Cas # 188 C 7-17-81

Government of Canada / Gouvernement du Canada

MEMORANDUM **NOTE DE SERVICE**

TO / A: Dr. C.T. Miller,
Co-ordinator,
Task Force for Re-assessment
of Chemical Safety

FROM / DE: N. Platonow,
Task Force for Re-assessment
of Chemical Safety

SECURITY CLASSIFICATION - DE SÉCURITÉ

OUR FILE - N / REFERENCE

YOUR FILE - N / REFERENCE

DATE: July 17, 1981

SUBJECT / OBJET: AUDIT AND VALIDATION OF THE IBT STUDY: "TERATOGENIC STUDY WITH SD-15418 BLADEX TECHNICAL IN ALBINO RABBITS"

NAME OF LABORATORY:	IBT
LABORATORY REPORT NUMBER:	8580-11112
REPORT DATE:	March 15, 1979
COMMON NAME OF COMPOUND:	Cyanazine
TRADE NAME:	Bladex; SD-15418
FORM OF TEST MATERIAL:	Technical, 98%, Batch 351803
PETITIONER:	Shell Canada
TYPE OF STUDY:	Teratology
SPECIES, BREED AND STRAIN:	New Zealand Albino Rabbits
FILE UNDER :	Cyanazine
RECOMMENDATION:	Audit & Validation: Study Invalid

OVERALL COMMENTS:

Incidence of pregnancy and especially incidence of pregnancy with litter was low in all groups.

Incidence of fatal respiratory and other infection was high, as was abortion and possible abortion.

Taking the above into account it is reasonable to question the quality of animal husbandry and consequently the quality of the study.

... Continued

OVERALL COMMENTS - CONTINUED

The sponsor's auditor recognizes that the incidence of pregnancy was low in all groups but claims that it is not compound related.

We concur with the statement that the incidence of pregnancy was low in all groups, but wish to point out that the incidence of pregnancy with litter at term was unacceptably low. It was as low as 33% in middle group i.e. 1 mg/kg/day. With figures as low as that it is difficult, if not impossible to make any meaningful teratogenic safety evaluation.

There are no indications on raw data that pathology examination and dissection of progeny was performed by or in the presence of pathologist/veterinarian as stated in the final report. Cause of deaths was not properly elucidated: gross pathology raw records contain statements such as "lung full of blood".

In view of the above omissions and deficiencies the present study should be delcared invalid.

NOTE: It should be noted that malformations such as talipomanous (clubfoot), forked rib, floating rib were observed in fetuses of treated groups and should be taken into account during the process of safety evaluation of the present compound.

AUDIT

1. REPORT'S TITLE: "Teratogenic Study with CD-15418 Bladex Technical in Albino Rabbits"

2. REPORT'S NUMBER & DATE: IBT No. 8580-11112
Dated March 15, 1979

3. DATES OF STUDY: Proposed starting date: September 8, 1977
Proposed termination date: December 8, 1977

4. SPONSOR: Shell Oil Company (USA)

5. PROTOCOL: Proposed protocol written by Shell Oil Co. (USA), dated August 26, 1977 is available on microfiche.

6. TEST MATERIAL: 9g of technical Bladex, batch 351803 (Code 8-21-0-0) 98% pure was received at the IBT on September 16, 1977.

7. TEST ANIMALS: Information on raw data is not available on origin, strain, and age of animals. Internal IBT memo states that rabbits were bred on October 18 and 19, 1977.

8. RAW DATA: Adequate in quality and quantity.

VALIDATION

1. DATES: Actual start of study: October 18-19, 1977 (0 day)
Actual termiantion of study: November 16-17, 1 (delivery by caesarian section)

2. PROTOCOL: Three groups (18 does/group) of artificially inseminated female rabbits were exposed orally to Bladex technical at levels of 0.32, 1.0 or 3.2 mg/kg during the period day 6 to 18 of gestation. Control group (18 does) treat with 1% methylcellulose and positive control (18 does) treated with thalidomide were also included. On day 29 of gestation females of all experimental groups were sacrificed and their reproductive status was determined. All fetuses were examined for external, visceral and skeletal alterations.

