Attachment 2

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY 014150
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES
FINIAND
TOXIC SUBSTANCES

Date: April 21, 2000

MEMORANDUM

SUBJECT: PP 7F04924 CLODINAFOP PROPARGYL (PC Code: 125203)
Toxicology Disciplinary Chapter for Registration Support Document

DP Barcode: D264028
PRAT Case#: 289249
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Company Designation: CGA 184927
Class: Herbicide 40 CFR: None
MRID: N/A

FROM: Stanley B. Gross, Ph.D., Toxicologist
HED-OPP / RAB 3 (7509C)

THROUGH: Stephen C. Dapson, Ph.D., Branch Senior Scientist
HED-OPP / RAB 3 (7509C)

Stephen C. Dapson
05/02/2000

TO: Anna Lowit, Ph.D., Toxicologist
HED-OPP / RAB 3 (7509C)

This document contains toxicology information regarding the hazard characterization for clodinafop propargyl (2-propynyl (R)-2-[4-(5-chloro-3-fluoro-2-pyridinyloxy)-phenoxy]-propionate)), as part of a human health risk assessment for new uses on wheat. as requested by Novartis Crop Protection, Inc. (Formerly Ciba Crop Protection, Inc.).

Clodinafop 2E has been previously registered in Canada. As part of the Agency's continued efforts at harmonization with Canada under NAFTA, OPP management has agreed to use Canada's reviews to the extent possible in making a U.S. registration decision. The EPA reviewers had reviewed and made comments on these Canadian reviews. Peer review summaries by HIARC (6/2/99), CARC (12/7/99), Q* memorandum (3/2/00), Mode of Action

on Liver Carcinogenicity (9/21/99), and FQPA SFC meeting reports (3/6/00 and 3/20/00) are included as attachments at the end of the document.

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1.0 HAZARD CHARACTERIZATION

Clodinafop 2E has been previously registered in Canada. As part of the Agency's continued efforts at harmonization with Canada under NAFTA, OPP management has agreed to use Canada's reviews to the extent possible in making a U.S. registration decision. The EPA reviewers had reviewed and made comments on these Canadian reviews. Peer review summaries by HIARC (6/2/99), CARC (12/7/99) and Mode of Action on Liver Carcinogenicity (9/21/99) committee are included as attachments at the end of the document. On April 29, 1999, the Health Effects Division's Hazard Identification Assessment Review Committee (HIARC) evaluated the toxicology database of clodinafop-propargyl (CGA 184927), established a Reference Doses (RfDs), selected toxicological endpoints for acute and chronic dietary as well as occupational and residential exposure risk assessments. The HIARC also addressed the potential enhanced sensitivity to infants and children as required by the Food Quality Protection Act (FQPA) of 1996.

The acute toxicity data indicates that clodinafop-propargyl (CGA 184927) has low acute oral, dermal, and inhalation toxicity. It is not a eye or skin irritant. However, it is a skin sensitizer.

In subchronic and chronic studies, the primary target organ was the liver for dogs, mice, and rats. Liver toxicity was evidenced by increased liver weight, elevated liver enzyme activities and abnormal histopathological findings. For dogs only, skin lesions (e.g. pustules, erythema, and crusts) were observed in the subchronic and chronic dog feeding studies. It was noted that the skin lesions were observed at doses as low as 50 ppm (1.73 mg/kg/day) in the subchronic feeding study while skin lesions were observed at higher doses (500 ppm or 15.2 mg/kg/day) in the one year feeding study.

In a rat reproductive toxicity study, there were no adverse effects on reproduction in adult animals (LOAEL => 64.2 mg/kg/day; NOAEL = 64.2 mg/kg/day). However, there were fetotoxic effects (LOAEL = 31.7 mg/kg/day; NOAEL = 3.2 mg/kg/day, reduced fetal viability, decreased pup body weight, and dilatation of renal pelvis) observed at doses that produced maternal toxicity (decreased body weight gain, increased liver and kidney weights with histopathological changes).

In a developmental toxicity study in rats (NOAEL = 5 mg/kg/day; LOAEL = 40 mg/kg/day) , fetotoxic effects were observed at doses lower than those that produced maternal toxicity. Fetal anomalies consisted of an increased incidence of the following: bilateral distension of the ureter and bilateral torsion of the ureter, hematoma to the head, absence of ossification in the sternebrae, incomplete ossification of the thoracic vertebral centra, absence of ossification in the caudal vertebral arches, unilateral 14th ribs, incomplete ossification of the metacarpals, and incomplete ossification of various cranial bones (parietal, interparietal,

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occipital, and squamosal). Also there was a significant but slight reduction (7%) in mean fetal body weights at the high-dose group compared to the control.

A developmental toxicity study in rabbits resulted in maternal deaths at the high and intermediate high dose levels of 125 and 175 mg/kg (NOAEL = 25 mg/kg/day; LOAEL = 125 mg/kg/day) without teratogenic effects (NOAEL = 125 mg/kg/day; LOAEL > 125 mg/kg/day).

Carcinogenicity studies indicated that treatment with clodinafop-propargyl increased the incidence of prostate and ovarian tumors in rats and hepatocellular tumors in mice. An increased incidence of hepatoma and hepatocellular carcinoma were observed at the high dose (29.6 mg/kg/day) in a mouse carcinogenicity study. A $Q^* = 1.29 \times 10^{-1}$. was calculated for these tumors.

The genetic toxicology studies indicate that clodinafop-propargyl is not mutagenic in bacteria (*Salmonella typhimurium*) or cultured mammalian cells (Chinese hamster V79 lung fibroblasts). There is also no evidence of clastogenicity *in vivo*. CGA 184927 did not induce unscheduled DNA synthesis (UDS) in primary rat hepatocytes. However, the acceptable studies do not satisfy the 1991 mutagenicity guideline requirements since the submitted *in vitro* cytogenetic assay has been classified as unacceptable. It was recommended, therefore, that an *in vitro* cytogenetic assay be conducted to fulfill guideline requirements.

There are no neurotoxicity studies available for clodinafop-propargyl. However, clinical signs indicative of neurotoxicity were observed in the dog, rat and rabbit studies. These signs included hypoactivity and circling behavior (subchronic mouse study, MRID 4439238); tremors and decreased activity (subchronic dog study, MRID 44399139); and decreased activity, paddling movements, abnormal gait and uncoordinated movements (chronic dog study, MRID 44399128). In order to further define the potential of neurotoxicity, the HIARC recommended acute and subchronic neurotoxicity studies to be conducted on this chemical.

Results from metabolism studies in rats indicated that clodinafop-propargyl was well absorbed from the intestinal tract (74.7% - 94.1% of administered dose, AD) and excreted via the urine; much smaller percentage was eliminated in the feces. The excretion rate was much faster for females, i.e. more than 80% of the AD was excreted within the first 24 hours, whereas for males, only about 75% of the AD was excreted by 96 hours post-dosing. Significant residues were evident in most tissues of the males, with highest levels seen in the fat, bone marrow, liver, and kidney. Tissue residues in females were significantly lower than males due to the higher elimination rate of the test material. Residues in females were highest in the fat, ovaries, uterus, and kidney. In the urine, the major metabolite was (R)-2-[4-(5-chloro-3-fluoro-2-pyridinyloxy)--phenoxy]--propionic acid (36.7-39.1% of AD). In addition, seven unidentified metabolites were isolated (0.1-5.2% of the AD). In the feces, the major metabolite corresponded to the major urinary metabolite, accounting for 15.7% to 16.9% of the AD. Six unidentified

metabolites were isolated, ranging from 0.3% to 1.4% of the AD. In the fat, all metabolites were reportedly acylglycerides, the majority of which were hybrid di- and triacylglycerides (3.5 and 17% of the AD, respectively). In the liver, kidney, and carcass, the metabolite pattern reflected the transformations seen in excreta and fat.

Two of the four structural analogs (i.e., haloxyfop-methyl and diclofop-methyl) were found to induce liver tumors in mice. Fluazifop-butyl and diclofop-methyl are not mutagenic.

The Registrant proposed that clodinafop-propargyl (CGA 184927) acts as a peroxisome proliferator and is directly involved with the onset of liver carcinogenesis in the rodent. However, the CARC determined that the submitted mechanistic studies do not support the proposed mode of action for the occurrence of prostate and ovarian tumors in rats or liver and blood vessel tumors in mice.

2.0 REQUIREMENTS

The requirements (40CFR 158.690) for food use for Clodinofop-propargyl are in Table 1.

Table 1. Requirements for food use.

Test		Technical	
		Required	Satisfied
870.1100	Acute Oral Toxicity	yes	yes
870.1200	Acute Dermal Toxicity	yes	yes
870.1300	Acute Inhalation Toxicity	yes	yes
870.2400	Primary Eye Irritation	yes	yes
870.2500	Primary Dermal Irritation	yes	yes
870.2600	Dermal Sensitization	yes	yes
870.3100	Oral Subchronic (Rat)	yes	yes
870.3150	Oral Subchronic (Dog)	yes	yes
870.3200	28-Day Dermal	no	yes
870.3250	90-Day Dermal	no	-
870.3465	90-Day Inhalation	no	-
870.3700a	Developmental Toxicity (Rat)	yes	yes
870.3700b	Developmental Toxicity(Rabbit)	yes	yes
870.3800	Reproduction	yes	yes

Table continued...

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Test		Technical	
		Required	Satisfied
870.4100a	Chronic Toxicity (Rat)	yes	1
870.4100b	Chronic Toxicity (Dog)	yes	yes
870.4200a	Oncogenicity (Rat)	yes	1
870.4200b	Oncogenicity (Mouse)	yes	yes
870.4300	Chronic/Oncogenicity (Rat)	yes	yes
870.5100	Mutagenicity—Gene Mutation - bacterial	yes	yes
870.5375	Mutagenicity—Gene Mutation - mammalian	yes	yes
870.5385	Mutagenicity—Structural Chromosomal Aberrations	yes	yes
870.5395	Mutagenicity—Micronucleus	no	-
870.6100a	Acute Delayed Neurotox. (Hen)	no	-
870.6100b	90-Day Neurotoxicity Hen)	no	-
870.6200a	Acute Neurotox. Screening Battery (Rat)	yes	no
870.6200b	90 Day Neuro. Screening Battery (Rat)	yes	no
870.6300	Develop. Neuro	yes	no
870.7485	General Metabolism	yes	yes
870.7600	Dermal Penetration	no	-
Special Studies for Mechanism		no	-

1 = a chronic/onocgenicity study is available for the rat, thus separate chronic and oncogenicity are not necessary.

3.0 DATA GAPS.

HIARC (6/99) recommended the following studies as data gaps:
Acute Neurotoxicity Study in Rats (81-8; OPPTS 870.6200)
Subchronic Neurotoxicity Study in Rats (82-7; OPPTS 870.6200)
In vitro cytogenetic assay (84-2; OPPTS 870.5375)
FQPA SFC (3/20/00) required Developmental Neurotoxicity Study.

4.0 HAZARD ASSESSMENT

Except for neurotoxicity studies and the cytogenicity study noted as data gaps, toxicity studies making up the hazard assessment of clodinafop-propargyl are considered adequate.

4.1 Acute Toxicity

The acute toxicity data (see Table 1) indicates that clodinafop-propargyl (CGA

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184927) has low acute oral, dermal, and inhalation toxicity. It is not an eye or skin irritant. However, it is a skin sensitizer.

Adequacy of data base for acute toxicity: Except for the requirement of an acute neurotoxicity study (870.6200a Acute Neurotox. Screening Battery in Rats), the data base for acute toxicity is considered complete.

Table 2. Acute Toxicity Profile for Clodinafop-propargyl (CGA 184927)

GDLN	Study Type	MRID	Results	Tox. Cat.
870.1100	Acute Oral- Rat	44399124	LD_{50} =1392(♂)/2271(♀) mg/kg	3
870.1200	Acute Dermal -Rabbit -Rat	44399125	LD_{50} > 2000 mg/kg (rat or rabbit)	3
870.1300	Acute Inhalation- Rat	44399126	LC_{50} >2.3 mg/L(♂&♀)	4
870.2400	Primary Eye Irritation- Rabbit	44399127	Slightly eye irritant	3
870.2500	Primary Skin Irritation- Rabbit	44399128	Non-irritant	4
870.2600	Dermal Sensitization-Rat	44399129	Skin sensitizer	NA

4.2 Subchronic Toxicity

Adequacy of data base for subchronic toxicity is considered complete.

Summary: In subchronic and chronic studies, the primary target organ was the liver for dogs, mice, and rats. Liver toxicity was evidenced by increased liver weight, elevated liver enzyme activities and abnormal histopathological findings. For dogs only, skin lesions (e.g. pustules, erythema, and crusts) were observed in the subchronic and chronic dog studies. It was noted that the skin lesions were observed at doses as low as 50 ppm (1.73 mg/kg/day) in the subchronic feeding study while skin lesions were observed at higher doses (500 ppm or 15.2 mg/kg/day) in the one year feeding study.

870.3100 28-day oral (gavage) toxicity study in rats.

In a 28-day oral (gavage) toxicity study in rats (MRID# 44399130), CGA 184927 (93.7% a.i.) suspended in water containing 0.5% carboxymethylcellulose and 0.1% Tween 80 was administered to Tif:RAIf (SPF) rats at dosage levels of 0, 5, 40 and 200 mg/kg/day for 28 consecutive days.

Mortality was excessive in males at the high dose level (all rats in this group died prior to term). One female in the high dose group died prior to term. Though clinical signs of toxicity were evident in all rats that died in the high dose group, such signs were not evident in the animals surviving to term. The primary target organ for toxicity was the liver, where findings such as liver enlargement, increased liver weight and hepatocellular hypertrophy were observed at all doses in males, i.e. at 5, 40 and presumably 200 mg/kg/day. A NOAEL for liver effects among males was not identified in this study. Among females, liver toxicity was clearly evident at 200 and 40 mg/kg/day based on increased liver weight and hepatocellular hypertrophy, and was considered to extend to the lowest dose based on non-statistically significant increases in liver weight of 15-17% in concert with more remarkable dosing related effects on this parameter at 40 and 200 mg/kg/day.

Additional treatment related effects included decreased body weight gain: males (200 and 40 mg/kg/day), females (200 mg/kg/day); decreased food consumption: males (200 and 40 mg/kg/day), females (200 mg/kg/day); hematology - increased platelets: males (40 and 5 mg/kg/day), females (200 and probably 40 and 5 mg/kg/day); clinical chemistry - increased alkaline phosphatase: males (40 and 5 mg/kg/day), increased urea: males (40 mg/kg/day), increased glucose: males (40 mg/kg/day), females (200 mg/kg/day), increased alanine aminotransferase: females (200 mg/kg/day), increased A/G ratio: females (200 and 40 mg/kg/day).

