CASWELL FILE

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

009486

MAY 7 1992

OFFICE OF
PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

SUBJECT: Review of a Chronic Toxicity and Carcinogenicity with Diuron in the Diet of Mice **[6(a)(2) data]**.

TO: Walter Waldrop/Carol Peterson PM-71
SRRD/Reregeristration (H7508W)

FROM: David S. Liem, Ph.D. David Liem 4/1/92
Section II, Toxicology Branch II/HED (H7509C)

THROUGH: K. Clark Swentzel, Section Head K. Clark Swentzel 4/2/92
Section II, Toxicology Branch II/HED (7509C)
and
Marcia van Gemert, Ph.D., Branch Chief
Toxicology Branch II/HED (H7509C) M. van Gemert 4/3/92

MRID No.: 421595-01
Caswell No.:410

DP Barcode No.: D173592
HED Project No.: 2-1176

ACTION REQUESTED

To review a study entitled "Diuron: Study for Chronic Toxicity and Carcinogenicity with NMRI Mice (Administration in Diet for 24 Months)".

CONCLUSIONS:

Technical diuron was administered in the diet of four groups of 60 mice of each sex/group at 0, 25, 250, and 2500 ppm for a period of 24 months.

Based on the data presented in the study report, the systemic LOEL is determined to be 2500 ppm based on the treatment-related effects observed including decreased body weight in the males, increased spleen and liver weight males, elevated leucocyte and reticulocyte counts in both sexes, increased incidence of hemosiderin deposits in liver cells in males, increased incidences of liver single cell necrosis and cell mitosis in both sexes, and others (see list on p. 16 and 17 of this DER). The NOEL is 250 ppm. This study is currently classified as core-supplementary, because numerous discrepancies and deficiencies were noted (see p. 20 and 21 of the attached DER).

The doses employed in this study were sufficient to produce a compound-related systemic effect and appear to be adequate to test the carcinogenic potential of the test material. Treatment-related increased incidences of mammary gland adenocarcinoma and ovarian luteoma were noted in the 2500 ppm females.

This study is considered to be a 6(a)2 study, because diuron affected the hematopoietic system and there is evidence of a carcinogenic effect.

The data of this study together with the other two rat chronic feeding/oncogenicity studies (MRID# 408865 and 00017764) will be presented to the HED Cancer Peer Review Committee.

RECOMMENDATIONS

The registrant is requested to address the items listed in the study deficiencies and discrepancies (p. 20 and 21 of this DER) and also to provide the historical control data of non-neoplastic and neoplastic lesions in NMRI mice conducted in the facility.

Upon satisfactory review of the requested information this study may be upgraded.

The testing facilities, Institute of Toxicology-Industrial Chemicals and the Institute of Toxicology-Pharmacology, both part of the Fachberreich Toxikologie, Bayer AG, Friedrich-Ebert-Strasse 217-333, D-5600 Wuppertal in Germany and the Histological Services LTD, Herefordshire, England, should be audited.

This study was conducted in accordance with OECD Guidelines for testing chemicals (Health effects, sec.4, no. 453, of 12th May, 1981) and under OECD GLP principles and standards. Since certain study record retention requirements under OECD may differ from the current FIFRA regulations, LDIAD/OCM/EPA or RD/OPP/EPA should determine whether or not this submission complies with 40 CFR 169.2(k) regulation.

CLASSIFICATION: Core-Supplementary. May be upgraded upon satisfactory review of the requested information.

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Primary Reviewer: **David S. Liem, Ph.D.** David Liem 4/1/92
Section II, Toxicology Branch II/HED
Secondary Reviewer: **K. Clark Swentzel, Section Head** K. Clark Swentzel 4/2/92
Section II, Toxicology Branch II/HED

DATA EVALUATION REPORT

6(a)2 DATA

Study Type: Chronic/Oncogenicity Study Guideline 83-5

Test Animal: NMRI Mice

EPA Identification No.s: MRID (Accession) No.: 421595-01
DP Barcode No.: D173592 Caswell No.:410
HED Project No.: 2-1176

Test Material: Diuron with a purity of 98.7%; batch No.23114080

Synonym: N'-(3,4-dichlorophenyl)-N,N-dimethyl urea

Dosages: 0, 25, 250, and 2500 ppm

Sponsor: Agricultural Products Department,
E.I. du Pont de Nemours & Co., Inc.

Study Number: Bayer AG T 4010922; Du Pont Report No. DIUR/TOX9

Study Period: October 1981 and October 1983

Testing Facilities: Institute of Toxicology-Industrial Chemicals (in-life study) and the Institute of Toxicology-Pharmacology (Clinical laboratory tests and pathology studies), Fachberreich Toxikologie, Bayer AG, Friedrich-Ebert-Strasse 217-333, Wuppertal, Germany.

Title of Report: Diuron: Study for Chronic Toxicity and Carcinogenicity with NMRI Mice (Administration in diet for 24 Months)

Author: Dr. R. Eiben

Report Issued: The study was completed in October 29, 1983; Study Director signature on the report dated May 24, 1990; Translation was completed in January 1991; Final Report submission date is December 19, 1991.

Conclusions:

Technical diuron was administered in the diet of four groups of 60 NMRI mice of each sex per group at 0, 25, 250, and 2500 ppm for 24 months.

Based on the data presented in the study report, the systemic LOEL is determined to be 2500 ppm based on the treatment-related effects observed including decreased body weight in the males, increased spleen and liver weight in males, elevated leucocyte and reticulocyte counts in both sexes, increased incidence of hemosiderin deposits in liver cells in males, increased incidences of liver single cell necrosis and cell mitosis in both sexes, and others (see list on p. 16 and 17 of this DER). The NOEL is 250 ppm.

The doses employed in this study were sufficient to produce a compound-related systemic effect and appear to be adequate to test the carcinogenic potential of the test material. Treatment-related increased incidences of mammary gland adenocarcinoma and ovarian luteoma were noted in the 2500 ppm females.

This study is considered to be a 6(a)2 study, because diuron affected the hematopoietic system and there is evidence of a carcinogenic effect.

The data of this study together with the other two rat chronic feeding/oncogenicity studies (MRID# 408865 and 00017764) will be presented to the HED Cancer Peer Review Committee.

Because numerous discrepancies and deficiencies were noted (see p. 20-21 of this DER), this study does not fully satisfy USEPA's Guideline 83-5 requirements for a chronic toxicity/oncogenicity study, and it is currently classified as core-supplementary data. If more information is provided and if the data satisfactorily address the questions posed in this DER, this study may be upgraded.

The registrant is requested to address the items listed in the study deficiencies and discrepancies (p. 20 and 21 of this DER) and to provide the historical control data of non-neoplastic and neoplastic lesions in NMRI mice conducted in the facility.

Since a large number of deficiencies and discrepancies were noted in the study report, it is recommended that the testing facilities, Institute of Toxicology-Industrial Chemicals (in-life study) and the Institute of Toxicology-Pharmacology (Clinical laboratory tests and pathology studies), both part of the Fachberreich Toxikologie, Bayer AG, Friedrich-Ebert-Strasse 217-333, D-5600 Wuppertal in Germany, and the Histological Services LTD, Herefordshire, England, be audited.

The current chronic/oncogenicity study in NMRI mice was conducted under OECD GLP principles and standards. Since certain study record retention requirements under OECD may differ from the current FIFRA regulations, LDIAD/OCM/EPA or RD/OPP/EPA should determine whether or not this submission complies with 40 CFR 169.2(k) regulation.

This study was conducted in accordance with OECD Guidelines for testing chemicals, section 4, health effects, no. 453, of 12th May, 1981.

Statements of No Confidentiality Claim (FIFRA sec. 10(d)(1)(A), (B), and (C), Good Laboratory Practice (40 CFR 160) and Flagging Statement (40 CFR 158.34) were provided with the study report.

CLASSIFICATION: Core-Supplementary. May be upgraded upon satisfactory review of the requested information.

