

CENTER FOR DRUG EVALUATION AND RESEARCH

APPROVAL PACKAGE FOR:

APPLICATION NUMBER

21-673

Pharmacology Review(s)

MEMORANDUM

Date: December 20, 2004
From: John K. Leighton, Ph.D., DABT
Supervisory Pharmacologist, HFD-150
To: File for NDA #21-673
Re: Approvability for Pharmacology and Toxicology
Clolar (clofarabine)

In support of the NDA application for clofarabine, the Sponsor submitted reports of nonclinical studies of its pharmacologic mechanism of action. Also submitted were reports of studies of the toxicology of clofarabine, including general toxicology (daily for 5 days and repeat dose up for 5 cycles), genetic toxicity (standard ICH battery), and reproductive toxicity. Pharmacokinetic and ADME studies were also provided. The reports of studies investigating the pharmacologic mechanism of action of clofarabine were obtained from the scientific literature; toxicology, pharmacokinetic and ADME studies were conducted by the sponsor.

Clofarabine, a purine analogue of adenine, is a cytotoxic agent with primary pharmacodynamic action as an inhibitor of DNA synthesis and repair. Clofarabine is a prodrug that is converted to the triphosphate before incorporation into DNA. Incorporation of clofarabine monophosphate into DNA is likely the principal route through which clofarabine exerts its pharmacodynamic action. However, Dr. Goheer points out in his review that other mechanisms of anticancer activity may also be important.

Clofarabine is active in growing and quiescent cells, an activity that remains to be fully explored. The evidence supporting this conclusion was discussed in Dr. Goheer's supplemental labeling review to the NDA. Additional exploratory research in this area does not impact the approvability of clofarabine for the proposed indication.

Toxicological findings for the most part were those expected of cytotoxic agents, primarily toxicity to rapidly dividing cells of the gastrointestinal and hematopoietic tissues. Of note, however, was cardiovascular toxicity that may be due to direct action of clofarabine on mitochondria. In a 5-day GLP cardiac biomarker study in male rats conducted by the Sponsor, a lack of statistical significance was observed with troponin I after clofarabine treatment in the presence of myocardial degeneration. These findings were discussed in the Executive Summary of the review. The relevance of the troponin findings is unclear and should be considered exploratory research as the assay used was not fully validated. The use of exploratory research in conjunction with a GLP toxicology study did not invalidate the main part of the GLP toxicology study in which histopathologic lesions were observed. Cardiovascular toxicity was addressed during clinical development.

Embryo-fetal developmental studies (Segment II) indicated that clofarabine was a developmental toxicant in rats and rabbits. The findings were observed in the absence of

maternal toxicity. Pregnancy Category D is recommended. These findings are discussed in detail in the review and in label recommendations.

Recommendations: I concur with Dr. Goheer's conclusion that pharmacology and toxicology data support the approval of NDA 21-673, Clolar for the treatment of pediatric patients with acute lymphoblastic leukemia (ALL) after at least two prior regimens. There are no outstanding nonclinical issues.

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/s/

John Leighton
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PHARMACOLOGIST



DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH

PHARMACOLOGY/TOXICOLOGY REVIEW AND EVALUATION

NDA NUMBER: 21-673
SERIAL NUMBER: 000
DATE RECEIVED BY CENTER: 9/26/2003
PRODUCT: Clofarabine
INTENDED CLINICAL POPULATION: Pediatric primary relapsed or refractory acute leukemia
SPONSOR: ILEX Products, Inc., 4545 Horizon Hill Blvd,
San Antonio, Texas 78229-2263
DOCUMENTS REVIEWED: Electronic submission
REVIEW DIVISION: Division of Oncology Drug Products (HFD-150)
PHARM/TOX REVIEWER: M. Anwar Goheer, Ph.D.
PHARM/TOX SUPERVISOR: John K. Leighton, Ph.D., D.A.B.T.
DIVISION DIRECTOR: Richard Pazdur, M.D.
PROJECT MANAGER: Christy Cottrell

Date of review submission to Division File System (DFS):

Recommendations on labeling

The sponsor proposed:

Mechanism of Action: Clofarabine is sequentially metabolized intracellularly to the 5'-monophosphate metabolite by deoxycytidine kinase and mono- and di-phosphokinases to the active 5'-triphosphate metabolite. Conversion of the



Clofarabine 5'-triphosphate also disrupts the integrity of mitochondrial membrane, leading to the release of the pro-apoptotic mitochondrial proteins, cytochrome C and apoptosis-inducing factor, leading to programmed cell death.



Reviewer: Anwar Goheer, Ph.D.

NDA No. 21-673

We recommend:

Mechanism of Action:

|

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The sponsor proposed:

Activity in Pharmacology Models:

L

J

We recommend:

[quiescent cancer cells in vitro.

] clofarabine is cytotoxic to rapidly proliferating and

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The sponsor proposed:



We recommend:

Deletion

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the approval package consisted of draft labeling

(P. 6)

We recommend:

WARNINGS

Pregnancy – Teratogenic Effects: Pregnancy Category D

Clofarabine may cause fetal harm when administered to a pregnant woman.

Clofarabine was teratogenic in rats and rabbits. Developmental toxicity (reduced fetal body weight and increased post-implantation loss) and increased incidences of malformations and variations (gross external, soft tissue, skeletal and retarded ossification) were observed in rats receiving 54 mg/m²/day (approximately equivalent to the recommended clinical dose on a mg/m² basis), and in rabbits receiving 12 mg/m²/day (approximately 23% the recommended clinical dose on a mg/m² basis).

There are no adequate and well-controlled studies in pregnant women using clofarabine. If this drug is used during pregnancy, or if the patient becomes pregnant while taking this drug, the patient should be apprised of the potential hazard to the fetus.

Women of childbearing potential should be advised to avoid becoming pregnant while receiving treatment with clofarabine.

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The sponsor proposed:

PRECAUTIONS

Pregnancy/Nursing

All patients should be advised to use effective contraceptive measures to prevent pregnancy. Female patients should be advised to avoid breast feeding during treatment with clofarabine.

Carcinogenesis, Mutagenesis, Impairment of Fertility

Pregnancy Category D

See Warnings :

Carcinogenesis

L

]

Mutagenesis

[

]

Impairment of Fertility

Studies in mice, rats, and dogs have demonstrated dose-related adverse effects on male reproductive organs. Seminiferous tubule and testicular degeneration and atrophy were reported in male mice receiving IP doses of τ \nearrow The testes of rats

receiving 25 mg/kg/day (150 mg/m²/day) in a 6-month IV study had bilateral degeneration of the seminiferous epithelium with retained spermatids and atrophy of interstitial cells. In a 6-month IV dog study, cell degeneration of the epididymis and degeneration of the seminiferous epithelium in the testes were observed in dogs receiving 0.375 mg/kg/day (7.5 mg/m²/day) and higher. Ovarian atrophy or degeneration and uterine mucosal apoptosis were observed in female mice at 75 mg/kg/day (225 mg/m²/day), the only dose administered to female mice. The effect on human fertility is unknown.

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We recommend:

PRECAUTIONS

Carcinogenesis, Mutagenesis, Impairment of Fertility

Carcinogenesis: Clofarabine has not been tested for carcinogenic potential.

Mutagenesis: Clofarabine showed clastogenic activity in the in vitro mammalian cell chromosome aberration assay (CHO cells) and in the in vivo rat micronucleus assay. It did not show evidence of mutagenic activity in the bacterial mutation assay (Ames test).

Impairment of fertility: Studies in mice, rats, and dogs have demonstrated dose-related adverse effects on male reproductive organs. Seminiferous tubule and testicular degeneration and atrophy were reported in male mice receiving IP doses of 3 mg/kg/day (9 mg/m²/day, approximately 17% of clinical recommended dose on a mg/m² basis). The testes of rats receiving 25 mg/kg/day (150 mg/m²/day, approximately 3 times of the recommended clinical dose on a mg/m² basis) in a 6-month IV study had bilateral degeneration of the seminiferous epithelium with retained spermatids and atrophy of interstitial cells. In a 6-month IV dog study, cell degeneration of the epididymis and degeneration of the seminiferous epithelium in the testes were observed in dogs receiving 0.375 mg/kg/day (7.5 mg/m²/day, approximately 14% of the clinical recommended dose on a mg/m² basis). Ovarian atrophy or degeneration and uterine mucosal apoptosis were observed in female mice at 75 mg/kg/day (225 mg/m²/day, approximately 4 fold of recommended human dose on a mg/m² basis), the only dose administered to female mice. The effect on human fertility is unknown.

Pregnancy

Teratogenic Effects: Pregnancy Category D. See WARNINGS

Nursing Mothers

It is not known whether clofarabine or its metabolites are excreted in human milk. Because of the potential for tumorigenicity shown for clofarabine in animal studies and the potential for serious adverse reactions, women treated with clofarabine should not nurse.

Recommendation: Accept sponsor's proposal

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/s/

Anwar Goheer
12/15/04 02:52:40 PM
PHARMACOLOGIST

John Leighton
12/20/04 09:40:14 AM
PHARMACOLOGIST



DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH

PHARMACOLOGY/TOXICOLOGY REVIEW AND EVALUATION

NDA NUMBER: 21-673
SERIAL NUMBER: 000
DATE RECEIVED BY CENTER: 9/26/2003
PRODUCT: Clofarabine
INTENDED CLINICAL POPULATION: Pediatric primary relapsed or refractory acute leukemia
SPONSOR: ILEX Products, Inc., 4545 Horizon Hill Blvd,
San Antonio, Texas 78229-2263
DOCUMENTS REVIEWED: Electronic submission
REVIEW DIVISION: Division of Oncology Drug Products (HFD-150)
PHARM/TOX REVIEWER: M. Anwar Goheer, Ph.D.
PHARM/TOX SUPERVISOR: John K. Leighton, Ph.D., D.A.B.T.
DIVISION DIRECTOR: Richard Pazdur, M.D.
PROJECT MANAGER: Amy Baird

Date of review submission to Division File System (DFS): 9/16/2004

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Executive Summary

I. Recommendations

- A. Recommendation on approvability: The non-clinical studies submitted to this NDA provide sufficient information to support the use of clofarabine (CLOLAR™) in pediatric patients with refractory or relapsed acute lymphoblastic leukemia (ALL) and acute myelogenous leukemia (AML).
- B. Recommendation for nonclinical studies: None
- C. Recommendations on labeling: None at this time.
A separate review will be conducted.

II. Summary of nonclinical findings

- A. Brief overview of nonclinical findings: Clofarabine is a second-generation purine nucleoside antimetabolite. It is structurally related to two approved purine analogs for hematological malignancies, fludarabine phosphate and cladribine (2-CdA). Clofarabine differs from these two compounds in that it has 2 halogen atoms incorporated into its structure. Its metabolism and mechanisms of action are significantly different from fludarabine and cladribine (Blood 2000; 96:3537-3543). Clofarabine is an efficient substrate for the activating enzyme deoxycytidine kinase.

The *in vivo* and *in vitro* studies have demonstrated the activity of clofarabine on various tumors and drug-resistant leukemias. Clofarabine is primarily metabolized intracellularly to its 5'-monophosphate by deoxycytidine kinase (dCK). Clofarabine monophosphate is converted to clofarabine diphosphate by a purine nucleotide monophosphate kinase. Diphosphokinase converts clofarabine diphosphate to the active 5'-triphosphate metabolite. Once transformed into the triphosphate form, adenine deoxynucleosides can induce DNA damage and trigger the apoptotic cascade (Proc Natl Acad Sci USA. 1992; 89:2970-2975). Conversion of monophosphate to diphosphate is the rate-limiting step resulting in cellular accumulation of both clofarabine mono- and tri-phosphate. The effects of clofarabine on the metabolism of cytochrome p450 substrates and drug-drug interaction studies have not been conducted.

The proposed mechanism of action of clofarabine (clofarabine triphosphate) is believed to be due to the induction of DNA damage, disruption of mitochondrial function, and activation of pro-apoptotic factors. Clofarabine induces apoptosis in cells by two separate pathways (i.e., a direct action on the mitochondria and an indirect pathway via DNA damage).