The principal findings in this study were excessive mortality in males at 200 mg/kg/day, at which dose all males died prior to term. One female died prior to term in the same high dose group which was attributed to the test material and liver toxicity. Over all, males were more remarkably affected in this study. Based primarily upon adverse effects on the liver, the systemic toxicity LOAEL was 5 mg/kg/day in males, conclusively, and in females, conservatively. A definitive NOAEL was not identified in this study for either sex.

This was classified as Acceptable/Guideline subchronic oral study in rats.

870.3100 13 Week Subchronic Study in Rats.

In a subchronic toxicity study in rats (MRID# 44399132), CGA 184927 (93.7%, a.i.) was administered to Tif: RAIf (SPF) albino rats (20/sex/group) via diet at dose levels of 0, 2, 15,

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120, or 1000 ppm (0, 0.13, 0.92, 8.24, or 70.0 mg/kg/day for males; and 0, 0.13, 0.94, 8.24, or 71.1 mg/kg/day for females, respectively) for a period of 92 to 94 days. At the end of the treatment period, 10 rats/sex/group were sacrificed; the remaining rats were retained for a 28-day recovery period, i.e., fed control diet, and were then sacrificed.

At the end of the treatment period, an increase in mean absolute and relative liver weights at 120 (males only) and 1000 ppm (both sexes), and increased alkaline phosphatase activity at 120 (males) and 1000 ppm (both sexes) were noted. The other findings at 1000 ppm were: decreased mean bodyweight and reduced mean absolute and relative thymus weight, in males. Histopathological findings considered to be treatment-related were hepatocytic hypertrophy seen in both sexes, and thymic atrophy observed in males only.

After a 28-day recovery period, it was demonstrated that the treatment-related findings were reversible.

The LOAEL is 120 ppm (8.24 mg/kg/day) for males and 1000 ppm (71.1 mg/kg/day) for females, based on increased liver weights and enzyme activity in males at 120 ppm and liver hypertrophy in both sexes at 1000 ppm. The NOAEL was determined to be 15 ppm (0.92 mg/kg/day) for males, and 120 ppm (8.24 mg/kg/day) for females.

This was classified as Acceptable/Guideline subchronic oral study in rats.

870.3100 13 week feeding study in mice.

In a subchronic toxicity study (MRID 44399138) CGA 184927 technical was administered to 10 albino mice/sex/dose in the diet at dose levels of 0, 2, 6, 50, or 400 ppm (Lot No./Batch No. P612003, purity 93.7%) for 90 days. These concentrations resulted in doses of 0, 0.28, 0.88, 7.27, or 53.0 mg/kg/day for males, and 0, 0.34, 1.05, 8.56, and 71.3 mg/kg/day for females. These concentrations are, however, unreliable due to inhomogeneous distribution of the test substance in the rodent diet during the last half of the study.

Treatment related deaths included only one male in the 400 ppm group; prior to death this animal exhibited ruffled fur, hunched posture, and hypoactivity. Clinical signs suggestive of neurotoxicity included circling behavior in one female exposed to 400 ppm; other exposure groups were observed to be similar to control groups. Treatment-related increases in body weight compared to controls were observed for males in the 400 ppm exposure group for weeks 4 - 13 (not significant except for week 7; $p< 0.05$), for females in the 50 ppm group at weeks 3, 4, and 6 ($p<0.05$ or $p<0.01$) and for females in the 400 ppm group for weeks 2 - 7 ($p<0.05$ or $p<0.01$). Food consumption for males remained essentially comparable to the controls, female mice fed 400 ppm, however had increased food consumption for weeks 9, and 11-13 ($p<0.05$).

Male mice treated with 400 ppm had increased water consumption for weeks 2-13 ($p<0.05$ or 0.01) and females had consistently increased water consumption throughout the study, though only significantly for weeks 1, 2, 5, and 10 ($p<0.05$ or 0.01), compared to their respective control groups.

Clinical chemistry analysis revealed elevated plasma levels of aspartate aminotransferase, alanine aminotransferase, and alkaline phosphatase in males treated with 400 ppm ($p<0.01$), in addition, males in the 50 ppm group and females in the 400 ppm group had elevated alkaline phosphatase levels ($p<0.01$) compared to control mice. Glucose levels were elevated in males of the 50 ppm group ($p<0.05$), and males and females of the 400 ppm groups ($p<0.05$). Sodium and chloride were elevated compared to the control for males in the 50 and 400 ppm groups ($p<0.01$). Plasma protein levels were also increased for males treated with 400 ppm ($p<0.01$).

At necropsy, males and females in the 50 and 400 ppm groups had increased liver weights ($p<0.05$ and 0.01, respectively) compared to controls and males in the 400 ppm group had increased adrenal weights (n.s.). Females in the 400 ppm group also had increased kidney weight ($p<0.05$). During necropsy the livers of 9/10 males and 2/10 females were noticeably enlarged compared to controls. Histopathological evaluation also revealed liver toxicity, including hypertrophy of hepatocytes in the 50 and 400 ppm groups along with hepatocellular necrosis, focal proliferation of the intrahepatic biliary ducts, and diffuse Kupffer cell hyperplasia in the 400 ppm males and females. Inflammatory cell infiltration was noted in all treated groups as well as in the controls.

Under the conditions of this study, the LOAEL was 50 ppm, 7.27 and 8.56 mg/kg/day for males and females, respectively. This was based on changes in clinical chemistries in males, liver enzyme, glucose, sodium and chloride increases, and liver toxicity manifested as hepatocellular hypertrophy in males and females. The NOAEL was 6 ppm, 0.88 and 1.05 mg/kg/day for males and females, respectively.

This subchronic toxicity study is classified as **unacceptable (guideline) and not upgradable.** This study does not satisfy the requirements for a subchronic oral toxicity study in mice **(EPA Subdivision F, 82-1a).** This is due to large variations of test substance concentrations in samples analyzed for target concentrations.

870.3150 90 day feeding study in dogs.

In a subchronic toxicity study in dogs (MRID# 44399139), CGA 184927 was administered to beagle dogs (4/sex/group). The test diets contained technical CGA 184927, purity 84.3% administered during study weeks 1 and 2, and purity 93.7% administered for the remainder of the study, at dietary concentrations of 0, 1/1000/500 (days 1-54/55-66/67-90), 10,

50 or 200 ppm (0.038/34.07/16.43, 0.346, 1.73, 7.91 mg/kg/day for males and 0.036/32.29/16.86, 0.390, 1.89 and 7.16 mg/kg/day for females, respectively) for a period of 90 days.

In the 50 ppm group (males only) and the 200 ppm group (females only) pustular formation was noted in the inguinal/abdominal areas. At 1000 ppm, more extensive skin lesions were observed, characterized as general erythema of the skin, pustule formation, conjunctivitis, purulent areas, alopecia and encrustation. In addition, tremors (8 days after dosing but were transient), decreased activity (persisted until necropsy), increased incidence of diarrhea and decreased bodyweight and food consumption were noted during the 1000 ppm dosing period. Decreased RBC count, Hgb and HCT were noted at 1/1000/500 ppm group. At 13 weeks, increased alkaline phosphatase (both sexes), alanine aminotransferase (females only) and aspartate aminotransferase (females only) were noted in the 1/1000/500 ppm (both sexes) group. Mean absolute and relative liver and kidney weights were increased in the 1/1000/500 ppm group, males only. However, these later findings were statistically non-significant and were not supported by histopathological changes in these organs. Histopathological changes consisting of acute hemorrhagic to purulent pneumonia, duodenal ulceration and acute focal ulcerative dermatitis, considered to be treatment-related were only observed in the male dog sacrificed in extremis on study day 71 (1/1000/500 ppm group). The only other finding considered to be treatment-related was vacuolated cell foci in the zona fasciculata of the adrenal cortex in all animals of the 1/1000/500 ppm group. This change was not observed in any other animal at any other dose level tested.

The LOAEL is 50 ppm (1.73 mg/kg/day) for males and 200 ppm (7.16 mg/kg/day) for females, based on occurrence of skin lesions. The NOAEL was determined to be 10 ppm for males (0.346 mg/kg/day), and 50 ppm for females (1.89 mg/kg/day).

This was classified as Acceptable/Guideline subchronic oral study in dogs.

870.3200 28-Day dermal toxicity in rats.

In this 28-day dermal toxicity study (MRID 44399141), forty rats (5/sex/dose) were assigned to four groups. Groups 1, 2, 3, and 4 received topical application of either 0, 50, 200, or 1000 mg/kg/day of CGA 184927 (2 mL/kg/day) to the shaved area six hours per day, 5 days/week for 4 weeks. No treatment-related mortality or dermal signs were observed. Dose-related clinical signs included piloerection and hunched posture at ≥ 200 mg/kg/day (males only) mainly during weeks 2, 3, and 4. Post-mortem examination revealed an increase in absolute and relative liver weight as well as in liver:brain weight ratio in males at ≥ 200 mg/kg/day and centrilobular hypertrophy in male rats treated at 1000 mg/kg/day. There was a decrease in thymus weight in males and females receiving 1000 mg/kg/day and atrophy of the thymus in

male rats receiving this dosage.

The systemic toxicity LOAEL is 200 mg/kg/day based on dose-related increases in liver weights and clinical signs (piloerection and hunched posture) in male rats. The systemic toxicity NOAEL is 50 mg/kg/day. The dermal toxicity NOAEL is 1000 mg/kg/day.

This 28-day dermal toxicity study is classified Acceptable/Guideline).

4.3 Prenatal Developmental Toxicity.

Adequacy of data base for prenatal developmental toxicity: The database is adequate and indicates positive developmental effects.

Summary: In a developmental toxicity study in rats (NOAEL = 5 mg/kg/day; LOAEL = 40 mg/kg/day) , fetotoxic effects were observed at doses lower than those that produced maternal toxicity. Fetal anomalies consisted of an increased incidence of the following: bilateral distension of the ureter and bilateral torsion of the ureter, hematoma to the head, absence of ossification in the sternebrae, incomplete ossification of the thoracic vertebral centra, absence of ossification in the caudal vertebral arches, unilateral 14th ribs, incomplete ossification of the metacarpals, and incomplete ossification of various cranial bones (parietal, interparietal, occipital, and squamosal). Also there was a significant but slight reduction (7%) in mean fetal body weights at the high-dose group compared to the control.

A development toxicity in rabbits resulted in maternal deaths at the high and intermediate high dose levels of 125 and 175 mg/kg (NOAEL = 25 mg/kg/day; LOAEL = 125 mg/kg/day) without teratogenic effects (NOAEL = 125 mg/kg/day; LOAEL > 125 mg/kg/day).

870.3700a Prenatal gavage development study in rats.

In a developmental toxicity study (MRID 44399145), pregnant Ico: OFA SD. (IOPS Caw) rats were dosed by gastric lavage with CGA 184927 (purity 93.70%), as a suspension in aqueous hydroxypropyl methylcellulose, at dose levels of 0 (vehicle control), 5, 40 and 160 mg/kg/day, 25 mated females per group, on days 6 to 15 of gestation, inclusive.

There were no mortality or treatment-related toxicity observed. At 160 mg/kg/day, a statistically non-significant decrease in mean maternal body weight gain was seen during GD 6-11 (21%) and during GD 6-16 (9%) of the dosing period. However, the corrected body eight gain was comparable to that of the control group. Therefore, the observed effect on body weight gain during GD 6-11 and 6-16 was caused by the intrauterine effect on fetuses. **Therefore, the**

maternal LOAEL is >160 based on lack of effect and the maternal NOAEL is 160 mg/kg/day.

Fetal anomalies considered to be treatment-related, consisted of increased incidences of the following: bilateral distension of the ureter and bilateral torsion of the ureter, 40 and 160 mg/kg/day; hematoma to the head, 160 mg/kg/day; absence of ossification in the sternebrae, 160 mg/kg/day; incomplete ossification of the thoracic vertebral centra, 160 mg/kg/day; absence of ossification in the caudal vertebral arches, 160 mg/kg/day; unilateral 14th ribs, 40 and 160 mg/kg/day; incomplete ossification of the metacarpals, 40 and 160 mg/kg/day; and incomplete ossification of various cranial bones (parietal, interparietal, occipital, and squamosal) at 40 and 160 mg/kg/day. Also there was a significant but slight reduction (7%) in mean fetal body weights at 160 mg/kg/day compared to the control. **Therefore, the developmental LOAEL is 40 mg/kg/day, based on increased incidences of bilateral distension and torsion of the ureters, unilateral 14th ribs, and incomplete ossification of the metacarpals and various cranial bones (parietals, interparietals, occipital, and squamosal). The developmental NOAEL is 5 mg/kg/day.**

This study is classified as **acceptable** and **satisfies** the guideline requirement for a developmental toxicity study (83-3A) in rats.

870.3700b Prenatal gavage development study in rabbits.

In a developmental toxicity study (MRID 44399144), hybrid albino (HyCr) rabbits were dosed by gavage with CGA 184927 (purity 93.7%), as a suspension in aqueous hydroxypropyl methylcellulose, at dose levels of 0 (vehicle control), 5, 25 and 125 mg/kg/day, 18 mated females per group, and 175 mg/kg/day, 14 mated females, on days 7 to 19 of gestation, inclusive.

Maternal toxicity was evident at 125 and 175 mg/kg/day which consisted of mortality (5/18 during GD 14-22 and 11/14 during GD 11-15, respectively), clinical signs (labored breathing, reduced activity, ataxia, pallor and nasal discharge) and body weight loss (in nonsurvivors only). Necropsy revealed ulceration of the stomach and hemorrahagic contents of the caecum and colon. One doe from 175 mg/kg/day group aborted on GD 23. Mortality and clinical signs were also noted in the range-finding study at 160 mg/kg/day. Consequently, the observed effect seen this study were considered to be treatment-related. **Therefore, the maternal LOAEL is 125 based on mortality, clinical signs and body weight loss and the maternal NOAEL is 25 mg/kg/day.**

Due to high rate of mortality at 175 mg/kg/day, the fetal data for this group was excluded from analyses. There was no treatment-related developmental toxicity observed in other dose groups. **Therefore, the developmental LOAEL is >125 mg/kg/day based on lack of**

developmental toxicity. The developmental NOAEL is ≥125 mg/kg/day.

This study is classified as **acceptable** and **satisfies** the guideline requirement for a developmental toxicity study (83-3b) in rats.

4.4 Reproductive Toxicity.

Summary: In a rat reproductive toxicity study, there were no adverse effects on reproduction in adult animals (LOAEL => 64.2 mg/kg/day; NOAEL = 64.2 mg/kg/day). However, there were mild fetotoxic effects (LOAEL = 31.7 mg/kg/day; NOAEL = 3.2 mg/kg/day, reduced fetal viability, decreased pup body weight, and dilatation of renal pelvis) were observed at doses that produced maternal toxicity (decreased body weight gain, increased liver and kidney weights with histopathological changes).