Study Title: Diuron: Study for Chronic Toxicity and Carcinogenicity with NMRI Mice (Administration in the diet for 24 months)

MRID#: 421595-01; DPBarcode#: D173592; HED Project#: 0-1839

Study No.: Bayer AG T 4010647; Du Pont Report No. DIUR/Tox 19

Test Material: Diuron with a purity of 98.7%; batch No.232114080; N'-(3,4-dichlorophenyl)-N,N-dimethyl urea

1. OBJECTIVE

This study was designed to evaluate the chronic toxicity and the carcinogenic potential of diuron following a lifetime dietary administration of technical diuron to NMRI mice.

2. MATERIALS AND METHODS

The in-life and necropsy phases of this study were conducted at the Institute of Toxicology-Industrial Chemicals, and the Clinical Chemistry, Hematology, and histopathologic evaluations were conducted at the Institute of Toxicology-Pharmacology, both part of Fachbereich Toxikologie, Bayer AG, Friedrich-Ebert-Strasse 217-333, D-5600 Wuppertal in Germany.

Summary study materials and protocol are as follows:

Test Material: Physical Description: solid, whitish test article
Source: Bayer AG
Storage: Cold Storage at 4°C.

Test Animals: Species: SPF-bred mice/Bor strain NMRI (SPF HAN)
Source: Winkelmann, Borchen (address not given)
Total Number: 240 males and 240 females
Age: About 7 weeks old at start of study
Starting Weight: Males = 27 g (range 19-33 g);
Females =23 g (range 19-27 g)
Caging: In individual Makrolon cages with dust-free wood chips, changed weekly
Acclimation period: About 7 days
Feed: Altromin 1321 basal diet from Altromin GmbH, Lage & water were provided ad libitum.

Environmental Parameter: Air temperature = 22 ± 2°C; Relative Humidity = about 50%; 12 hours dark/light cycle (6:00 AM to 6:00 PM light); animal room was cleaned with disinfectant solutions, Gevisol, Rapidosept or Zephirol.

Study Duration

The study lasted 24 months (October 1981 to October 1983) of compound administration. Ten animals/sex from each group were necropsied after 12 months of compound administration. All surviving rats were terminated during month 24.

Dose Selection

On p. 17 of the study report it was noted that the doses were selected based on the results of a 4-week preliminary study using 20 males each dosed at 0, 1000, 2500 and 5000 ppm diuron in the diet. It was stated that the NOEL was 1000 ppm and that the only treatment-related effect was the enlargement of the spleen at the 2500 ppm dose level. This reviewer can not verify this results because this 4-week preliminary study was not submitted to this Agency.

Group Arrangement

At the start of the study mice were randomly assigned to the study as follows:

Test Group	Dosage (ppm)	Main Males/Females	Satellite@ Males/Females
Control	0	50/50	10/10
Low Dose	25	50/50	10/10
Mid Dose	250	50/50	10/10
High Dose	2500	50/50	10/10

@ = All rats from satellite groups were sacrificed after 12 months on study

Diet Preparation

The diets containing diuron were prepared weekly. Concentration of the test material was not adjusted during the course of the study.

Diet Analyses

Prior to the study initiation, the homogeneity and stability of the compound in the diet were determined. The test material content in the diet was analyzed at the initiation of the study, and approximately every three months thereafter, throughout the study.

Clinical Observations

The rats were checked at least twice daily (once on weekends and on public holidays) for mortality, moribundity and signs of toxicity. Detailed physical examinations were conducted weekly.

Body Weights

Individual body weights were taken at the start of the study, prior to scheduled sacrifices, weekly through week 27, then every two weeks through week 37, and weekly thereafter to termination.

Food Consumption

Group food consumption was recorded weekly.

Compound Intake

The test compound intakes were calculated from the food consumption values, i.e. mean weekly food intake per animal per day and per kg body weight per day.

Clinical Pathology Evaluation

Blood samples were collected from the tail vein for glucose determination and from the orbital sinus (under anesthesia) of mice for other determinations. Bleeding was conducted at 6, 12, 18, and 24 months.

a. Hematology

Hematology tests were conducted at 6, 12, 18, and 24 months. Parameters evaluated were hemoglobin concentration, mean cell volume (MCV), erythrocyte and leucocyte counts, hematocrit: mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC), reticulocyte count, erythrocyte morphology and the differential blood count from blood smears. On p. 25 of the study report it was not mentioned that thrombocytes will be evaluated.

b. Clinical Chemistry

Clinical chemistry tests were conducted at 6, 12, 18 and 24 months. At each interval, blood serum from the same mice selected for hematologic study was used. Parameters evaluated were as follows: alanine aminotransferase, aspartate aminotransferase, glucose, alkaline phosphatase, total protein, creatinine, blood urea nitrogen (BUN), bilirubin and cholesterol. The following parameters were not determined: albumin/globulin (A/G) ratio, albumin, globulin, inorganic phosphate, calcium, potassium, sodium, chloride, and creatine phosphokinase. Lactic acid dehydrogenase was only determined at the end of the study.

c. Urinalysis

Urinalysis testing was not conducted in this study.

Ophthalmologic Examination

Mice on study were not subjected to ophthalmologic examination

Gross Macroscopic Examinations

All mice which died or were sacrificed in extremis and all mice sacrificed at the scheduled necropsies were subjected to gross macroscopic examination. At twelve months, an interim necropsy of 10 animals of each sex in each group was conducted. All survivors were sacrificed at 24 months. Mice sacrificed at scheduled necropsy were anesthetized with diethylether, killed by exsanguination, and then necropsied. All moribund mice were also necropsied after they were killed by the same procedure described above. Tissues harvested from all mice were fixed in 10% buffered formalin.

USEPA's guideline's required tissues (✓) for a combined chronic and carcinogenicity study:

✓ eyes	✓ liver
✓ brain	✓ spleen
✓ pituitary gland	✓ pancreas
✓ thyroid/parathyroid	✓ kidneys
✓ salivary gland	✓ adrenal glands
✓ thymus	✓ urinary bladder
✓ mandibular lymph node	✓ testes
✓ larynx	✓ epididymides
✓ trachea	✓ prostate gland
✓ lung	✓ seminal vesicles
✓ esophagus	✓ ovaries
✓ stomach	✓ uterus
✓ duodenum	✓ oviduct
✓ jejunum	✓ vertebral column with spinal cord
✓ ileum	✓ femur with skeletal musculature
✓ colon	and sciatic nerve
✓ caecum	✓ gall bladder
✓ rectum	✓ Heart
✓ skin and mammary gland	✓ Harderian Gland
✓ mandibular lymph nodes	✓ sternum
✓ mesenteric lymph node	✓ all gross lesions

All tissues listed above except the oviduct and mesenteric lymph node were routinely harvested and were subjected to routine histopathological evaluations.

Organ Weights

The heart, lung, liver, spleen, kidneys, adrenals, and testes from all animals killed at the interim (12 months) and terminal necropsies were weighed. The brain and ovaries as required for a chronic toxicity/carcinogenicity study were not weighed.

Morphometric Measurement of the Uteri

Corpus uteri which were at least 3 mm in diameter or uteri horns which were at least 2 mm in diameter were harvested and recorded at terminal sacrifice.

Densitometric Measurements of the Spleens

Hemosiderin content determination of the male mice spleen was conducted by staining the tissues with Turnbull's Blue and then densitometrically measured using the IBAS-II system. Evaluations were carried out by ZF-TPE 6, Bayer AG, Uerdingen.

Histopathological Evaluation

The fixed tissues were trimmed, embedded in paraplast, sectioned, and stained with hematoxylin-eosin. In addition, certain sections (specific tissue not mentioned) were stained with VAN GIESON and MASSON, Turnbull's Blue (to demonstrate iron deposits), Congo red and the PAS reaction.

On p. 29 of the study report, it was noted that histological sections of the tissues harvested at terminal sacrifice were processed at the Histological Services LTD, Herefordshire, England. Histological sections of tissues harvested at the 12 months interim sacrifice and other tissues which showed gross lesions of the 25 ppm and 250 ppm dose groups were processed and evaluated histopathologically at the Institute of Toxicology-Pharmacology, Fachbereich Toxicologie, Bayer AG, Friedrich-Ebert-Strasse 217-333, D-5600 Wuppertal in Germany.

