose-dependent responses and was considered incidental. Gross pathology showed increased incidence of erosion/ulcer in the glandular stomach in females at 1000 and 8000 ppm. However, the microscopic evaluation of the stomach showed that these erosions/ulcers also occurred with equal frequency in the control group and was not considered treatment-related.

Under conditions of this study, the LOAEL was not observed. The NOAEL is 8000 ppm (equivalent to 2289/3010 mg/kg/day in M/F).

This study is classified **acceptable/guideline** and satisfies the guideline requirements (OPPTS 870.3100; OECD 408) for a 90-day oral toxicity study in the mouse.

c. Subchronic Toxicity Study in Dogs

In a subchronic oral toxicity study (MRID 45902205), BAS 670H (95.2-95.8% a.i., Batch/Lot #s: N17 & N26) was administered to Beagle dogs (5/sex/dose) in a diet at doses of 0, 3000, 9000, or 25,000 ppm (equivalent to 0/0, 182/205, 535/624, or 1511/1712 mg/kg/day [M/F]) for up to 90 days.

No adverse treatment-related effects were observed on mortality, clinical signs, food consumption, hematology, clinical chemistry, gross or histopathology parameters.

Decreased body weight gains were observed in males at 25,000 ppm throughout treatment with the terminal body weight decreased by 10% ($p \leq 0.05$). Food efficiency was decreased by 28-338% compared to controls in the 25000 ppm males throughout the study. No significant differences in body weights were observed at any dose in females; decreased body weight gains were noted at all doses; however, no dose response was seen.

Urinalysis showed increased ketone level in the urine of all treated animals of both sexes. This finding may be a false positive due to excretion of p-hydroxyphenylpyruvic acid (a keto-acid) which interferes with the reagent of the test strip. Increased incidence of crystal (identified as magnesium complex of the parent compound) was seen in urine sediments of 25000 ppm males.

Absolute brain weight was decreased ($p \leq 0.01$) by 15-16% in the $\geq$9000 ppm females; however, the relative brain weight were comparable in all doses. In addition, as there was no corroborative histopathological evidence in the brains of these animals, this finding is of equivocal toxicological importance. Histopathology revealed inflammation in the urinary bladder of two male dogs at the 25000 ppm. All other histopathological findings were either incidentally as single case or were equally distributed over the dose groups.

The NOAEL for males is 9000 ppm (equivalent to 535 mg/kg/day), and the LOAEL is 25,000 ppm (equivalent to 1511 mg/kg/day) based on decreased body weight gain, impaired food efficiency, and inflammation of the urinary bladder. The NOAEL for females is 25000 ppm (equivalent to 1712 mg/kg/day), the LOAEL for females is not established.

This study is classified as **acceptable/guideline** and satisfies the guideline requirements (OPPTS 870.3150; OECD 409) for a 90-day oral toxicity study in the dog.

5. Chronic Toxicity

1. Chronic Toxicity Study in Rats

In a chronic toxicity study (MRID 45902217), 20 Wistar-(CrlGlxBrlHan:WI) rats/sex/dose were exposed to BAS 670H (95.2-95.8 % a.i.; Lot/Batch Nos.: N17 and N26) in the diet at concentrations of 0, 6, 60, 600, or 6000 ppm (equivalent to 0/0, 0.4/0.5, 3.9/5.3, 42.0/53.2, and 422.6/535.0 mg/kg/day in males/females) for up to 364 days.

No treatment-related effect was observed on mortality, body weight, body weight gains, food consumption, food efficiency, hematology, clinical chemistry, urinalysis, or neoplasia.

Clinical observations showed an increased incidence of corneal opacity in the right and/or left eye in the ≥60 ppm groups of both sexes. During ophthalmoscopic examination, an increased incidence of corneal pannus was observed in the ≥60 ppm groups of both sexes. Increased incidence of corneal opacity was observed in the ≥60 ppm females and in the ≥600 ppm males. Incidence and initial time observed was generally dose-dependent. Macroscopic examination showed a dose-dependent increased incidence of cloudiness of the cornea in the≥60 ppm groups of both sexes.

Microscopic examination revealed a dose-dependent increase in incidence and severity of minimal to marked chronic keratitis, which typically corresponded to cloudiness of the cornea, in the ≥60 ppm groups of both sexes. In the thyroid, increased incidence of minimal to slight diffuse hypertrophy and focal follicular cell hyperplasia were observed in the ≥60 ppm groups of both sexes. The thyroid hypertrophy was increased in incidence and severity with dose; however, the hyperplasia was not clearly a dose-dependent effect, and severity was not reported. In the pancreas, a dose-dependent increase in incidence and severity of minimal to moderate diffuse degeneration was observed in ≥600 ppm males.

The LOAEL is 60 ppm (equivalent to 3.9/5.3 mg/kg/day in males/females), based on corneal opacity and pannus and chronic keratitis in both sexes, and thyroid hypertrophy in males. The NOAEL is 6 ppm (equivalent to 0.4/0.5 mg/kg/day in males/females).

This study is classified as **acceptable/guideline** and satisfies the guideline requirements for a chronic oral study [OPPTS 870.4100, OECD 452] in rats.

2. Carcinogenicity Study in Rats

In a carcinogenicity study (MRID 45902222), 50 Wistar rats/sex/dose were exposed to BAS 670H (95.2-95.8% a.i.; Batch/Lot Nos.: N17 and N26) in the diet at concentrations of 0, 6, 60, 600, or 6000 ppm nominally (equivalent to 0/0, 0.4/0.5, 3.6/4.7, 36.4/50.8, and 381.5/524.1 mg/kg/day in males/females) for up to 24 months.