870.3800 2 Generation feeding reproduction study in rats.

In a two-generation study (MRID 44399146) was conducted using Sprague Dawley Crl: CD (SD)BR SPF rats, fed test diets containing 0, 5, 50, 500 or 1000 ppm (0, 0.33, 3.21, 31.69, and 64.24 mg/kg/day in males and 0, 0.41, 3.77, 37.54, and 73.60 mg/kg/day in females, respectively) CGA 184927 continuously throughout the study period, 25 rats per sex per group. Each female in each generation was mated to produce one litter only.

There were no treatment-related mortality or clinical signs of toxicity observed. The mean bodyweight gains were significantly but slightly lower during the premating/ mating period for mid- and high-dose males in both generations. The mean food consumption was decreased during the premating period for males in the 500 and 1000 ppm groups, and during lactation for females in the 500 and 1000 ppm groups.

Absolute and relative liver and kidney weights were increased in the 500 and 1000 ppm groups, one or both sexes, in both generations. The mean testicular weight was decreased for F1 males in the 1000 ppm group only.

Necropsy revealed an increased incidence of dilatation of the renal pelvis in the 500 and 1000 ppm groups in both sexes in F1 generation only.

Histopathological findings observed in the 1000 ppm group of both generations and sexes, and considered to be treatment-related were as follows: liver - hepatocytic hypertrophy; kidney - hyaline casts, parenchymal atrophy, pigment deposits in tubules (P), pelvis dilatation (F1), tubular dilatation (F1), and loss of tubular epithelium with gray masses in tubule lumina (F1).

There were no adverse, treatment-related effects on reproductive performance.

For F1a pups, viability index on day 21 was lower in the 1000 ppm group and the mean pup weight was decreased in the 500 and 1000 ppm groups on days 14 and 21 of lactation. For F2a pups, mean pup weight was decreased in the 1000 ppm group throughout the lactation period and in the 500 ppm group on days 7, 14 and 21 of lactation.

Minor delays noted in development of F1a and F2a pups in the 500 and 1000 ppm groups consisted of pinna unfolding and/or incisor eruption and/or eye opening. However, these were no longer evident at weaning and so were considered transient. There were no treatment-related effects on functional tests at weaning (pupillary reflex, auditory response).

An increase in the incidence of dilatation of renal pelvis was observed in the 500 and 1000 ppm groups, F2a pups only.

The LOAEL for parental systemic toxicity is 500 ppm (31.7 mg/kg/day) based on decrease in body weight gain, reduced food consumption, increased liver and kidney weights and histopathological changes in the liver (hepatocytic hypertrophy, and renal tubules (hyaline casts, parenchymal atrophy, pigment deposits, dilatation and loss of tubular epithelium). The NOAEL for parental systemic toxicity is 50 ppm (3.2 mg/kg/day).

The LOAEL for reproductive toxicity is ≥1000 ppm (≥64.2 mg/kg/day) based on lack of reproductive effects. The NOAEL for reproductive toxicity is 1000 ppm (64.2 mg/kg/day).

The LOAEL for developmental toxicity is 500 ppm (31.7 mg/kg/day) based on reduced viability, decreased pup body weight and dilatation of renal pelvis. The NOAEL is 50 ppm (3.2 mg/kg/day).

This study is classified as **acceptable/guideline** and **satisfies** the guideline requirement for a reproductive toxicity study (83-4) in rats.

4.5 Chronic Toxicity.

Adequacy of Data Base is considered complete.

Summary. The primary target organs were the liver for dogs, mice, and rats. Liver toxicity was evidenced by increased liver weight, elevated liver enzyme activities and abnormal histopathological findings. For dogs only, skin lesions (e.g. pustules, erythema, and crusts) were observed in the subchronic and chronic dog studies. It was noted that the skin lesions were

observed at doses of 500 ppm or 15.2 mg/kg/day in the one year feeding study.

870.4100b 1 year Chronic Feeding Study in Dogs (44399128).

In a 52-week chronic toxicity study (MRID# 44399142), beagle dogs (4/sex/group) were fed diets containing 0, 10, 100 or 500 ppm (0, 0.32, 3.38 or 15.2 mg/kg/day for males and 0, 0.32, 3.37 or 16.7 mg/kg/day for females, respectively) CGA 184927 (93.7% a.i.).

There was no effect on the survival of animals. At 500 ppm, skin lesions (pustules, crusts, scales, erythema, increased severity of alopecia, fissures, reddened sclera and ocular exudates), clinical signs of toxicity (decreased activity, paddling movements, abnormal gait or uncoordinated movements, pallor, dyspnea, diarrhea), and decreased body weight gain (-12% and -35% in males and females, respectively) were noted. In addition, hematology revealed decreased platelet count and increased thromboplastin time in females and reduced iron concentration in males.

The LOAEL is 500 ppm (15.2 and 16.7 mg/kg/day for males and females, respectively), based on occurrence of skin lesions, clinical signs and reduced body weight gain and food consumption. The NOAEL was determined to be 100 ppm (3.38 and 3.37 mg/kg/day for males and females, respectively).

This study is classified as Acceptable/Guideline.

4.6 Carcinogenicity.

The data base for carcinogenicity is adequate as indicates that the agent is a carcinogen.

Summary Carcinogenicity studies indicated that treatment with clodinafop-propargyl increased the incidence of prostate and ovarian tumors in rats and hepatocellular tumors in mice. An increased incidence of hepatoma and hepatocellular carcinoma were observed at the high dose (29.6 mg/kg/day) in a mouse carcinogenicity study (see CARC report, in Attachments). A $Q^* = 1.29 \times 10^{-1}$. was calculated for these tumors (see memorandum by Lori Brunsman included in the Attachments).

870.4200b 18-month carcinogenicity study in mice.

In an 18-month carcinogenicity study (MRID# 44399143), Tif:MAGf (SPF) albino mice (60/sex/group) were fed diets containing 0, 1, 10, 100 or 250 ppm (0, 0.113, 1.10, 11.0 or 29.6 mg/kg bw/day for males and 0, 0.129, 1.25, 12.6 and 33.1 mg/kg bw/day for females, respectively) CGA 184927 (93.7% a.i.).

At 250 ppm, among males, there was increased mortality (12/60; 20%) during the last month of the study; a high proportion of them (38/60; 63%) developed hepatocellular tumors. The mean final body weight (5-6%) and mean overall body weight gain (11%) were lower in males only. At ≥100 ppm, there were increases in liver enzyme activity and liver weights in both sexes. At necropsy, an increased incidence of enlarged livers and liver nodules/masses were noted in males at 100 ppm and in males and females at 250 ppm. Histopathology revealed non-neoplastic changes including hepatocytic hypertrophy, kupffer cell pigmentation and intrahepatic bile duct hyperplasia in males at 100 ppm and in males and females at 250 ppm; hepatocytic necrosis in males at 100 and 250 ppm and necrosis in females at 250 ppm. In addition, an increased incidence of pre-neoplastic foci was noted at 100 and 250 ppm in males only; an increase in the severity of thymic atrophy was seen in both sexes.

The LOAEL for systemic toxicity is 11.0 and 12.6 mg/kg/day for males and females, respectively, based on increases in liver enzyme activity and liver weights. The NOAEL was estimated to be 1.10 and 1.25 mg/kg/day for males and females, respectively.

This study was classified as Acceptable/Guideline oncogenicity study in mice.

870.4300 2- year Chronic/Oncogenicity Feeding study in Rats.

In a combined chronic/carcinogenicity study (MRID# 44399147), CGA 184927 (93.7% a.i.) was administered in diet to male and female Tif: RAIf (SPF) albino rats (80/sex/group) for a period of 24 months. The test diets contained technical CGA 184927 at dietary concentrations of 0, 1, 10, 300 or 750 ppm (0, 0.031, 0.32, 10.18, 26.28 mg/kg/day for males; and 0, 0.034, 0.36, 11.31 and 29.48 mg/kg/day for females, respectively). At interim sacrifice, during week 53, 10 rats/sex/dose were sacrificed.

An increase in liver enzyme levels was noted in both sexes at ≥300 ppm. At interim and final sacrifices, absolute and/or relative liver and kidney weights increased in one or both sexes at ≥300 ppm. Necropsy revealed increased incidence of enlarged, or mottled liver in males at ≥300 ppm and in females at 750 ppm. In addition, there was dose-related increase in the incidence of microscopic changes in the liver including hepatocytic hypertrophy in males at ≥10 ppm as well as focal or nodular hyperplasia and fibrosis in one or both sexes at ≥300 ppm. Thyroid hypertrophy of follicular epithelium was noted in females at 750 ppm. Kidney changes noted in both sexes consisted of increased incidence of chronic progressive nephropathy and tubular pigmentation at ≥10 ppm. Increased incidence of ovarian medullary tubular hyperplasia was noted at ≥300 ppm.

Under the conditions of this study, treatment with CGA 184927 increased the incidence of prostate and ovarian tumors in rats at 750 ppm. For males, an increased incidence of prostate adenoma was seen in the high-dose group, i.e., incidence rates were

8/80 (10.0%), 9/80 (11.25%), 12/80 (15.0%), 13/80 (16.25%) and 19/80 (23.75%) in the 0, 1, 10, 300 and 750 ppm groups, respectively. At 750 ppm, one of 80 males developed hepatocarcinoma.

For females, an increased incidence of tubular adenomas of the ovary was noted in the high-dose group, i.e., incidence rates of 2/80 (2.5%), 1/80 (1.25%), 1/80 (1.25%), 1/80 (1.25%) and 9/80 (11.25%) for the 0, 1, 10, 300 and 750 ppm groups, respectively. The chemical was administered at a dose sufficient to test its carcinogenic potential.

The EPA reviewer does not concur with the NOAEL/LOAEL established by the Canadian and determines that the LOAEL for systemic toxicity is 10 ppm (0.32 and 0.36 mg/kg/day in males and females, respectively) based on hepatocytic hypertrophy, chronic progressive nephropathy and tubular pigmentation. The systemic NOAEL is 1 ppm (0.031 and 0.034 mg/kg/day in males and females, respectively).

4.7 Mutagenicity.

The data base for mutagenicity is considered adequate.

Summary: The genetic toxicology studies indicate that clodinafop-propargyl is not mutagenic in bacteria (*Salmonella typhimurium*) or cultured mammalian cells (Chinese hamster V79 lung fibroblasts). There is also no evidence of clastogenicity *in vivo*. CGA 184927 did not induce unscheduled DNA synthesis (UDS) in primary rat hepatocytes. However, the acceptable studies do not satisfy the 1991 mutagenicity guideline requirements since the submitted *in vitro* cytogenetic assay has been classified as unacceptable. It was recommended, therefore, that an *in vitro* cytogenetic assay be conducted to fulfill guideline requirements.

870.5100 Gene Mutation Salmonella and Eschericia/liver microsome test.

In independently conducted microbial reverse gene mutation assays (MRID No. 44399153), *Salmonella typhimurium* strains TA1535, TA1537, TA98 and TA100 were exposed to Clodinafop-propargyl technical as CGA-184927 (93.7%) doses ranging from 20-5000 µg/plate in both the presence and the absence of S9 activation. The S9 fraction was derived from Aroclor 1254-induced Tif:RAIf (SPF) male rat livers and the test material was delivered to the test system in acetone.

Compound precipitation was reported at levels ≥313 µg/plate; cytotoxicity was not seen at any dose. All strains responded in the expected manner to the appropriate positive control. **There was, however, no evidence that Clodinafop-propargyl technical induced a**

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mutagenic effect in any strain.

This study is classified as Acceptable and satisfies the guideline requirements for a bacterial gene mutation assay (84-2).

870.5200 Gene Mutation Mutation test with Chinese Hamster cells V70.

In independently performed *in vitro* forward gene mutation assays (MRID No: 44399152), Chinese hamster V79 cells were exposed to Clodinafop-propargyl as CGA-184927 technical (93.7% at concentrations of 25.0, 50.0, 100, 200, 300, 400 or 500 µg/mL without S9 activation and 2.5, 5.0, 10.0, 20.0, 30.0, 40.0 or 50.0 µg/mL with S9 activation (Trial 1) or **2.1, 4.1, 4.2, 16.4, 24.6, 32.8 or 41 µg/mL for Trial 2 (based on analytical determinations).** The S9 fraction was derived from Aroclor 1254 induced rat livers and the test material was delivered to the test system in dimethyl sulfoxide.

The highest dose(s) tested (500 µg/mL -S9; ≈41-50 µg/mL +S9) were cytotoxic. The positive controls induced the expected increases in the mutation frequency. **There was, however, no evidence that CGA-184927 induced a mutagenic effect at any dose with or without S9 activation in either trial.**

This study is classified as **Acceptable** and satisfies the guideline requirement for an in vitro gene mutation assay in cultured mammalian cells.

870.5315 Chromosome Studies Human Lymphocytes in vitro.

In an *in vitro* mammalian cell cytogenetic assay (MRID No: 44399154), human lymphocytes derived from a healthy donor were exposed to Clodinafop-propargyl technical as CGA 184927 (93.7%) doses of 53.13 to 850 µg/mL without S9 activation or 5.5 to 88.0 µg/mL with S9 activation for 3 hours. Cytogenetic analysis was performed on cultures treated with nonactivated doses of 53.13, 106.5, 212.5 or 425 µg/mL or S9-activated levels of 5.5, 11, 22, 44 or 88 µg/mL. Reanalysis of the slides prepared for the nonactivated phase of the study was also performed. The S9 liver homogenate was derived from Aroclor 1254 induced rat livers and the test material was delivered to the test system in dimethyl sulfoxide.

Cytotoxicity, as indicated by a ≥50% reduction in the mitotic indices (MIs), was reported at nonactivated levels ≥850-1000 µg/mL (Initial reading) or ≥106.25 µg/mL (reanalysis) or at ≥125 µg/mL +S9. The conflict in the cytotoxicity data relative to the nonactivated assay is of concern. The positive controls induced the expected high yield of cells with chromosome aberrations in the lymphocytes derived from the male and female donors. There was a slight increase in the frequency of cells with chromosome aberrations

at 425 µg/mL-S9 (3%--initial reading; 3.7%--reanalysis)), however, both values fell within the historical control background range. There was also an increase in the frequency of cells with rare chromatid exchanges at this nonactivated dose (2% both analyses versus 1% in the control). A similar increase in cells with aberrations that was within the background level and in chromatid exchanges (1%) compared to the concurrent control was noted at 44 µg/mL +S9. Owing to the conflicting results from the cytotoxicity assessment and the presence of rare complex chromosome aberrations both with and without S9 activation, the study is considered inconclusive.

This study is classified as **Unacceptable** and does not satisfy the guideline requirement for an in vitro cytogenetic assay.

