SUBJECT: Prowl - requesting the establishment of a temporary tolerance for combined negligible residues of the herbicide Prowl (N-(1-ethylpropyl)-2,6-dinitro-3,4-xylidine) and its metabolite 4[(1 ethylpropyl)amino]-2-methyl-3,4-dinitrobenzyl alcohol in or on corn grain at 0.1 ppm.

TO: Mr. Lee TerBush
Acting Chief
Coordination Branch

Pesticide Petition No.: 4G1451 - American Cyanamid Co.
Agriculture Division
P.O. Box 400
Princeton, N.J. 08540

Formulation:

Prowl 3E Herbicide

Active Ingredient: ZW/W

Prowl (93%) ......................... 38.1

Inert Ingredients:

Prowl 4E Herbicide

Active Ingredient:

Prowl (93%) ......................... 47.7
Use Pattern:

The following weed species are susceptible to preemergence treatments of Prowl at the rates recommended according to soil type.

- Green foxtail (*Setaria viridis*)
- Yellow foxtail (*Setaria lutescens*)
- Giant foxtail (*Setaria faber*)
- Barnyardgrass (*Echinochloa crus-galli*)
- Fa-1 panicum (*Panicum dichotomiflorum*)
- Large crabgrass (*Digitaria sanguinalis*)
- Lambsquarter (*Chenopodium album*)
- Pigweed (*Amaranthus retroflexus*)
- Velvetleaf (*Abutilon theophrasti*)
- Smartweed (*Polygonum pensylvanicum*)

For control of the following weeds, Prowl 4E should be used in combination with either atrazine or Bladex at the rates recommended according to soil type.

- Ragweed (*Ambrosia spp.*)
- Jimsonweed (*Datura stramonium*)
- Morningglory (*Ipomoea spp.*)
- Cocklebur (*Xanthium pensylvanicum*)
- Mustard (*Brassica spp.*)

**Prowl 4E Weed Control Recommendation Table**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Rate per Acre</th>
<th>Soil Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prowl 4E</td>
<td>1 1/2 qts.</td>
<td>2 to 3.5% O.M.</td>
</tr>
<tr>
<td>Prowl 4E</td>
<td>2 qts.</td>
<td>greater than 3.5% O.M.</td>
</tr>
<tr>
<td>Prowl 4E + atrazine 80W</td>
<td>1 qt. + 1.3 lbs.</td>
<td>2 to 3.5% O.M.</td>
</tr>
<tr>
<td>Prowl 4E + atrazine 80W</td>
<td>1 1/2 qts. + 1.9 lbs.</td>
<td>more than 3.5% O.M.</td>
</tr>
<tr>
<td>Prowl 4E + Bladex 80W</td>
<td>1 qt. + 1.9 lbs.</td>
<td>2 to 3.5% O.M.</td>
</tr>
<tr>
<td>Prowl 4E + Bladex 80W</td>
<td>1 1/2 qts. + 2.5 lbs.</td>
<td>more than 3.5% O.M.</td>
</tr>
</tbody>
</table>
Toxicology:

Acute Studies:

Technical

- Rats Oral - LD<sub>50</sub> 1250 mg/kg
- Dogs Oral - LD<sub>50</sub> > 5000 mg/kg
- Mice Oral - LD<sub>50</sub> > 1620 mg/kg (m) > 1340 mg/kg (f)
- Rabbits Dermal - LD<sub>50</sub> > 5000 mg/kg
- Rabbits Skin irritation - not irritating
- Rabbits Eye irritation - slight transient conjunctivitis
- Rat Inhalation - LC<sub>50</sub> > 320 mg/L
- Mice 30 Day oral gave NEL of 500 ppm
- Rats 30 Day oral gave NEL of 1600 ppm
- Dog 30 Day oral gave NEL of 1 g/kg

Formulation

Prowl 3E:

- Rat oral LD<sub>50</sub> - 4900 mg/kg
- Rabbit Dermal LD<sub>50</sub> -> 5230 mg/kg
- Rabbit Skin irritation - considered only slightly toxic score 3.9
- Rabbit Eye irritation - irritating to eyes
- Rat Inhalation - LD<sub>50</sub> 2230 mg/kg (m), 1770 mg/kg (f).