Dose-dependent increase of mortality was observed in males at Week 105: 6 (12%), 60 (16%), 600 (16%), and 6000 (24%) ppm vs 6% in the concurrent controls; however, survival rates are within guideline requirement. No treatment-related effect on mortality was observed in females. Clinical observations showed corneal opacity in animals of both sexes at doses ≥60 ppm (20-88% vs 0% in control). Ophthalmoscopic examination on Days 267 (females) and 282 (males) showed corneal pannus and opacity at ≥600 ppm in both sexes and in 60 ppm females (only animals with clinically observed corneal opacity were examined). On Days 722 (females) and 728 (males), increased incidence of corneal pannus and opacity was observed in the 60 ppm group of both sexes vs 0% in the controls and 6 ppm group (only the controls and the 6 and 60 ppm groups were examined).

Decreased body weight and body weight gains were observed in males at ≥60 ppm, these decreases occurred late in the study, first observed on Day 595 (6000 ppm) and Day 651 (≥60 ppm). Statistically significant decreases of body weight and body weight gains in males at 60 and 600 ppm groups were observed on several days during the study. In females, decreased body weight and body weight gains were observed at 600 and 6000 ppm.

Treatment-related increases ($p \leq 0.01$) of absolute and/or relative organ weight were observed in the following organs: (i) liver in ≥60 ppm males and ≥600 ppm females; (ii) kidneys in ≥60 ppm males and ≥600 ppm females. Gross pathology showed the following treatment-related gross lesions: (i) skin decubitus in all treated male groups (42-56% treated vs 30% controls); (ii) enlarged iliac and popliteal lymph node in all treated male groups (22-50% treated vs 16-18% controls); (iii) cloudiness in the cornea in ≥60 ppm males and females (16-88% treated vs 0% controls); (iv) thyroid gland mass in ≥600 ppm males (18-24% treated vs 4% controls) and enlarged thyroid in the 6000 ppm males (26% treated vs 12% controls); (v) decreases in testes, epididymides, seminal vesicle, and prostate size (10-24% treated, each lesion vs 0-4% controls) at 6000 ppm; and (vi) liver focus in the 6000 ppm females (68% treated vs 38% controls).

Non-neoplastic microscopic pathology revealed: (i) focal follicular cell hyperplasia in the thyroid gland in all treated male groups (46-64% treated vs 36% controls) and female groups (26-56% treated vs 16% controls); (ii) minimal to slight diffuse follicular cell hypertrophy in the thyroid

gland in all treated male groups (42-76% treated vs 32% controls) and female groups (28-58% treated vs 22% controls); (iii) minimal to severe loss of sperm in the epididymides of all treated male groups (10-18% treated vs 4% controls); (iv) minimal to marked chronic keratitis in the ≥60 ppm males (16-82% treated vs 0% controls) and females (14-88% treated vs 0% controls); (v) minimal to severe hematopoiesis in the spleen in ≥60 ppm males (38-54% treated vs 32% controls); and (vi) minimal to marked diffuse degeneration in the pancreas in ≥600 ppm males (46-66% treated vs 0% controls) and ≥60 ppm females (10-38% treated vs 0% controls). In addition, increases were observed in the incidence of minimal to marked ulcerative skin inflammation in males (42-56% treated vs 32% controls) and pars distalis hyperplasia in females (16-26% treated vs 10% controls); however, the exact incidence at 6, 60, and 600 ppm is unknown, because all animals were not examined (incidence calculated as # affected/50 for all groups). Additionally, increased incidences of the following lesions were observed at 6000 ppm: (i) in the liver: focal necrosis in both sexes, hematopoiesis (equivocal) and centrilobular hypertrophy in males, and pigment storage in females; (ii) male reproductive system abnormalities: diffuse degeneration in the testes, seminal vesicles atrophy, and prostate gland atrophy and inflammation; (iii) bone marrow activation in the femur in both sexes and in the sternum of males; (iv) in the adrenal cortices, accessory adrenal tissue in both sexes and a focal fatty change in males; (v) pars distalis hyperplasia in males; (vi) focal degeneration in the sciatic nerves in both sexes; and (vii) mandibular lymph node hyperplasia in males. Because all animals in the 6, 60, and 600 ppm groups were not examined, it was unclear if the following lesions were observed at a higher incidence than controls at ≤600 ppm: adrenal cortices, male pituitary, sciatic nerves, mandibular lymph node, and female bone marrow activation. For the same reason, it could not be confirmed that ulcerative skin inflammation was a dose-dependent effect.

Because all animals in the 6, 60, and 600 ppm groups were not examined, it was unclear if the following lesions were observed at a higher incidence than controls at <6000 ppm: adrenal cortices, male pituitary, sciatic nerves, mandibular lymph node, and female bone marrow activation. For the same reason, it could not be confirmed that ulcerative skin inflammation was a dose-dependent effect.

For systemic toxicity, the LOAEL is 60 ppm (equivalent to 3.6/4.7 mg/kg/day), based on increased incidence of corneal opacity, decreased body weight and body weight gains in males and histopathological findings in the thyroid, pancreas, and eyes. The NOAEL was 6 ppm (equivalent to 0.4/0.5 mg/kg/day).

Neoplastic pathology examination showed treatment-related increases of neoplastic lesions in the thyroid. Increased incidences of follicular cell adenoma were 8/50 (16%), 12/50 (24%), 13/50 (26%), 18/50 (36%), or 23/50 (46%) for males and 2/50 (4%), 4/50 (8%), 7/50 (14%), 8/50 (16%), or 13/50 (26%) for females at 0, 6, 60, 600, or 6000 ppm, respectively with historical controls of 28% in males and 10% in females. Dosing was considered adequate based on numerous indications of toxicity, such as corneal opacity and chronic keratitis in both sexes.