During traditional toxicity assessment, clofarabine was administered to rodents (mice, rats) and non-rodent (dogs) for 1, 5, 21, and 163 (d x 5 q 28 d for 5 cycles; d x 5, then 17 days off) days. Single and multi-dose administration of clofarabine induced adverse

effects on rapidly proliferating tissues of the GI, testes, lymphoid tissues, and bone marrow of mice, rats and dogs. Based upon their respective MTD values, the dog (15 mg/m²/day) was the most sensitive species, followed by the rat (75 mg/m²/day) and mouse (225 mg/m²/day). High dose rats and dogs had unilateral ocular findings (superficial focal white corneal deposit) in both eyes. Similar findings have been observed for other drugs in this class (Ophthalm Plast Reconstr Surg 2003, 19:216-24). Clofarabine administration in rats induced adverse histopathological effects on the heart (myocardial degeneration, necrosis, inflammation, fibrosis, and compensatory hypertrophy) in chronic studies. These adverse effects correlated well with the mechanism of action of clofarabine, induction of apoptosis through a direct and indirect action on the mitochondria (Blood 2000; 96:3537-3543).

Cardiovascular safety pharmacology study conducted in male Fischer rats at 300 mg/m²/day clofarabine (~6 fold the recommended pediatric dose) for 5 days showed biologically significant myocardial degeneration without any statistically significant affect on troponin I values. CK-MB was also examined in this study and supported histopathological evidence of myocardial degeneration.

Clofarabine was not mutagenic in the Ames test in the presence and absence of metabolic activator (S-9). Clofarabine did show clastogenic activity in the *in vitro* mammalian cell chromosome aberration assay (CHO) in the presence and absence of metabolic activator (S-9) and in the *in vivo* rat micronucleus assay. The clastogenic findings were expected based upon the mechanism of anticancer activity for clofarabine.

Teratological effects (embryo-fetal development) of clofarabine were examined in both Sprague Dawley rats and New Zealand White rabbits. There were significant increases in major skeletal or visceral malformations due to clofarabine treatment to rats and rabbits at 54 and 12 mg/m²/day, respectively. The recommended pediatric (up to 21 years old) dose is 52 mg/m² daily for 5 consecutive days. Therefore, it is important that sexually active pediatric patients be aware that there is no safety margin and should take precautions to avoid pregnancy.

B. Pharmacologic activity: Clofarabine is a prodrug that is converted by the cytosomal enzyme deoxycytidine kinase (dCK) into its active clofarabine triphosphate. The active clofarabine triphosphate competes with dATP for incorporation into the DNA chain by polymerase α . Once clofarabine triphosphate is incorporated into the DNA chain, it disrupts DNA chain elongation and induces DNA strand breaks.

C. Nonclinical safety issues relevant to clinical use: Cardiac toxicity in male Fischer rats and GI toxicity during long term toxicity studies in rats and dogs were observed.

2.6 PHARMACOLOGY/TOXICOLOGY REVIEW**2.6.1 INTRODUCTION AND DRUG HISTORY**

NDA number: 21-673
Review number: 1
Sequence number/date/type of submission: 001 / 9-26-03 / NDA
Information to sponsor: Yes () No (X)
Sponsor and/or agent: ILEX™ Products, Inc.,
 4545 Horizon Hill Blvd.
 San Antonio, Texas 78229-2263.

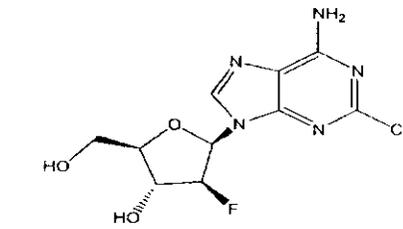
Manufacturer for drug substance:

Reviewer name: M. Anwar Goheer, Ph.D.
Division name: Division of Oncology Drug Products
HFD #: 150
Review completion date: September 16, 2004

Drug:

Trade name: CLOLAR
Generic name: Clofarabine, clofarex
Code name: N/A
Chemical name:
 2-chloro-9-(2'-deoxy-2'-fluoro-β-D-arabinofuranosyl)-9H-purine-6-amine,
 Cl-F-ara-A, 2-chloro-2'-fluoro-deoxy-9-β arabinofuranosyladenine,
 2-chloro-9-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-adenine (Cl-F-ara-A),
 2-chloro-2'-arabino-fluoro-2'-deoxyadenosine, 2-chloro-2'-ara-
 fluoro-deoxyadenosine (CaFdA),
 2-chloro-2'-fluoro-deoxyadenosine (CaFdA), or
 FP028 (ASI Internal Designation Code)

CAS registry number:
Molecular formula/molecular weight: C₁₀H₁₁ClFN₅O₃ / 303.68
Structure:



Relevant INDs/NDAs/DMFs: INDs 63,641 & 43,275

Drug class: Purine nucleoside antimetabolite
Intended clinical population: Pediatric primary relapsed or refractory acute leukemia.
Clinical formulation: Intravenous solution
Route of administration: Intravenous infusion
Proposed use: "The recommended pediatric dose and schedule is 52 mg/m² administered by IV infusion over 1 to 2 hours daily for 5 consecutive days. Treatment cycles are repeated every 2 to 6 weeks following recovery or return to baseline organ function".

Disclaimer: Tabular and graphical information are constructed by the reviewer unless cited otherwise.

Studies reviewed within this submission:

2.6.2 PHARMACOLOGY

2.6.2.2 Primary pharmacodynamics

In Vitro Pharmacodynamic and Mechanism of Action

1. Effects of 2-chloro-9-(2-deoxy-2-fluoro-β-D-arabinofuranosyl) adenine on K562 Cellular Metabolism and the Inhibition of Human Ribonucleotide Reductase and DNA Polymerases by its 5'-triphosphate.
2. Comparison of the Mechanism of Cytotoxicity of 2-chloro-9-(2-deoxy-2-fluoro-β-D-arabino-furanosyl) adenine, 2-chloro-9-(2-deoxy-2-fluoro-β-D-ribofuranosyl) adenine, 2-chloro-9-(2-deoxy-2,2-difluoro-β-D-ribofuranosyl) adenine in CEM Cells.
3. Deoxyadenosine Analogues Induce Programmed Cell Death in Chronic Lymphocytic Leukemia Cells by Damaging the DNA and by Directly Affecting the Mitochondria.
4. DNA Repair Initiated in Chronic Lymphocytic Leukemia Lymphocytes by 4-hydroperoxycyclophosphamide is Inhibited by Fludarabine and Clofarabine.
5. Comparison of Cytotoxicity of 2-chloro-2'-arabino-fluoro-2' deoxyadenosine (clofarabine) with Cladribine in Mononuclear Cells from Patients with Acute Myeloid and Chronic Lymphocytic Leukemia.

In Vivo Pharmacodynamic and Drug Activity Related to Proposed Indication

6. Synthesis and Biologic Activity of 2'-Fluoro-2-halo derivatives of 9 β-D-arabinofuranosyladenine.

7. Oral Antilymphocyte Activity and Induction of Apoptosis by 2-chloro-2'-arabino-fuoro-2'-deoxyadenosine.
8. Preclinical Antitumor Activity of 2-chloro-9-(2-deoxy-2-fluoro- β -D-arabino-furanosyl)-adenine (Cl-F-ara-A).
9. Antitumor Activity of 2-chloro-9-(2-deoxy-2-fluoro- β -D-arabinofuranosyl) adenine, A Novel Deoxyadenosine Analog, Against Human Colon Tumor Xenografts by Oral Administration.

2.6.2.4 Safety pharmacology

1. Five- Day Intravenous GLP Cardiac Biomarker Study of Clofarabine in Male Fischer 344 Rats. Study Number: 0204- 04.

2.6.4 PHARMACOKINETICS/TOXICOKINETICS

2.6.4.4 Distribution:

1. Tissue Distribution in Male Fischer 344 Rats Following a Five Day Intravenous Dose Regimen of 14 C-Clofarabine. Report No. QKAN-2002-0783-ADM.

2.6.4.5 **Metabolism:** See under excretion and general toxicity studies in rats and dogs.

2.6.4.6 Excretion

1. Clofarabine Mouse Studies (Report MDACC 1).
2. Clofarabine Dog studies (Report MDACC 2).

2.6.4.8 Other Pharmacokinetic Studies

1. In Vitro Metabolism of Clofarabine in Rat, Dog, and Human Cryopreserved Hepatocytes. [Report No.: [2002-0542-BIO, [Study No.: BA020050, and [Project No.: DAA00710.100.]]]

2.6.6 TOXICOLOGY

2.6.6.3 Repeat-dose toxicity

1. A Multi-Cycle (Five Days' Dosing Plus 23 Days' Recovery per Cycle] Intravenous Toxicity and Toxicokinetics Study in Fischer 344 Rats. — Study No. 5471, ILEX Study Number: 0204-03.
2. A Multi-Cycle (Five Days' Dosing Plus 23 Days' Recovery per Cycle) Intravenous Toxicity and Toxicokinetics Study in Beagle Dogs. — Study No. 2758, ILEX Study No. 0203-01.

2.6.6.4 Genetic toxicology

1. Bacterial Reverse Mutation Report. Study No. AA53PR.503. — CLO.00.07.02.
2. In Vitro Mammalian Chromosome Aberration Test. Study No. AA53PR.331. — , CLO.00.07.03.
3. Mammalian Erythrocyte Micronucleus Test. Study No. AA53PR.125. — , CLO.00.48.

2.6.6.6 Reproductive and developmental toxicology

1. Intravenous Dosage-Range Developmental Toxicity Study of Clofarabine in Rats. Sponsor's Study Number: 0202-01.
2. Intravenous Developmental Toxicity Study of Clofarabine in Rats. Sponsor's Study Number: 0204-01.
3. Intravenous Dosage-Range Developmental Toxicity Study of Clofarabine in Rabbits. Sponsor's Study Number: 0202-02.
4. Intravenous Developmental Toxicity Study of Clofarabine in Rabbits. Sponsor's Study Number: 0204-02.

Studies not reviewed within this submission:**2.6.2 PHARMACOLOGY****2.6.2.2 Primary pharmacodynamics**

These studies are discussed in the pharmacology tabulated summary.

1. Clofarabine PO or IP Against HT29 Human Colorectal Carcinoma in Nude Mice. ILEX Study Number: DE111802- 02.
2. Response of SC CCRF-CEM Leukemia to Combination Treatment with Clofarabine and ARA-C. Report: ILEX 12.
3. Response of SC NCI-H460 Lung Tumor to Treatment with Clofarabine. Report: ILEX 16.
4. Response of SC HT29 Colon Tumor to Combination Treatment with Clofarabine and Irinotecan. Report: ILEX 19.
5. Response of SC HT29 Colon Tumor to Combination Treatment with Clofarabine and Oxaliplatin. Report: ILEX 24.
6. Response of SC HT29 Colon Tumor to Combination Treatment with Clofarabine and Camptosar. Report: ILEX 25.
7. Response of SC HT29 Colon Tumor to Combination Treatment with Clofarabine and 5-FU. Report: ILEX 8.
8. Response of SC PC-3 Prostate Tumor to Combination Treatment with Clofarabine and Taxotere. Report: ILEX 9.

2.6.4 PHARMACOKINETICS/TOXICOKINETICS

2.6.4.2 Methods of Analysis:

1. Validation of a Method for the Determination of Clofarabine in Rat Plasma Using
L
1
2. Validation of a Method for the Determination of Clofarabine in Dog Plasma
Using C
1

2.6.4.4 Distribution

1. Distribution of 2'-Chloro-2'-deoxyadenosine, 2-Chloro-2'-arabino-fluoro-2'-deoxyadenosine, Fludarabine, and Cytarabine in Mice: A Whole Body Autoradiography Study.

2.6.4.5 Metabolism

1. In Vivo Metabolism of Clofarabine in Rat Myocardium, Liver, Urine, Feces, and Plasma Samples Collected from Protocol PK-917
2. Metabolism and Actions of 2-Chloro-9-(2-deoxy-2-fluoro- β -D-arabinofuranoyl)-adenine in Human Lymphoblastoid cells.