870.5395 Micronucleus Test (Mice).

In a mouse micronucleus assay (MRID No. 44399151), groups of eight male and eight female Tif:MAGF(SPF) NMRI-derived mice received single oral gavage administrations of 1667 or 5000 mg/kg Clodinafop-propargyl as CGA-184927 technical (93.7%). The test material was delivered to the animals in 0.5% carboxy methyl cellulose. Five males and five females per group were sacrificed at 24, 48 and 72 hours postadministration and harvested bone marrow cells were examined for the incidence of micronucleated polychromatic erythrocytes (MPEs).

One high-dose male and seven high-dose females died prior to the scheduled sacrifice; no other toxic signs were reported. No bone marrow cytotoxicity was noted at any dose or sacrifice time. The positive control induced the expected high yield of MPEs in males and females. **There was, however, no clear evidence that Clodinafop-propargyl induced a clastogenic or aneugenic effect in either sex at any dose or sacrifice time.**

The study is classified as Acceptable and satisfies the requirements for FIFRA Test Guideline 84-2 for in vivo cytogenetic mutagenicity data.

870.5550 Unscheduled DNA Repair, Human Fibroblasts.

In independently performed *in vitro* unscheduled DNA synthesis (UDS) assays (MRID No. 44399155), cultured human fibroblasts were exposed to Clodinafop-propargyl technical as CGA-184927 technical (93.7%) doses of 42.7, 128, 320, 800, 2000 or 5000 μg/mL in both trials. Fibroblasts harvested 5 hours from all treatment groups were scored for silver grains/nucleus. The test was not performed in the presence of a metabolic activator. The test material was delivered to the test system in dimethyl sulfoxide.

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Compound precipitation was seen at doses ≥320 μg/mL: there was, however, no indication of a cytotoxic effect at any dose. The positive control induced the expected marked increases in UDS. **There was, however, no evidence that CGA- 184927 in the absence of S9 activation induced a genotoxic response in either trial.**

This study is classified as **Unacceptable** and does not satisfy the guideline requirement for a UDS assay (84-4).

870.5550 Unscheduled DNA Repair Rat Hepatocytes.

In independently performed *in vitro* unscheduled DNA synthesis (UDS) assays (MRID No. 44399156), primary rat hepatocytes were exposed to Clodinafop-propargyl technical as CGA-184927 technical (93.7%) doses of 0.04, 0.2, 1, 5, 25 or 50 μg/mL (Trial 1). Actual doses used in Trial 2, based on analytical determinations, were 0.6, 3.2, 16.05, 32.1, 48.2 or 60.2 μg/mL for solution targeted to contain 0.75, 3.73, 18.67, 37.33, 56 or 70 μg/mL, respectively. Hepatocytes harvested 16-18 hours after treatment were scored for net nuclear grains/nucleus. The test material was delivered to the test system in dimethyl sulfoxide.

Compound precipitation was noted at levels ≥4000 μg/mL. Lethality was apparent in the preliminary cytotoxicity test at 94.8 μg/mL. The positive control induced the expected marked increases in UDS. **There was, however, no evidence that CGA 184927 induced a genotoxic response in either trial.**

This study is classified as Acceptable and satisfies the guideline requirement for a UDS assay (84-4).

4.8 Neurotoxicity.

The data base for neurotoxicity is considered inadequate.

Summary. There are no neurotoxicity studies available for clodinafop-propargyl. However, clinical signs indicative of neurotoxicity were observed in the dog, rat and rabbit studies. Some of these signs included hypoactivity and circling behavior (subchronic mouse study, MRID 4439238); tremors and decreased activity (subchronic dog study, MRID 44399139); and decreased activity, paddling movements, abnormal gait and uncoordinated movements (chronic dog study, MRID 44399128). In order to further define the potential of neurotoxicity, the HIARC recommended acute and subchronic neurotoxicity studies to be conducted on this chemical. The FQPA Committee recommended a developmental

neurotoxicity study also be conducted.

870.6100a Acute Delayed Neurotox. (Hen) Not required.

870.6100b 90-Day Neurotoxicity Hen) Not required.

870.6200a Acute Neurotox. Screening Battery (Rat) Data Gap.

870.6200b 90 Day Neuro. Screening Battery (Rat) Data Gap.

870.6300 Develop. Neuro Toxicity Study. Data Gap.

4.9 Metabolism.

The data submitted for metabolism is considered adequate.

Summary: Results from metabolism studies in rats indicated that clodinafop-propargyl was well absorbed from the intestinal tract (74.7% - 94.1% of the administered dose, AD) and excreted via the urine; much smaller percentage was eliminated in the feces. The excretion rate was much faster for females, i.e. more than 80% of the AD was excreted within the first 24 hours, whereas for males, only about 75% of the AD was excreted by 96 hours post-dosing. Significant residues were evident in most tissues of the males, with highest levels seen in the fat, bone marrow, liver, and kidney. Tissue residues in females were significantly lower than males due to the higher elimination rate of the test material. Residues in females were highest in the fat, ovaries, uterus, and kidney. In the urine, the major metabolite was (R)-2-[4-(5-chloro-3-fluoro-2-pyridinyloxy)-phenoxy]-propionic acid (36.7-39.1% of AD). In addition, seven unidentified metabolites were isolated (0.1-5.2% of the AD). In the feces, the major metabolite corresponded to the major urinary metabolite, accounting for 15.7% to 16.9% of the AD. Six unidentified metabolites were isolated, ranging from 0.3% to 1.4% of the AD. In the fat, all metabolites were reportedly acylglycerides, the majority of which were hybrid di- and triacylglycerides (3.5 and 17% of the AD, respectively). In the liver, kidney, and carcass, the metabolite pattern reflected the transformations seen in excreta and fat.

870.7485 Metabolism and pharmacokinetics, Rat gavage study.

In a metabolism study (MRID 44399159), two ^{14}C labeled variants of CGA 184927 (one labeled on the 2 pyridil carbon and the other universally labeled on the phenyl ring, purity >98%) were administered to groups of five male Tif:RAI f (SPF) rats, approximately 7 weeks of age and weighed about 200 g at time of dosing, by gavage at concentrations of 25.2 mg/kg ([2-^{14}C]pyridil) and 24.6 mg/kg ([U-^{14}C]phenyl). For dosing, the test material was dissolved in

polyethylene glycol 200/ethanol/water (7/5/2) and 0.7 ml/rat was administered to deliver the dose via gavage. Individual urine and fecal samples were collected at 24 hour intervals for 7 days post-dosing, after which animals were sacrificed by cervical dislocation. Samples of liver, kidney and fat, and the remaining carcass, were retained for residue analysis.

After a single oral dose of about 25 mg/kg/b.w , it is estimated that approximately >70.0% of the dose was absorbed from the intestinal tract. Recovery of the radiolabel was excellent and ranged from 97-105%. In [U-^{14}C]phenyl CGA 184927 treated rats, the excretion of radioactivity in the urine and feces were 48.4% and 22.3% of the administered dose (AD), respectively. Of this, only 15.4% (urine) and 11.2% (feces) of the AD had been eliminated within the first 24 hours. Carcass and tissues contained 25.1% of the AD. Residues in fat, liver, kidney and carcass were 3.8%, 0.8%, 0.2% and 20.3% of the AD, respectively. In [2-^{14}C]pyridil CGA 184927 treated rats, the excretion of radioactivity in the urine and feces were 51.1% and 23.6% of the AD, respectively. Of this, only 14.8% (urine) and 13.3% (feces) of the AD had been eliminated within the first 24 hours. Carcass and tissues contained 22.9% of the AD. Residues in fat, liver, kidney and carcass were 3.9%, 0.9%, 0.2% and 17.9% of the AD, respectively. These results indicate that the CGA 184927 is slowly excreted and most of the residual radioactivity remained in the carcass (18%-20%) and results from both labelling study were almost identical indicating that the diarylether bond of CGA 184927 is not cleaved to any significant extent.

Metabolic profiles were determined by thin layer chromatography (TLC), liquid chromatography (LC), high performance liquid chromatography (HPLC) and high voltage electrophoresis (HVE). Metabolite patterns (urine, fecal and tissue) for [U-^{14}C]phenyl-, and [2-^{14}C]pyridil-labelled CGA 184927 were almost identical. In the urine, the major metabolite (fraction U7) was determined to be (R)-2-[4-(5-chloro-3-fluoro-2-pyridinyloxy)-phenoxy]-propionic acid, reference material CGA 193469, accounting for 36.7% to 39.1% of the AD. In addition, seven unidentified metabolites were isolated, ranging from 0.1% to 5.2% of the AD. Metabolite fraction U3 hydrolysed to yield fraction U7 (i.e., CGA 193469), when treated with NaOH or HCl. Unchanged CGA 184927 was not identified. In the feces , the major metabolite (fraction F*7) corresponded to the urinary metabolite U7 (CGA 193469), accounting for 15.7% to 16.9% of the AD. Metabolite fraction F*8 was determined to be unchanged CGA 184927, accounting for 0.4% to 1.7% of the AD. Six unidentified metabolites were isolated, ranging from 0.3% to 1.4% of the AD. In the fat, all metabolites were reportedly acylglycerides, the majority of which were hybrid di- and triacylglycerides, i.e., approximately 3.5% and 17.0% of the AD, respectively. In the liver, kidney and carcass, the metabolic pattern reflected the transformations seen in excreta and fat.

This metabolism study in the rat is classified **acceptable/guideline** and does satisfy the current guideline requirement for a metabolism study (85-1) in rats.

870.7485 Metabolism and pharmacokinetics, Rat gavage study.

In the absorption, distribution and excretion study (MRID 44399160), [U- ^{14}C-] Phenyl labeled CGA- 184927 {2-propynyl (R)-2-[4-(5-chloro-3-fluoro-2-pyridinyloxy)-phenoxy]-propionate, purity > 99%, radiochemical purity >98%) was administered by gavage to adult male and female Tif:RAI f(SPF) rats at single dose level of 0.5 , 50.0 mg/kg/b.w or 14 repeated 0.5 mg/kg/b.w dose level followed a radioactive 0.5 mg/kg/b.w dose. For dosing, [U-^{14}C-] phenyl CGA- 184927 was dissolved in polyethylene glycol 200/ethanol/water, ratio of 7/5/2, v/v. The animals were grouped and dosed with CGA- 184927 (dose volume of 0.7 or 0.8 ml/rat) by gavage as follows:

i) Group A: left unassigned due to the low solubility in aqueous media, [U-^{14}C-] phenyl CGA- 184927 was not administered intravenously

ii) Group B: Single low dose, 5 rats per sex. The actual dose administered was 0.54 mg/kg b.w, 5.1 μCi. for males and 0.53 mg/kg b.w, 5.1 μCi for females.

iii) Group C: Fourteen daily doses of unlabelled by gavage, followed by a radioactive single oral low dose, 5 rats per sex. The actual dose administered was 0.48 mg/kg b.w, 5.8 μCi for males and 0.55 mg/kg b.w, 5.8 μCi for females.

iv) Group D: Single oral high dose, 5 rats per sex. The actual dose administered was 52.4 mg/kg b.w, 49.7 μCi for males and 53.4 mg/kg bw, 49.7 μCi for females.

Treated animals were individually housed in all-glass metabolism cages, suitable for the collection of urine, feces and expired CO_2. Urine and fecal samples were collected from each animal at intervals for up to 168 hours post-dosing and expired air up to 24 hours post-dosing (high dose group only). Animals were sacrificed 7 days post-dosing and various organs and tissue samples were collected and retained for analysis of radioactivity.

Average total radioactivity recovery ranged from 94.8% to 101.1%. From the results of the study, it is estimated that about 74.7% to 94.1% of the AD was absorbed from the gastrointestinal tract into the systemic circulation after oral administration. After rats received a single low oral dose (0.5 mg/kg bw), single high oral dose (50.0 mg/kg bw) or 14 daily oral low doses (0.5 mg/kg bw) of CGA 184927, it was determined that the major portion of the test material was excreted via the urine, i.e., for males, 44.09% to 64.70%; for females, 87.86% to 91.46%. A lower percentage was eliminated in the feces, i.e., for males, 19.65% to 21.23%; for females, 2.23% to 5.93%. The excretion rate was much faster for females, i.e., more than 80% of the AD had been excreted within the first 24 hours post-dosing in all groups, whereas for males, more than 75% of the AD had only been excreted by 96 hours post-dosing. It is evident that excretion rate and route are highly sex-dependent, but are independent of dose level. After 14 daily oral low doses, urinary excretion was accelerated and increased for both sexes; fecal excretion was not affected. Less than 0.1% of the AD was detected in volatiles and expired air. Significant residues were evident in most tissues of the male animals, with highest levels seen in the fat, bone marrow, liver and kidneys. Tissue residues in females were significantly lower than

for males due to the higher elimination rate of the test material. Residues were highest in fat, ovaries, uterus and kidney. In general, tissue residues were accordingly higher at the high-dose level for both sexes. The mean absolute and relative liver weights were increased for the high-dose group, males only, when compared to the concurrent control group.

The major metabolite in urine and feces was determined to be CGA 193469, accounting for about 36% to 47% of the AD for males, and 80% to 85% of the AD for females. In addition, 11 minor metabolite fractions were isolated from urine and feces. Three were further identified as reference materials CGA 193468, CGA 214111 and unchanged CGA 184927. The remaining (8) minor fractions were not identified. There were significant sex-dependent differences in the urinary metabolite profile, and the quantitative distribution was influenced by pretreatment. In contrast, the fecal metabolic profile was not affected by sex, dose level or pretreatment. The metabolite CGA 193469 was identified in blood and lung tissue. In addition, triacylglycerides were isolated from lung extract.

This metabolism study in the rat is classified **acceptable/guideline** and does satisfy the current guideline requirement for a metabolism study (85-1) in rats.

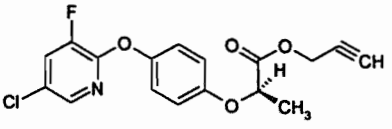
4.10 Other/Special Studies.

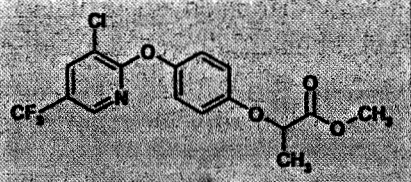
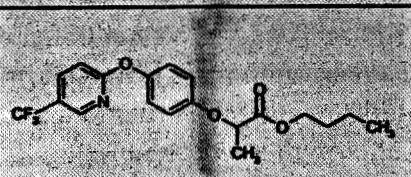
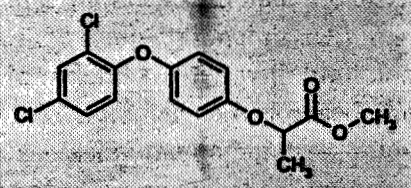
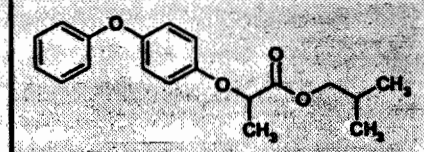
The following issues are discussed in some detail in the CARC document provided as an attachment.