Prowl 4E:

- Rat Oral - LD<sub>50</sub> 3380 mg/kg
- Rabbit Dermal - LD<sub>50</sub> > 5495 mg/kg
- Rabbit Skin irritation - considered only slightly toxic score 5.0
- Rabbit Eye irritation - Irritating to eyes

Prowl + Atrazine 80W:

- Rat Oral - LD<sub>50</sub> > 5 g/kg
- Rabbit Skin irritation - relatively non toxic score 0.80
- Rabbit Eye irritation - not irritating to eye

Prowl + Bladex 4F:

- Rat Oral - LD<sub>50</sub> > 5 g/kg
- Rabbit Skin irritation - relatively non toxic score 1.4
- Rabbit Eye irritation - not irritating to eye
Subacute Studies:

- Rabbit 21 day Dermal - 30 male and 30 female New Zealand rabbits were given repeated dermal applications of technical Prowl and a similar group of rabbits were given the same repeated dermal application with a final formulation of 3 lbs active per gallon. Rabbits were treated according to the following schedule:

<table>
<thead>
<tr>
<th>Group No.</th>
<th>Male</th>
<th>Female</th>
<th>Test Material</th>
<th>Treatment Level</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>6</td>
<td>6</td>
<td>Corn Oil/E.C.formulation</td>
<td>2.0 ml/kg</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>4</td>
<td>TG* 92553</td>
<td>250 mg/kg</td>
</tr>
<tr>
<td>3</td>
<td>4</td>
<td>4</td>
<td>TG* 92553</td>
<td>500 mg/kg</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>4</td>
<td>TG* 92553</td>
<td>1000 mg/kg</td>
</tr>
<tr>
<td>5</td>
<td>4</td>
<td>4</td>
<td>FF** 92553</td>
<td>0.5 ml/kg</td>
</tr>
<tr>
<td>6</td>
<td>4</td>
<td>4</td>
<td>FF** 92553</td>
<td>1.0 ml/kg</td>
</tr>
<tr>
<td>7</td>
<td>4</td>
<td>4</td>
<td>FF** 92553</td>
<td>2.0 ml/kg</td>
</tr>
</tbody>
</table>

TG* = technical grade material  
FF** = final formulation

NOTE: The control animals (Group No. 1) were sham-treated with the E.C. formulation and vehicle corn oil used in the final formulation at a volume equivalent to that given the high level test groups.

Results of this study are as follows:

Food and water intake, elimination and general behavior revealed no adverse or pharmacologic effects.

Urine analyses for all rabbits were normal.

Hemograms for all groups were comparable to controls.

Groups 2, 3 and 4 receiving the technical compound produced minimal edema and erythema, which lessened by 21 days. Groups 5, 6 and 7 produced severe erythema and edema, which in most cases remained until termination of the experiment after 21 days of treatment. The control group also developed moderate to severe edema and erythema therefore it was concluded that the skin irritation was due to the carrier and not the active ingredient.
Two rabbits died during the second week, one control after blood sampling and one (1000 mg/kg) died secondary to pancreatic focal necrosis, acute splenitis and lymphadenitis.

A dose-related, superficial pustular hyperkeratosis and acanthosis with a variable inflammatory response was seen microscopically in those animals in groups 5, 6 and 7.

- **Beagle Dog 90 day oral** - 16 male and 16 female beagle dogs were divided into three test groups each consisting of 4 male and 4 female dogs. The low dose level test group was fed 62.6 mg/kg as a dietary admixture 7 days each week. Due to incompatibility of the test material at higher doses the 250 and 1000 mg/kg levels were given by oral intubation in a 50% aqueous suspension 5 days each week.

Daily observations were recorded for each dog on appetite, elimination, general appearance, behavior and gross signs of systemic toxicity.

Clinical Laboratory Studies were twice initially, at 1 and 3 months of the study for each dog as follows:

Hematology: Hgb, Hct, Sed. rate, RBC, differential count, reticulocyte count, prothrombin time, and Lee-White clot time.

Blood chemistry: Blood glucose, urea nitrogen, SGPT, SGOT and SAP.

Urine Analyses: appearance, sp. gravity, pH, occult blood, protein, bilirubin and ketones.

Terminal Studies were conducted on all animals that died as well as all survivors sacrificed at termination of this 90-day study. Histological examinations were conducted on H & E stained slides of the following:

gall bladder  liver  spleen  
stomach  small intestine  large intestine  
pancreas  adrenals  kidneys  
urinary bladder  pituitary  thyroid  
gonad  salivary gland  thymus  
heart  aorta  lymph nodes  
marrow  skin  lungs  
skeletal muscle  spinal cord  mammary glands  
eye  bone  brain  
gross lesions
Results:

One female died during the study because of a technical error in dosing the high dose level animals during the third week of study.