2.6.4.6 Excretion

1. Radiokinetic and ^{14}C Excretion/Mass Balance Study of ^{14}C -Clofarabine in Fischer 344 Rats.

2.6.6 TOXICOLOGY

2.6.6.3 Repeat-dose toxicity

1. Five- Day Intravenous GLP Toxicity Study of Clofarabine in Male Fischer 344 Rats. Study Number: 0106- 01.
2. Five- Day Intravenous Infusion GLP Toxicity Study of Clofarabine in Male Fischer 344 Rats. Study Number: 0110- 02.
3. Three Weekly Oral and Intravenous Dose Bridging Toxicity Study of Clofarabine in Fischer 344 Rats. Study Number: 0204-05.
4. Twenty-One Day Oral Range-Finding Study of Clofarabine in FISHER 344 Rats. Study Number: 0110-01.
5. Twenty- One Day Oral Range- Finding Study of Clofarabine in Beagle Dogs Study Number: 0109- 01.

2.6.2 PHARMACOLOGY

2.6.2.1 Brief summary: In nonclinical pharmacology studies, clofarabine has demonstrated antitumor activity against a variety of human cancer cell lines (AML, CLL) and tumor of xenografts (solid and hematologic tumors). Clofarabine inhibited DNA synthesis in K562 cells by inhibiting ribonucleotide reductase and DNA polymerase α . The data in support of these findings are presented in Pharmacology Tabulated Summary below.

2.6.2.2 Primary pharmacodynamics:

In Vitro Pharmacodynamic and Mechanism of Action

1. Effects of 2-chloro-9-(2-deoxy-2-fluoro- β -D-arabinofuranosyl) adenine on K562 Cellular Metabolism and the Inhibition of Human Ribonucleotide Reductase and DNA Polymerases by its 5'-triphosphate.

Clofarabine inhibited K562 cell (chronic myelogenous leukemia cell line) growth with an IC_{50} value of 5 nM following 72 hours of continuous exposure. Clofarabine inhibited DNA synthesis but did not affect the incorporation of [3H]Urd into RNA or [3H]leucine into protein (Cancer Research 1991; 51:2386-2394).

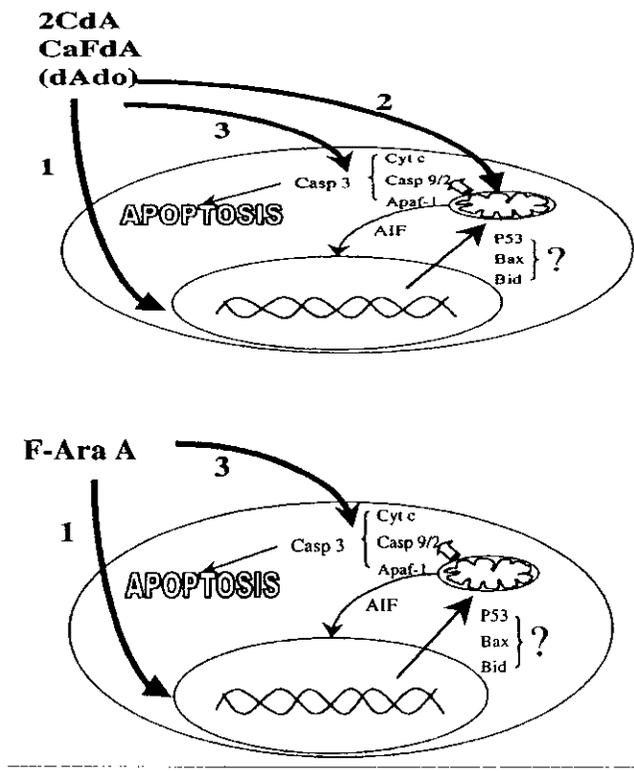
2. Comparison of the Mechanism of Cytotoxicity of 2-chloro-9-(2-deoxy-2-fluoro- β -D-arabino-furanosyl) adenine, 2-chloro-9-(2-deoxy-2-fluoro- β -D-ribofuranosyl) adenine, 2-chloro-9-(2-deoxy-2,2-difluoro- β -D-ribofuranosyl) adenine in CEM Cells.

The biochemical pharmacology of three structurally similar compounds with different antitumor activities is reported in this paper. Clofarabine was 50-fold more potent as inhibitor of CEM cell growth than the other two compounds as shown below. Clofarabine was 10- to 20-fold more potent in its ability to inhibit DNA synthesis with little or no effect on RNA or protein synthesis (Molecular Pharmacol 1999; 55:515-520).

Compound	IC_{50} (nM)
Clofarabine	6
2-Chloro-9-(2-deoxy-2-fluoro- β -D-ribofuranosyl) adenine	313
2-Chloro-9-(2-deoxy-2,2-difluoro- β -D-ribofuranosyl) adenine	317

3. Deoxyadenosine Analogues Induce Programmed Cell Death in Chronic Lymphocytic Leukemia Cells by Damaging the DNA and by Directly Affecting the Mitochondria.

Clofarabine (2-chloro-2'fluorodeoxyadenosine, CaFdA), cladribine (2-chloro-2'-deoxyadenosine, 2CdA), 2-deoxyadenosine (dAdo), and fludarabine (9-β-D-arabinofuranosyl-2-fluoroadenine, F-Ara-A) induced DNA damage in quiescent lymphocytes. The cytotoxicity of these drugs depends mainly on the accumulation of their triphosphate metabolites in lymphocytes. Once transformed into the triphosphate form, adenine deoxynucleosides can induce DNA damage and trigger the apoptotic cascade (Proc Natl Acad Sci USA. 1992; 89:2970-2975). Clofarabine and cladribine activated caspase 3 and caspase 9 after 12 hours of incubation. The nucleoside-induced damage led to the release of the pro-apoptotic mitochondrial proteins cytochrome c and apoptosis-inducing factor. Fludarabine took 24-hour incubation to activate caspase 3 and caspase 9 but did not affect mitochondrial function (i.e., membrane potential). According to the authors, "interference with mitochondrial integrity may be a factor in the potent cytotoxic effects of clofarabine and cladribine toward nondividing lymphocytes." (Blood 2000; 96:3537-3543).

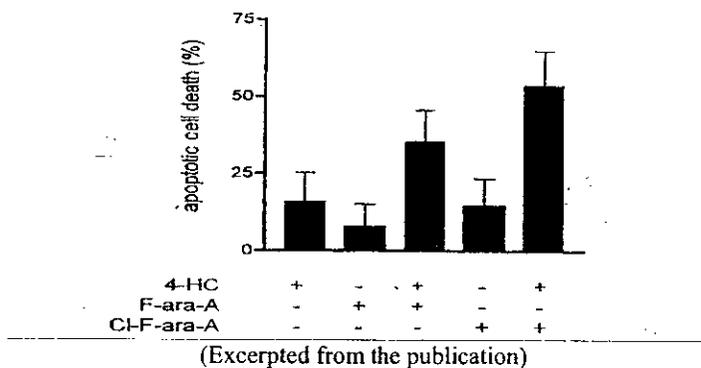


(Excerpted from the publication)

Mechanisms of action: There are 3 proposed modes of action of the adenine deoxynucleoside analogs in nonproliferating CLL cells. In the cells, the drugs are transformed into their triphosphate form, which induce DNA damage, mitochondrial dysfunction, and binding to APAF- 1 (cytosolic apoptosis activating factor) and activation of the caspase pathway. F-Ara-A (fludarabine) kills cells mainly by DNA damage and Apaf-1 activation. 2CdA (cladribine) and CaFdA (clofarabine) also disrupted mitochondrial function leading to the release of the pro-apoptotic mitochondrial proteins cytochrome c and apoptosis-inducing factor in B-CLL cells. The different effects of the adenine deoxynucleosides in various lymphoid malignancies may reflect the relative importance of the 3 mechanisms in apoptosis regulation.

4. DNA Repair Initiated in Chronic Lymphocytic Leukemia Lymphocytes by 4-hydroperoxycyclophosphamide is Inhibited by Fludarabine and Clofarabine.

Lymphocytes from 50 patients with chronic lymphocytic leukemia (CLL) were treated with a cyclophosphamide prodrug (4-hydroperoxycyclophosphamide, 4-HC) with or without prior incubation with fludarabine nucleoside (F-ara-A) or with clofarabine (Cl-F-ara-A). The purpose of this experiment was to evaluate the effect of the nucleoside analogs on the DNA repair process after initiating damage with 4-HC. DNA damage repair kinetics and cytotoxicity were determined. Results are shown below. Preincubation with nucleoside analogues inhibited DNA repair initiated by 4-HC (Clin Cancer Res 2001; 7:3580-89).



Increase in 4-HC induced cytotoxicity by the addition of nucleoside analogues.

5. Comparison of Cytotoxicity of 2-chloro-2'-arabino-fluoro-2' deoxyadenosine (clofarabine) with Cladribine in Mononuclear Cells from Patients with Acute Myeloid and Chronic Lymphocytic Leukemia.

Mononuclear cells isolated from 52 patients with chronic lymphocytic (CLL) and acute myeloid leukemia (AML) were incubated with drugs for 48 hours and activity of deoxycytidine kinase (dCK) and deoxyguanosine kinase (dGK), and cytotoxicity were evaluated.

The *in vitro* median EC₅₀ values for clofarabine and cladribine were 0.12 and 0.15 μM, respectively, indicating slightly higher cytotoxicity with clofarabine than with cladribine. Clofarabine was phosphorylated more efficiently than cladribine. There was no significant difference in enzyme activity between the refractory/relapsed and untreated CLL or AML patients for either dCK or dGK (Haematologica 2003; 88:324-332).

In Vivo Pharmacodynamic and Drug Activity Related to Proposed Indication

6. Synthesis and Biologic Activity of 2'-Fluoro-2-halo derivatives of 9 β-D-arabinofuranosyladenine.

Cd2F1 mice were implanted IP with 10⁶ P388 leukemia cells. Tumor bearing mice were treated with clofarabine QD, days 1-5 at 100 or 200 mg/kg/dose and Q3Hx8, days 1, 5, and 9 at 20 or 25 mg/kg/dose. Clofarabine increased the life span (38-220 %) of mice inoculated with P388 leukemia. The best results were obtained when the compounds were administered q3H x 8 on days 1, 5, and 9 after implantation of leukemia cells [J Med Chemistry 1992; 35:397-401].

7. Oral Antilymphocyte Activity and Induction of Apoptosis by 2-chloro-2'-arabino-fluoro-2'-deoxyadenosine.

SCID mice were injected IP with 5 x 10⁷ CLL cells. One week after implantation, the mice were treated with PO cladribine or clofarabine (1 mg/mL in drinking water) and results are shown below [Proc. Natl. Acad. Sci. USA. 1992; 89:2970-2974].

Treatment Group	Number of Mice	Plasma Drug Level (nM)	% of CLL Cells Surviving Mean ± SE
Control Vehicle	8	0	83.0 ± 2.8
Cladribine	9	48	46.9 ± 2.0
Clofarabine	9	562	8.9 ± 6.6

SE: standard error

(Excerpted from the publication)

8. Preclinical Antitumor Activity of 2-chloro-9-(2-deoxy-2-fluoro- β -D-arabino-furanosyl)-adenine (Cl-F-ara-A).

Clofarabine exhibited cytotoxicity against a variety of human and murine tumors (colon, renal, lung, prostate) and compared with antitumor activity of fludarabine is shown in the Pharmacology Tabulated Summary (page 19). Clofarabine activity against P388 leukemia in vivo is shown below [Nucleosides, Nucleotides and Nucleic Acids 2000; 19:447-460].

Activity of clofarabine against P388 leukemia in vivo is shown below.

Schedule	Route	Dosage range (mg/kg/dose)	Optimal dosage (<LD ₁₀) (mg/kg/dose)	Total dose (mg/kg/dose)	Median % ILS ^a (dying mice only)	Net log ₁₀ cell kill ^b	Tumor-free survivors/total
Days 1-5	ip	300-100	200	1000	+59	+1.6	0/3
q3h x 8, Days 1, 5, 9	ip	30-10	20	480	+220	+6.6	1/6
q4h x 3, Days 1-9	ip	20-8.9	20	540	+300	+6.0	3/6
q6h x 4, Days 1, 5, 9	po	150-45	67	804	+104	+1.7	0/6

^a Median day of death of tumored control mice was 10-11 days.