A. Structure Activity Relationship

Two of the four structural analogs shown in Table 3 (SAR Compounds) (i.e., haloxyfop-methyl and diclofop-methyl) were found to induce liver tumors in mice. The diphenyl ether like structure in the molecule may be responsible for the carcinogenic potential of these compounds. Fluazifop-butyl and diclofop-methyl are not mutagenic. The mutagenicity data on other compounds were not available.

Table 3. Structural Activity Relationships.

Compound	Structure	Carcinogenic Effect
Clodinafop-propargyl CAS 105511-96-4 PC 125203		- Prostate and ovarian tubular tumors in rats. - Liver tumors in mice

Haloxyfop-Methyl CAS 69806-40-2 PC 125201		- Classification: B2 - Liver tumor (adenoma and carcinomas). - B6C3F1 mice.
Fluazifop-Butyl CAS 69806-50-4 PC 122805		- No evidence of increased liver tumors; however, the dose levels may be inadequate.
Diclofop-Methyl (Hoelon) CAS 51338-27-3 PC 110902		- Classification: Group C; possible human carcinogen - Hepatocellular adenoma and/or carcinomas - NMRKf (SPF) mice.
Clofop-Isobutyl CAS 51337-71-4		- No data is available. - The chemical has been voluntarily canceled.

B. Mechanistic Studies

The Registrant proposed that clodinafop-propargyl (CGA 184927) acts as a peroxisome proliferator and is directly involved with the onset of liver carcinogenesis in the rodent. Peroxisome proliferation is a transcription-mediated process that involves activation by the peroxisome proliferator of a nuclear receptor in rodent liver called the peroxisome proliferator-activated receptor (PPARα), a member of the steroid hormone receptor superfamily. Upon

activation by peroxisomal proliferators, PPARα forms a heterodimer with the retinoid-X-receptor. This dimer binds to peroxisome proliferator response elements (PPRE) in the promoter region of target genes known to be regulated by PPARα. PPARα induces mitogenesis and cell proliferation which can lead to the formation of hepatocellular tumors. Oxidative stress appears to play a significant role in this increased cell proliferation. It triggers the release of Tumor Necrosis Factor (TNFα) by Kupffer cells, which in turn acts as a potent mitogen in hepatocytes.

The CARC determined that the mechanistic studies do not support the proposed mode of action for the occurrence of prostate and ovarian tumors in rats or liver and blood vessel tumors in mice.

1. Special Study: Giannone, C. (1991). Determination Of Residues As CGA 193469 In Abdominal Fat After A 3-Month Oral Toxicity Study In Rat. MRID 44399134

In a subchronic oral toxicity study in Tif:RAIf albino rats(MRID 44399132) at necropsy, samples of abdominal fat were analysed and the content of CGA 184927 was calculated by determining the content of CGA 193469 using HPLC. In this study male and female rats (20/sex/group) received CGA 184927 at dietary levels of 0, 2, 15, 120 and 1000ppm.

There was a dose-dependent increase in CGA 184927 residues in fat samples from both sexes taken at the end of treatment (14 weeks) and after the 4-week recovery period (18 weeks). Concentrations of CGA 184927 were higher in male rats at all dose levels tested. With the exception of low-dose group males, for all remaining groups, residues in the fat at 18 weeks had decreased by between 40% - 51.5% of the 14 week value.

This is classified as **acceptable/nonguideline**.

2. Special Study Determination Of Residues As CGA 193469 In Abdominal Fat After 12 Months In Study. MRID 44399135

In a chronic toxicity study in rats treated with CGA 193469, at interim sacrifice (53 weeks) and terminal sacrifice (105 weeks), samples of abdominal fat were analysed and the content of CGA 184927 was calculated by determining the content of CGA 193469 using HPLC.

There was a dose-dependent increase in CGA 184927 residues in fat samples from both sexes taken after 12 and 24 months on treatment.

After 12 months, at 1 ppm and 10 ppm, the concentration of CGA 184927 in the abdominal

fat was higher in males when compared to females. At 300 and 750 ppm, the concentration of CGA 184927 in the abdominal fat was comparable between males and females.

After 24 months, at 1 ppm, CGA 184927 residues were negligible. At 10, 300 and 750 ppm, the concentration of CGA 184927 in abdominal fat was higher in males when compared to females.

The results of this study also indicate that the CGA 193469 residue in fat is reduced after 1 year of treatment compared to 3 month treatment. (EPA Toxicologist's note: It is not clear based on the DER how this relates to CGA184o27)

This is classified as **acceptable/nonguideline**.

3. Special Study: Waechter, F. (1991). The Effect Of CGA 184927 On Selected Biochemical Parameters In The Rat Liver Following Subchronic Administration. MRID 44399137

In a 3-month subchronic study, CGA 184927 increased the liver weights in both sexes of rat at 1000 ppm and in males at 120 ppm. Recovery in liver weights was noted after a treatment-free period. This study was intended to characterize biochemically the liver effects of CGA 184927 and their reversibility in selected animals. In a subchronic toxicity study, liver samples were obtained from the control (10 rats/sex/group), 2, 15, 120 and 1000 ppm groups (6 rats/sex/group) at week 14. (after 4 week recovery period), additional liver samples were taken from the control and 1000 ppm groups (8 rats/sex/group). Subcellular fractions were prepared to measure various biochemical parameters including microsomal enzymes. These parameters included protein content (supernatant, microsomal and cytosolic), microsomal cytochrome P-450, microsomal ethoxycoumarin O-de-ethylase, microsomal metabolism of R-warfarin, microsomal hydroxylation of lauric acid, microsomal epoxide hydrolase, microsomal UDP-glucuronosyltransferase, cytosolic glutathione S-transferase, β-oxidation of [1-^{14}C]palmitoyl-CoA. The microsomal fractions were also subjected to SDS PAGE and immunoblot analysis.

At 14 weeks, there was increase in all above parameters at one or more dose levels with the exception of decrease in glutathione S-transferase in males at 1000 ppm.

At 18 weeks, there was statistically significant but slight increase in microsomal protein, cytochrome P-450 and fatty acyl-CoA β-oxidation, seen in the high-dose group, males only. However, these data indicate partial recovery toward normal levels. All other parameters measured had recovered to normal control levels.

Immunoblot analyses reported an induction of cytochrome p-452 in both sexes at 1000

ppm and in males at 2, 15,and 120 ppm. Increased enoyl-CoA hydratase-3-hydroxyacyl-CoA dehydrogenase (peroxisomal bifunctional enzyme) was noted in both sexes at 1000 ppm. The major phenobarbital-inducible cytochrome P-450 isozymes b and e as well as the major polycyclic aromatic hydrocarbon-inducible cytochrome P-450 isozymes c and d were not induced.

The effects of CGA 184927 on selected liver enzymes in the rat were similar to the effects seen after subchronic treatment with known peroxisome proliferators (hypolipidemic compounds, phenoxyacetic acid derivatives). Hence, CGA 184927 was considered to most likely be a peroxisome proliferator in the rat liver.

This is classified as **acceptable/nonguideline**.

4. Special Study: Gerber, H. et al. (1986). Apparently clonal thyroid adenomas may contain heterogeneously growing and functioning cell subpopulations. New Frontiers in Thyroidology, p. 901-905, 1986 (Published). MRID 44399148

The purpose of this study was to investigate the hypothesis that the thyroid tumors might be the late consequence of the persistence, in the normal adult thyroid gland, of a few clones with intrinsically high replication rate, which commonly occurs in fetal thyroids. The author further studied whether the adenomas growing in long-standing mice goiters bear the criteria of a monoclonal tumor.

Forty-two ICR mice were fed a low iodine diet and treated for 20 weeks with 1% sodium perchlorate and 1% sucrose in drinking water. The animals were then injected with 3 x 10 uci 3H-thymidine daily for 2 weeks prior to sacrifice. Proliferation of the follicular epithelium was studied by autoradiography of serial sections. In a second experiment, sodium perchlorate was discontinued and 0.5 ug/ml T4 was given in drinking water for one week. Two hours prior to sacrifice, 10 mU of bovine TSH were injected intraperitoneally. Endocytotic droplets were examined in periodic acid Schiff staining. The average number of droplets/cell and percentage of droplets containing cells in individual adenomas was evaluated. In the third experiment,12 ICR mice received drinking water containing 0.15% methimazole and 1% sucrose (only during last two days, they were injected with 3H-thymidine 3 x 10 uCi per day). In the fourth experiment, the fraction of 3H-thymidine labeled cells growing autonomously in T4-treated newborn mice, was compared to that of equally TSH-suppressed adult mice.

The results of the three experiments are as follows:

Forty out of 42 mice treated with sodium perchlorate developed follicular and papillary tumors consisting of morphologically homogenous cell population, distinct from normal

follicular cells. The fraction of 3H-thymidine labeled cells varied but was significantly higher than the normal hyperplastic tissue. Also a subsets of cells with different replication rate had reappeared within the adenomas.

In the second experiment, in mice treated with sodium pechlorate for 20 weeks followed by one week with T4, the endocytotic response to TSH varied. The percentage of droplet-containing cells ranged from 54±22% to 90±10% in the adenomas as compared to 89±14% in normal hyperplastic tissue.

In the third experiment, after methimazole treatment for 2 weeks and 3H-thymidine labeling for two days, all glands contained rapidly replicating cell clusters. In one out of 12 glands, a clearly delimited adenoma grew after two weeks of stimulation; 100% of all cells appeared to be labeled suggesting the existence of a fast growing cell clone.

In the fourth experiment, an FLC (fraction of 3H-thymidine labeled cells) of 24% was found in T4-treated newborn mice labeled with 3H-thymidine from day 7 to 12 of life compared to 1% in identically treated, nine week old mice. Thus, a higher fraction of follicular cells replicated despite TSH suppression in the thyroid of newborn mice than in the adult mice.

The results of this study support the view that heterogeneous growth and adenoma formation are inevitable late consequences of chronic moderate growth stimulation of a tissue having components with different built-in replication rates.

The asynchronous growth rate of subsets of cells within the old adenomas as well as the intercellular heterogeneity of the endocytotic response to TSH suggests that clonal thyroid adenomas may acquire new qualities and can modify gene expression via much debated mechanism. The author concludes that the growth of benign thyroid tumors and progression does not require a change in genomic expression in any cell. The apparent heterogeneity of a tumor does not necessarily exclude its monoclonal origin.

This study is classified as acceptable/nonguideline.

5. Special Study: Zbinden, G. (1987). Assessment of hyperplastic and neoplastic lesions of the thyroid gland. TIPS, Vol. 8, p. 511-514., dated December 1987. (Published). MRID 44399149.

Focal hyperplastic and neoplastic changes of the thyroid are commonly seen in long-term studies with rodents. In most cases, these lesions are caused by the stimulation of a preexisting small population of cells by thyrotropin, TSH, or other growth factors, followed by clonal

expansion and tumor formation. Mechanisms responsible for TSH elevation include interference with iodine uptake and oxidation, thyroxine synthesis and release and stimulation of hepatic thyroxine catabolism. Some chemicals probably act by increasing the number of follicular epithelia capable of responding to endogenous growth factors.

Thyroid enlargement and tumors are mainly seen in rodents. For example, the sulfonamide sulfamethoxazole treatment induced thyroid nodules in rats at 50 mg/kg/day for 1 year. However, in rhesus monkeys given 300 mg/kg/day for the same duration, there were no increases in thyroid weight or morphological alterations seen. This species difference may be due to the lack of thyroxine-binding globulin in rodents resulting in shorter plasma half-life in rats (10-fold shorter than humans). Interference with thyroxine metabolism, although a transient one, thus leads to a more rapid depletion of the hormone in the rat. Also the species differ in response of the thyroid towards TSH. At least in cell cultures, TSH does not induce proliferation of human thyroid cells, but does stimulate the growth of cells obtained from rat and dog thyroids. The author believes that conventional procedures of evaluating carcinogenicity tests by simply counting tumors in rodents treated with high doses, and by mathematical extrapolation to the low doses to which humans are exposed, are not suitable for the proliferative reactions of the thyroid gland. In assessing the human risk, relevant conclusions can only be drawn if the physiological factors of growth control are known, and if the biological mechanisms by which chemicals initiate focal proliferation and support their progression to tumors are considered.

This study is classified as acceptable/nonguideline.

6. Special Study: Stott, W.T. (1988). Chemically induced proliferation of peroxisomes: Implications for risk assessment. Regulatory Toxicology and Pharmacology, Vol. 8, p. 125-159, 1988. MRID 44399150 (Published). MRID 44399150.

An increasing number of drugs as well as industrial and agricultural chemicals have been found to cause a dose-related hepatomegaly in rodents which is associated with the proliferation of the peroxisome. In chronic bioassays, the prolonged proliferation of hepatocellular peroxisomes and the enhanced production of the peroxisomal metabolic byproduct, hydrogen peroxide, have been hypothesized to account for the tumorigenicity of these compounds, most of which are nongenotoxic in in-vitro and in-vivo assays.

The pronounced differences in interspecies sensitivity to peroxisomal proliferators indicate that the proliferation and the induction of its enzymes is primarily restricted to rodent species. Responses in higher mammals are generally weaker and only occur at much higher doses than in rodents. It appears that a toxicologically significant degree of peroxisomal proliferation is unlikely to occur in humans, even upon chronic administration at high doses of known

proliferators. Thus, rodent species do not represent an appropriate model for the potential toxicity including hepatocarcinogenicity of this class compounds in humans. Rather, the data obtained in nonrodent species, preferably the dog or monkey, would be useful in establishing relevant no-observable-effect levels of exposure and in interspecies risk assessments for peroxisome proliferative agents. The author concludes that based on the present understanding of the mechanism of action of compounds which induce hepatic peroxisome proliferation in rodents, there is a need for the reevaluation of the concept of "maximum tolerated dose level" (MTD) as it pertains to chronic oncogenicity bioassays for peroxisome proliferators. A more appropriate MTD of a peroxisome proliferative agent in sensitive species would appear to be based upon evidence of the proliferation of peroxisomes and the induction of peroxisomal enzymes capable of producing an increased intracellular oxidative stress. Exceeding these dosages will only result in a predictable sequence of events leading, ultimately, to tumor formation due to physiological adaptation of the animal to the administered compound rather than from the direct effects of the compound itself. Only the appropriate application of animal data from well-designed rodent studies, in combination with mechanistic and interspecies sensitivity data, may provide a scientifically sound and defensible assessment of the potential risk of these compounds to humans.

This study is classified as acceptable/nonguideline.