Statistical analysis

The arithmetic group means, standard deviations, upper and lower confidence limits ($P < 0.05$ and $P < 0.01$) were calculated. The combined values were tested and compared with combined control using the Mann and Whitney U test and Wilcoxon's method at $P < 0.05$ and $P < 0.01$. Mortality data were evaluated at a significant level at 1% and 5% using Fischer's exact test.

Compliance Statements

- o A signed Statement of Data Confidentiality Claims was provided
- o A signed GLP Compliance Statement was provided
- o A signed Flagging Statement was provided

RESULTS

a. Analyses of Test Article and Diet

Analysis of diet samples collected on the day of diet preparation indicated that the diets containing diuron for all dose levels at various intervals were prepared at or near the intended concentrations, i.e. between 98% and 101% of nominal. Diuron in the low (25 ppm) and high (2500 ppm) dose diet mixtures were sampled from the animal food containers kept at room temperature were stable for 17 days and the recovery rates were between 98% to 108%. The homogeneity of test article in the diet analyzed at the start of study was 85% and approximately 100% of theoreticals for the 25 ppm and 2500 ppm diets, respectively.

b. Mortality

The cumulative mortality data of mice that died spontaneously or were sacrificed in extremis during the study and the number of mice sacrificed at terminal necropsy are as follows:

	0 ppm		25 ppm		250 ppm		2500 ppm	
	M	F	M	F	M	F	M	F
#/Start of Study	50	50	50	50	50	50	50	50
Week 26/survival % Mortality	49 2%	48 4%	50 0%	49 2%	50 0%	49 2%	48 4%	48 4%
Week 52/survival % Mortality	48 4%	40 20%	46 8%	43 14%	44 12%	49 2%	45 10%	47 6%
Week 78/survival % Mortality	42 16%	31 38%	37 26%	32 36%	41 16%	35 30%	36 28%	33 34%
Week 102/survival % Mortality	18 64%	11 78%	23 54%	11 78%	25 50%	11 78%	21 58%	15 70%

The above table show that the percent cumulative mortality in the high-dose group was increased in males as compared to the control on week 78 of study, but no trend was evident. The mortality data did not show any treatment-related effects.

c. Clinical Signs Observations

No treatment-related clinical signs were evident (p. 34 and p. 382-419 of the study report).

d. Body weight data

The mean body weights are presented in Appendix A. Consistent statistically significant reductions in body weights were noted

in the high-dose males (starting from week 26) as compared to the control. Body weight reductions were observed in the high-dose females starting from week 39 but statistically significant values were sporadic. The percent mean absolute body weight difference between the high-dose groups as compared to the controls was between - 6.7% to -20.7% for the males, and -3.7% to -7.5% for the females. The body weight gain mean percent differences between the control and the high-dose mice, and the mean percent body weight gains at different intervals were calculated by this reviewer as follows:

Intervals Weeks	Control Male/Female	High-dose Male/Female	Mean % Difference Male/Female
0 - 2	16.1/7.4	11.0/7.3	-5.1/-0.1
0 - 4	26.4/13.9	17.5/13.4	-8.9/-0.5
0 - 6	32.6/19.7	27.1/16.4	-5.5/-3.3
0 - 10	38.8/31.0	29.6/24.7	-9.2/-6.3
0 - 14	45.8/30.6	38.9/33.3	-6.9/+2.7
0 - 18	49.3/36.3	38.9/34.9	-10.4/-1.4
0 - 26	62.3/48.5	47.9/42.8	-14.4/-5.7
0 - 39	71.1/55.0	49.7/44.6	-21.2/-10.4
0 - 52	70.3/59.5	55.4/51.9	-19.9/-7.6
0 - 78	70.3/67.2	53.9/59.4	-16.4/-7.8
0 - 102	54.9/64.1	49.6/55.8	- 5.3/-8.3

As seen from the above Table, starting from week 18 the mean percent body weight gain differences between the control and the high-dose males were greater than 10%. Therefore, the body weight reductions of the high-dose males are judged to be related to treatment.

e. Food Consumption Data

Summary food intake per group and per animal/day are presented in Appendix B. The high-dose groups consumed slightly more feed as compared to the control (17% more for ♂ and 12% more for ♀). The food intake values for other groups are comparable. The investigators did not calculate the mean food intake efficiency nor were they discussed in the study report.

f. Compound Intake

Summary calculated compound intakes over the course of the

study are presented in Appendix C. It shows that females ingested more diuron (7.5, 77.5 and 869.0 mg/kg BW/day) as compared to the males (5.4, 50.8, and 640.1 mg/kg BW/day).

g. Clinical Pathology

The results of hematological and clinical chemistry data at 6, 12, 18, 24 months are presented in Appendices D1, D2 and E.

1. Hematology (Appendices D1 and D2)

The hematology measurements were conducted at 6, 12, 18, and 24 months. As seen from Appendix D1, leucocyte counts were elevated in the high dose groups in both sexes as compared to the control at all intervals. These elevated values were not statistically significant in either sex at the 12-month or in males at the 24-month interval.

The mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH except the 24 month interval) were elevated in the high-dose rats in both sexes as compared to the controls. MCV values in all treated males at 6 months and the high-dose females at 12-, 18-, 24-month intervals were statistically significant. MCH in the high-dose males at 6- and 12- month, and females at 12- (low-dose), 18- (low- and high-dose), and 24- (high-dose) month intervals, were also statistically significant as compared to the controls.

All reticulocyte counts (RETI) in the high-dose groups were elevated and all values (except in females at 24-month interval) were statistically significant. The reticulocyte count at the 6- month interval was not provided and no explanation was given. The differential blood count data did not show any evidence of treatment related effects.

Elevations of the leucocyte counts, mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH), and reticulocyte counts in the high-dose male and female mice are judged to be related to treatment. All other scattered instances of statistically significant differences of hematological data, between the treated groups as compared to the control were judged to be artifactual and unrelated to treatment.

2. Clinical Chemistry (Appendix E)

Clinical chemistry parameters were conducted at 6, 12, 18 and 24 months. Alanine aminotransferase was elevated in the high-dose group males at all intervals tested, but in the high-dose females, elevation of alanine aminotransferase was noted at 6- and 24-month intervals, as compared to the control. These elevations may be related to treatment.

Bilirubin was elevated in the high-dose males at all intervals tested, but in the high-dose female elevation of bilirubin was noted after the 12-month interval, as compared to the control. Elevation of bilirubin in both sexes is judged to be related to treatment at the high-dose. Other scattered statistically significant differences between the control and the treated groups are judged to be unrelated to treatment.

Gross Macroscopic Findings

All mice which died or were sacrificed in extremis or at scheduled necropsies were subjected to gross macroscopic examinations. On p. 48 of the study report, the investigators noted that "organ changes (in the 12 month period) observed were classified as spontaneous mutations characteristic of mice of this age". This statement could not be verified, because findings during the first year and the number of mice that died or were sacrificed in extremis prior to the 12-month interim sacrifice were not tabulated separately. Macroscopic findings were listed in the individual Table on p.771-1532 of the study report.

The investigators provided summarized gross macroscopic data for mice scheduled for 24 month treatment on p. 422-429 of the study report. It showed totals of 45, 48, 47, and 46 males and 46, 37, 48, and 45 females that were examined. These totals excluded interim sacrificed as well as autolyzed mice, but included moribund-sacrificed and found-dead mice during the first 12 months of treatment. Based on the data presented on this Table, there was an increased incidence of clear lobule in the liver, lung discoloration in all treated males, and liver discoloration in all treated females as compared to the controls. Comparisons of macroscopic changes that occurred in the first year as compared to those that occured in the second year could not be made, because macroscopic findings of moribund-sacrificed and found-dead mice during the first 12 months of treatment as well as those of interim sacrificed mice were not tabulated in separate Tables.