^b Net log₁₀ reduction in the tumor cell population between the beginning and the end of therapy, based on the median day of death of the mice that died.

(Excerpted from the publication)

9. Antitumor Activity of 2-chloro-9-(2-deoxy-2-fluoro- β -D-arabinofuranosyl) adenine, A Novel Deoxyadenosine Analog, Against Human Colon Tumor Xenografts by Oral Administration.

Clofarabine showed antiproliferative activity against four human colon tumor cell lines (HCT116, HT-29, DLD-1, WiDr). It also showed antitumor activity against four human colon tumor xenograft models (HT-29, WiDr, Co-3, COLO-320M) in a 5-day daily administration schedule [Cancer Chemother Pharmacol 1999; 43:233-240].

2.6.2.3 Secondary pharmacodynamics: See primary pharmacodynamics.

2.6.2.4 Safety pharmacology

Neurological effects: Not studied

Cardiovascular effects:**1. Five- Day Intravenous GLP Cardiac Biomarker Study of Clofarabine in Male Fischer 344 Rats. Study Number: 0204- 04.**

Key study findings: The lowest lethal dose (LLD) and MTD were 50 and 25 mg/kg/day, respectively, for IV administration of clofarabine for five consecutive days. CK-MB was a biomarker indicative of myocardial degeneration in this study.

Study no.: 0204- 04
Volume #, and page #: Electronic submission
Conducting laboratory and location: ILEX™ Products, Inc.,
 4545 Horizon Hill Blvd., San Antonio
 Texas 78220- 2263, USA.
Date of study initiation: April 17, 2002
GLP compliance: Yes
QA report: yes (X) no ()
Drug, lot #, and % purity: Clofarabine, lot # 010188, — purity
Vehicle: 25% PEG 400 in 0.9% sodium chloride for injection, USP.

Methods

Doses:

Group No.	Compound	Route	Clofarabine Dosage (mg/kg/day)	Clofarabine Dosage (mg/m ³ /day)	Clofarabine Concentration (mg/mL)	No. of Rats
1	Vehicle	IV	0	0	0	8
2	Clofarabine	IV	12.5	75	1.25	8
3	Clofarabine	IV	25	150	2.5	8
4	Clofarabine	IV	50	300	5.0	8

(Excerpted from the sponsor's submission)

Species/strain: Fischer 344 rats
Number/sex/group or time point (main study): 8 • /group
Route, formulation, volume, and infusion rate: IV in the tail vein daily for 5 consecutive days, 10 mL/kg.
Satellite groups used for toxicokinetics or recovery: No
Age: 10 weeks
Weight: 190-258 g
Unique study design or methodology: Cardiac troponin I was measured by

C

3

assay () Creatine kinase isoenzyme MB was determined by gel electrophoretic separation of total creatine kinase () and analyzed for individual isozyme bands by scanning in fluorescence mode. Troponin I and CK-MB measurements were performed at ()

Observation times and results

Observation (times)	Results
Mortality: (daily)	G4 (50 mg/kg) – 2 died (25 %) on day 4 and 5 (63 %) euthanized in extremis on day 4
Clinical signs: Once daily	G4 - Rough fur, decreased motor activity, and salivation
Body weights: Daily	G4 – 18% ↓ on day 8 compared to day 1
Food consumption	Not measured
Cardiac biomarkers: Pre and on days 1, 5, and 8 at 2 and 8 hours after dosing	G4 - ↑Troponin I (statistically not significant) on day 4. CK-MB values ↑ for high dose animals as compared to control, 12.5, and 25 mg/kg animals (see tables below).
EKG	Not measured
Urinalysis (not done)	n/a
Organ weights (not done)	n/a
Gross pathology (D)	G4 – Lungs, jejunum and thymus discolored red.
Histopathology: heart slides only	G4 – Myocardial degeneration in left ventricle myocardium in a band subjacent to the endocardial surface, interventricular septum, and left atrial myocardium. Five of the eight rats had proliferation of endomysial cells. Pulmonary congestion was also present.

Group Mean Troponin I (ng/mL)

Dosage (mg/kg/day)		Pre-dose	Day 1		Day 4	Day 5		Day 8
			2 hour	8 hour		2 hour	8 hour	
0	Mean	0.1	0.1	0.0	-	0.1	0.0	0.2
	SD	0.1	0.1	0.1	-	0.2	0.1	0.1
	n	8	8	8	0	8	8	7
12.5	Mean	0.2	0.1	0.1	-	0.1	0.0	0.2
	SD	0.1	0.1	0.1	-	0.1	0.0	0.1
	n	8	8	8	0	8	8	8
25	Mean	0.0	0.1	0.2	-	0.0	0.1	0.4
	SD	0.1	0.1	0.1	-	0.0	0.1	0.8
	n	8	8	8	0	8	8	8
50	Mean	0.2	0.3	0.4	0.1	0.0	0.2	0.0
	SD	0.2	0.1	0.1	0.1	-	-	-
	n	8	8	8	5	1	1	1

(Excerpted from the sponsor's submission)

There was no statistically significant effect on troponin I values, even five high dose animals euthanized in extremis on day 4 had no increases in troponin I when compared to control.

However, the pre-dose troponin I value for animal no.4106, dosed at 50 mg/kg/day was 0.6 ng/mL (see table below). It may be considered spurious outlier because this value exceeded the control and all treated dose range values. The same animal had \leq (below the limit of quantitation, \leq ng/mL) and \leq ng/mL troponin I values on day 1 after 2 and 8 hours clofarabine administration, respectively. This animal was found dead on day 4 with myocardial edema, degeneration, lesions concentrated in left ventricle and interventricular septum. All eight high dose animals showed myocardial degeneration.

Measurement of cardiac troponin I is very method specific (Clinica Chemica Acta 2000; 298:13-28, Clin Chem 2002; 48:1626-7 & Clin Chem 2004; 50:327-32) and individual values for each animal at different timing are shown below.

Individual Troponin I (ng/mL)

Dosage (mg/kg/day)	Animal No.		Pre-dose	Day 1		Day 4	Day 5		Day 8
	ID	Ear Tag		2 hour	8 hour		2 hour	8 hour	
0	1101	AAA-062	0.3						
	1102	AAA-029	0.0						
	1103	AAA-049	0.0						
	1104	AAA-051	0.3						
	1105	AAA-032	0.0						
	1106	AAA-055	0.0						
	1107	AAA-080	0.0						
	1108	AAA-052	0.2						
			Mean	0.1	0.1	0.0	-	0.1	0.0
		SD	0.1	0.1	0.1	-	0.2	0.1	0.1
12.5	2101	AAA-045	0.2						
	2102	AAA-036	0.0						
	2103	AAA-050	0.2						
	2104	AAA-056	0.0						
	2105	AAA-061	0.4						
	2106	AAA-048	0.2						
	2107	AAA-063	0.2						
	2108	AAA-031	0.0						
			Mean	0.2	0.1	0.1	-	0.1	0.0
		SD	0.1	0.1	0.1	-	0.1	0.0	0.1
25	3101	AAA-053	0.0						
	3102	AAA-034	0.2						
	3103	AAA-030	0.0						
	3104	AAA-043	0.0						
	3105	AAA-058	0.0						
	3106	AAA-057	0.0						
	3107	AAA-028	0.0						
	3108	AAA-041	0.0						
			Mean	0.0	0.1	0.2	-	0.0	0.1
		SD	0.1	0.1	0.1	-	0.0	0.1	0.8
50	4101	AAA-042	0.0						
	4102	AAA-037	0.0						
	4103	AAA-040	0.2						
	4104	AAA-059	0.0						
	4105	AAA-038	0.0						
	4106	AAA-044	0.6						
	4107	AAA-054	0.2						
	4108	AAA-035	0.2						
			Mean	0.2	0.3	0.4	0.1	0.0	0.2
		SD	0.2	0.1	0.1	0.1	-	-	-

Troponin values < 0.2 represented as 0.

(Excerpted from the sponsor's submission)

CK-MB (U/L)			
Dosage (mg/kg/day)		Day 4	Day 8
0	Mean	-	1
	SD	-	2
	n	0	7
12.5	Mean	-	0
	SD	-	0
	n	0	8
25	Mean	-	0
	SD	-	0
	n	0	8
50	Mean	20	0
	SD	5	-
	n	5	1

(Excerpted from the sponsor's submission)

Conclusion: While troponin I values may not be statistically significant at 2 and 8 hour time point, there appears to be an increase in these values over control and pretreatment levels in most HD animals. An increase is also seen at MD 8 hour values. The time points chosen and/or method for evaluation may not be optimal for this serum biomarker.

The CK-MB value of 5 high dose animals sampled on day 4 exceeded the group mean value of the control on day 8. These high CK-MB levels are supported by microscopic evidence of myocardial degeneration in these animals.

Pulmonary effects: Not studied

Renal effects: Not studied

Gastrointestinal effects: Not studied

Abuse liability: Not studied

Other: None

2.6.2.5 **Pharmacodynamic drug interactions:** Not studied

2.6.3 PHARMACOLOGY TABULATED SUMMARY

In vitro techniques were used to compare the antineoplastic activity of clofarabine (clofarex) with that of fludarabine and cladribine.

Summary of In Vitro Activity

	In Vitro Cytotoxicity of Clofarex and Fludarabine Phosphate Against Human Cell Lines								
	IC ₅₀ Values (µM)								
	ACHN	SNB-7	NCI-H23	DLD-1	SK-MEL-28	K562	WI-38	CAK-1	CCRF-CEM
Clofarex	0.11	0.29	0.29	11	0.67	0.028	6.8	0.29	0.15
Fludarabine phosphate	6.4	54	45	70	39	0.58	5.1	41	0.29

ACHN: human renal cell carcinoma, SNB-7: human CNS tumor, H23: human non-small cell lung adenocarcinoma, DLD-1: human colon adenocarcinoma, SK-MEL-28: human melanoma, K562: human chronic myelogenous leukemia, WI-38: normal human fibroblasts, CAK-1: human renal carcinoma, CCRF-CEM: human acute lymphoblastic leukemia.

(Excerpted from the sponsor's submission)

Comparative Antineoplastic Activity of Clofarabine (Clofarex), Fludarabine, and Cladribine Against Human Colon Tumor Cell Lines

Cell Line	Antiproliferative Activity IC ₅₀ (µM) ^a		
	Clofarex	Fludarabine	Cladribine
HCT116	0.12	3.1	0.12
HT-29	0.77	30	3.4
DLD-1	0.07	18	0.09
WiDr	0.67	72	9.5

^aCells were exposed to various concentrations of the drugs for 72 hours.

(Excerpted from the sponsor's submission)

Activity of Clofarabine against Drug-Resistant P388 Leukemias in Vivo

Expt. No.	Resistant Leukemia ^a	Optimal IP dosage ^b (cLD ₁₀ mg/kg/dose)	Therapeutic Response						Cross-Resistance
			Parental P388 Leukemia			Resistant Leukemia ^a			
			Median %ILS	Net Log ₁₀ Cell Kill ^c	60-day Survivors/Total	Median %ILS	Net Log ₁₀ Cell Kill ^c	60-day Survivors/Total	
1	P388/ADR	13.3	+252	+6.6	2/5	+170	+6.8	0/5	No
3	P388/VP-16	13.3	+240	+7.0	1/5	+107	+4.1	0/5	Marginal
1	P388/Taxol	13.3	+252	+6.6	2/5	+162	+6.6	3/5	No
3	P388/L-PAM	13.3	+240	+7.0	1/5	-	-	5/5	No
2	P388/ARA-C	13.3	+305	+6.7	3/5	-30	-2.3	0/5	Yes
2	P388/MTX	13.3	+305	+6.7	3/5	+287	+6.4	3/5	No

^a ADR = doxorubicin, VP-16 = etoposide, Taxol = paclitaxel, L-PAM = melphalan, ARA-C = 1-β-D-arabino-furanosylcytosine, MTX = methotrexate.

^b Clofarex was injected every 4 h x 3 on days 1-9 using an injection volume of 0.25 mL/10 g animal body weight.