This study is classified as **acceptable/guideline** and satisfies the guideline requirements (OPPTS 870.4200a; OECD 451) for a carcinogenicity study in rats.

3. Carcinogenicity Study in Mice:

In a carcinogenicity study (MRID 45902221), 50 C57BL/6J Rj mice/sex/dose were exposed to BAS 670H (95.2-95.8% a.i.; Batch/Lot Nos.: N17 and N26) in the diet at concentrations of 0, 80, 800, or 8000 ppm nominally (equivalent to 0/0, 19/26, 194/256, and 1903/2467 mg/kg/day in males/females) for up to 78 weeks.

There were no treatment-related effects on clinical signs, food efficiency, ophthalmology, organ weights, or gross and histological pathology.

At ≥80 ppm, generally dose-dependent increases in magnitude and frequency of body weight decreases ($p \leq 0.05$) were observed (decr 2-8%), and corresponded to decreases ($p \leq 0.05$) in body weight gains (decr 9-86%). Although significant decreases ($p <= 0.05$) in body weights in the 80 ppm males were slight in magnitude (decr 2-4%) and less common than at the higher doses, slight decreases (often not statistically significant) were generally observed throughout the study, which contributed to the clearly adverse effect on body weight gain. At 80 ppm, body weight gain was decreased ($p \leq 0.05$) by ≥12% for more than half the study, by 43% on Day 14, and by 17% at termination on Day 546.

At 8000 ppm, other effects were observed that were not clearly treatment-related or the toxicological significance was unclear. Survival was decreased in the females (76% treated vs 92% controls). However, four animals died very early in the study (between Days 19-22) and one died during the necropsy period (Day 552). These deaths may have been incidental; therefore, the effect on mortality is equivocal. Food consumption was generally decreased ($p \leq 0.05$) in the females at Day 7 through Day 119 (decr 9-27%, a transient effect); only sporadic, inconsistent differences ($p \leq 0.05$) were observed thereafter. Morphological variations were observed in the leukocytes of the males, including metamyelocytes, changes in the nucleus of neutrophils, juvenile lymphocytes, and monoblasts; however, the toxicological significance of these findings was unclear.

An increased incidence of central hepatocellular hypertrophy was observed in males only as follows: controls (6%), and 80 (2%), 800 (22%), and 8000 (38%) ppm. There was no increase in absolute liver weight in males; however, minor increases ($p \leq 0.05$) in relative to body liver weights was observed in all treated groups. In females, increased absolute liver weight and relative to body liver weights were observed at 800 and 8000 ppm (↑7-11%); however, no corroborated gross or histological hepatic lesions was observed.

The LOAEL is 80 ppm (equivalent to 19/26 mg/kg/day in males/females), based on decreased body weight and body weight gains in males. The NOAEL was not established.

At the doses tested, there was an increase in tumor incidence in the hemolymphoreticular system when compared to controls. Malignant lymphoma was observed in males (4%, 8%, 8%, or 8% for 0, 80, 800, or 8000 ppm, respectively) and in females (12%, 20%, 14%, or 14% for 0, 80, 800, or 8000 ppm, respectively). No dose-response relationship was found in both sexes. Histiocytic sarcoma was observed in males (0%, 2%, 4%, or 6% for 0, 80, 800, or 8000 ppm, respectively) and in females (6%, 0%, 2%, or 16% for 0, 80, 800, or 8000 ppm, respectively).

Dosing was considered adequate based on decreased body weight and body weight gain in males, increased liver and kidney weight of both sexes and an increased incidence of central hepatocellular hypertrophy in males.

This study is **acceptable/guideline** and satisfies guideline requirement for a carcinogenicity study [OPPTS 870.4200b; OECD 451] in mice.

4. Chronic Toxicity Study in Dogs

In a chronic toxicity study (MRID 45902215), BAS 670H (95.8% a.i., Batch No.: N26) was administered to beagle dogs (5/sex/dose) in the diet at doses of 0, 3000, 9000, or 25,000 ppm for 209 days and 0, 2600, 7800, or 22,000 for study days 210-364 (equivalent to 0/0, 81/92, 248/287, and 688/780 mg/kg/day in males/females over the entire study). Because a NOAEL was not established in the initial study, a supplementary study (MRID 45902216) was performed. The same test material (Batch No.: N26) was administered to 5 beagle dogs/sex/dose in the diet at doses of 0, 100, or 500 ppm (equivalent to 0/0, 2.9/3.1, and 15.3/15.4 mg/kg/day in males/females) for up to 12 months.

Two 25,000 ppm males died, one on Day 137 and the other on Day 280. The cause of death for both dogs were attributed to necrotizing cystitis and postrenal uremia. Observed clinical signs were limited to these two dogs that die. No other treatment-related adverse effects were seen on ophthalmology, food consumption, hematology, and clinical chemistry.

Slight decreases (not statistically significant [NS]) in body weight were frequently observed throughout the studies in males at ≥500 ppm and in females at ≥3000 ppm. This effect was clearly dose-dependent by Day 126 in the initial study and throughout the supplementary study. Cumulative body weight gains were decreased in all treated animals throughout the studies. Overall body weight gains (Day 0-364) were decreased in males at 3000 (↓9%, NS), 9000 (↓25%, NS), 25000 (↓34%, NS) ppm and in females at 3000 (↓14%, NS), 9000 (↓55%, $p \leq 0.05$), and 25,000 (↓29%, NS) ppm. Significant decreases ($p \leq 0.05$) were often observed in the 9000

ppm females from Day 77 to termination and in the 25,000 ppm females on Day 238. The effect on body weight gain in the females was not clearly dose-dependent.