Organ Weights

The heart, lung, liver, spleen, kidneys, adrenal glands, and testes from all animals sacrificed on scheduled necropsies were weighed. The brain and ovaries were not weighed. The data are summarized in Appendices F and G.

a. 12 Months Interim Sacrifice (Appendix F)

The mean absolute and relative (to body weights) spleen and liver weights in the male high-dose group were significantly increased as compared to the control. Only the absolute liver weight of the high-dose males was not statistically significant. These increases are judged to be related to treatment. Other

statistically significant increases in organ weights are not related to treatment.

b. 24 Months Terminal Sacrifice (Appendix G)

Both the absolute and relative spleem and liver weights in the high-dose males were significantly increased as compared to the control, but only the liver weight increase was statistically significant. Increased spleen and liver weights in the high-dose males are judged to be related to treatment. Other statistically significant increases or decreases of other absolute and relative organ weights are not judged to be related to treatment.

Histopathological Evaluation

a. Non-neoplastic Lesions in the 12-month Sacrificed Mice

Pertinent non-neoplastic lesions for 12 month sacrificed mice are summarized as follows:

	Control	Low-dose	Mid-dose	High-Dose
	M/F	M/F	M/F	M/F
No. Mice Examined	10/10	10/10	10/10	10/10
Liver +Fatty infiltration	6/10	8/8	9/9	9/10
Kidney +Round-cell infiltrates	3/10	10/9	8/9	4/9
+Nephropathy	6/4	5/5	6/3	9/3
Spleen +Iron deposit	1/0	ne/ne	ne/ne	5/6
Urinary Bladder +Mucosal Hyperplasia	0/0	ne/ne	ne/ne	0/5
Thyroid +Follicular Cysts	1/1	ne/ne	ne/ne	2/1
Uterus +Cysts	na/1	na/ne	na/1[a]	na/4

M = Male; F = Female; ne = Not examined; - = Not applicable;
@ = Three mice were evaluated; from p. 484-500 of study report.

The above table shows that increased incidences of mucosal hyperplasia in the urinary bladder and uterine cysts were noted in the high-dose females and iron deposit in the spleen was present in high-dose males and females. Kidney nephropathy was slightly increased in the high-dose males. These findings appeared to be related to treatment.

b. Non-neoplastic Lesions in the 24-month Sacrificed Mice

Pertinent non-neoplastic lesions in the 24-month sacrificed mice are as follows:

Tissues	Control	Low-dose	Middose	High-Dose
	M/F	M/F	M/F	M/F
Liver				
No of Mice Evaluated	45/46	48/38	46/48	46/46
+Enlarged degenerative	0/0	0/1	0/3	0/10**
+Hepatopathy	1/0	0/0	0/0	15**/0
+Increased mitosis	1/0	2/3	0/0	8**/4*
+Single cell necrosis	3/12	2/7	5/10	7*/19**
+Kupffer cells	6/9	6/9	8/9	11*/9
No of Mice Evaluated	35/ne	ne/ne	46/ne	45/ne
+Hemosiderin[a] in:				
-Phagocytes				
-Grade 0-1	97%/ne	ne/ne	93%/ne	82%/ne
-Grade 2-3	3%/ne	ne/ne	7%/ne	18%/ne
-Hepatocytes				
-Grade 0-1	86%/ne	ne/ne	83%/ne	73%/ne
-Grade 2-3	14%/ne	ne/ne	17%/ne	27%/ne
Kidney				
No. of Mice Examined	45/46	48/38	47/48	46/46
+Goldenbrown pigment[a]	0/0	0/1	0/1	0/5*
No. of Mice Examined	36/45	48/ne	47/48	45/46
+Percent Hemosiderin in tubular epithelia[a]	12/33	21/ne	33/32	26/34
-Grade 0-1	89%/60%	88%/ne	83%/56%	84%/46%
-Grade 2-4	11%/40%	12%/ne	17%/44%	16%/54%
Spleen				
No. of Mice Examined	45/46	48/38	46/48	46/46
+Goldbrown pigments[a]	1/6	0/2	1/6	14**/19**
No. of Mice Examined	37/45	ne/ne	47/46	45/41
+Hemosiderin deposit@	70%/87%	ne/ne	81%/83%	84%/90%
Urinary Bladder				
No. of Mice Examined	44/46	47/36	47/45	46/44
+Epithelial Hyperplasia	14/5	12/5	13/3	12/23**
+Edema	0/0	0/0	0/0	0/17**
+Thickened Mucosa	0/0	0/0	0/0	0/5**

a = Hemosiderin deposit; M/F = Male/Female; ne = Not examined; - = Not applicable; @ =Relative density indicated by Turnbullblue staining; Derived from p. 55 and 487 of study report.

The table on the previous page shows that increased single cell necrosis and cell mitosis of the liver were noted in the high-dose of both sexes. Significant increase of enlarged degenerative liver cells in the high-dose females, hepatopathy, Kupffer cells, and hemosiderin deposits in liver cells (18% versus 3% in phagocytes and 27% versus 14% in hepatocytes) were noted in the high-dose males as compared to the control, and they are considered to be related to treatment. These findings together with the elevation of the alanine aminotransferase activity indicate signs of liver toxicity.

A statistically significant increase of intracellular golden brown pigments (hemosiderin) in cortical renal tubules was noted in the high-dose females as compared to the control. Special staining with Turnbullblue only revealed a slight increase of hemosiderin deposit in the high-dose females.

A statistically significant increase of golden brown pigment (hemosiderin) in the H&E sectioned spleen was noted in the high-dose males and females as compared to the control. Special staining with Turnbullblue, however, only revealed a slight increase of hemosiderin deposits in the high-dose groups as compared to the control. Densitometric measurement of Turnbull-blue stained spleen tissues confirmed the significant increase of hemosiderin in the high-dose males. Increased hemosiderin in the high-dose group is judged to be related to treatment. The systemic presence of hemosiderin deposits in the liver, spleen and kidneys together with a significant increase of mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), reticulocyte and a depressed erythrocyte count clearly indicate destruction of red blood cells.

Statistically significant increase of epithelial hyperplasia, edema, and thickened mucosa of the urinary bladder was noted in the high-dose females as compared to the control and this is judged to be related to treatment.

Since the uterine horn appeared to be enlarged, its diameter was measured. The results (p. 66 of the study report) show that increased incidence of enlarged (with > 2 mm diameter) uterine horn was noted in the high-dose group (in 24 mice) versus the control (in 16 mice), and this appears to be related to treatment.

Approximately 50% of the females were affected by fibrosis of the bone marrow, generally mild to medium-grade in severity, but no dose-related trend was evident.

The oviduct and mesenteric lymph node were not routinely harvested, hence were not subjected to routine histopathological evaluations as required for a chronic toxicity/carcinogenicity.

c. Neoplastic Lesions for the 12 Month Interim Sacrificed Mice

Neoplastic incidences for the 12 month sacrificed mice are summarized in Appendix H. As seen from this Appendix, there is no evidence of dose-related effects in any tissue evaluated during the 12 months of study.

d. Neoplastic Lesions for the 24 Month Interim Sacrificed Mice

Pertinent neoplastic incidences for found dead and unscheduled sacrificed, and 24-month sacrificed mice are summarized in Appendix I. Increased incidences of mammary gland adenocarcinoma and ovarian luteoma were noted in the high-dose females and showed a statistically significant trend using Peto's trend test analysis. These findings are judged to be related to treatment.

Incidences of liver adenoma were increased in all treated (25, 250, and 2500 ppm) males but none was statistically significant, also, a dose related trend was not evident. This finding is judged to be unrelated to treatment.

Uterine stromal sarcoma was noted in two mid- and in two high-dose females, but the toxicological significance is unknown. Other observed tumor incidences appear to be of no toxicological importance. The historical control data for the noted neoplastic lesions in NMRI strain mouse were not provided.