^c Net log₁₀ reduction in the tumor cell population between the beginning and the end of therapy, based on the median day of death of mice that died.

^d In these studies, the degree of resistance of a drug-resistant subline in comparison to the parental line was as follows: ADR, 5-log₁₀ units; VP-16 9-log₁₀ units; Taxol, 2-log₁₀ units; L-PAM, 7-log₁₀ units; ARA-C, 8-log₁₀ units; and MTX, 2-log₁₀ units.

(Excerpted from the sponsor's submission)

In vivo studies were also conducted with a variety tumors implanted in mice and results are shown below.

Summary of In Vivo Activity

Tumor	Optimal IP Dose (mg/kg/dose) [(Q4H x 3) x 8 Days]	Clofarabine	Fludarabine Phosphate
		Growth Delay [(T-C)/C, %]	Growth Delay [(T-C)/C, %]
Human colon DLD-1	20	181	21
Human colon HCT-15	20	356	101
Human colon HCT-116	20	198	48
Human colon HT29	20	164	81
Human colon KM20L2	20	310	150
Human colon SW620	13.3	179	67
Human renal A498	20	245	73
Human renal RXF 393	20	35	13
Human lung A549	13.3	165	91
Human lung NCI-H322M	20	65	27
Human lung NCI-H23	20	146	37
Human lung NCI-H460	20	127	22
Human prostate DU-145	20	116	74
Human prostate LNCAP	13.3	98	108
Human prostate PC-3	13.3	497	470
Murine mammary 16/C	20	88	35

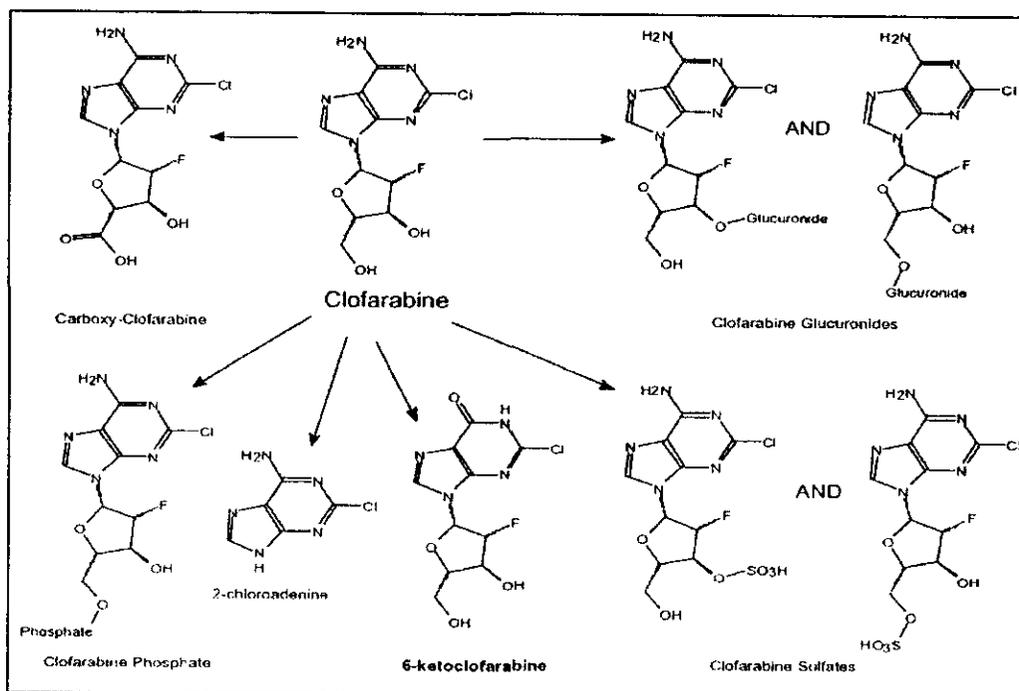
(Excerpted from the sponsor's submission)

In conclusion, in vivo and in vitro studies showed the activity of clofarabine in various cell lines and xenograft models including drug-resistant leukemias.

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2.6.4 PHARMACOKINETICS/TOXICOKINETICS

2.6.4.1 **Brief summary:** The pharmacokinetics of clofarabine has been studied *in vivo* in mice, rats, and dogs. The proposed metabolites identified are shown below.



(Excerpted from the sponsor's submission)

2.6.4.2 **Methods of Analysis:** See under excretion and general toxicity studies in rats and dogs.

2.6.4.3 **Absorption:** Intravenous drug

2.6.4.4 Distribution**Tissue Distribution in Male Fischer 344 Rats Following a Five Day Intravenous Dose Regimen of ¹⁴C-Clofarabine. Report No. QKAN-2002-0783-ADM.**

Key study findings: The highest concentrations of ¹⁴C clofarabine were found in the urine, urinary bladder, spleen, kidney, cecum, and thymus. Low concentrations of radioactivity were detected in the tissues of the central nervous system through the 2- hour time point, but declined quickly and were not quantifiable at 6 hours postdose. Distribution to bone marrow was not particularly notable.

Study no: PK-916
Volume #, and page #: Electronic submission
Conducting laboratory and location: Quintiles, Inc, 10245 Hickman Mills Drive, Kansas City, Missouri.
Date of study initiation: June 27, 2002
GLP compliance: Yes
QA report: yes (X) no ()
Drug, lot #, radiolabel, and % purity: Clofarabine, lot # CSL-01-142-60-33, 165 µ Ci/mg (50.1 mCi/mmol), ...

Methods: Animals were sacrificed and blood collected prior to dosing on day 5, and 0.5, 2.0, 6.0, and 30 hours after the final dosing on day 5. Two animals were sacrificed at each time. Therefore, stated concentration values are the mean of two animals at each sampling time. Radioactivity concentrations were determined by quantitative whole body autoradiography (QWBA) and liquid scintillation counting (LSC).

Dosing:

Species/strain: Fischer 344 rats
#/sex/group or time point (main study): 10 males/group
Satellite groups used for toxicokinetics or recovery: No
Age: 9-11 weeks
Weight: 200-250 mg
Doses in administered units: 90 µ Ci/kg/day daily for 5 consecutive days

Group No.	n	Administered Materials	Nominal Dose (mg/kg)	Route of Administration	Measured Concentration (mg/ml) ^a	Dose Volume (ml/kg)
1	10	¹⁴ C-clofarabine and clofarabine	25	Intravenous	2.44	10

^a Measured concentrations are shown as the mean of Day 1 and Day 5 values.

Route, form, volume, and infusion rate: Slow intravenous (~3 minutes) injection into femoral vein.

Results:

Mortality: None
Clinical signs: Transient labored breathing

Average percent radioactivity recovered

Matrix	Total	% of Total Radioactivity Attributed to:	
		Clofarabine	Metabolites
Urine	77.1	87.2	12.8
Cage wash	6.0	---	---
Feces	10.8	6.9	93.1
Carcass	<1%	---	---
Total Recovered	95.3	76.0	21.7

Legend: --- denotes metabolic profiling not done.

(Excerpted from the sponsor's submission)

Maximum ¹⁴C-clofarabine concentrations in the blood and plasma were observed at 0.5 hours after the final dosing as shown below.

Mean concentration of total radioactivity in blood and plasma

Sacrifice Time	Mean Concentration (µg equiv/g)		
	QWBA-Derived	LSC-Derived	
	Blood	Blood	Plasma
Day 4, 24 hours	BLQ	0.112	0.100
Day 5, 0.5 hours	10.529	10.436	9.198
Day 5, 2 hours	3.703	3.097	2.682
Day 5, 6 hours	0.616	0.537	0.448
Day 5, 30 hours	BLQ	0.122	0.080

BLQ indicates below the limit of quantitation.

(Excerpted from the sponsor's submission)

Tissue distribution of total radioactivity

Tissue Type	Tissue	Concentration (µg equiv/g)				
		Time Point				
		Day 4, 24 hours	Day 5, 0.5 hours	Day 5, 2 hours	Day 5, 6 hours	Day 5, 30 hours
Dermal	Hair	23.713	49.083	38.812	24.672	22.717
	Skin	BLQ	18.786	11.063	1.522	BLQ
Gonads	Bulbo-urethral gland	0.637	15.762	7.428*	2.121	0.442*
	Epididymis	BLQ	11.212	4.007	0.765	BLQ
	Preputial gland	BLQ	10.570	0.443*	0.402	BLQ
	Prostate	0.958	15.089	7.245	2.011	1.429*
	Seminal vesicle	0.516	3.229	2.382	0.865	0.807
	Testes	BLQ	6.304	3.954	0.608	BLQ
Muscular	Diaphragm	0.756	22.373	10.473	3.413	0.552
	Muscle	0.553	17.970	7.121	1.758	0.354
	Myocardium	0.641	25.486	14.978	4.215	0.490
Unclassified	Bone	BLQ	4.260	1.220	0.378	BLQ
	Bone marrow	BLQ	30.238	8.247	1.166	BLQ
	Lung	BLQ	14.369	5.727	1.033	BLQ
	Nasal turbinates	BLQ	5.186	3.119	0.353	BLQ
	Pancreas	0.424	21.969	6.927	1.505	BLQ
	Spleen	0.502	123.215	44.185	4.702	0.383
	Trachea	0.399*	11.442	3.996	0.703	BLQ
	Urinary Bladder	12.750	253.850	174.907	32.689	5.247

Tissue Type	Tissue	Concentration (µg equiv/g)				
		Time Point				
		Day 4, 24 hours	Day 5, 0.5 hours	Day 5, 2 hours	Day 5, 6 hours	Day 5, 30 hours
Vascular/Lymphatic	Aorta	BLQ	30.980	12.294	2.069	0.378*
	Blood	BLQ	10.529	3.703	0.616	BLQ
	Lymph nodes	0.377	48.343	22.583	3.093	BLQ
Excretory/Metabolic	Kidney	0.737	82.336	21.596	3.155	0.389
	Liver	0.684	41.661	19.740	4.222	0.571
	Renal cortex	0.738	82.045	21.142	3.162	0.393
	Renal medulla	0.819	82.010	25.842	3.002	0.320
	Urine	27.773	439.812	240.837	111.206	12.967
CNS	Cerebellum	BLQ	0.824	0.446	BLQ	BLQ
	Cerebrum	BLQ	0.634	0.503	BLQ	BLQ
	Medulla	BLQ	0.307*	0.286*	BLQ	BLQ
	Spinal cord	BLQ	0.492	0.433	BLQ	BLQ
Endocrine	Adrenal gland	0.610	19.862	10.572	1.804	0.457
	Pituitary gland	BLQ	13.347	4.269	0.701	BLQ
	Thymus	0.452	61.589	27.648	3.638	0.382
	Thyroid	0.418*	20.985	8.080	1.451	0.326*
Secretory	Exorbital lacrimal gland	0.324*	17.777	6.565	1.211	BLQ
	Harderian gland	0.524	23.755	22.598	4.558	0.449
	Intra-orbital lacrimal gland	0.384*	17.444	7.667	2.046	BLQ
	Salivary gland (submaxillary)	0.327	20.969	8.618	1.883	0.306*
Fatty	Fat (abdominal)	BLQ	3.549	1.886	0.396	BLQ
	Fat (brown)	0.677	28.238	15.573	4.523	0.579

(Excerpted from the sponsor's submission)

Tissue distribution of total radioactivity

Tissue Type	Tissue	Concentration (µg equiv/g)				
		Time Point				
		Day 4, 24 hours	Day 5, 0.5 hours	Day 5, 2 hours	Day 5, 6 hours	Day 5, 30 hours
Alimentary Canal	Cecum	3.993	65.234	49.205	37.180	3.185
	Cecum contents	12.180	68.966	107.997	125.590	10.641
	Esophageal contents	0.831	10.396	3.936	9.340	0.636
	Esophagus	0.521	20.472	8.938	2.550	0.308
	Gastric mucosa	0.540	31.619	16.295	2.197	BLQ
	Large intestinal contents	23.017	111.772	94.636	169.555	18.237
	Large intestine	1.993	39.910	24.110	10.377	0.942
	Small intestinal contents	1.584	49.807	96.955	7.150	0.898*
	Small intestine	0.648	70.962	95.974	4.311	0.404*
	Stomach contents	4.664	6.758	9.845	1.350	BLQ
	Stomach	0.807	22.264	13.505	2.132	0.346
Ocular	Eye	BLQ	2.925	1.327	0.344	BLQ

*Data derived from one animal only.