In the supplementary study, slight decrease (NS) of body weight and body weight gains were observed in 500 ppm males compared to the control during the entire treatment period. Interestingly, increased body weight and body weight gains (NS) were observed in females at 500 ppm. Overall (Days 0-364) body weight gain was increased (NS) in the 100 and 500 ppm females by 5-46%. This increase (NS) may have been incidental as the effect of treatment in these studies is generally a decrease in body weight gain. For these reasons, the effect on body weight gain in the 100 and 500 ppm females was not considered adverse.

Although food consumption was limited and the ration was generally entirely consumed, food efficiency was affected as evidenced by the variation in body weight gains. The average food efficiency (calculated by the reviewer) was decreased dose-dependently in the males of both studies; however, the decrease in the 100 ppm males was minor. Food efficiency was decreased in the ≥3000 ppm females, but the effect was not clearly dose-dependent. Food efficiency was not decreased in the 100 and 500 ppm females.

No significant treatment-related adverse effects were observed in hematology and chemistry parameters; however, serum tyrosine level was not measured in this study. Urinalysis showed increased ketone level in the urine in all treated animals. This finding may be a false positive due to excretion of p-hydroxyphenylpyruvic acid (a keto-acid) which interferes with the reagent of the test strip. Decreased urine pH values (pH<6) were observed in all treated animals compared with the controls (pH>6). Examination of urinary sediment revealed crystals (it was identified as magnesium complex of the parent compound). The study authors stated that due to the known solubility characteristics of the compound which is heavily dependent on pH level, it is possible that the limit of solubility was surpassed, as indicated by crystals of the compound in urine sediment. This finding was supported by high concentration of BAS 670H detected in the urine of treated animals. The quantity detected in the urine was not linearly proportional to the administered dose.

Histopathology examinations showed increased incidences of minimal to moderate thyroid C-cell hyperplasia in males (3-4/5 treated vs 1/5 controls). No dose-response was observed in females (3-5/5 treated vs 5/5 controls). It is known from other concurrently submitted studies that the compound affects the thyroid. Therefore, the thyroid c-cell hyperplasia is considered treatment-related.

Significant microscopic lesions were observed only in the two males of the 25000 ppm group that died: (i) renal/urinary system toxicity as evidenced by ureter dilation; urinary bladder cystitis; and pyelonephritis, perinephritis, and concretion in the kidney; (ii) ilium Peyer patch atrophy; (iii) brain glia cell reaction; (iv) indications of reproductive organ toxicity including

hyperemia in the epididymides and prostatitis; (v) starry sky appearance of the thymus; (vi) serositis in the mesenteric lymph node; and (vii) hyperemia in the auxiliary lymph node.

One male dog of the 9000 ppm group showed multiple hemorrhages in the urinary bladder wall which corroborates with its gross pathological finding. This may be an indication for early toxic damage of the structures of the urinary bladder wall. The loss of the integrity of the transitional epithelium could lead to bacterial infection and other secondary effects.

All other histopathological findings were considered incidental as it occurred in single case or were equally distributed over the treated and control groups.

For males, the NOAEL is 100 ppm (equivalent to 2.9 mg/kg/day), and the LOAEL is 500 ppm (equivalent to 15.3 mg/kg/day) based on increased incidence of thyroid C-cell hyperplasia. For females, the NOAEL is 500 ppm (equivalent to 15.4 mg/kg/day), and the LOAEL is 3000 ppm (equivalent to 92 mg/kg/day) based on decreased body weights, body weight gains, and food efficiency. No serum tyrosine level was measured.

This study is classified as **acceptable/guideline** and satisfies the guideline requirement (OPPTS 870.4100b, OECD 452) for a chronic oral toxicity study in dogs.

6. Mode of Action

The Registrant had proposed a mechanism of thyroid tumors formation by BAS 670H. Formation of thyroid tumor was a consequence of the impairment of pituitary-thyroid hormone levels which was caused by liver enzyme induction. In 1998, EPA has established a policy for data requirement to demonstrate anti-thyroid activity and will be discussed in detail in the next section. Studies were submitted to support the proposed mode of action (MRID 45902223,45902224). **See Appendix A for evaluation of the proposed mode of action.**

V. COMMITTEE'S ASSESSMENT OF THE WEIGHT-OF-EVIDENCE

The Committee's assessment of the weight-of-the-evidence (WOE) is discussed below:

1. Carcinogenicity

Rat

- In male rats, the incidence of thyroid follicular cell adenomas, adenocarcinomas, and combined adenomas and/or adenocarcinomas for the control, 6, 60, 600, and 6000 ppm dose groups was as follows:

Adenomas	8/49 (16%), 12/50 (24%), 13/50 (26%), 18/50 (36%), 23/49 (47%)
Adenocarcinomas	2/47 (4%), 1/46 (2%), 1/46 (2%), 1/46 (2%), 2/42 (5%)
Combined	10/49 (20%), 13/50 (26%), 13/50 (26%), 19/50 (38%), 23/49 (47%)

Male rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 6000 ppm dose group with the controls, for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, all at $p < 0.01$. There were significant differences in the pair-wise comparisons of the 600 ppm dose group with the controls for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, both at $p<0.05$. The incidence of thyroid follicular cell adenomas at 600 ppm (36%) and 6000 ppm (47%) exceeded the laboratory historical control mean of 28%. Therefore, the CARC considered the thyroid follicular cell tumors in male rats, driven by adenomas, to be treatment-related.