7. Special Study: Bieri, F. (1991). The Effect of CGA 193469, the Free Acid Derivative of Cga 184927, on Peroxisomal B-oxidation in Primary Cultures of Rat, Mouse, Marmoset and Guinea Pig Hepatocytes. MRID 44399157.

Following oral administration, CGA 184927, a peroxisome proliferator, is rapidlyhydrolysed to CGA 193469 (acid derivative) and propargyl alcohol. Since propargyl alcohol is a known cytotoxic agent, the CGA 193469 derivative is considered the component most likely to trigger the peroxisomal proliferation. Thefore, the effect of CGA 193469 on peroxisomal beta-oxidation was investigated in rat, mouse, marmoset and guinea pig hepatocytes cultured in vitro.

This study characterized and compared the in vitro effects of CGA 184927 on selected parameters (i.e., cytotoxicity and induction of peroxisomal beta-oxidation) in primary hepatocytes from various species.

The monolayer cultures were treated with medium containing CGA 184927, CGA 193469 or propargyl alcohol at the appropriate concentrations (0.1 to 100 µg/ml), or solvent controls and incubated for three days. Hepatocytes were then examined for morphological alterations and cell viability. The lactate dehydrogenase (LDH) activity was measured as an indicator of cytotoxicity. In addition, protein content of hepatocytes were measured to determine the

membrane damage. Peroxisomal beta-oxidation was measured in hepatocyte homogenates treated with [1-14]palmitoyl-CoA, a peroxisomal enzyme marker.

CGA 184927-induced cytotoxicity through propargyl alcohol released following its enzymatic hydrolysis. In cultured hepatocytes from all species, CGA 193469, the free acid derivative of CGA 184927, was noncytotoxic at concentrations of ≤100 µg/ml as examined by light microscopy and quantified for LDH activity. There was no change in the total LDH activity and the protein contents were unaffected indicating that there was no cell leakage of LDH. CGA 193469 induced in the cultured rat and mouse hepatocytes the effect characteristically produced by other peroxisomal proliferators. In rat hepatocytes, it induced-peroxisome beta-oxidation in a dose-dependent manner. This enzyme activity was marginal in the guinea pig hepatocytes and was non-measurable in marmoset hepatocytes.

The results of this study suggest that guinea pig and primates are less susceptible to the liver effects by this class of compounds.

This is classified as **acceptable/nonguideline**.

8. Special Study: Guyomard, C. (1992). Effects of CGA 193469, the acid derivative of CGA 184927, on the peroxisomal beta-oxidation in human hepatocytes. MRID 44399158.

Monolayer cultures of human lymphocytes were grown in a medium containingCGA193469 (at concentrations of 0, 0.1, 1, 10 or 100 μM in 0.1% DMSO), benzafibric acid (a reference material with a strong peroxisomal beta-oxidation inducing activity in vivo and in vitro) or solvent control and incubated for 72 hours. After 24, 48 and 72 hours incubation, hepatocytes were examined for morphological alterations and cell viability. The lactate dehydrogenase (LDH) activity was measured as an indicator of cytotoxicity. In addition, protein content of hepatocytes were measured to determine the membrane damage. Peroxisomal beta-oxidation was measured in hepatocyte homogenates treated with palmitoyl-CoA, a peroxisomal enzyme marker, at study initiation and at the end of incubation.

There were no alterations in morphology, nor in intracellular LDH levels after incubation with CGA 193469, or benzafibric acid, at any concentration tested. The protein contents were unaltered. An increase (123%) in peroxisome beta-oxidation was noted in hepatocytes incubated with CGA 193469 for 72 hours at 100 μM. However, this finding was not statistically significant, and in the absence of a dose-response relationship was not considered to be a treatment-related finding. Treatment with CGA 193469 or bezafibric acid, at all concentrations tested, was not cytotoxic to human hepatocytes, in vitro.

Under the conditions of this study, neither CGA 193469 nor bezafibric acid induced peroxisomal beta-oxidation in human hepatocytes, in vitro. However, in the absence of a

known concurrent human positive control to validate the test system, i.e., a substance known to elicit peroxisomal beta-oxidation in human hepatocytes, this cannot be definitely concluded.

This is classified as **acceptable/nonguideline**.

9. Special Study: Trendelenburg, C. (1999) Effects on selected plasma concentrations and biochemical parameters in the liver upon subchronic administration to male adult rats. MRID 443767401

In a special study (MRID no. 44767401), CGA 184927 (94.3% a.i.) was administered to 10 male Tif:RAIf (SPF) rats/dose at dietary dose of 0 or 750 ppm (0 or 53.5 mg/kg/day) for 14 days. During the treatment period, food consumption, body weight and clinical signs were recorded. On day 4 of treatment and prior to sacrifice, blood samples were collected. At necropsy, liver, endocrine glands and accessory sex organs were frozen and stored at -80°C. The biochemical as well as hormonal parameters were determined in the appropriate subcellular liver fractions or in blood plasma according to published procedures. The parameters examined included liver protein content, liver microsomal regio-and stereoselective testosterone hydroxylation, liver microsomal cytochrome P450 CYP19A1 (aromatase), liver peroxisomal fatty acid β-oxidation, plasma dihydrotestosterone, free and total plasma testosterone, plasma total estradiol, and plasma follicle stimulating hormone, prolactin and luteinizing hormone.

Treatment with CGA 194927 caused no clinical signs of toxicity. Rats in the 750 ppm exhibited decreased feed consumption relative to the control group during the first 4 days of dosing which resulted in decreased body weights which persisted for the duration of the study. Although absolute and relative liver weights increased (134% and 139% of control, respectively). There were no changes in organ weights of endocrine and sex hormone dependent organs.

After 14 days of treatment, there was significant increase in liver peroxisomal fatty acid β-oxidation activity (807% of control) as well as a significant increase in liver microsomal cytochrome P450 isoenzymes CYP4A1/A3 and CYP4A2 (242% and 618% of control, respectively), thus supporting the proposed mechanism of peroxisomal proliferation in the rat liver by CGA 184927.

The treatment with CGA 184927 significantly ($p < 0.05$) decreased the total liver microsomal testosterone oxidation rate to 34% of control. Hydroxylation rates at positions 2α and 16α decreased to 19% and 20% of control (significantly; $p < 0.05$), respectively, and the oxidation rate to androstenedione was decreased to 43% of control. The reduced hydroxylation rates at positions 2α and 16α and the conversion of testosterone to androstenedione indicated

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depletion of the male specific cytochrome P450 isoenzyme CYP2C11. Decrease in testosterone hydroxylation rates at positions 2 β, 6α, 6 β and 7α (63, 84, 70 and 50%, respectively) were indicative of partial depletion of cytochrome P450 isoenzymes of subfamilies CYP2A and CYP3A. The treatment resulted in significant increases in hepatic aromatase activity ($p < 0.05$; 169%) and plasma estradiol concentration (not significant; 179% and 140% (CYP19A1) for 3 and 14 days, respectively) compared to control. The total and free plasma testosterone concentrations were not effected by treatment with CGA-184927. Plasma 5α-dihydrotestosterone level increased (not significantly; 198% of control) after 14 days of treatment. No change in plasma concentrations of prolactin, follicular stimulating and luteinizing hormone were noted.

These findings indicate that in the male rat, CGA 184927 acts as a peroxisomal proliferating agent and alters monooxygenase activity in subfamilies of cytochrome P450 which are known to be involved in the synthesis or catabolism of steroid hormones.

The submitted special study is classified as **acceptable/nonguideline**

Special Study: Roberts R. (1999). CGA-193469 (free acid derivative of CGA-184927) peroxisome proliferators: species difference in the regulation of gene expression (MRID 44923101).

Promoter-reporter gene assays were employed to determine the ability of CGA 193469, the free acid derivative of clodinafop-propargyl (CGA 184927), to activate the rodent and human acyl CoA oxidase (ACO) gene promoters. The data show that CGA 183469 is able to activate the rodent ACO gene promoter-reporter construct as expected for a rodent peroxisome proliferator. In contrast, CGA 193469 was unable to activate the human ACO promoter-reporter construct.

10. Special Study: Staubli, W. and Bentley, P. (1989). Electron Microscopical Study, PS 3.3 Cell Biology, dated April 25, 1989. MRID NUMBER: Not assigned

In a subchronic study at necropsy, liver samples were taken from CGA 184927 treated animals at 14 weeks (2/sex/dose from the control and high-dose groups and during recovery period, at 18 weeks (3/sex/dose from the control and 1000 ppm dose groups). These samples were processed and examined by electron microscopy.

At 14 weeks, an increase in the number and size of peroxisomes with matrical inclusion bodies was noted in the 1000 ppm group, males only. At 18 weeks, the number and size of peroxisomes in the 1000 ppm group, both sexes, were comparable to the control group.

However, an increased number of matrical granules was observed in hepatic mitochondria indicating that these mitochondrial changes were not fully reversible during the 4-week recovery phase.

The results of electron microscopic examination of liver tissue support the conclusion that CGA 184927 is a peroxisome proliferator in the rat liver.

5.0 HAZARD ENDPOINT SELECTION

On April 29, 1999, the Health Effects Division's Hazard Identification Assessment Review Committee (HIARC) evaluated the toxicology database of clodinafop-propargyl (CGA 184927), established Reference Doses (RfDs), selected toxicological endpoints for acute and chronic dietary as well as occupational and residential exposure risk assessments. The doses and toxicological endpoints selected are summarized in a table in Section 9.2. The hazard identification rationales for RfD selections are also given below.

A. Dietary Exposure:

Acute Dietary Exposure: An acute reference dose (RfD) was selected for the subpopulation of females 13-50 years old. This acute RfD of **0.05 mg/kg/day** is based on the NOAEL of 5 mg/kg/day selected from a developmental toxicity study in rats (MRID no. 44399145) where an increased incidence of bilateral distension and torsion of the ureters, unilateral 14th ribs, and incomplete ossification of the metacarpals and various cranial bones (parietals, interparietals, occipital, and squamosal) were observed at 40 mg/kg/day (LOAEL). (The NOAEL of 5 mg/kg/day is divided by uncertainty factors (UF) for inter-species extrapolation (10x) and intra-species variability (10x).) Based on the conservative assumption that developmental toxicity could occur following a single exposure to a pregnant female, this endpoint is appropriate for acute risk assessment for females 13-50.

An acute RfD for the general population was also selected by the HIARC. The acute RfD of 0.25 mg/kg/day is based on a NOAEL of 25 mg/kg (NOAEL) from a developmental toxicity study in rabbits (MRID 44399144) where maternal toxicity (increased mortality, clinical signs and body weight loss) was observed at 125 mg/kg/day. This study is suitable for general population because mortality, clinical signs, and maternal body weight loss occurred on the first measurement time point (days 2 after dosing). It is reasonable to assume that the effects could occur after a single dose.

Chronic Dietary: The HIARC selected a **chronic RfD of 0.0003 mg/kg/day** (NOAEL =

0.03 mg/kg/day; Uncertainty Factor = 100). This chronic RfD is based on a two year combined chronic/oncogenicity study in rats (MRID No. 44399147). In this study, the NOAEL of 0.03 mg/kg/day was based on observations of hepatocytic hypertrophy, chronic progressive nephropathy and tubular pigmentation at 0.3 mg/kg/day (LOAEL). The Uncertainty Factor accounts for both interspecies extrapolation (10X) and intraspecies variability (10X).

B. Occupational/Residential Exposure

There are no residential uses; however, there is potential for residential exposure to spray drift resulting from aerial application. Based on the use pattern, there is potential for short-term exposures (private- one field) and intermediate-term exposure (commercial- several fields) during mixing, loading, application, and post-application activities. Long-term exposure is not expected to occur.

Short and Intermediate Term Dermal Endpoints: Short- and intermediate-term dermal endpoints of 50 mg/kg/day (NOAEL) were selected from a 28-day dermal toxicity study (MRID 44399141) where dose-related increases in liver weights and clinical signs (piloerection and hunched posture) in male rats were observed at 200 mg/kg/day (LOAEL). This study is selected because its duration and route of exposure are appropriate for short and intermediate term dermal exposure.

Short-Term Inhalation Risk Assessment: A short-term inhalation endpoint of 5 mg/kg/day (NOAEL) was selected from a developmental toxicity study in rats (MRID no. 44399145) where an increased incidences of bilateral distension and torsion of the ureters, unilateral 14th ribs, and incomplete ossification of the metacarpals and various cranial bones (parietals, interparietals, occipital, and squamosal) were observed at 40 mg/kg/day (LOAEL). Only an acute inhalation toxicity study has been submitted to the Agency. There were no inhalation toxicity studies appropriate for risk assessment in the toxicology database. Consequently, the oral values should be used for inhalation exposure risk assessment; the route-to-route extrapolation should be done as shown below.

Intermediate-Term Inhalation Risk Assessment: A intermediate-term inhalation endpoint of 0.9 mg/kg/day (NOAEL) was selected from a subchronic oral toxicity in rats (MRID no. 44399132) where increases in liver weights and enzyme activity were observed in males at 8.2 mg/kg/day (LOAEL). Only an acute inhalation toxicity study has been submitted to the Agency. There were no inhalation toxicity studies appropriate for risk assessment in the toxicology database. Consequently, the oral values should be used for inhalation exposure risk assessment; the route-to-route extrapolation should be done as follows:

Convert the inhalation exposure component (i.e. μg a.i./day) using a 100% absorption rate (default value) and an application rate to an equivalent oral dose (mg/kg/day) and compare it to the oral value of 5 mg/kg/day for short-term and oral values of 0.9 mg/kg/day for intermediate-term exposure to calculate the MOEs.

Long-Term Dermal and Inhalation Risk Assessments: Based on the proposed use patterns, no long-term dermal or inhalation exposure is expected to occur. Therefore, no endpoints were selected.

Margins of Exposures for Occupational Exposure Risk Assessments

A MOE of 100 is required for occupational exposure risk assessment.

5.1 See Section 9.2 for Endpoint Selection Table Summary.

5.2 Dermal Absorption

No dermal absorption study was submitted. The HIARC estimated the % dermal absorption for CGA 184927 to be 2.5%. This dermal absorption rate was derived by taking the ratio of the LOAEL from the 28-day oral (gavage) toxicity study in rats (5 mg/kg/day) and the 28-dermal toxicity study in rats (200 mg/kg/day) based on the common endpoint of liver toxicity.

5.3 CLASSIFICATION OF CARCINOGENIC POTENTIAL

See attached CARC document (Section 9.5).

On September 29, 1999, the Cancer Assessment Review Committee (CARC) of the Health Effects Division of the Office of Pesticide Programs met to evaluate the carcinogenic potential of clodinafop-propargyl (CGA-184927). The studies evaluated included a 24-month chronic toxicity/carcinogenicity study in Tif: RAIf (SPF) albino rats and an 18-month carcinogenicity study in Tif:MAGf (SPF) albino mice as well as mechanistic studies submitted by the Registrant to support a non-linear mode of action for induction of prostate, ovarian, and liver tumors.