SUMMARY AND DISCUSSIONS

Based on data submitted in the study report, administration of technical diuron at 25, 250, and 2500 ppm in the diet of NMRI mice for 24 months resulted in the following treatment related effects:

- o Body weight decrease in the 2500 ppm males.
- o Increased spleen and liver weight in the 2500 ppm males.
- o Elevated leucocyte and reticulocyte counts in the 2500 ppm mice of both sexes.
- o Elevated mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) in the 2500 ppm mice of both sexes.
- o An elevated bilirubin in the 2500 ppm mice of both sexes.
- o Increased incidences of intracellular golden brown pigments in the cortical renal tubules in the 2500 ppm females, and in the spleen of the 2500 ppm males and females
- o An increased incidence of hemosiderin deposits in liver cells of the 2500 ppm males.

- o Increased incidences of liver single cell necrosis and cell mitosis in the 2500 ppm mice of both sexes.
- o An increased incidence of enlarged degenerative liver cells in the 2500 ppm females,
- o Increased incidences of hepatopathy and Kupffer cells in the 2500 ppm males.
- o Increase incidences of urinary bladder edema, epithelial hyperplasia, and thickened mucosa in the 2500 ppm females.
- o An increased incidence of enlarged (with > 2 mm diameter) uterine horn in the 2500 ppm females.
- o Increased incidences of ovarian luteoma and mammary gland adenocarcinoma in the 2500 ppm females.

In another chronic toxicity/oncogenicity study in Wistar rats (MRID# 408865-01) fed with Diuron with a purity of 98.7% at dose levels of 25, 250, and 2500 ppm conducted at the Institute of Toxicology, Bayer AG, Wuppertal, Friedrich-Ebert-Strasse 217-333, West Germany and Institute of Experimental Pathology, Hanover Medical University, West Germany (reviewed by this Agency) showed similar findings such as increased weight and hemosiderin deposit in the spleen, increased swelling of the liver and increased hemosiderin deposits in the liver, increased urinary bladder wall thickening in the males, and a significant increase in erythrogenic activities as indicated by an increase of hematopoietic and a decrease of fat marrow surface areas of the bone marrow. The increase of hematopoietic marrow and reticulocytes, and the decrease of fat marrow suggest an increase in erythrogenic activity of the bone marrow. It also showed increased incidences of urinary bladder wall thickening in the 250 ppm and 2500 ppm rats, and urinary bladder and renal pelvis epithelial papillomas and carcinomas in the 2500 ppm rats were observed. Therefore, diuron appears to be carcinogenic in this study based on increased incidences of urinary bladder and renal pelvis epithelial papillomas and carcinomas in the 2500 ppm rats. This study was classified as core-supplementary because the systemic NOEL could not be determined and because numerous discrepancies were noted (see DER dated December 3, 1990).

In yet another chronic feeding/carcinogenic study in rats (strain not specified) fed with diuron conducted by Hodge in 1963 (reviewed EPA, DER dated 2/7/89), slight anemia, enlarged spleens, increased erythrogenic activity in the bone marrow and abnormal pigments in the blood were seen. The systemic NOEL was determined to be 25 ppm and the systemic LOEL was 125 ppm. There was no evidence of carcinogenicity up to the 2500 ppm dose level tested.

CONCLUSIONS

Based on the data presented in the study report, the systemic LOEL is determined to be 2500 ppm based on the treatment-related effects observed including decreased body weight in the males, increased spleen and liver weight in males, elevated leucocyte and reticulocyte counts in both sexes, increased incidence of hemosiderin deposits in liver cells in males, increased incidences of liver single cell necrosis and cell mitosis in both sexes, and others (see list on p. 16 and 17 of this DER). The NOEL is 250 ppm.

The doses employed in this study were sufficient to produce a compound-related systemic effect and appear to be adequate to test the carcinogenic potential of the test material. Treatment-related increased incidences of mammary gland adenocarcinoma and ovarian luteoma were noted in the 2500 ppm females.

This study is considered to be a 6(a)2 study, because diuron affected the hematopoetic system and there is evidence of a carcinogenic effect.

The data of this study together with the other two rat chronic feeding/oncogenicity studies (MRID# 408865 and 00017764) will be presented to the HED Cancer Peer Review Committee.

Because numerous discrepancies and deficiencies were noted (see p. 20-21 of this DER), this study does not fully satisfy USEPA's Guideline 83-5 requirements for a chronic toxicity/oncogenicity study, and it is currently classified as core-supplementary data. If more information is provided and if the data satisfactorily address the questions posed in this DER, this study may be upgraded.

The registrant is requested to address the items listed in the study deficiencies and discrepancies (p. 20 and 21 of this DER) and to provide the historical control data of non-neoplastic and neoplastic lesions in NMRI mice conducted in the facility.

Since a large number of deficiencies and discrepancies were noted in the study report, it is recommended that the testing facilities, Institute of Toxicology-Industrial Chemicals (in-life study) and the Institute of Toxicology-Pharmacology (Clinical laboratory tests and pathology studies), both part of the Fachberreich Toxikologie, Bayer AG, Friedrich-Ebert-Strasse 217-333, D-5600 Wuppertal in Germany, and the Histological Services LTD, Herefordshire, England, be audited.

The current chronic/oncogenicity study was conducted under OECD GLP principles and standards. Since study record retention requirements under OECD may differ from the current FIFRA regulations, LDIAD/OCM/EPA or RD/OPP/EPA should determine whether or not this submission complies with 40 CFR 169.2(k).

This study was conducted in accordance with OECD Guidelines for testing chemicals, section 4, health effects, no. 453, of 12th May, 1981.

Statements of No Confidentiality Claim (FIFRA sec. 10(d)(1)(A), (B), and (C), Good Laboratory Practice (40 CFR 160) and Flagging Statement (40 CFR 158.34) were provided with the study report.

CLASSIFICATION: Core-Supplementary. May be upgraded upon satisfactory review of the requested information.

STUDY DEFICIENCIES:

Substantive Deficiencies

- Body weight gains were not presented in the study report.
- The mean food intake efficiency data (body weight gain divided by food consumed multiplied by 100) were not calculated nor were they discussed in the report.
- The oviduct and the mesenteric lymph node were not harvested and were not subjected to histopathological evaluations.
- No explanation was given whether the summary gross macroscopic data presented on p. 422-429 of the study report were findings of all mice on study throughout the 24 month period or findings of mice after the 12 month study period Separate summary tables are required for interim sacrificed mice and for those that were found dead or sacrificed in extremis within the first 12 months of study.
- Historical control data for neoplastic lesions in the strain of mouse were not provided with the study report.
- The brain and ovaries were not weighed.
- The reticulocyte count at the 6 month interval was not provided in the summary Table on p. 44 of the study report, and no explanation was given.
- The following clinical chemistry parameters were not determined: albumin, inorganic phosphate, calcium, sodium, potassium, chloride, and creatine phosphokinase.
- Urinalysis was not determined.

Minor Deficiencies

- The study report is an English translation of the original German language version. This translation is not well written and is difficult to read. Some technical terms used in this study report are not quite accurate or are not commonly used in a toxicological report. Some terms did not make sense such as "tolerated harmlessly", "manufactured", "nodose", "nodose substance", "nodose surface", and "bosselated" on p. 11, 29, 423, 424, 425, and 1110 of the study report.
- No summary nor individual clinical sign observation data were presented in the study report.
- Typo on p. 33 of the study report: Table 1 (page 33) should read Table 1 (page 34).

- A summary table for the 12 month gross macroscopical data was not provided.

- The numbers of animals in the historical controls on hematology and clinical chemistry data (p. 81-82) were not provided. Only the historical control values after 1981 (the year when this study was conducted) were provided.

GLP-RELATED DISCREPANCIES AND DEFICIENCIES

- The Study Director and the Pathologists signed the report in November 1990 (7 years after the completion the study and 3 months before the translation was completed in January 1991) - see p.9 of the study report.

- It was stated that the study was completed in October 1983 (reviewer's comment: the in-life study only). The record showed that the QA unit inspected the study between October 30, 1981 and October 10, 1983 only, and it is assumed that the pathology evaluations and data were not QAed.