The IBT in a letter of September 16, 1977 indicated to the Shell Oil Co. that 7-8 month old rabbits were used, rather than 5-6 months old as originally stated, that pathologist will be present at autopsy and during dissection, and that randomization will be used if inseminations are not done on one day (they were done on 2 days).

There is no record indicating that pathologist and/or veterinarian was present during autopsy and/or dissectio

3. PERSONNEL:

The following individuals participated in the conduct of the study and/or preparation of the report:

Dale Fletcher, B.S. - Study Director, Robert Ladd - Group Leader, Agnes Debevec - Senior Technician, Bernard Szyszko, B.A.M.T. - Supervi Quality Assurance.
Specific participation of the above individual will be indicated in the text that follows.

EXECUTION OF THE STUDY

1. MATERNAL PARAMETERS

(i) Dose administration: The record on calculations for preparation of test material - vehicle mixture for different dose levels used is available. The daily doses calculated from the body weights on days 6, 9, 12, 15 and 18 of gestation are recorded for individual animals.

(ii) Body Weights: Raw data are basically similar to results given in the final report. Original individual records are dated and signed by R. Ladd & A. Debenek.

(iii) Reaction & Behavior: Daily observations were recorded, dated and signed by R. Ladd & A. Debenek.

(iv) Mortality: Mortality indicated in the final reports is supported by the raw data.

(v) Reproductive Effects: Individual animal records for corpora lutea, implanat & resorption sites, and number of live fetuses are available on raw data. These parameters correspond to results given in the final reports. Records are dated and signed by R. Ladd and A. Debenek.

(vi) Pathology: Histopathological examination was not required by the protocol a: it was not performed.

Gross pathology was apparently performed on each animal, but individual dat: are not properly tabulated and are not properly reported in the final repor:

The following animals/group were affected with lung infection, most often specified as "lung full of blood":

Control - 2
Positive Control - 6
0.32 mg/kg - 3
1.0 mg/kg - 3
3.2 mg/kg - 2

Final report did not clearly outline number of abortion and number of possible abortion (i.e. blood under cage, doe not pregnant, presence or absence of implantation and resorption sites not reported in raw data). Retabulation of these parameters in shown below.

GROUP	CONTROL	POSITIVE CONTROL	0.32 MG/KG	1 MG/KG	3.2 MG/KG
Inseminated	18	18	18	18	18
Pregnant	10 (55%)	5 (28%)	9 (50%)	8 (44%)	9 (50%)
Pregnant with litter	8 (44%)	3 (17%)	8 (44%)	6 (33%)	8 (44%)
Mortality	1	5	3	2	2
Abortion*	-	1	3	-	1
Possible Abortion	2	1	1	1	1

* Includes: Died Aborting

2. FETAL PARAMETERS:

(i) Body Weights: Individual data are dated and signed by R. Labb & A. Debenek. Raw data basically support results given in the final report.

(ii) Sex Distribution: Fetuses were not sexed.

(iii) Fetal Viability and Body Measurements: Individual data are dated and signed by R. Ladd & A. Debenek. Raw data basically support results given in the final report.

(iv) External and Soft Tissue Development: Individual data are dated and signed by R. Ladd & A. Debenek. Raw data basically support results given in the final report.

(v) Skeletal Development: Raw individual data were compared with results given in the final report and no significant discrepancies were noted.

OVERALL COMMENTS

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OTHER AUDIT & VALIDATION

The sponsor's staff i.e. P.K. O'Sullivan, B.S., A.K. Schweitzer and C.C. Lu, Ph.D., performed audit but no conclusion was drawn whether the study is valid or invalid.

N. Platonow

P. Nawrot

C.T. Miller