In the studies below, CGA-184927 was administered in the diet to rats (80/sex/group) at 0, 1, 10, 300 or 750 ppm (0, 0.031, 0.32, 10.18, or 26.28 mg/kg/day for males; and 0, 0.034, 0.36, 11.31, or 29.48 mg/kg/day for females, respectively) and to mice (60/sex/group) at 0,

1, 10, 100 or 250 ppm (0, 0.113, 1.10, 11.0 or 29.6 mg/kg/day for males and 0, 0.129, 1.25, 12.6 or 33.1 mg/kg/day for females, respectively).

• **CGA-184927 was carcinogenic to rats** because: 1) In males there were significant increases in the pair-wise comparisons of the high-dose group (750 ppm or 26.28 mg/kg/day) with controls for prostate gland adenomas ($p<0.05$) and combined adenomas/carcinomas ($p<0.01$). There were also significant increasing trends for prostate adenomas and combined adenomas/carcinomas; 2) Females had a statistically significant ($p<0.05$) increased incidence at 750 ppm (29.5 mg/kg/day) by pair-wise comparison with the controls for ovarian tubular adenomas. There was also a statistically significant ($p<0.01$) increasing trend for these tumors. The dosing at the highest dose in males was considered to be adequate based on increased mortality, increased liver weights and non-neoplastic changes in various organs. The CARC considered the prostate and ovarian tumors to be treatment-related.

• CGA-184927 was carcinogenic to mice because: 1) Males had a statistically significant ($p<0.01$) increase in the pair-wise comparisons of the high dose group (250 ppm or 29.6 mg/kg/day) with the controls for hepatomas and combined hepatomas/carcinomas. The incidences of these tumors exceeded the range of historical controls. The increased incidence of carcinomas in high-dose males was considered by the CARC to be biologically significant. There were also statistically significant increasing trends for hepatomas ($p<0.01$), carcinomas ($p<0.05$), and combined hepatomas/carcinomas ($p<0.01$); 2) In females, the incidences of liver tumors were not significant by pair-wise comparison with controls and were within the historical control range. However, there were statistically significant increasing trends in hepatomas ($p<0.01$), and combined hepatomas/carcinomas ($p< 0.05$); 3) In females, there was a borderline increase in hemangiomas and angiosarcomas compared with controls. The combined incidence of these tumors was outside the range of historical controls. These tumors are considered to be uncommon and therefore, the CARC concluded they could not be discounted. There was a split vote among Committee members regarding adequacy of dosing. However, the majority concluded that the dosing at the highest dose was adequate and not excessive in both sexes based on increased liver weight and presence of non-neoplastic changes.

The acceptable genetic toxicology studies indicated that CGA 184927 was not mutagenic in *Salmonella typhimurium* and cultured Chinese hamster V79 lung fibroblast cells. It was not clastogenic *in vivo* and did not induce unscheduled DNA synthesis (UDS) in primary rat hepatocytes. The submitted *in vitro* cytogenetic assay was unacceptable. The CARC therefore, recommended that an *in vitro* cytogenetic assay be conducted to fulfill the guideline requirements. This recommendation was strengthened by the evidence from the literature that propargyl alcohol, a possible metabolite of CGA 184927, induced

chromosome aberration *in vitro*.

Structurally-related compounds, haloxyfop-methyl and diclofop-methyl are hepatocarcinogens in mice while fluazifop-butyl and diclofop-methyl are non mutagens.

The Committee determined that the mechanistic studies do not support the proposed mode of action for the occurrence of prostate and ovarian tumors in rats or liver and blood vessel tumors in mice.

In accordance with the EPA Draft Guidelines for Carcinogen Risk Assessment (July, 1999), the CARC classified clodinafop-propargyl (CGA 184927) as **"likely to be carcinogenic to humans "** by the oral route based on the occurrence of prostate tumors in male rats, ovarian tumors in female rats and liver tumors in both sexes of mice, as well as blood vessel tumors in female mice. For the quantification of human cancer risk, the Committee recommended a linear low-dose extrapolation approach based on the most potent of these tumor types. This approach is supported by possible genotoxic potential and the lack of confirmation of the mode of action of CGA 184927.

The unit risk, Q_1^* $(mg/kg/day)^{-1}$, of CGA 184927 based upon female rat ovarian tubular adenoma tumor rates is 1.29×10^{-3} in human equivalents. The dose levels used from the 106-week dietary study were 0, 1, 10, 300, and 750 ppm of CGA 184927. The corresponding tumor rates were 2/67, 1/65, 1/70, 1/68, and 9/66, respectively.

6.0. FQPA CONSIDERATIONS

The Health Effects Division (HED) FQPA Safety Factor Committee met on March 6, 2000 and again on March 20, 2000 to evaluate the hazard and exposure data for clodinafop-propargyl and recommended that the FQPA safety factor (as required by the Food Quality Protection Act of August 3, 1996) be retained at *up to* 10x in assessing the risk posed by this chemical.

Application of the Safety Factor - Population Subgroups / Risk Assessment Scenarios

When assessing **Acute Dietary Exposure,** the safety factor is **Retained at 10x** for the **Females 13-50 Population Subgroup** since there are data gaps in the toxicology database for clodinafop-propargyl including a developmental neurotoxicity study and there is quantitative evidence of increased susceptibility to the young following *in utero* exposure to clodinafop-propargyl in the prenatal developmental study in rats.

The safety factor can be **Reduced to 3x** for the **Infants and Children Population Subgroups** when assessing acute dietary exposure since the increased susceptibility observed following *in utero* exposure is only of concern for the developing fetus, leaving only the uncertainty due to the data gap for the developmental neurotoxicity study. See also the requirement for a developmental neurotoxicity study (Section 6.2, below).

When assessing the **Chronic Dietary Exposure,** the safety factor should be **Retained at 10x** for **All Population Subgroups** since there is concern for qualitative increased susceptibility of the young demonstrated after repeated oral exposures in the 2-generation reproduction study and since there are data gaps in the toxicology database including a developmental neurotoxicity studies in rats.

6.1 Special Sensitivity to Infants and Children

The HIARC concluded that there is concern for the increased susceptibility of the young to exposure to clodinafop-propargyl based on the developmental toxicity study in rats where increased skeletal effects were observed at doses much lower (40 mg/kg/day) than the maternal NOAEL (160 mg/kg/day). There was no evidence of increased susceptibility in the prenatal developmental toxicity study in rabbits since no treatment-related developmental toxicity was observed.

Although there was no evidence of reproductive toxicity, a fetotoxic effect was noted in the two-generation reproduction study in rats since reduced fetal viability, decreased pup body weight, and dilatation of renal pelvis were observed in the offspring at doses that produced relatively minimal parental toxicity (decreased body weight gain, increased liver and kidney weights with histopathological changes).

6.2 Recommendation for a Developmental Neurotoxicity Study

The HIARC has requested acute and subchronic neurotoxicity studies in order to further define the neurotoxic potential of this chemical due to the observation of clinical signs indicative of neurotoxicity in the developmental, subchronic, and chronic studies conducted in dogs, rats, and rabbits. At the April 29, 1999 meeting, the HIARC reserved its recommendation for a developmental neurotoxicity study for clodinafop-propargyl pending submissions and review of the acute and subchronic neurotoxicity studies (HED Doc. No.013425).

On March 20, 2000, the FQPA Safety Factor Committee recommended that a developmental neurotoxicity study be conducted with clodinafop-propargyl based on the

evidence of potential endocrine disruption in the special studies submitted by the Registrant including the Trendelenburg *in vivo* study (1999; MRID 44767401). These mechanistic studies were not evaluated by the HIARC but were included in the September 29, 1999 evaluation of clodinafop-propargyl by the HED Carcinogenicity Assessment Review Committee (HED Doc. No. 013893). **The FQPA SFC recommended that a developmental neurotoxicity study be conducted to evaluate hormonal responses associated with the developing fetal nervous system following pre- and postnatal exposure.**

7.0 OTHER ISSUES

None at this time.

8.0 REFERENCES in MRID order Studies by MRID number: .

A. Technical Agent Studies.

Study Type	MRID#
Acute oral toxicity- Mouse	44399123
Acute oral toxicity- Rat	44399124
Acute dermal toxicity- Rat	44399125
Acute inhalation toxicity- Rat	44399126
Primary eye irritation- Rabbit	44399127
Primary dermal irritation- Rabbit	44399128
Dermal sensitization- Guinea pig	44399129
28-day oral (gavage)- Rat	44399130
Subchronic oral (feeding)- Rat	44399132
Subchronic oral (feeding)- Mouse	44399138
Subchronic oral (feeding)- Dog	44399139
28-day dermal toxicity- Rat	44399141
Chronic oral (feeding)- Dog	44399142
Carcinogenicity- Mouse	44399143
Developmental toxicity- Rabbit	44399144
Developmental toxicity- Rat	44399145
Two Gen reproduction- Rat	44399146
Combine chronic/carcinogenicity- Rat	44399147

Mutagenicity (mouse micro-nucleus)	44399151
Mutagenicity (gene mutation in mammalian cells, Chinese hamster cells)	44399152
Mutagenicity (Salmonella /mammalian microsome mutagenicity assay)	44399153
Mutagenicity (in vitro cytogenetic with human lymphocyte	44399154
Mutagenicity (UDS in human fibroblasts)	44399155
Mutagenicity (UDS in rat hepatocytes)	44399156
Metabolism- Rat	44399159
Metabolism- Rat	44399160
Special study	44399134
Special study	44399135
Special study	44399137
Special study	44399148
Special study	44399149
Special study	44399150
Special study	44399157
Special study	44399158
Special study	44767401
Special studies	No MRID assigned

B. End-use Product (Clodinafop 2E Herbicide)

Study Type	MRID#
Acute oral- Rat	44387304
Acute dermal- Rat	44387305
Acute inhalation- Rat	44387306
Primary eye irritation- Rabbit	44387307
Primary dermal irritation- Rabbit	44387308
Dermal sensitization- Guinea pig	44387309
Subchronic oral (gavage)- Rat	44399131
Subchronic oral (feeding)- Rat	44399133
Subchronic oral (feeding)- Dog	44399140
21-day dermal toxicity- Rat	44387310

9.0 APPENDICES.

9.1 Toxicity Profile Summary Tables.

9.1.1 Acute Toxicity Table (see Section 4.1)

9.1.2 Subchronic, Chronic and Other Toxicity Studies.

TABLE 4. Toxicity Summary Table for Clodinafop-propargyl (CGA 184927)

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
870.3100/ 28-Day oral toxicity in rats, gavage	44399130 (1988) / 0, 5, 40, 200 mg/kg/day Acceptable Study, None- Guideline	NOAEL < 5 mg/kg LOAEL = 5 mg/kg for M and F based on liver toxicity (enzyme changes., At higher doses, mortality,decr. body wt; liver changes -- ALP, ALT, increased liver wt., hepatocellular hypertrophy; incr. BUN and glucose.
870.3100/ 13 week oral toxicity in rats,. feeding study.	44399132 (1988)/ 0, 2, 15, 120, 1000 ppm M: 0, 0.1, 0.9, 8.2, 70.0 mg/kg/day F: 0, 0.1, 0.9, 8.2, 71.1 mg/kg/day. Acceptable Study.	NOAEL = M:: 15 ppm, 0.9 mg/kg; F: 120 ppm 8.2 mg/kg/day LOAEL = M: 120 ppm (8.2 mg/kg/day); F: 1000 ppm (71.1 mg/kg/day) decreased body wt; based on increased liver weights and enzymes (AlPtase); decr. thymus wt. (atrophy). Reversed after 28 days recovery period.
870.3100/ 13 week oral toxicity in mice,. dietary	44399138 (1989)/ 0, 2, 6, 50, 400 ppm M: 0,0.3, 0.9, 7.3, 53 mg/kg/day F: 0,0.3, 1.1, 8.6, 71.3 mg/kg/day **UNACCEPTABLE** (variability in concentrations)	NOAEL = 6 ppm (0.9(♂)/1.1(♀)) LOAEL = 50 ppm (7.3(♂)/8.6(♀)) based on clinicl chemistries (liver enzymes-- Alk Ptase); glucose, sodium and chloride increases and hepatocellular hypertrophy in males and females.

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
870.3150-- 90-Day oral toxicity in dogs, feeding	44399139 (1989)/ 0, 10, 50 and 200 ppm for 90 days and 1/1000/500 ppm for days 1-54/55-66/67-90. M: 0,0.35, 1.7, 7.9, 0.04/34.7.16 mg/kg/day(♂) 0,0.4, 1.9, 7.2, 0.04/32.3/16.9 mg/kg/day(♀) Acceptable Study	The NOAEL was determined to be 10 ppm for males (0.346 mg/kg/day), and 50 ppm for females (1.89 mg/kg/day). The LOAEL is 50 ppm (1.73 mg/kg/day) for males and 200 ppm (7.16 mg/kg/day) for females, based on occurrence of skin lesions.
870.3200-- 28-Day dermal toxicity in rats.	44399141 (1987)/ 0, 50, 200 and 1000 mg/kg/day. Acceptable Study	Systemic NOAEL = 50 mg/kg/day LOAEL = 200 mg/kg **based on dose-related increases in liver weights and clinical signs (piloerection and hunched posture) in male rats.** **The dermal toxicity NOAEL is 1000 mg/kg/day.**
870.3700a Prenatal developmental in rats, oral gavage. French	44399145 (1989)/ 0, 5, 40, 160 mg/kg/day(GD 6-15) Acceptable Study	**Maternal** NOAEL = 160 mg/kg/day LOAEL > 160 mg/kg/day based on **lack of effect.** **Developmental** NOAEL = 5 mg/kg/day LOAEL = 40 mg/kg/day **based on increased incidences of bilateral distension and torsion of the ureters, unilateral 14th ribs, and incomplete ossification of the metacarpals and various cranial bones (parietals, interparietals, occipital, and squamosal).**
870.3700b Prenatal developmental in rabbits	44399144 (1989)/ 0, 5, 25, 125, 175 mg/kg/day(GD 7-19) Acceptable Study	**Maternal** NOAEL = 25 mg/kg/day LOAEL = 125 mg/kg/day based on **mortality, clinical signs and body weight loss and the maternal** **Developmental** NOAEL = 125 mg/kg/day LOAEL >125 mg/kg/day based on **based on lack of developmental toxicity.**

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
870.3800 2 Generation Dietary Reproduction and fertility effects in rats.	44399146 (1991)/ 0, 5, 50, 500 or 1000 ppm 0, 0.33, 3.21, 31.69, and 64.24 mg/kg/day in males 0, 0.41, 3.77, 37.54, and 73.60 mg/kg/day in females. Acceptable Study	**Parental/Systemic NOAEL 50 ppm (3.2 mg/kg/day). LOAEL = 500 ppm (31.7 mg/kg/day) based on decrease in body weight gain, reduced food consumption, increased liver and kidney weights and histopathological changes in the liver (hepatocytic hypertrophy), and renal tubules (hyaline casts, parenchymal atrophy, pigment deposits, dilatation and loss of tubular epithelium).** **Offspring NOAEL = 50 ppm (3.2 mg/kg/day). LOAEL = 500 ppm (31.7 mg/kg/day) based on reduced viability, decreased pup body weight and dilatation of renal pelvis.** **Reproductive NOAEL = 1000 ppm (64.2 mg/kg/day). LOAEL ≥1000 ppm (≥64.2 mg/kg/day) based on lack of reproductive effects.**
870.4100b 1 year chronic toxicity dogs, Feeding	44399128 (1990)/ 0, 10, 100 or 500 ppm 0, 0.3, 3.4, 15.2 mg/kg/day (♂) 0, 0.3, 3.4, 16.7 mg/kg/day(♀) Acceptable Study	NOAEL = 100 ppm (3.38 and 3.37 mg/kg/day for males and females, respectively). LOAEL = 500 ppm (15.2 and 16.7 mg/kg/day for males and females, respectively), based on occurrence of skin lesions, clinical signs and reduced body weight gain and food consumption.