APPENDIX A: Summary Mean Body Weights at Selected Intervals (derived from p. 128-141 of the study report)

APPENDIX B: The Total (g/group) and Mean (g/Animal/day) Food Intake, and the Total (g/kg BW) and the Mean (g/Animal/day per kg Body Weight) food intake per Group (Copied from p. 41 of the study report)

APPENDIX C: The Total (g/group) and the Mean (g/Animal/day) Test Article Intake, and the Total (g/kg BW) and the Mean (g/Animal/kg BW/day) Test Article Intake per Group (Copied from p. 41 of the study report)

APPENDIX D1: Hematology Data for 6, 12, 18, and 24 Month Intervals (Copied from p. 44 of the study report)

APPENDIX D2: Differential Blood Count Data for 6, 12, 18, and 24 Month Intervals (Copied from p. 45 of the study report)

APPENDIX E: Clinical Chemistry Data for 6, 12, 18, and 24 Months Intervals (Copied from p. 47 of the study report)

APPENDIX F: Summary the Absolute and Relative Organ Weights for the 12-month Scheduled Sacrificed Mice (Copied from p. 50 of the study report).

APPENDIX G: Summary of Absolute and Relative Organ Weights for the 24-month Scheduled Sacrificed Mice (Copied from p. 51 of the study report).

APPENDIX H: Neoplastic Incidence for the 12 Month Sacrificed Mice (Copied from p. 70 of the study report).

APPENDIX I: Neoplastic Incidence for the 24 Month Sacrificed Mice (Derived from p. 72 of the study report).

APPENDIX A
SUMMARY MEAN BODY WEIGHTS (G) AT SELECTED INTERVALS@

Week	Control 0 ppm M/F	Low-Dose 25 ppm M/F	Mid-Dose 250 ppm M/F	High-Dose 2500 ppm M/F
0	27.3/22.9	27.8/23.0	28.1/23.1	28.0/23.1
1	29.6/24.0	29.6/23.3**	29.0/23.8	28.6**/22.8**
2	31.7/24.6	32.2/24.5	31.6/24.9	31.2/24.8
4	34.5/26.1	34.3/26.2	34.9/26.1	32.9/26.2
6	36.2/26.8	36.0/27.9**	36.4/26.9	35.6/27.6
10	37.9/29.0	37.6/29.0	38.3/28.4	37.3/28.8
14	39.8/29.9	39.8/30.9	40.2/29.8	38.9/30.8*
18	40.7/30.9	40.0/31.0	41.2/30.4	38.9*/31.5
22	41.4/31.8	41.2/32.3	42.5/31.3	40.3/31.9
26	44.3/33.0	44.2/32.9	44.2/31.9	41.4**/33.0
31	44.9/34.0	44.3/34.2	44.2/32.6*	41.3**/33.2
35	46.6/35.0	45.1/35.5	45.9/34.3	43.3**/34.0
39	46.9/35.5	46.3/35.3	46.6/33.3**	41.9**/33.4**
43	46.8/35.9	46.7/36.3	47.2/35.2	44.1**/33.9**
47	46.5/36.3	45.6/36.1	45.7/34.8	44.1**/34.9
52	46.5/36.3	46.1/36.5	46.7/35.7	43.5**/35.1
58	46.5/37.1	45.9/36.7	47.4/36.5	43.6**/34.4*
64	46.5/36.3	45.9/36.5	47.1/36.3	44.2*/35.4
70	46.5/35.9	45.7/38.1	47.0/36.2	44.0*/35.5
78	46.5/38.3	45.3/38.2	45.8/38.5	43.1**/36.6
84	47.1/39.2	46.0/39.0	46.6/39.4	44.3**/37.4
90	44.1/37.8	44.8/37.8	45.4/38.7	41.9*/35.7
96	44.9/39.1	44.3/39.2	45.3/38.6	42.5*/37.1
102	42.3/37.6	42.1/39.8	44.0/38.9	41.9/36.0

@ = Derived from p.128-141 of the study report: * = significant at $p < 0.05$; ** = significant at $p < 0.01$.

APPENDIX B

The Total (g/group) and Mean (g/Animal/day) Food Intake, and the Total (g/kg BW) and the Mean (g/Animal/day per kg Body Weight) food intake per Group (copied from p. 41 of the study report)

Futteraufnahme / food intake

gruppenweise Bestimmung / determination per group

Dosis dose	Sex Appl	Tage days	g /Tier g /animal gesamt total	g /Tier g /animal pro Tag per day	g/kg Koerpergewicht g/kg body weight gesamt total	g/kg Koerpergewicht g/kg body weight pro Tag per day
0 PPM	m/m PO	723	6593	9.1	151118	209.0
25 PPM	m/m PO	723	6798	9.4	157448	217.8
250 PPM	m/m PO	723	6444	8.9	146901	203.2
2500 PPM	m/m PO	723	7717	10.7	185127	256.1
0 PPM	w/f PO	723	7547	10.4	216468	299.4
25 PPM	w/f PO	723	7599	10.5	215750	298.4
250 PPM	w/f PO	723	7718	10.7	224022	309.9
2500 PPM	w/f PO	723	8466	11.7	250743	346.8

6.

APPENDIX C

The Total (g/group) and the Mean (g/Animal/day) Test Article Intake, and the Total (g/kg BW) and the Mean (g/Animal/kg BW/day) Test Article Intake per Group (copied from p. 41 of study report)

Substanzaufnahme / test compound intake

gruppenweise Bestimmung / determination per group

Dosis dose	Sex Appl sex adm	Tage days	mg /Tier mg /animal gesamt total	mg /Tier mg /animal pro Tag per day	mg /kg Koerpergewicht mg /kg body weight gesamt total	mg /kg Koerpergewicht mg /kg body weight pro Tag per day
0 PPM	m/m PO	723	0	0.0	0	0.0
25 PPM	m/m PO	723	170	0.2	3936	5.4
250 PPM	m/m PO	723	1611	2.2	36725	50.8
2500 PPM	m/m PO	723	19293	26.7	462818	640.1
0 PPM	w/f PO	723	0	0.0	0	0.0
25 PPM	w/f PO	723	190	0.3	5396	7.5
250 PPM	w/f PO	723	1929	2.7	56005	77.5
2500 PPM	w/f PO	723	21159	29.3	626858	867.0

APPENDIX D1

Hematology Data for 6, 12, 18, and 24 Month Intervals
(Copied from p. 44 of the study report)

BLUTUNTERSUCHUNG / HEMATOLOGY

ppm	LEU GIGA/L	THROM GIGA/L	ERY TERA/L	HGB G/L	MCV FL	HCT L/L	MCH PG	MCHC G/L	ERY	RETI 0/00
6 MONATE / 6 MONTHS										
MAENNLICH/MALE										
0	4.1	713	8.59	152	62	0.53	17.7	287		
25	5.4*	853*	8.20	150	64*	0.53	18.5	289		
250	5.0*	856*	9.00	161*	64*	0.57**	17.9	281		
2500	6.8**	902*	8.59	158	66**	0.56	18.5**	282		
WEIBLICH/FEMALE										
0	4.1	768	8.12	152	62	0.50	18.8	304		
25	4.2	716	8.17	164**	63	0.51	20.2*	321*		
250	3.5	810	8.31	154	61	0.51	18.6	306		
2500	5.3*	889	7.63**	149	63	0.48*	19.6	310		
12 MONATE / 12 MONTHS										
MAENNLICH/MALE										
0	6.3	814	8.27	143	51	0.42	17.2	336		21
25	7.0	948	8.24	145	51	0.42	17.6	345		22
250	4.2*	816	8.12	143	51	0.42	17.6	343		23
2500	8.3	890	8.07	148	53	0.42	18.4**	351*		34**
WEIBLICH/FEMALE										
0	4.9	750	8.00	146	54	0.42	18.4	344		24
25	4.9	789	8.23	150	53	0.43	18.2	345		22
250	4.2	824	8.31	147	52	0.43	17.6*	337		20
2500	5.8	884*	7.80**	147	55*	0.43	18.9*	342		35**
18 MONATE / 18 MONTHS										
MAENNLICH/MALE										
0	6.1	1238	8.48	150	51	0.43	17.7	348		30
25	6.2	1157	7.95	139	53	0.41	17.7	338**		30
250	6.1	1196	8.23	145	52	0.43	17.6	341*		37*
2500	9.0**	1319	8.20	149	52	0.43	18.2	350		45**
WEIBLICH/FEMALE										
0	4.7	941	7.96	142	52	0.41	17.9	344		38
25	4.6	905	7.28	136	53	0.38**	18.8*	354		39*
250	4.5	923	7.82	140	51	0.40	17.9	350		42*
2500	7.1**	948	7.61	141	55**	0.42	18.5*	336		47**
VERSUCHSENDE / END OF STUDY										
MAENNLICH/MALE										
0	4.9	969	7.42	121	50	0.37	16.3	328		33
25	5.0	1430**	8.11	132	50	0.41	16.3	323		38
250	5.1	1329**	8.71*	139*	50	0.43**	16.0	321		31
2500	6.3	1461**	8.18	130	50	0.41	15.9	317*		36*
WEIBLICH/FEMALE										
0	4.3	1055	8.38	129	48	0.40	15.4	319		35
25	4.6	1140	7.65	122	51	0.38	16.1	318		40
250	3.8	918	8.42	134	49	0.42	16.0	323		32
2500	8.4**	1032	7.88	134	51*	0.40	17.0**	332**		57