BLQ indicates below the limit of quantitation

(Excerpted from the sponsor's submission)

The highest concentrations of radioactivity after 5 days 25 mg/kg/day intravenous administration of ¹⁴C-clofarabine were found in the urine (440 µg equiv/g), urinary bladder (254 µg equiv/g), spleen (123 µg equiv/g), kidney (82.3 µg equiv/g), cecum (65.2 µg equiv/g), and thymus (61.6 µg equiv/g). Low concentrations (approximately 7- to 34-fold less than blood) of ¹⁴C-clofarabine-derived radioactivity were detected in the tissues of the central nervous system through the 2-hour time point, but declined quickly and were not quantifiable at 6 hours postdose. After the final administration, radioactivity concentrations generally continued to decline, but the elimination was not complete by

post 30 hours administration. Not surprising was the high radioactivity in bladder and kidney as renal appears to be the route of elimination.

2.6.4.5 Metabolism: See under toxicology studies.

2.6.4.6 Excretion: See under toxicology studies.

2.6.4.7 Pharmacokinetic drug interactions: Not studied

2.6.4.8 Other Pharmacokinetic Studies

In Vitro Metabolism of Clofarabine in Rat, Dog, and Human Cryopreserved Hepatocytes. [] . Report No.: [] -2002-0542-BIO, []
Inc. Study No.: BA020050, and [] Project No.: DAA00710.100.

Key study findings:

- The amount of [¹⁴C] clofarabine remaining after 6-h incubation in rat, dog, and human cryopreserved hepatocyte cells were 96, 95, and 99.8%, respectively.
- Carboxy- or methoxy-clofarabine was the principal metabolite in rats and dogs.
- The only metabolite observed in human hepatocytes was clofarabine sulfate

Study no: [] Study No.: BA020050
Volume #, and page #:
Conducting laboratory and location: []
GLP compliance: Yes
QA report: yes () no (X)
Drug, lot #, radiolabel, and % purity: [¹⁴C] Clofarabine, lot CSL-01-142-60-33, []

Methods: ¹⁴C-Clofarabine (20 μM) and live cells (2 x 10⁶) were incubated in a 6-well plate. Incubations were terminated at 2 and 6 hours. Clofarabine and metabolite samples were analyzed by LC/MS.

Results:

Recovery of radioactivity from acetonitrile extraction of the hepatocyte samples

Sample ID	Pre-extraction dpm	Post extraction dpm	% Recovery
Rat T0	2179612	2013540	92
Rat T2	2240035	2091601	93
Rat T2	2243991	2105685	94
Rat T6	2207605	2099282	95
Rat T6	2214820	2079849	94
Rat neg	2236573	2143289	96
Dog T0	2075812	1980773	95
Dog T2	1837381	2026950	110
Dog T2	2234466	2083684	93
Dog T6	2166725	2043319	94
Dog T6	2203049	2018602	92
Dog neg	2181698	2051712	94
Human T0	2054672	1961200	95
Human T2	2157677	2042227	95
Human T2	2150615	2076103	97
Human T6	2109835	2031845	96
Human T6	2166370	2106062	97
Human neg	2170083	2085460	96

(Excerpted from the sponsor's submission)

Metabolite profiles of rat, dog and human hepatocytes

Sample	% Radioactivity in the Chromatogram										
	P1 ^a (12-14 min) ^b	P2 (16-17 min)	P3 (18 min)	P4 (18-21 min)	P5 (24 min)	P6 (26-26 min)	P7 (29 min)	P8 (30 min)	P9 (32 min)	P10 (38-39 min)	P11 (40 min)
Rat hep T0	ND ^c	0.20	ND	ND	ND	ND	ND	ND	ND	99.81	ND
Rat hep neg control	ND	0.19	ND	ND	ND	ND	ND	ND	ND	99.81	ND
Rat hep T2 ^d	ND	0.33	0.54	0.17	0.57	0.15	0.18	0.15	0.20	97.54	0.20
Rat hep T6 ^d	ND	0.36	0.46	0.39	0.70	1.22	0.87	0.11	0.53	95.76	0.23
Dog hep T0	ND	ND	ND	ND	ND	ND	ND	ND	ND	100.00	ND
Dog hep neg control	ND	0.21	ND	ND	ND	ND	ND	ND	ND	99.79	ND
Dog hep T2 ^d	0.19	0.27	ND	0.21	0.33	0.78	0.56	ND	0.23	97.90	ND
Dog hep T6 ^d	0.37	0.54	ND	0.62	ND	2.51	0.81	ND	0.42	94.75	ND
Human hep T0	ND	ND	ND	ND	ND	ND	ND	ND	ND	100.00	ND
Human hep neg control	ND	ND	ND	ND	ND	ND	ND	ND	ND	100.00	ND
Human hep T2 ^d	ND	ND	ND	ND	ND	ND	ND	ND	ND	100.00	ND
Human hep T6 ^d	ND	ND	ND	ND	ND	ND	ND	ND	0.18	99.82	ND

- a. Radioactive peak region.
- b. Elution time.
- c. ND: not detected.
- d. Mean of n = 2.

(Excerpted from the sponsor's submission)

Summary of the molecular masses

Regions	<i>m/z</i>	Description
P1	ND ^a	Unknown
P2	390 (rat) 170 (rat and dog)	Unknown 2-Chloroadenine
P3	343 (rat)	Unknown
P4	392 (rat)	Unknown
P5	ND	Unknown
P6	318 (dog)	Carboxy- or methoxy-clofarabine
P7	480 (rat and dog)	Clofarabine-glucuronide
P8	480 (rat)	Clofarabine-glucuronide
P9	384 (rat, dog and human)	Clofarabine-sulfate
P10	304	Clofarabine
P11	ND	Unknown

a. ND: not determined.

(Excerpted from the sponsor's submission)

2.6.4.9 Discussion and Conclusions: The pharmacokinetics of clofarabine has been studied *in vivo* in mice, rats, and dogs after single and multiple dosing. The highest doses used in 6-months toxicity studied in rats and dogs were in the same range as doses being evaluated in pediatric patients with refractory or relapsed ALL and AML. Using a five day intravenous dose regimen of radiolabeled clofarabine, 77%, 11%, 6%, and <1% of the administered dose to rats was found in the urine, feces, cage wash, and carcass, respectively. In the mass balance study in rats, 6-keto-clofarabine was the major metabolite in plasma, urine, feces, heart, and liver. The enzyme responsible for this metabolism is unknown. The effect of clofarabine on the metabolism of cytochrome p450 substrates has not been studied. Clofarabine sulfate was observed after 6-hour incubation of cryopreserved hepatocytes of rat, dog and human. In separate studies, clofarabine protein binding has been reported to be low in rat plasma (~13%) as compared to human plasma (~47%) and human serum albumin (~27%) [Cancer Chemother Pharmacol 1994; 33:484-488 & J Liq Chromatogr 1995; 18(6):1123-1135].

2.6.4.10 Tables and figures to include comparative TK summary

Comparison of Toxicokinetics data

Species (dose)		Rat (75 mg/m ²)		Dog (30 mg/m ²)		Human (52 mg/m ²)	
Parameter	Unit	Day 1	Day 145	Day 5	Day 145	Day 1	Day 5
C _{max}	ng/mL	2627	6262	1238	1125	403	559
AUC*	ng.h/mL	7489	10912	2942	2586	1223	1757
Half-life	hours	1.7	1.7	1.8	1.8	6.2	6.2

*AUC_{0-8h} for human,AUC_{0-10h} for ratAUC_{0-12h} for dog

The area under the plasma concentration-time curve increase in a dose-dependent manner in rats and dogs. The elimination half-life in rats, dogs, and humans did not change after repeated dosing. However, C_{max} and AUC values increased after repeated dosing in rats and humans indicating accumulation of clofarabine. The PK values for dogs on day 1 are not provided. In conclusion, the PK of clofarabine has been reasonably characterized in rats and dogs and can be compared with humans.

2.6.5 PHARMACOKINETICS TABULATED SUMMARY: See 2.6.4.10 above.

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2.6.6 TOXICOLOGY

2.6.6.1 Overall toxicology summary:

General toxicology:

A Multi-Cycle (Five Days' Dosing Plus 23 Days' Recovery per Cycle) Intravenous Toxicity and Toxicokinetics Study in Fischer 344 Rats. Male and female rats were divided into three phases and treated up to six months on multi-cycle basis to evaluate the toxicity and toxicokinetics of clofarabine. Each complete cycle consisted of daily administration of the drug for 5 consecutive days followed by a 23-day treatment-free period. The dose levels of clofarabine investigated in this study were 6.25, 12.5 and 25 mg/kg/day by slow bolus (30-60 seconds) intravenous (tail vein) injection. The phase A rats were treated for 2 complete cycles of treatment and wash-out/observation. At the end of the second cycle, they were dosed for an additional 5 days and euthanized on day 65. The Phase B animals were subjected to 5 complete cycles of treatment and wash-out/observation. At the end of the fifth cycle, they were dosed for an additional 5 days and sacrificed on day 149. The Phase C rats were treated for six, 5-day treatment periods and euthanized on day 163.

Piloerection, eyes partially or fully closed, absent or small amount of feces, salivation, decreased activity, cold to touch, tremors, hunched back, righting reflex slow, labored/irregular respiration were observed during the dosing period. Heavy mortality among high dose animals (100 % HD • ; 80 % HD •) at the end of third period of dosing suggests the development of a cumulative adverse effect of clofarabine. Food intake and body weight gain were reduced in a dose-dependent manner in male rats. Food consumption did not track changes in body weight as females decreased food consumption without changes in body weight. Histopathological examination revealed decreased cellularity in the lymphoid tissues (lymph nodes, spleen, thymus and lymphoid tissue of small intestine), apoptotic bodies in the crypts in the small intestine, and retained spermatids in the testes. Myocardial degeneration/necrosis, inflammation, and fibrosis were also observed in high dose animals. In conclusion, most of the preterminal deaths were due to cardiac lesions. The C_{max} and AUC_{0-10} values on day 145 increased by approximately 3 and 1.5 folds, respectively, as compared to day 1 values. These values were higher in females than males. There were no clear changes in half-life ($T_{1/2}$) after multiple dosing.

A Multi-Cycle (Five Days Dosing Plus 23 Days Recovery Per Cycle) Intravenous Toxicity and Toxicokinetics Study in Beagle Dogs: Male and female Beagle dogs were divided into three phases and treated with clofarabine for six months on multi-cycle basis. Each complete cycle consisted of daily administration of the drug for 5 consecutive days followed by a 23-day treatment-free period. The clofarabine doses initially administered were 0.75, 1.5 and 3.0 mg/kg/day. Due to severe toxicity (multiple mucosal hemorrhages throughout the small and large intestines) at 3.0 mg/kg/day, the animals were reassigned and the study continued at dose levels of 0.375, 0.75 and 1.5 mg/kg/day.

The phase A dogs were treated for 2 complete cycles of treatment and wash-out/observation. At the end of the second cycle, they were dosed for an additional 5 days and euthanized on day 65. The Phase B dogs were subjected to 5 complete cycles of treatment and wash-out/observation. At the end of the fifth cycle, they were dosed for an additional 5 days and sacrificed on day 149. The Phase C dogs were treated for six, 5-day treatment periods and euthanized on day 163.

The animals at 3 mg/kg/day have vomiting, diarrhea, dehydration, weight loss, hypothermia and reduced motor activity. All high dose animals (3 mg/kg/day) either died or became moribund and euthanized. Four animals at 1.5 mg/kg/day became moribund and sacrificed. A cytotoxic effect of clofarabine on the gastrointestinal tract was supported by histopathological findings in these animals. There was no mortality among the low dose (0.375 mg/kg/day) or mid dose (0.75 mg/kg/day) dogs.