- In female rats, the incidence of thyroid follicular cell adenomas, adenocarcinomas, and combined adenomas and/or adenocarcinomas for the control, 6, 60, 600, and 6000 ppm dose groups was as follows:

Adenomas	2/50 (4%), 4/50 (8%), 7/50 (14%), 8/50 (16%), 13/50 (26%)
Adenocarcinomas	0/50 (0%), 0/50 (0%), 2/50 (4%), 2/50 (4%), 0/50 (0%)
Combined	2/50 (4%), 4/50 (8%), 9/50 (18%), 10/50 (20%), 13/49 (26%)

Female rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 6000 ppm dose group with the controls, for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, all at $p < 0.01$. There were significant differences in the pair-wise comparisons of the 600 ppm dose group with the controls for thyroid follicular cell adenomas and adenomas and/or adenocarcinomas combined, and of the 60 ppm dose group with the controls for thyroid follicular cell adenomas and/or adenocarcinomas combined, all at $p<0.05$. The incidence of thyroid follicular cell adenomas at 600 ppm (16%) and 6000 ppm (26%) exceeded the laboratory historical control mean of 10%. Therefore, the CARC considered the thyroid follicular cell tumors in female rats, driven by adenomas, to be treatment-related.

- In male and female rats, the CARC concluded that dosing at the high dose was considered to be adequate and not excessive based on increased mortality in males, decreased body weight (↓9%/↓8%, males/females) and body weight gains (↓11%/↓10%, males/females), clinical observations (corneal opacity and chronic keratitis) and histopathological findings.

Mouse

- In female mice, the incidence of histiocytic sarcomas was 3/48 (6%), 0/47 (0%), 1/48 (2%), and 7/44 (16%) for the control, 80, 800, and 8000 ppm dose groups, respectively. Female mice had a significant increasing trend in histiocytic sarcomas of the hemolymphoreticular system at $p<0.01$. There were no significant differences in the pair-wise comparisons of the dosed groups with the controls. The incidence of histiocytic sarcomas at 8000 ppm (16%) was within the range of the historical control from the conducting laboratory (15.6%). No preneoplastic lesions were noted. This tumor is known to be common in mice, associated with aging. Therefore, the CARC did not consider the histiocytic sarcomas in female mice to be treatment-related.

- There were no significant treatment-related tumors in male mice.

- The CARC considered dosing at the high dose, which was greater than the limit dose, in both sexes to be adequate and not excessive. In males, this was based on decreased body weight and body weight gain, increased relative liver weight and increased incidence of hepatocellular hypertrophy. In females, this was based on increased mortality, however, no significant decreases in body weight and body weight gains, or histopathological findings were observed.

2. Mutagenicity

- There is no mutagenicity concern for BAS 670H based on *in vivo* or *in vitro* assays.

3. Structure Activity Relationship (SAR)

- The very limited SAR data was inconclusive and not useful in the weight-of-the-evidence analysis.

4. Mode of Action

- The Registrant provided data showing effects of BAS 670H on thyroid hormones (T4 and TSH), thyroid weights and thyroid histopathology (hypertrophy and hyperplasia). Overall, there is evidence of concordance between the effects on thyroid homeostasis and thyroid tumor formation. The CARC concluded that the mode of action for thyroid

follicular cell tumors in rats has been established. Evidence for the demonstration of antithyroid activity as required by the Agency's Policy Document (EPA/630/R-97/002) is described in detail in Appendix A.

VI. CLASSIFICATION OF CARCINOGENIC POTENTIAL

In accordance with the EPA Final Guidelines for Carcinogen Risk Assessment (March 29, 2005), the CARC classified BAS 670H as **"Not likely to be carcinogenic to humans at doses that do not alter rat thyroid hormone homeostasis"**. This decision was based on the following considerations:

(i) No treatment-related tumors were seen in male or female mice when tested at doses (greater than the limit dose) that were adequate to assess carcinogenicity;

(ii) Treatment-related thyroid follicular cell tumors were seen in both male and female rats at 600 and 6000 ppm, which were considered to be adequate, and not excessive, to assess carcinogenicity;

(iii) There is no mutagenicity concern from *in vivo* or *in vitro* assays;

(iv) The non-neoplastic toxicological evidence (i.e., thyroid growth, thyroid follicular cell hypertrophy/hyperplasia, thyroid hormonal changes) indicated that BAS 670H was inducing a disruption in the thyroid-pituitary hormonal status.

VII. QUANTIFICATION OF CARCINOGENIC POTENTIAL

The mechanistic data for thyroid follicular cell tumor formation meet the criteria established by the Agency for the use of a margin of exposure approach for human cancer risk assessment. However, the CARC determined that quantification of human cancer risk is not required since the NOAEL (0.4 mg/kg/day) for non-cancer risk assessment is not expected to alter thyroid hormone homeostasis nor result in thyroid tumor formation.