Ophthalmoscopic examination of the dogs during the study revealed no changes associated with the administration of this material. Weekly palpation of the mammary glands revealed no gross pathology.

Slight body weight losses were observed in 2 dogs in the 250 mg/kg group and 6 in the 1000 mg/kg group. However, these changes did not exceed a 2 kg weight loss.

Hematology studies revealed that the tested parameters were within the normal limits and compared favorably with the control animals. At 1 and 3 months there was a slight increase in percent reticulocytes.

Blood chemistry revealed an elevated BUN in one male animal in the high level group during the last month of the test and an increase SGPT in 1 female in the 250 mg group at the 4 week period. All other parameters were within normal limits and comparable to the control animals.

Terminal observations revealed slight increases in organ/body weight ratios in the livers of male test animals according to the petitioner. However, the following table shows that both male and female had liver weight gains at all levels.

<table>
<thead>
<tr>
<th></th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>2.70</td>
<td>2.83</td>
</tr>
<tr>
<td>62</td>
<td>3.21</td>
<td>3.44</td>
</tr>
<tr>
<td>250</td>
<td>3.62</td>
<td>3.20</td>
</tr>
<tr>
<td>1000</td>
<td>3.50</td>
<td>3.20</td>
</tr>
</tbody>
</table>

Gross necropsy revealed only slight reddening of the small intestine and colon which microscopically revealed only mild patchy congestion. Focal calcifications in the kidney collecting tubules were seen in both control and test animals. However, it is felt that these increases are not dose related or significant. The NEL would therefore be 1000 mg/kg.

Retard for liver increase due to increase of NEL 62.5 mg/(kg/day) hyper trophy?
18 Month carcinogenicity study

Three hundred mice received a control diet while 150 mice per group (75 male, 75 female) received diets containing 100, 500 or 2500 ppm. At the beginning of the 8th week of treatment the concentration in the high dose group was increased from 2500 ppm to 5000 ppm.

At 36 week interim there were no differences in body weight of male mice but at the 5000 ppm level female body weights were significantly less than the control group. However, there was no difference in food consumption of either male or female group at any level fed. Amber urine noted in the 500 and 2500 ppm groups.

At 79 week interim the mean body weights were similar to those reported in the 36 week report. Mortality over the course of the study revealed no difference between control and treated group. No significant difference was found in SGPT, AP, GL, BUN, RBC ChE, Hgb, Hct, RBC or WBC. Amber urines noted at the six month interim were evident only in the 5000 ppm group in the 79 week report. Signs occurring in all groups including the control with no apparent dose relationship included alopecia of the cervical region, staining of the abdominal fur, lacrimation, exophthalmos, ruptured eyes and eye opacities. At this time gross observations of terminally sacrificed mice revealed no differences in tissue mass incidence between control and treated groups.

90 Day male rat feeding study for mammary effect.

Twenty weanling Sprague-Dawley received Prowl in their diet at 2500 ppm for the first 6 weeks and 5000 ppm for the next 7 weeks. A similar group of 20 were maintained as controls. After 60 days 5 rats from each group were examined for microscopic mammary changes. At the conclusion of the study the survivors were examined likewise.

All animals were weighed weekly for the first 6 weeks and bimonthly thereafter.

Results: None of the animals exhibited any gross or microscopic pathology of the mammary glands in either the controls or the test animals.

3 and 24 month oral toxicity and carcinogenicity in rats.

Five groups of 60 male and 60 female Long-Evans rats were fed 100, 500, 2500/5000 ppm and 2 controls. The high dosage group was started at 2500 ppm and on day 35 increased to 5000 ppm.
Parameters tested are as follows:

General: Daily for physical appearance, signs of local or systemic toxicity, pharmacologic effects or mortality.

Body Weight: Weekly, beginning one week prior to treatment, and terminally (after fast).

Food Consumption: Weekly, beginning one week prior to treatment.

Laboratory Studies:

Time Intervals and Number of Animals: Month 3, 3 rats/sex/each control group, and 6 rats/sex/dose group (rats sacrificed at 3 months).

Hematology: Clotting time, hemoglobin, hematocrit, erythrocytes, total and differential leucocytes, erythrocyte morphology.