Leukocytopenia was the principal hematological effect of clofarabine. Histopathological findings in the large intestine of pre-terminal sacrificed animals included mucosal congestion and hemorrhage, apoptotic necrosis, regenerative hyperplasia, occasional ulceration or erosion and mucosal inflammation with dilatation of glands. Small intestine changes included mucosal inflammation (with crypt dilatation), congestion and hemorrhage, apoptotic necrosis, and regenerative hyperplasia. Testes (multinucleated spermatids, focal, minimal) and liver inflammation were also seen in mid- and high-dose animals. Values for C_{max} and AUC_{0-12} demonstrated dose-proportionality and there were no gender differences and no accumulation over successive treatment periods in toxicokinetic findings. The C_{max} and AUC_{0-12} values on the first day of successive 5-day treatment period are not presented. Therefore, we cannot compare the PK values on days 1 and 5. In conclusion, gastrointestinal tract may be dose-limiting organ of toxicity of clofarabine in dogs.

A comparison of the C_{max} and AUC determined for rats and dogs in their respective 6-month toxicity studies is presented below.

Dose	Rat	Dog	Comment
MTD mg/kg/day:	12.5	0.75	Rat = 17X greater
mg/m ² /day:	75.0	15	Rat = 5X greater
C_{max} (ng/mL): ¹	6,114	563	Rat = ~11X greater
AUC(ng*h/mL):	10,585	1,484	Rat = ~7X greater

¹Male animals on Day 145

(Excerpted from the sponsor's submission)

Gastrointestinal toxicity in dogs did not allow higher administration of clofarabine in dogs as compared to rats. Lower systemic exposure and/or C_{max} in dogs when compared to rats may explain the cardiotoxicity seen in rats and not in dogs.

Genetic toxicology: Clofarabine showed clastogenic activity in the *in vitro* mammalian cell chromosome aberration assay (CHO cells) and in the *in vivo* rat micronucleus assay. It did not show mutagenic activity in the bacterial mutation assay (Ames test). The clastogenic findings in the *in vitro* CHO cells and in the *in vivo* rat micronucleus assay were expected based upon the mechanism of anticancer activity for clofarabine.

Carcinogenicity: Studies not conducted

Reproductive toxicology: The effects of clofarabine on fertility and early embryonic development to implantation (before mating through mating and implantation) and perinatal and postnatal development, including maternal function (implantation through weaning) have not been studied in animals.

Intravenous Developmental Toxicity Study of Clofarabine in Rats: Female rats were dosed with 0 (vehicle), 1, 3, and 9 mg/kg/day clofarabine by slow bolus intravenous administration from DG 7 to 17. The dosages were selected based on a dose-range finding study. The no observable adverse effect level (NOAEL) and lowest observable adverse effect level (LOAEL) for the pregnant female rats were 1 and 3 mg/kg/day, respectively. The 9 mg/kg/day dosage of clofarabine was associated with reduced food consumption and body weight gain. Postimplantation loss in high dose animals may be an effect of clofarabine. Fetal gross external and soft tissue malformations in high dose animals included short trunk, tail malformations, digit malformations, whole body edema (anasarca), limb defects, thin transparent skin, and alterations in urogenital development. Variation in brain development (slight dilation of the lateral or third ventricle), vessel formation (presence of the umbilical artery to the left of the urinary bladder) and kidney development (moderate dilation of the renal pelvis) were also attributed to the drug. Skeletal examination showed multiple malformations of the axial skeleton, sternum, and cervical vertebrae. Similar growth retardation, tail malformation and fetal skeletal alteration had been shown by cyclophosphamide (Cancer Chemother Rep 1967; 51:363-376). In summary, teratogenic effects of clofarabine in the rat are demonstrated in this study.

Intravenous Developmental Toxicity Study of Clofarabine in Rabbits. Intravenous administration of clofarabine at 0.1, 0.3, and 1.0 mg/kg/day to NZW rabbits from DG 6 to 18 did not cause any mortality. The dosages selected for this study were based on the dosage range-finding study. Maternal body weight and food consumption were unaffected by clofarabine up to 1.0 mg/kg/day. There were a decrease in fetal body weight and an increase in postimplantation losses in high dose animals. Fetal gross external malformations attributable to the 1.0 mg/kg/day dosage of clofarabine included: craniofacial malformations, limb defects, digit malformations, short trunk, tail malformations, defects of abdominal wall closure and spinal defects. Multiple soft tissue malformations included hydrocephaly, displaced adrenals and alterations in

cardiovascular, lung, urogenital and hepatic development. Variations in brain development (great vessel formation) and lung development were also observed at 1.0 mg/kg/day dosage of clofarabine.

Skeletal examination of the 1.0 mg/kg/day dosage group fetuses revealed multiple malformations of the skull, axial and appendicular skeleton, as well as variations of the skull, hyoid, pectoral and pelvic girdles, ribs and sternum. The NOAEL dose of clofarabine for developmental toxicity in rabbit was 0.1 mg/kg/day.

Similar adverse embryotoxic and teratogenic effects have been reported for other nucleoside analogues, such as cladribine (Teratology 2002; 66:6-18), 5-Aza-2'-deoxycytidine (Teratology 2002; 65:180-90), and gemcitabine (Teratology 1993; 48:365-81).

Special toxicology: Not studied

In conclusion, based upon its cellular activity, clofarabine showed adverse effect in general toxicity (damage to proliferating tissues such as bone marrow, lymphoid tissue and gastrointestinal tract), genetic toxicity (clastogenic), and developmental toxicity (teratogenic) in rats and rabbits.

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2.6.6.3 Repeat-dose toxicity

Study title: Clofarabine: A Multi-Cycle (Five Days' Dosing Plus 23 Days' Recovery per Cycle) Intravenous Toxicity and Toxicokinetics Study in Fischer 344 Rats.

Key study findings:

- Multi-cycle administration (d x 5 q 28 d for 5 cycles) of IV clofarabine at 25 mg/kg/day produced 100 % (Days 87-141) and 80 % (Days 32-116) mortalities in male and female animals, respectively.
- Liver, kidneys, eye and especially the heart were the main organ of toxicity, primarily based on histopathology.
- Clinical chemistry evaluation was unremarkable.
- Hematology: WBC more affected than RBC, bone marrow histopathology generally unremarkable.

Study no.: 0204- 03 (- 5471)
Volume #, and page #: Module 5.4.11.4
Conducting laboratory and location: []
Date of study initiation: 4/4/02
GLP compliance: Yes
QA report: yes (x) no ()
Drug, lot #, and % purity: clofarabine; lot # 010203; — % (HPLC)

Methods

Doses: 6.25, 12.5, 25 mg/kg/d
Schedule:
 Phase A: d x 5 q 28 d for 2 cycles; d x 5, then 3 days off (64 d total)
 Phase B: d x 5 q 28 d for 5 cycles; d x 5, then 3 days off (148 d total)
 Phase C: d x 5 q 28 d for 5 cycles; d x 5, then 17 days off (163 d total)
Species/strain: Fischer 344 rat
Number/sex/group or time point (main study): 5/sex
 Note that all available animals were included in the evaluation; e.g., except for decedents, till d 64, n = 15 for body wt evaluations for all dose groups
Route: iv
Formulation: 25% PEG 400 in 0.9% saline
Volume: 10 mL/kg
Infusion rate: slow bolus (30-60 s)
Satellite groups used for toxicokinetics or recovery: TK: 15/sex (no control); Recovery, see Phase C above
Age: 10 weeks • and 12-16 weeks • •
Weight: 200 g • and 175 g • •
Unique study design or methodology: Cardiac troponin I was measured by

[

] assay [

] Troponin I measurements were performed at [

]

Observation (times)	Results
Mortality (daily)	<ul style="list-style-type: none"> • • 10/10 HD D87-141 • • 1/10 LD D95; 1/10 MD D78; 8/10 HD D32-116
Clinical signs (2x/d during dosing, daily off dosing)	<p>MD • death attributed to sampling</p> <p>Control: eyes partially or fully closed*; absent or small amount of feces; dry brown and/or beige material on the face*</p> <p>LD: above; clear or red/pink lacrimation; thin body condition, skin turgor slow; fur ungroomed, matted; piloerection; hunched back; walking on tip toes</p> <p>MD: above; salivation</p> <p>HD: above; decreased activity, red conjunctiva; cold to touch, pale in color; animal lying on its cage floor; tremors; righting reflex slow; gasping and/or increased and/or decreased and/or shallow and/or labored/irregular respiration</p> <p>* most commonly reported events</p>
Body weights (pretreatment, D1, 3, 5 of dosing, wkly during non dosing period)	<ul style="list-style-type: none"> • • HD sig ↓ starting cycle 3; some recovery during the non dosing phase but still sig ↓ vs C (range 9-24%); MD sig ↓ starting cycle 4; LD sig ↓ cycle 6; see graph; LD and MD similar to C during the recovery period (no HD animals) • • unremarkable <div style="text-align: center;"> <p>The graph plots Body Weights (g) on the y-axis (150 to 350) against Day on the x-axis (0 to 150). Four data series are shown: Control (solid line), Low dose (dashed line), Mid dose (dotted line), and High dose (dash-dot line). All groups start at approximately 200g. The Control group reaches ~300g by day 150. The Low dose group reaches ~280g. The Mid dose group reaches ~260g. The High dose group reaches ~240g. All groups show a sharp decline in weight around day 60, followed by a recovery phase.</p> </div>
Food consumption (pretreatment, D1, 3, 5 of dosing, wkly off dosing)	<p>Dose dependent ↓ • • • MD and HD during treatment period; generally recovered to control during non dosing periods; average changes (range %) for all dosing cycles were:</p> <ul style="list-style-type: none"> • • MD 17% (3-29); HD 43% (20-69) • • MD 15% (8-28); HD 28% (18-34)
Ophthalmoscopy (pretreatment, week before necropsy for phase B, prior to recovery phase C)	unremarkable
EKG (not conducted)	n/a

Hematology (unfasted D 8, 22, 64, 78, 148, 162)	<p>D8, 64, 148*</p> <p>WBC: • ; • ↓ HD ~40-50%, MD 20-40%; all lineages affected to some degree but particularly eosinophils and lymphocytes</p> <p>RBC: D64, 148 ↓ of 5-10% in • ; • ; D8, 64, 148 reticulocyte% (and absolute counts) were ↓ 70-80% v C, generally in HD both sexes, with MD occasionally affected.</p> <p>D22, 78, 162**</p> <p>Some recovery in all parameters but still ↓ v C</p>
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3 days following the end of the 1st, 3rd and 6th 5-day dosing period*

** nondosing period

Coagulation/Troponin I (D65, 149, 163)	Coagulation unremarkable Troponin I – below the limit of quantitation (0.2 ng/mL)
Clinical chemistry (unfasted D8, 22, 64, 78, 148, 162)	unremarkable
Urinalysis (not done)	n/a
Organ weights (D65, 149, 163)	<p>D65</p> <p>Spleen: ↓ HD • abs (40%) and rel wt (35%) vs C ↓ HD • abs (62%) and rel wt (58%) vs C</p> <p>D149</p> <p>Ovaries: ↓ HD abs (39%) and rel wt (39%) vs C</p> <p>Heart: ↑ MD and LD • (no HD •) rel wt (10 and 25%, respectively; not dose related) ↑ HD • abs (41%) and rel (43%) vs C</p> <p>Other organ wt findings not consistent between genders, were associated with bw loss, or not temporally consistent. Heart wts were still elevated after recovery period.</p>

Gross pathology (D65, 149, 163)

Tissue	Decedents		D65		Recovery	
	• •	• •	• •	• •	• •	• •
Spleen: small	6/10 HD	4/8 HD		3/4 HD		
Thymus: small	8/10 HD	4/8 HD	5/5 HD 3/5 MD	4/4 HD 2/5 MD 1/5 LD		
emaciated				4/4 HD		
Epididymides: small	3/10 HD					
Heart: myocardial fibrosis brown pigment			5/5 HD	2/4 HD		
Liver: pale	5/10 HD	4/8 HD				
Prostate: small	5/10 HD					
Seminal vesicles	5/10 HD					
Stomach: dark foci/material	4/10 HD	3/8 HD				
Thoracic cavity: fluid	8/10 HD	3/8 HD				1/1 HD

Terminal D149 unremarkable

Histopathology	C, HD, decedents, target organs all doses
Fasted: Days 65, 149 and 163 for the relevant animals, laboratory investigations (coagulation)	

Histopathology: Adequate Battery: yes (X), no ()—explain
 Peer review: yes (), no (X)

Preterminal sacrifice and decedents:

High dose (12.5 mg/kg) – Adrenal glands (vacuolation, congestion), bone marrow sternum (degeneration/necrosis, decreased cellularity), epididymides (depletion, necrosis), eye (cornea mineralization, atrophy), femur, marrow and joint (degeneration/necrosis, decreased cellularity), heart (fibrosis, hypertrophy, degeneration/necrosis), kidneys (glomerulonephropathy, mineralization), liver (eosinophilia, hypertrophy, hyperplasia, degeneration/necrosis), lung (macrophage, congestion), lymph node (decreased cellularity, necrosis), mammary gland (necrosis), ovaries (atrophy, depletion), pancreas (edema, hypertrophy), prostate (edema, atrophy), Jejunum & ileum (apoptotic bodies), spleen (decreased cellularity, necrosis), testes (seminiferous degeneration, atrophy), thymus (necrosis, hemorrhage, decreased cellularity).