APPENDIX D2

Differential Blood Count Data for 6, 12, 18, and 24 Month Intervals (Copied from p. 45 of the study report)

DIFFERENTIALBLUTBILD / DIFFERENTIAL BLOOD COUNT %								
ppm	BASO	EOSIN	IPC/ JGL	STAB	SEGM	LYM	MONO	PLAS
6 MONATE / 6 MONTHS								
MAENNLICH/MALE								
0	0.0	5.3	0.0	0.0	14.3	80.3	0.1	0.0
25	0.0	4.8	0.0	0.1	16.1	78.9	0.1	0.0
250	0.0	3.6	0.0	0.1	16.5	79.7	0.1	0.0
2500	0.0	4.4	0.0	0.0	17.7	77.9	0.0	0.0
WEIBLICH/FEMALE								
0	0.0	4.3	0.0	0.0	8.8	86.9	0.0	0.0
25	0.0	2.9	0.0	0.0	12.8	84.3	0.0	0.0
250	0.0	2.9	0.0	0.0	12.0	85.1	0.0	0.0
2500	0.0	2.2	0.0	0.1	10.5	87.3	0.0	0.0
12 MONATE / 12 MONTHS								
MAENNLICH/MALE								
0	0.0	4.7	0.0	0.0	19.7	75.6	0.0	0.0
25	0.0	4.4	0.0	0.0	17.5	78.1	0.0	0.0
250	0.0	3.6	0.0	0.0	19.0	77.4	0.0	0.0
2500	0.0	2.6	0.0	0.0	22.1	75.3	0.0	0.0
WEIBLICH/FEMALE								
0	0.0	2.2	0.0	0.0	24.0	73.8	0.0	0.0
25	0.0	2.4	0.0	0.0	17.0	80.6	0.0	0.0
250	0.0	1.7	0.0	0.0	21.1	77.2	0.0	0.0
2500	0.0	1.3	0.0	0.0	17.7	81.0	0.0	0.0
18 MONATE / 18 MONATE								
MAENNLICH/MALE								
0	0.0	4.3	0.0	0.0	29.8	65.9	0.0	0.0
25	0.0	3.5	0.0	0.4	26.4	69.7	0.0	0.0
250	0.0	4.4	0.0	0.4	24.0	71.2	0.0	0.0
2500	0.0	3.5	0.0	0.0	26.1	70.4	0.0	0.0
WEIBLICH/FEMALE								
0	0.0	4.1	0.0	0.2	30.2	65.5	0.0	0.0
25	0.0	2.0	0.0	0.1	23.9	74.0*	0.0	0.0
250	0.0	1.6*	0.0	0.0	26.6	71.8	0.0	0.0
2500	0.0	3.3	0.0	0.1	20.0**	76.6*	0.0	0.0
VERSUCHSENDE / END OF STUDY								
MAENNLICH/MALE								
0	0.0	4.9	0.0	0.0	34.5	60.4	0.2	0.0
25	0.0	3.5	0.0	0.1	40.1	56.1	0.2	0.0
250	0.0	6.3	0.0	0.1	41.0	52.4	0.2	0.0
2500	0.0	3.6	0.0	0.2	36.9	59.3	0.1	0.0
WEIBLICH/FEMALE								
0	0.0	1.5	0.0	0.0	32.6	65.9	0.0	0.0
25	0.0	1.8	0.0	0.0	30.1	68.0	0.1	0.0
250	0.0	1.5	0.0	0.1	33.2	65.2	0.0	0.0
2500	0.0	1.4	0.0	0.1	25.9	72.5	0.1	0.0

ABBREVIATIONS OF HEMATOLOGICAL AND CLINICAL CHEMISTRY TERMS USED IN APPENDICES D1, D2, AND E

Hematology

ANISOCYT= Anisocytes
BASO = Basophile
EOSIN = Eosinophile
ERY = Erythrocyte
HCT = Hematocrit
HGB = Hemoglobin
HIK = Heinz Bodies
IPC/JGL = Immature polymorphonuclear neutrophils
LEU = Leucocyte count
LYM = Lymphocytes
MCH = Mean Corpuscular Hemoglobin
MCHC = Mean Corpuscular Hemoglobin Concentration
MCV = Mean Cell Volume
MONO = Monocytes
PLAS = Plasma cells
RETI = Reticulocyte Count
SEGM = Segmented Granulocyte (Mature Neutrophils)
STAB = Band Granulocytes (Immature Neutrophils)
THROM = Thrombocyte

Clinical Chemistry

AP = Alkaline phosphatase
ALAT = Alanine aminotransferase
ASAT = Aspartate aminotransferase
BILI = Bilirubin
CHOL = Cholesterol
CREA = Creatinine
LDH = Lactate dehydrogenase
PROT = Total protein

Statistical Notations

* = $P < 0.05$
** = $P < 0.01$

APPENDIX E

Clinical Chemistry Data for 6, 12, 18, and 24 Months Intervals (Copied from p. 47 of the study report)