VIII. REFERENCES

MRID	Citation
45902202	Mellert, W., K. Deckardt, K. Küttler, and B. Hildebrand (2000) BAS 670H - Subchronic oral toxicity study in C57BL mice: administration in the diet for 3 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Rhein, Germany. Laboratory Project ID: 60C0124/98096, October 17, 2000. Unpublished.
45902203	Mellert, W., K. Deckardt, Dr. Küttler, et al. (2001) Range finding study in Wistar rats; administration in the diet for 4 weeks. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project Id.: 30S0362/98137, October 8, 2001. Unpublished
45902204	Kaspers, U., K. Deckardt, C. Gembardt, et al. (2003) BAS 670H - subchronic toxicity study in Wistar rats; administration in the diet for 3 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project Id.: 50S0124/98168, March 10, 2003. Unpublished.
45902205	Kaspers, U., K. Deckardt, W. Kaufmann, et al. (2002) BAS 670H - Subchronic oral toxicity study in Beagle dogs: Administration in the diet for 3 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Rhein, Germany. Laboratory Project ID: 31D0124/98110, May 23, 2002. Unpublished.
45902215	Kaspers, U., K. Deckardt, W. Kaufmann, *et al.* (2002) BAS 670H: chronic oral toxicity study in beagle dogs: administration in the diet for 12 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: 33D0124/98123, December 4, 2002. Unpublished.
45902216	Kaspers, U., K. Deckardt, W. Kaufmann, *et al.* (2002) BAS 670H: Supplementary chronic oral toxicity study in beagle dogs: administration in the diet for 12 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: 33D0124/98144, December 20, 2002. Unpublished.
45902217	Kaspers, U., K. Deckardt, K. Küttler, et. al. (2002) BAS 670H - Chronic toxicity study in Wistar rats administration via the diet for 12 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Rhein, Germany. Laboratory Project Id.: 70S0124/98098, December 12, 2002. Unpublished.

45902221 Mellert, W., K. Deckardt, K. Küttler, et al. (2002) BAS 670H: carcinogenicity study in C57BL mice: administration in the diet for 18 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: 76C0124/98112, December 6, 2002. Unpublished.

45902222 Kaspers, U., K. Deckardt, K. Küttler, *et al.* (2003) BAS 670H: Carcinogenicity study in Wistar rats: administration in the diet for 24 months. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: 82S0124/98099, January 8, 2003. Unpublished.

45902223 Mellert, W., K. Deckardt, C. Gembardt, *et al.* (2002) BAS 670H - thyroid hormone study in Wistar rats: administration in the diet for 4 weeks and recovery periods up to 13 weeks. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID: Project No. 37C0124/98159, BASF Registration Document No. 2002/1014188, December 6, 2002. Unpublished.

45902224 Mellert, W., D.B. Beimborn, W. Kaufmann, et al. (2002) BAS 670H - Hepatic enzyme induction study in Wistar rats: administration in the diet for 4 weeks. Experimental Toxicology and Ecology, BASF Aktiengesellschaft, Rhein, Germany. Laboratory Project No.: 99C0124/98161. November 19, 2002. Unpublished

45902225 Engelhardt, G. and Hoffmann, H. D. (1999). *Salmonella typhimurium/ Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with BAS 670 H (= Reg. -Nr. 375 080). Department of Toxicology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 40M0124/984210, Document No. 1999/11499; Study Completion Date: October 29, 1999. Unpublished.

45902226 Engelhardt, G. and Leibold, E. (2002). *Salmonella typhimurium/ Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with BAS 670 H. Experimental Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 40M0124/984232, Document No. 2002/1014113; Study Completion Date: November 25, 2002. Unpublished.

45902227 Engelhardt, G. and Leibold, E. (2002). *Salmonella typhimurium/ Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with BAS 670 H. Experimental Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification

40M0124/984234, Document No. 2002/1014199; Study Completion Date: December 9, 2002. Unpublished.

45902228 Engelhardt, G. and Leibold, E. (2003). *Salmonella typhimurium/ Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with BAS 670 H. Experimental Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 40M0124/984235, Document No. 2003/1014309; Study Completion Date: January 28, 2003. Unpublished.

45902229 Engelhardt, G. and Hoffmann, H.D. (2001). *Salmonella typhimurium/ Escherichia coli* Reverse Mutation Assay (Standard Plate Test and Preincubation Test) with Reg. No. 388 010. Experimental Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 40M0362/984231, Document No. 2001/1021847; Study Completion Date: May 29, 2001. Unpublished.

45902230 Engelhardt, G. and Hoffmann, H.D. (2000). *In Vitro* Gene Mutation Test With BAS 670 H in CHO Cells (HPRT Locus Assay). Experimental Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 50M0124/984183, Document No. 2000/1018642; Study Completion Date: November 10, 2000. Unpublished.

45902231 Wollny, H.-E. (2001). Gene Mutation Assay (HPRT) in Chinese Hamster V79 Cells *In Vitro* With Reg. -No. 388 010. RCC Cytotest Cell Research GmbH, Roßdorf, Germany; Laboratory Project No. 50M0362/989168, BASF Registration Document No. 2001/1021846; Study Completion Date: February 20, 2001. Unpublished.

45902232 Engelhardt, G. and Hoffmann, H.D. (1999). *In Vitro* Chromosome Aberrations Assay With BAS 670 H in V79 Cells. Department of Toxicology of BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 32M0124/984174, Document No. 1999/11688; Study Completion Date: December 7, 1999. Unpublished.

45902233 Engelhardt, G. and Leibold, E. (2002). *In Vitro* Chromosome Aberration Assay With BAS 670 H in V79 Cells. Experimental Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification: 32M0124/984233, Document No. 2002/1013986; Study Completion Date: November 21, 2002. Unpublished.

45902234 Engelhardt, G. and Hoffmann, H.D. (1999). Cytogenetic Study *In Vivo* With BAS 670 H in the Mouse Micronucleus Test After Two Intraperitoneal Administrations. Department of Toxicology of BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 26M0124/984175, Document No. 1999/11683; Study Completion Date: November 29, 1999. Unpublished.

45902301 Engelhardt, G. and Hoffmann, H.D. (2001). Cytogenetic Study *In Vivo* With Reg. No. 388 010 in the Mouse Micronucleus Test Single Intraperitoneal Administrations. Department of Toxicology and Ecology BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 26M0362/984230, Document No. 2001/1021845, Study Completion Date: May 14, 2001. Unpublished.