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
870.4200b 18 mo. carcinogencity mice, feeding.	44399143 (1992)/ 0, 1, 10, 100 or 250 ppm 0, 0.113, 1.10, 11.0 or 29.6 mg/kg bw/day for males and 0, 0.129, 1.25, 12.6 and 33.1 mg/kg bw/day for females, Acceptable Study	The Systemic NOAEL was estimated to be 1.10 and 1.25 mg/kg/day for males and females, respectively. **LOAEL for systemic toxicity is 11.0 and 12.6 mg/kg/day for males and females, respectively, based on increase in liver enzyme activity and liver weights.**
870.4300 2-YR Chronic/ Oncogencity Feeding Study in the Rat.	44399147 (1992)/ 0, 1, 10, 300 or 750 ppm 0,0.03, 0.3, 10.2, 26.3 mg/kg/day(♂) 0, 0.03, 0.4, 11.3, 10.2, 29.5 mg/kg/day(♀) Acceptable Study	Systemic NOAEL = 1 ppm [0.03 (♂) and(♀)] mg/kg/day Systemic LOAEL = 10 ppm [0.3(♂)/0.4(♀) mg/kg/day] based on hepatocytic hypertrophy, chronic progressive nephropathy and tubular pigmentation. **Under the conditions of this study, treatment with CGA 184927 increased the incidence of prostate and ovarian tumors in rats at 750 ppm. For males, an increased incidence of prostate adenoma was seen in the high-dose group, i.e., incidence rates were 8/80 (10.0%), 9/80 (11.25%), 12/80 (15.0%), 13/80 (16.25%) and 19/80 (23.75%) in the 0, 1, 10, 300 and 750 ppm groups, respectively. At 750 ppm, one of 80 males developed hepatocarcinoma.** **For females, an increased incidence of tubular adenomas of the ovary was noted in the high-dose group, i.e., incidence rates of 2/80 (2.5%), 1/80 (1.25%), 1/80 (1.25%), 1/80 (1.25%) and 9/80 (11.25%) for the 0, 1, 10, 300 and 750 ppm groups, respectively. The chemical was administered at a dose sufficient to test its carcinogenic potential.**

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
870.5100 Gene Mutation Salmonella and Eschericia/liver microsome test.	44399153/ (1990) Acceptable Study	Neg. for mutagenicity.
870.5200 Gene Mutation Mutation test with Chinese Hamster cells V79	44399152 (1987)/ Acceptable Study	Neg. for mutagenicity.
870.5315 Chromosome Studies Human Lymphocytes in vitro.	44399154 (1988) Unacceptable Study	**Owing to the conflicting results from the cytotoxicity assessment and the presence of rare complex chromosome aberrations both with and without S9 activation, the study is considered inconclusive.**
870.5395 Micronucleus Test (Mice)	44399151 (1987)/ Acceptable Study	**No clear evidence that Clodinafop-propargyl induced a clastogenic or aneugenic effect in either sex at any dose or sacrifice time.**
870.5550 DNA Repair Human Fibroblasts.	44399155 (1988)/ Unacceptable Study No cytotoxicity at any dose used.	Compound precipitation was seen at doses ≥320 μg/mL: there was, however, no indication of a cytotoxic effect at any dose. The positive control induced the expected marked increases in UDS. **There was, however, no evidence that CGA- 184927 in the absence of S9 activation induced a genotoxic response in either trial.**
870.5550 DNA Repair Rat Hepatocytes	44399156 (1987)/ Acceptable Study	Compound precipitation was noted at levels ≥4000 μg/mL. Lethality was apparent in the preliminary cytotoxicity test at 94.8 μg/mL. The positive control induced the expected marked increases in UDS. **There was, however, no evidence that CGA 184927 induced a genotoxic response in either trial.**

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Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
870.7485 Metabolism and pharmaco-kinetics, Rat gavage study.	44399159 (1989)/ [3- 14C] Quinoline CGA-185072 Acceptable Study	The main metabolite was CGA 19469 (propionic acid , 76% in male urine, Additional 5% was in the form of taurine conjugate of CGA 193469. Similar distribution was found in feces.
870.7485 Metabolism and pharmacokinetics, Rat gavage study.	44399160 (1990)/ Acceptable Study	The major metabolite in urine and feces was determined to be CGA 193469, accounting for about 36% to 47% of the AD for males, and 80% to 85% of the AD for females. In addition, 11 minor metabolite fractions were isolated from urine and feces. Three were further identified as reference materials CGA 193468, CGA 214111 and unchanged clodinofop-propargyl.
Special Study: Determination Of Residues As CGA 193469 In Abdominal Fat After A 3-Month Oral Toxicity Study In Rat.	44399134 14 week treatment, 0, 2, 15, 120 and 1000ppm	There was a dose-dependent increase in CGA 184927 residues in fat samples from both sexes taken at the end of treatment (14 weeks) and after the 4-week recovery period (18 weeks). Concentrations of CGA 184927 were higher in male rats at all dose levels tested. With the exception of low-dose group males, for all remaining groups, residues in the fat at 18 weeks had decreased by between 40% - 51.5% of the 14 week value.
Special Study: Determination Of Residues As CGA 193469 In Abdominal Fat After 12 and 24 months of treatment.	44399135 1, 10, 300 and 750 ppm. Acceptable Non-guideline study.	**Residues in fat at 12 and 24 month of 1 and 10 ppm feeding level indicated concentration of CGA 184927 in the abdominal fat was higher in males when compared to females. At 300 and 750 ppm, the concentration of CGA 184927 in the abdominal fat was comparable between males and females. The results indicated that residues at 12 months were lower than residues at 3 months.**

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
Special Study: The Effect Of CGA 184927 On Selected Biochemical Parameters In The Rat Liver Following Subchronic Administration.	44399137	**The effects of CGA 184927 on selected liver enzymes in the rat were similar to the effects seen after subchronic treatment with known peroxisome proliferators (hypolipidemic compounds, phenoxyacetic acid derivatives). Hence, CGA 184927 was considered to most likely be a peroxisome proliferator in the rat liver.**
Special Study: Apparently clonal thyroid adenomas may contain heterogeneously growing and functioning cell subpopulations. New Frontiers in Thyroidology, p. 901-905, 1986 (Published Article)	44399148	The asynchronous growth rate of subsets of cells within the old adenomas as well as the intercellular heterogeneity of the endocytotic response to TSH suggests that clonal thyroid adenomas may acquire new qualities and can modify gene expression via much debated mechanism. The author concludes that the growth of benign thyroid tumors and progression does not require a change in genomic expression in any cell. The apparent heterogeneity of a tumor does not necessarily exclude its monoclonal origin.
Special Study: Assessment of hyperplastic and neoplastic lesions of the thyroid gland. TIPS, Vol. 8, p. 511-514., dated December 1987. (Published)	44399149	In cell cultures, TSH does not induce proliferation of human thyroid cells, but does stimulate the growth of cells obtained from rat and dog thyroids. Conventional procedures of evaluating carcinogenicity tests by simply counting tumors in rodents treated with high doses, and by mathematical extrapolation to the low doses to which humans are exposed, are not suitable for the proliferative reactions of the thyroid gland. In assessing the human risk, relevant conclusions can only be drawn if the physiological factors of growth control are known, and if the biological mechanisms by which chemicals initiate focal proliferation and support their progression to tumors are considered.

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
Special Study Stott, W.T. (1988). Chemically induced proliferation of peroxisomes: Implications for risk assessment. Regulatory Toxicology and Pharmacology, Vol. 8, p. 125-159, 1988. (Published).	44399150	The concept of "maximum tolerated dose level" (MTD) as it pertains to chronic oncogenicity bioassays for peroxisome proliferators. A more appropriate MTD of a peroxisome proliferative agent in sensitive species would appear to be based upon evidence of the proliferation of peroxisomes and the induction of peroxisomal enzymes capable of producing an increased intracellular oxidative stress. Exceeding these dosages will only result in a predictable sequence of events leading, ultimately, to tumor formation due to physiological adaptation of the animal to the administered compound rather than from the direct effects of the compound itself.
Special Study: Stott, W.T. (1988). Chemically induced proliferation of peroxisomes: Implications for risk assessment. Regulatory Toxicology and Pharmacology, Vol. 8, p. 125-159, 1988. (Published)	44399150	The author concludes that based on the present understanding of MTD) as it pertains to chronic oncogenicity bioassays for peroxisome proliferators. A more appropriate MTD of a peroxisome proliferative agent in sensitive species would appear to be based upon evidence of the proliferation of peroxisomes and the induction of peroxisomal enzymes capable of producing an increased intracellular oxidative stress. Exceeding these dosages will only result in a predictable sequence of events leading, ultimately, to tumor formation due to physiological adaptation of the animal to the administered compound rather than from the direct effects of the compound itself.

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
Special Study Bieri, F. (1991). The Effect of Cga 193469, the Free Acid Derivative of Cga 184927, on Peroxisomal B-oxidation in Primary Cultures of Rat, Mouse, Marmoset and Guinea Pig Hepatocytes.	44399157	**CGA 184927-induced cytotoxicity through propargyl alcohol released following its enzymatic hydrolysis. In cultured hepatocytes from all species, CGA 193469, the free acid derivative of CGA 184927, was noncytotoxic at concentrations of ≤100 µg/ml as examined by light microscopy and quantified for LDH activity. There was no change in the total LDH activity and the protein contents were unaffected indicating that there was no cell leakage of LDH. CGA 193469 induced in the cultured rat and mouse hepatocytes the effect characteristically produced by other peroxisomal proliferators. In rat hepatocytes, it induced-peroxisome beta-oxidation in a dose-dependent manner. This enzyme activity was marginal in the guinea pig hepatocytes and was non-measurable in marmoset hepatocytes.**
Special Study Guyomard, C. (1992). Effects of CGA 193469, the acid derivative of CGA 184927, on the peroxisomal beta-oxidation in human hepatocytes.	44399158 0, 750 ppm (0, 53.5 mg/kg/day) for 14 days	Under the conditions of this study, neither CGA 193469 nor bezafibric acid induced peroxisomal beta-oxidation in human hepatocytes, in vitro. However, in the absence of a known concurrent human positive control to validate the test system, i.e., a substance known to elicit peroxisomal beta-oxidation in human hepatocytes, this cannot be definitely concluded.
Special Study: Roberts R. (1999). CGA-193469 (free acid derivative of CGA-184927) peroxisome proliferators: species difference in the regulation of gene expression	44923101	CGA 183469 is able to activate the rodent ACO gene promoter-reporter construct as expected for a rodent peroxisome proliferator. In contrast, CGA 193469 was unable to activate the human ACO promoter-reporter construct.

Guideline No./ Study Type	MRID No. (year)/ Doses/ Classification	Results
Special Study: Staubli, W. and Bentley, P. (1989). Electron Microscopical Study, PS 3.3 Cell Biology, dated April 25, 1989.	MRID NUMBER: Not assigned From subchronic study: control and 1000 ppm.	At 14 weeks, an increase in the number and size of peroxisomes with matrical inclusion bodies was noted in the 1000 ppm group, males only. At 18 weeks (4 week recovery period), the number and size of peroxisomes in the 1000 ppm group, both sexes, were comparable to the control group. CGA 184927 is a peroxisome proliferator in the rat liver.
Special Study: Trendelenburg, C. (1999) Effects on selected plasma concentrations and biochemical parameters in the liver upon subchronic administration to male adult rats.	443767401 Acceptable non-guideline.	CGA 184927 acts as a peroxisomal proliferating agent and alters monooxygenase activity in subfamilies of cytochrome P450 which are known to be involved in the synthesis or catabolism of steroid hormones.

9.2 Summary of Toxicological Dose Endpoints.

TABLE 5. Doses and Toxicological Endpoints Selected for Various Exposure Scenarios on Clodinafop-propargyl (CGA 184927)

EXPOSURE SCENARIO	DOSE (mg/kg/day)	ENDPOINT	STUDY
Acute Dietary (For females 13+)	NOAEL=5 (UF=100)	Increased incidences of bilateral distension and torsion of the ureters, unilateral 14th ribs, and incomplete ossification of the metacarpals and various cranial bones.	Developmental toxicity study in rats
	Acute RfD (females 13+) = 0.05 mg/kg/day		

ute Dietary or general opulation)	NOAEL= 25 (UF=100)	Maternal toxicity (increased mortality, clinical signs and body weight loss)	Developmental toxicity study in rabbits
	Acute RfD (general population) = 0.25 mg/kg/day		
onic Dietary	NOAEL=0.03 (UF=100)	Hepatocytic hypertrophy, chronic progressive nephropathy and tubular pigmentation	Chronic Toxicity -Rat
		Chronic RfD = 0.0003 mg/kg/day	
hort-term (Dermal)	Dermal NOAEL=50	Increased liver weight and clinical signs (piloerection and hunched posture) in males	28-Day Dermal Toxicity-Rats
nediate-Term (Dermal)			
term (Dermal)	Not Applicable	Based on the current use pattern, no long-term dermal exposure is expected to occur.	
hort-term nhalation)	Oral NOAEL= 5[a]	See acute dietary	Developmental toxicity in rats
mediate-Term nhalation)	Oral NOAEL=0.9[a]	Increased liver weight and enzyme activity in males	Subchronic oral toxicity study in rats
ong-term nhalation)	Not Applicable	Based on the current use pattern, no long-term inhalation exposure is expected to occur.	

route to route extrapolation

HIARC Memorandum (6/2/99).

FQPA Memoranda (3/6/00 and 3/20/00).

CARC Report (12/7/99).

Q* Memorandum (3/2/00).