Clinical Chemistry: Serum glutamic pyruvic transaminase, alkaline phosphatase, fasting blood sugar, blood urea nitrogen.

Urinalysis: Gross appearance, protein, glucose, pH, refractive index, ketones, bilirubin, occult blood.

At termination the number of animals sacrificed were 5 rats/sex from each control group and 10 rats/sex/dose: group.

Tissues Fixed for Histopathologic Examination:

- Adrenals
- Aorta
- Bone marrow (sternum)
- Brain (2 sections)
- Gonad
- Heart (with coronary vessels)
- Eye (with optic nerve)
- Intestine (colon, duodenum, ileum)
- Kidneys
- Liver
- Lung

- Lymph node (mesenteric)
- Mammary gland
- Pancreas
- Pituitary
- Salivary gland
- Prostate
- Spinal cord (thoracic)
- Skin
- Spleen
- Stomach (fundic, pyloric)
- Thyroids
- Urinary bladder (with neck)
- Uterus
- Skeletal muscle with nerve
- Any unusual tissue or lesion
Number of Animals Examinined Histologically: Forty rats - 5 rats/sex from each control group, and 10 rats/sex/dose group (only the kidneys, liver, lung, heart and mammary gland examined in low and mid-dose groups).

Results:

No effects were attributed to the administration of Prowl in evaluations of mortality, symptomology, food consumption, hematology and urinalysis data. However, at the 5000 ppm level there were significant decreases in body weight in both male and females. Alkaline phosphatase levels in all female treated groups and males in the mid and high dose groups were significantly lower than the controls.

Mean liver and thyroid weights increased in all treated groups, the kidney weights and ratios of the treated males, and the kidney/body weight ratio of high-dose females were adjudged to be dose related. However, histological examination revealed no significant change.

Mammary gland hyperplasia in male rats, occasionally with dysplastic changes, occurred with greater degree of change and higher incidence in animals receiving Prowl than those of the control group. However, these changes were not considered to be dose dependent. No morphologic changes were observed in the mammary gland tissue considered to be related to the administration of Prowl. However, a few glands exhibited cystic and degenerative changes with or without associated inflammation.

- Three generation rat reproduction study:

A partial report on this reproduction study was submitted. Three groups of 20 male and 20 females were dosed at levels 0, 500 and 5000 ppm. In this study the mating schedule was as follows:

```
F0 parents \rightarrow 1st mating \rightarrow 2nd mating \rightarrow 3rd mating \rightarrow discarded
\downarrow
F_{1a} offspring sacrificed at weaning
\downarrow
F_{1b} offspring sacrificed at weaning
\downarrow
F_{1c} offspring
\downarrow
\text{Offspring} \rightarrow 1st mating \rightarrow 2nd mating \rightarrow discarded
\downarrow
F_{2a} \quad F_{2b}
```
Results submitted are as follows:

**F₁a Offspring** - The mean live offspring weights of the high-dose treatment group were lower than the weights of the control groups.

**F₂b Offspring** - A decrease in offspring survival was observed in the litters of the high-dose group delivered during the first few days of the paturition period. The diet of this group was removed from dams which already had litters, and from other dams two days prior to their expected day of delivery.

**F₁c Offspring** - The diet of the high-dose group was removed from the dams two days prior to their expected day of delivery. No significant findings were observed.

**F₂a Offspring** - The diet of the high-dose group was not removed from the dams. The weaning meal live offspring weight of the high-dose group was lower than the control mean weight.

**F₂b Offspring** - The diet of the high-dose group was removed from the dams two days prior to their expected day of delivery. No significant findings were observed.

It would appear from these results that the reproduction no-effect level will be somewhere between 500 and 5000 ppm.

- Dominant Lethal Study in Rats

Three groups of 15 albino male rat weanlings were fed Prowl at 0, 500 and 2500 ppm. All rats were fed their respective diets ad libitum with free access to fresh tap water. Following feeding the test material for 60 days these male rats were mated 1:1 with untreated virgin females. Impregnation in the female was determined on the basis of a vaginal plug and this was considered day 0 of gestation. On day 13 of gestation, caesarian sections were conducted on all females mated. The mating schedule was repeated with additional untreated virgin female rats mated 1:1 with the control or treated male rats for 8 consecutive weeks.