45902302 Engelhardt, G. and Hoffmann, H.D. (1999). *In Vitro* Unscheduled DNA Synthesis (UDS) Assay With BAS 670 H in Primary Rat Hepatocytes. Department of Toxicology of BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany; Laboratory Project Identification 81M0124/984184, Document No. 1999/11404; Study Completion Date: October 12, 1999. Unpublished.

45902305 Leibold, E., and B. van Ravenzwaay (2002) ^{14}C-BAS 670H - Study of biokinetics in rats. Experimental Toxicology and Ecology. BASF Aktiengesellschaft, Ludwigshafen/Rhein, Germany. Laboratory Project ID.: Project No. 02B0022/996002. BASF Registration Document No. 2002/1006961. May 28, 2002. Unpublished.

45902306 Tilting, N., and H. E. Knoell (2002) Metabolism of ^{14}C-BAS 670H in rats. BASF Aktiengesellschaft, BASF Agricultural Center Limburgerhof, Crop Protection Division, Ecology and Environmental Analytics, Limburgerhof, Germany. Laboratory Project ID.: Study Code 55908. BASF Registration Document No. 2002/1011852. December 6, 2002. Unpublished.

---------- Brunsman, L. (2005). BAS 670H: Qualitative Risk Assessment Based On Wistar Rat and C57BL/6J Rj Mouse Dietary Studies. February 25, 2005. TXR No. 0053149.

---------- U.S. EPA. 1998. Assessment of Thyroid Follicular Cell Tumors. United States Environmental Protection Agency. Office of Research and Development. Risk Assessment Forum. Washington, DC. EPA/630/R-97/002. March 1998.

APPENDIX A: Consideration of the Use of the Non-linear Extrapolation Approach for BAS 670H

Appendix A: Consideration of the Use of the Non-linear Extrapolation Approach for BAS 670H

A. Introduction

The quotations which follow are taken from the Agency's Policy Document entitled "Assessment of Thyroid Follicular Cell Tumors", March 1998 (EPA/630/R-97/002):

> "Tumors of the thyroid gland follicular cells are fairly common in chronic studies of chemicals in rodents. Experimental evidence indicates that the *mode of action* for these rodent thyroid tumors involves (a) changes in the DNA of thyroid cells with the generation of mutations, (b) disruption of thyroid-pituitary functioning, or (c) a combination of the two. The only verified cause of human thyroid cancer is ionizing radiation, a *mutagenic* insult to which children are more sensitive than adults.
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> ...Treatments of rodents that cause *thyroid-pituitary disruption* result in chronic reduction in circulating thyroid hormone levels, increase in TSH levels and the development of increased cell division, increased size and numbers of thyroid cells, increased thyroid gland weight and, finally, tumors of the thyroid. In some cases, there is also an increase in tumors of the pituitary cells that produce TSH. Cessation of treatment early in the process before tumor development results in reversal of processes back towards normal."
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> When assessing tumors of the thyroid, "For those cases where thyroid tumors arise from chemically induced disturbances in thyroid-pituitary functioning, tumors are considered to be secondary to the adverse effects on the thyroid gland function that precede them. As exposures to such agents decrease, the likelihood of cancer decreases; risks may be seen as minimal at doses where there is no effect on thyroid-pituitary homeostasis. Generally, homeostasis is considered to apply when serum T4, T3 and TSH levels and thyroid and pituitary morphology and growth are within their normal limits."

In the Science Policy Guidance section of this document, factors that should be considered in making this determination are discussed.

> "Most of the focus in implementing this policy is devoted to answering the following questions: (1) Does an agent that shows thyroid carcinogenic effects have antithyroid activity? (2) Can modes of action other than thyroid-pituitary carcinogenic effects have antithyroid activity? (3) How can one express thyroid dose-response relationships?" The occurrence of tumors in tissues other than the thyroid is also considered in determining mechanism of carcinogenesis.

B. Determination of whether neoplasms are due to thyroid-pituitary imbalance

The Science Policy Guidance discusses the types of information necessary to characterize the mechanism of thyroid carcinogenesis. These are addressed as they apply to BAS 670H, as follows:

i. Consideration of whether the thyroid tumors associated with administration of BAS 670H can be attributed to disruption of the thyroid-pituitary hormonal balance (demonstration of antithyroid activity). In addressing this point, the Policy lists eight areas of inquiry for evidence demonstrating antithyroid activity (for additional details on the results described below, see individual study summaries presented earlier in this document or attached DERs for carcinogenicity and mechanistic studies):

a. Increases in cellular growth *in vivo* (evidence required):

In the 24-month feeding study in rats (MRID 45902222), increased incidence of focal follicular cell hyperplasia and hypertrophy in the thyroid gland were seen in all BAS 670H treated groups of both sexes. Similar observations of diffuse hypertrophy and follicular cell hyperplasia in the thyroid were seen in the 12-month chronic feeding study in rats (MRID 45902217). **EVIDENCE ESTABLISHED**

b. Hormone changes (e.g., reduced thyroid hormones T3, T4 and increased TSH; evidence required):

In a thyroid hormone study (MRID 45902223), BAS 670H was administered in the diet to Wistar rats at dose levels of 0, 6, 60, 600, or 6000 ppm for 4 weeks and observed for recovery in a period of 4 or 13 weeks. No treatment-related changes in T3 concentration were seen in both sexes. Decreased total T4 levels ($p \leq 0.05$) were observed in ≥ 60 ppm male groups on treatment Days 14 and 28 (↓13-26%). TSH levels were increased ($p \leq 0.05$) in the ≥ 600 ppm males on Day 28 (↑20-54%). After cessation of test substance administration, serum T4 and TSH concentrations returned to the control level during the recovery period of 4 or 13 weeks. **EVIDENCE ESTABLISHED**

c. Site of action (intra thyroidal, peripheral tissues, liver or other sites; evidence required):