DOSE/ DOSIS mg/kg	AP U/L	ASAT (GOT) U/L	ALAT (GPT) U/L	LDH U/L	BILI MICMOL/L	PROT G/L	UREA/ HST MMOL/L	CREA MICMOL/L	CHOL MMOL/L	GLUCOSE MMOL/L
6 MONATE / 6 MONTHS										
MAENNLICH/MALE										
0	65	46.2	41.1		2.3	53.8	10.98	65	3.60	6.01
25	73	60.5	42.7		2.5	55.8	10.02	55	3.15	6.77*
250	82	58.7*	54.6		2.5	54.4	11.38	61	3.19	7.44**
2500	64	47.5	67.0		2.9*	55.4	10.08	61	3.02*	7.57**
WEIBLICH/FEMALE										
0	110	46.5	37.3		2.2	54.6	7.91	58	2.73	6.02
25	128	51.1	37.6		2.1	52.8	8.24	49	2.22*	6.56
250	105	67.7**	40.1		2.2	52.1	10.37**	48	2.33	6.95**
2500	111	65.5*	61.8*		2.8	52.4*	8.56	52	2.40	6.99**
12 MONATE / 12 MONTHS										
MAENNLICH/MALE										
0	88	97.3	83.5		4.7	59.7	10.29	71	4.28	6.93
25	66*	100.8	110.8		4.1	60.5	10.14	59	3.94	6.76
250	66	82.3	92.6		4.5*	58.5	11.05	56	3.88	7.31
2500	87	74.6	96.5		5.5**	53.1**	9.46	57	3.75*	7.07
WEIBLICH/FEMALE										
0	148	61.3	70.9		3.5	63.3	8.42	57	3.15	6.09
25	227	100.1*	83.3		3.9	61.3	9.47	60	2.83	6.80
250	134	64.3	58.1		5.1**	62.3	10.06	48	3.19	6.38
2500	145	70.6	58.8		5.2**	61.3	9.69	59	3.06	6.71
18 MONATE / 18 MONTHS										
MAENNLICH/MALE										
0	75	49.0	57.4		2.4	57.0	9.12	38	3.98	6.27
25	99	79.9	80.9		3.0	59.4	9.92	42	3.69	5.96
250	76	51.7	63.5		3.2	59.6	10.51	41	4.09	7.21*
2500	78	58.2	97.5		3.7*	59.1	9.04	55**	3.56	6.31
WEIBLICH/FEMALE										
0	137	61.0	51.9		2.9	55.4	7.71	61	3.32	6.30
25	350*	70.5	61.4		4.1	53.7	8.47	56	2.78	6.09
250	195	60.5	55.6		3.2	53.9	8.77	43**	3.05	6.76
2500	266	60.0	55.9		3.8	56.0	8.18	53	3.43	5.59*
VERSUCHSENDE / END OF STUDY										
MAENNLICH/MALE										
0	104	54.2	74.5	631	2.8	59.9	9.44	43	3.68	5.98
25	135	62.8	72.8	628	3.2	59.3	9.34	39	3.68	5.44
250	115	49.2	51.9*	444	2.9	59.8	9.93	32	4.05	6.49
2500	123	74.4	185.1*	632	4.1**	60.5	9.62	40	3.13	5.82
WEIBLICH/FEMALE										
0	110	74.3	66.7	497	3.7	57.1	8.76	39	3.25	5.87
25	245	119.3	105.8	1055	4.6*	58.4	10.09	39	2.62	5.88
250	236*	71.8	58.8	430	4.0	60.2	13.46*	30**	3.02	5.63
2500	201*	69.9	87.5	448	4.1	58.9	9.28	31	3.66	4.98

APPENDIX H
Neoplastic Incidence for the 12 Month Sacrificed Mice
(Copied from p. 70 of the study report)

Inzidenzen von Neoplasien/incidences of neoplasms
(Zwischensektion/interim kill)§

Geschlecht/sex	männlich/male				weiblich/female			
Dosis/dose (ppm)	0	25	250	2500	0	25	250	2500
HARDERSCHE DRUESE/HARDERIAN GLANDS								
Cystadenom/cystadenoma (b) (bilateral)	1	-	-	0	0	-	-	0
HARNBLASE/URINARY BLADDER								
Sarkom/sarcoma (m)	0	-	-	0	0	-	-	1
LEBER/LIVER								
Carcinom/carcinoma (m)	0	0	1	0	0	0	0	0
LUNGE/LUNGS								
Adenom/adenoma (b)	0	1	1	1	0	0	1	0
PANKREAS/PANCREAS								
Inselzelladenom/islet-cell adenoma (b)	1	-	-	0	0	-	-	0
OVARIEN/OVARIES #								
Luteom/luteoma (bilateral) (b)	-	-	-	-	1	-	0	0
Haemangiom/hemangioma (b) (bilateral)	-	-	-	-	0	0	0	1
UTERUS/UTERUS #								
Mesenchymom/mesenchymoma (b)	-	-	-	-	0	1	0	0
Anzahl der untersuchten Tiere/ number of animals investigated	10	*	*	10	*	*	10	10
Anzahl der Neoplasien number of neoplasms	3	1	2	1	2	1	1	3
Anzahl der malignen Neoplasien number of malignant neoplasms	0	0	1	0	0	0	0	1
Anzahl der benignen Neoplasien number of benign neoplasms	3	1	1	1	2	1	1	2

b = benigne/benign m = maligne/malignant
* nicht alle Organe routinemaessig untersucht/not examined routinely
- nicht untersucht/not examined
Organ nicht routinemaessig untersucht/not examined routinely
§ bilateral tumors were counted twice

APPENDIX I
Neoplastic Incidence for the 24 Month Sacrificed Mice
(Derived from p. 72 of the study report).

Tissues	Control	Low-dose	Middose	HighDose
	M/F	M/F	M/F	M/F
# of Mice Evaluated	49/50	50/47	49/49	49/50
Total # of Neoplasm	50/55	58/48	52/70	43/67
# of Benign Neoplasm	13/29	25/26	22/32	12/33
# of Malignant Neoplasm	37/26	33/22	30/38	31/34
Liver				
+Adenoma (b)	2/0	5/0	5/1	6/0
+Carcinoma (m)	2/0	0/0	0/0	0/0
+Hemangiosarcoma (m)	1/0	0/1	0/0	0/0
+Sarcoma (m)	0/2	0/0	0/0	0/0
+Hamartoma (b)	0/0	0/0	0/0	0/1
Pancreas				
+Adenoma (b)	0/0	0/2	0/0	0/0
+Islet-cell Adenoma (b)	0/0	0/0	3/0	0/C
Lung				
+Adenoma (b)	14/4	14/4	13/9	12/4
+Carcinoma (m)	4/2	4/2	9/2	4/1
Spleen				
+Hemangioma (b)	1/0	0/0	0/0	0/0
+Hemangiosarcoma (m)	0/0	0/1	0/0	0/0
Ovaries				
+Granulosatheca-cell Tumor (b)	na/8	na/5	na/13	na/7
+Unilateral Luteoma (m)	na/3	na/6	na/2	na/7**
Uterus				
+Stromal Sarcoma (m)	na/1	na/1	na/2	na/2
+Leiomyosarcoma (m)	na/0	na/0	na/0	na/1
Urinary Bladder				
+Suburothelial Islet Malignant Tumor (m)	0/0	0/0	0/0	1/0
Hemolymphoreticular System				
+Lymphoma (m)	4/19	14/16	7/26	5/18
Mammary Gland				
+Adenocarcinoma	na/2	na/1	na/1	na/6*
+Anaplastic Carcinoma	na/0	na/1	na/0	na/0

M/F = Male/Female; b = Benign; m = Malignant; ne = Not examined; na = Not applicable; @ = Stained by Turnbulblue; * = P < 0.05; ** = P < 0.01; Derived from p. 55 and 487 of study report.

US ENVIRONMENTAL PROTECTION AGENCY
OFFICE OF PESTICIDES/HED/SACB/TOX
ONELINERS

TOXCHEM NO.: 410 (Diuron Technical with a 98.7% purity)

CITATION	MATERIAL	ACCESSION/ MRID #	RESULTS	TOX. CAT.	CORE GRADE/ DOCUMENT #
Guideline: 83-5 Chronic/Oncogenicity Study Species: NMRI Mice Institute of Toxicology Bayer, Wuppertal, Germany Study#: AG4010922 Date: October 1983	Diuron with a purity of 98.7%	421595-01	Dosages: 0, 25, 250, and 2500 ppm in diet Based on the data presented, the systemic toxicity LOEL was determined to be 2500 ppm based on findings listed below. The systemic NOEL is 250 ppm. However, because numerous deficiencies and discrepancies were noted (see DER), this study is currently classified as core-supplementary. Treatment-related findings, all noted in the 2500 ppm (HDT) dose groups included: -decreased body weight (♂) -increase spleen and liver weights (♂) -elevated reticulocytes, leucocytes, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and bilirubin (♂ & ♀) -increased hemosiderin deposits in liver (♂) -increased hemosiderin deposits in kidneys (♀) -increased hemosiderin deposits in spleen (♂ & ♀) -increased incidences of hepatopathy and Kupffer cells (♂) -increased incidences of liver cell mitosis and necrosis (♂ & ♀) -increased incidence of enlarge degenerative liver cells (♀) -increased incidences of urinary bladder edema, epithelial hyperplasia and thickened mucosa (♀) -increased incidences of ovarian luteoma and mammary gland adenocarcinoma, and enlarged uterine horns (♀) The doses employed in this study were sufficient to produce compound-related effects and appear to be adequate to test the carcinogenic potential of the test material. This study is classified as core-supplementary and may be upgraded upon satisfactory review of the requested information.		Supplementary

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upgraded to core

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