Results: No significant difference in weight gain was found between the control and test animals. The group average for the female reproduction performance are as follows:
<table>
<thead>
<tr>
<th>Implant Carpora sites / Lutea</th>
<th>Live Fetuses / Implant Sites</th>
<th>Mutagenic Index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>95.2</td>
<td>97.0</td>
</tr>
<tr>
<td>500 ppm</td>
<td>96.0</td>
<td>97.6</td>
</tr>
<tr>
<td>2500 ppm</td>
<td>96.3</td>
<td>98.2</td>
</tr>
</tbody>
</table>

The reported results state that this test indicates no-effect for either of the test groups. However, the mutagenic index indicates there is some effect but not statistically significant at the 2500 ppm level.

- Acute Toxicity to Fish using Prowl and DDT

<table>
<thead>
<tr>
<th>Fish</th>
<th>LT$_{50}$</th>
<th>Prowl</th>
<th>DDT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bluegill</td>
<td>96 hr</td>
<td>0.199 mg/L</td>
<td>0.008 mg/L</td>
</tr>
<tr>
<td>Rainbow Trout</td>
<td>LT$_{50}$</td>
<td>96 hr</td>
<td>0.138 mg/L</td>
</tr>
<tr>
<td>Channel Catfish</td>
<td>LT$_{50}$</td>
<td>96 hr</td>
<td>0.418 mg/L</td>
</tr>
</tbody>
</table>

- Mallard Ducks - 8 day toxicity comparison with Dieldrin

<table>
<thead>
<tr>
<th>LC$_{50}$ with Prowl</th>
<th>10,388 ppm</th>
</tr>
</thead>
<tbody>
<tr>
<td>LC$_{50}$ with Dieldrin</td>
<td>158 ppm</td>
</tr>
</tbody>
</table>

- Acute studies of Prowl Metabolites

- o-Toluic Acid, 4[(1 ethylpropyl)amino]-3,5-dinitro
  - Mouse oral LD$_{50}$ 1440 mg/kg

- o-Toluic Acid, 4[(1 ethyl-2-hydroxypropyl)amino]-3,5-dinitro
  - Mouse oral LD$_{50}$ 1650 mg/kg

- o-Toluic Acid, 4[(ethyl-3-hydroxypropyl)amino]-3,5-dinitro
  - Mouse LD$_{50}$ 2330 mg/kg

- o-Toluic Acid, 4 [(1 carboxymethyl)propyl]amino]-3,5-dinitro
  - Mouse LD$_{50}$ > 5000 mg/kg

- Benzyl Alcohol, 4[(10ethylpropyl)amino]-2 methyl-3,5-dinitro
  - Mouse LD$_{50}$ 2140 mg/kg
Teratogenic study (Charles River Rats)

Three groups of 20 females were dosed with Prowl at levels 0, 500 and 1000 mg/kg/day from days 6 through 15 of gestation. The following results were obtained during the investigation.

Females in the 1000 mg/kg group exhibited a weight loss of 14 grams during the first 3 days of treatment. Three females of this group died after 2 doses, no other deaths or abnormal reactions were noted in either of the test groups. Only 12 of the 18 remaining females in the high group had signs of pregnancy when sacrificed on gestation day 20. This compared to 19 of 20 in the 500 mg/kg test group and to 16 of 20 in the control group.

The mean values of implantation sites, resorption sites, viable fetuses, and corpora lutea were essentially the same for test and control females. Fetal body weights of the 1000 mg/kg group were slightly reduced as compared to the 500 mg/kg and control groups. No significant differences were found in sex ratios of the pups.

One runt fetus with a hematoma was found in the 1000 mg/kg group, all others appeared normal. Fetuses from dams of the 1000 mg/kg group exhibited an increase in the percent of fetuses with incomplete or non-ossified sternum sections. The runt fetus exhibited a bifurcated sternum. Three fetuses among those obtained from both the 500 and 1000 mg/kg groups revealed angulated, thickened, fragmented ribs. It was reported that although this control group did not exhibit angulated ribs, previous control groups have contained fetuses with this type of skeletal anomaly. From this it was concluded that Prowl is not a teratogenic compound.

Conclusions:

The data submitted with this petition support the proposed negligible tolerance. TB recommends that this temporary tolerance be established as proposed in or on corn grain at 0.1 ppm.

Robert P. Schmidt, D.V.M.
Toxicology Branch
Registration Division

cc: CB
    EEB
    Division File
    Branch Reading File
    PP# 4G1451

R/D Init:GEWhitmore:1/28/74
RPSchmidt:sss:1/31/74
Init:GEWhitmore