The available data suggest that the primary site of action may be the liver, and to a lesser extent, the thyroid. In a hepatic enzyme induction study (MRID 45902224), BAS 760H was administered in the diet to Wistar rats at nominal dose levels of 0 or 6000 ppm (equivalent to 0/0 or 557.9/593.6 mg/kg bw/day [M/F]) for 28 days. There were no effects of treatment on survival, clinical signs, body weights, body weight gains, food consumption, food efficiency, or thyroid weight. Hepatic activities of pNP-GT (p-

nitrophenol-glucuronyltransferase) or HOBI-GT (4-hydroxybiphenyl-glucuronyltransferase) showed no treatment-related effects. Increased (not significant) MUF-GT (4-methylumbelliferone-glucuronyltransferase, phase II enzyme) activity was seen in the treated males (↑71%) and females (↑29%) compared to controls. Relative (to body) liver weights were increased (↑8%; $p \leq 0.05$) in the treated males compared to controls. There were no treatment-related effects on liver weights in the females. The CARC noted the weakness in data for establishing the site of action, specifically no dose-response data on enhanced hepatic clearances were presented. The CARC determined that, although a dose-response was not established for the site of action, hepatic clearance is not a critical factor since the NOAEL (6 ppm; 0.4 mg/kg/day for the inhibition of T4) used for chronic risk assessment would address the concerns that the thyroid tumors are likely to be due to increased metabolism of thyroid hormone in the liver.

d. Dose correlations (evidence required):

The available data (rat chronic toxicity, carcinogenicity and special mechanistic studies) indicate that the increase in thyroid follicular cell tumors is correlated with perturbance of thyroid hormone levels, hypertrophy and hyperplasia in both sexes. Increases in thyroid tumors were only observed at dose levels causing these effects. **EVIDENCE ESTABLISHED.**

e. Reversibility (evidence required):

Thyroid hormone study (MRID 45902223) demonstrated that 4 weeks after cessation of treatment with BAS 670H for 28 days at a dietary concentration of 6-6000 ppm, the treatment-related increases in T4 and TSH were reversed. However, increases of thyroid weight did not reverse to control level even after 13 weeks recovery. The study did not evaluate reversibility of the increase in liver microsomal enzymes. In this study, microscopic examination did not demonstrate follicular cell hypertrophy/hyperplasia in the treatment or in recovery periods. **EVIDENCE ESTABLISHED.**

f. Lesion progression (evidence desirable):

Some evidence exists for lesion progression (hypertrophy/ hyperplasia to adenoma to adenocarcinoma). In the thyroid hormone study (MRID 45902223), increased thyroid weights were observed after 28 days of treatment. Histopathology did not demonstrate follicular cell hypertrophy/hyperplasia in the treatment and recovery periods (4-13 weeks). In the rat 90-day toxicity study, no thyroid hypertrophy/hyperplasia was observed. In a 12-month chronic toxicity study, thyroid hypertrophy/hyperplasia was observed at end of the study but no thyroid tumor was observed. In a rat 2-year carcinogenicity study, increased incidences of thyroid follicular cell hypertrophy/ hyperplasia was observed in all treated groups of both sexes. First

thyroid adenoma in males was observed at week 80, dose 600 ppm and in females at week 79, dose 6000 ppm. **EVIDENCE ESTABLISHED.**

g. Structure-activity analysis (evidence desirable):

The CARC determined that there are too few chemicals and too little structural similarity to draw any firm conclusion toward the structure-activity relationship.

h. Other studies (evidence desirable): Not available.

ii. Consideration of the extent to which genotoxicity may account for the observed tumor effects.

BAS 670 H was not mutagenic in *Salmonella typhimurium* or *Escherichia coli* in a series of reverse mutation assays using the standard plate incorporation and preincubation protocols. BAS 670 H was also not mutagenic in independently performed gene mutation assays in Chinese hamster ovary (CHO) cells up to insoluble and cytotoxic concentrations. BAS 670 H was also negative for induction of unscheduled DNA synthesis in primary rat hepatocytes. BAS 670 H induced a clastogenic response in cultured mammalian cells but this activity is not expressed in whole animals. The lack of a clastogenic effect *in vivo* casts doubts on the relevance of the *in vitro* response to a possible mutagenic mode of action for BAS 670H.

iii. Consideration of the occurrence of tumors in other tissues in addition to the thyroid.

In the mouse 18-month carcinogenicity study, a significant increasing trend in histiocytic sarcomas of the hemolymphoreticular system was observed in female mice. There were no significant differences in the pair-wise comparisons of the dosed groups with the controls. The incidence of histiocytic sarcomas at 8000 ppm (16%) was within the range of the historical control from the conducting laboratory (15.6%). No preneoplastic lesions were noted. This tumor is known to be common in mice, associated with aging. Therefore, the CARC did not consider the histiocytic sarcomas in female mice to be treatment-related.

iv. Consideration of the dose-response.

In the chronic toxicity/carcinogenicity study in rats, thyroid effects were observed at the same dose levels at which increases in thyroid tumors were observed in both sexes.

v. Conclusions:

The Registrant provided data showing effects of BAS 670H on thyroid hormones (T4 and TSH), thyroid weights and thyroid histopathology (hypertrophy and hyperplasia). Overall, there is evidence of concordance between the effects on thyroid homeostasis and thyroid tumor formation. The CARC concluded that the mode of action for thyroid follicular cell tumors in rats has been established.