Terminal Sacrifice, Phase C

Sex Dosage (mg/kg) Number of animals	Male		Female		
	6.25	12.5	6.25	12.5	25.0
	5	5	5	5	1
Tissue / observation					
Adrenal glands, vacuolation, zona fasciculata, minimal	2	2			1
Bone & marrow, sternum, increased cellularity, slight					1
Eye, mineralization, cornea, focal, minimal	3	4	3	5	1
Heart, degeneration/necrosis, myocardial, minimal dilatation, ventricles, moderate	0	2	1	1	1
Kidneys, basophilic tubules, focal, minimal	0	2	1	1	0
Liver, eosinophilia, hepatocellular, centrilobular degeneration/necrosis, hepatocellular, single cell hypertrophy, hepatocellular, diffuse	5 5 5	5 5 5	1 1 5	3 5	1 1
Lung, decreased cellularity, cortex, pigment, brown, macrophages, minimal	0 0	2 2			
Lymph node, mesenteric, increased cellularity, macrophages, sinus	1	2			1
Ovaries, depletion, corpora lutea, moderate				1	1
Spleen, decreased cellularity, marginal zone, marked				0	1
Thymus, hemorrhage, congestion, focal	4	4			

Toxicokinetics: D1 & D145 at 0, 5 min, 1, 2, 6 and 10 h (n = 3);

Summary of Clofarabine Toxicokinetic Parameters Following Multiple Intravenous Doses to Rats

Parameter	Dose Group (µg/kg)					
	6.25		12.5		25.0	
	Day 1	Day 145	Day 1	Day 145	Day 1	Day 145
Male Rats						
C _{max} , ng/mL	1277.4	3687.6	2347.5	6114.2	6721.7	NC
AUC(0-10), ng·h/mL	2991.0	4395.8	6585.2	10585.1	13227.1	NC
T _{1/2} , h	1.7	1.7	1.6	1.6	1.9	NC
Female Rats						
C _{max} , ng/mL	1476.7	4020.7	2905.3	6409.3	14978.5	NC
AUC(0-10), ng·h/mL	3517.8	6026.7	8393.0	11239.7	22682.3	NC
T _{1/2} , h	1.9	1.9	1.8	1.8	1.7	NC

NC - Not calculated due to insufficient plasma concentrations

(Excerpted from the sponsor's submission)

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Study title: **Clofarabine: A Multi-Cycle (Five Days' Dosing Plus 23 Days' Recovery per Cycle) Intravenous Toxicity and Toxicokinetics Study in Beagle Dogs.**

Key study findings:

- Six-month, multi-cycle administration of clofarabine to Beagle dogs produced cytotoxic effect on gastrointestinal tract of high (3.0 mg/kg/day) and mid (1.5 mg/kg/day) dose animals.
- Bone marrow, eye, lymphoid tissue, and testes were affected.
- The C_{max} and AUC_{0-12} values increased in a dose-dependent manner and there were no apparent gender differences and no-accumulation of drug over successive cycles.

Study no.: Study No. 2758, ILEX Study No. 0203-01.
Volume #, and page #: Electronic Module
Conducting laboratory and location:

Date of study initiation: April 8, 2002.
GLP compliance: Yes
QA report: yes (X) no ()
Drug, lot #, and % purity: Clofarabine, lot # 010203, — % purity

Methods

Doses: Dogs were originally to be treated as shown below.

Treatment Groups	Clofarabine Dose Level (mg/kg/h)	Dose Rate (mL/kg/h)	Clofarabine Concentration (mg/mL)	Number of Animals					
				Phase A		Phase B		Phase C	
				M	F	M	F	M	F
1. Control*	0	1	0	2	2	2	2	2	2
2. Low Dose	0.75	1	0.75	2	2	2	2	2	2
3. Mid Dose	1.5	1	1.5	2	2	2	2	2	2
4. High Dose	3.0	1	3.0	2	2	2	2	2	2

M: Males F: Females

(Excerpted from the sponsor's submission)

Schedule:

Phase A: d x 5 q 28 d for 2 cycles; d x 5, then 3 days off (total of 3, 5-day treatment periods; 64 days total)

Phase B: d x 5 q 28 d for 5 cycles; d x 5, then 3 days off (total of 6, 5 day treatment periods; 148 days total)

Phase C: d x 5 q 28 d for 5 cycles; d x 5, then 14 days off (total of 6, 5 day treatment periods; 162 days total)

Due to severe toxicity (mucosal hemorrhage, inappetence, reduced activity, diarrhea and dehydration) during first treatment cycle in high dose Phase C animals, the study was reorganized as shown below.

Treatment Groups	Clofarabine Dose Level (mg/kg/h)	Dose Rate (mL/kg/h)	Clofarabine Concentration (mg/mL)	Number of Animals					
				Phase A		Phase B		Phase C	
				M	F	M	F	M	F
1. Control*	0	1	0	0	0	3	3	3	3
5. Low Dose	0.375	1	0.375	0	0	2	2	2	2
2. Mid Dose	0.75	1	0.75	2	2	2	2	2	2
3. High Dose	1.5	1	1.5	0	0	3	3	3	3

M: Males; F: Females
(Excerpted from the sponsor's submission)

Schedule: Phases A, B, and C as given above.

Species/strain: Beagle dogs
 Number/sex/group or time point (main study): see table above.
 Route, formulation, volume, and infusion rate: Intravenous
 Satellite groups used for toxicokinetics or recovery: No
 Age: 7-8 months
 Weight: • 6.7-10.4 kg, • 5.7-9.4 kg

Observations (times) and Results

Observation (times)	Results
Mortality (daily)	•• G3 (1.5 mg/kg) – 3 D 7-8 G4 (3 mg/kg) – 4 D 5-8 •• G3 (1.5 mg/kg) – 1 D 8 G4 (3 mg/kg) – 4 D 5-7
Clinical signs (2x/d during dosing, daily off dosing)	Small amount of feces, no feces, vomiting, excessive salivation, decreased activity, cold to touch, weakness and partial recovery during wash out period in a dose-related manner
Body weights (pretreatment, D1, 3, 5 of dosing, wkly off dosing)	HD (1.50 mg/kg) – • 13% ↓, Reduced appetite and food intake.
Food consumption (daily during 1-week pretreatment and through treatment)	MD (0.75 mg/kg) – • 52% ↓, • 36% ↓ HD (1.50 mg/kg) – • 80% ↓, • 43% ↓
Ophthalmoscopy (pretreatment, week before necropsy)	MD (0.75 mg/kg) – 1 • & 1 • & HD (1.50 mg/kg) – 1 • & 1 • had superficial focal white corneal deposits. Minor corneal changes in a dose-unrelated manner.
EKG (pretreatment, final week of cycle 3 and week before necropsy)	No clinical significant changes
Hematology (unfasted D 8, 22, 64, 78, 148, 162)	HD (1.50 mg/kg) – leukocytes (• 43% ↑), erythrocytes (• 19% ↓), platelets (• 69% ↑, • 167% ↑)

Observation (times)	Results
Clinical chemistry (unfasted pretreatment and D 8, 22, 64, 78, 148, 162)	No effect
Urinalysis (not done)	n/a
Organ weights	No effect on organ weights
Gross pathology (D65, 149 163)	No macroscopic effect in terminal sacrificed animals. Discoloration in adrenal gland, gall bladder, large and small intestine, spleen, and stomach in pre-terminally euthanized animals.

Histopathology: Adequate Battery: yes (X), no ()

Peer review: yes (), no (X)

Pre-terminal sacrifice and decedent

	• (mg/kg/d)		• (mg/kg/d)	
	1.5	3.0	1.5	3.0
Number of animals / Finding	3	4	1	4
Adrenal glands, depletion, cytoplasmic vacuolation, minimal slight hypertrophy	1	0	0	1
	2	2	1	0
	0	2		
Bone marrow, congestion, slight, moderate decreased cellularity, granulocytic series, marked severe	3	2	0	3
	0	2	1	1
	1	2		
	0	2	1	2
Epididymides, depletion, sperm, bilateral, minimal severe degenerate germ cells, minimum slight	1	0		
	2	3		
	0	2		
	1	0		
Eyes, mitotic index, increased, minimum	0	2		
Injection sites, thrombosis, minimal	0	1	1	2
Kidneys, vacuolation, proximal tubule, minimal mineralization, inner medulla, focal, minimal	1	1		
			0	1
Large intestine, colon, hemorrhage, minimal, moderate congestion, slight degeneration, marked hyperplasia, regenerative, slight inflammation, mucosal, slight	2	2	1	0
	0	1	1	1
	0	2	0	3
	0	1	1	2
	1	2	1	0
	0	1	0	1
	0	1	0	1
Large intestine, cecum, congestion, minimal, moderate necrosis, apoptotic, glands, minimal slight	2	1	0	1
	0	1	0	1
	2	1	1	2
	0	2		
Large intestine, rectum, hemorrhage, moderate necrosis, apoptotic, glands, slight	0	1	1	2
	0	1	0	4
Liver, hyperplasia, bile duct, minimal	0	2		

	• (mg/kg/d)		• (mg/kg/d)	
	1.5	3.0	1.5	3.0
Number of animals / Finding	3	4	1	4
congestion, slight	0	1	1	1
Lungs, congestion, minimal	0	1	0	1
Lymph node, mandibular, decrease cellularity, cortex, marked	0	2	0	2
Lymph node, mesenteric, decreased cellularity, cortex, moderate	0	2	1	2
Oesophagus, depletion, secretion, mucous gland, marked	0	2	0	2
Ovaries, parovarian cyst, unilateral			0	1
Pituitary gland, cyst, present	1	2	0	1
Prostate gland, necrosis, single cell, minimal	0	2		
Small intestine, duodenum, hemorrhage, focal, slight	0	2	0	3
congestion, slight	0	1	0	2
hyperplasia, regenerative, moderate	2	1	1	0
atrophy/fusion, moderate	0	1	1	3
Small intestine, jejunum, congestion, minimal	3	2	1	1
degeneration/cell debris, moderate	0	2	0	1
Small intestine, ileum, congestion, moderate	0	1	1	2
Spleen, decreased cellularity, white, slight	2	2	1	2
Stomach, hemorrhage, focal, minimal	2	4		1
Testes, incomplete maturation	1	3		
Thymus, , decreased cellularity, cortex, severe	0	2	1	2
Urinary bladder, necrosis, single cell, epithelium, slight			0	1
Vagina, congestion, minimal			1	0

Terminal Sacrifice, Phase C

	• (mg/kg/day)			• (mg/kg/day)		
	0.375	0.75	1.5	0.375	0.75	1.5
Number of animals / Finding	2	2	3	2	2	3
Epididymides, degenerate germ cells, slight	0	0	2			
Injection sight, inflammation, dermal, focal, min.		1	1			1
Kidneys, mineralization, inner medulla, focal, min.				1	2	3
Liver, inflammation, mix cell, focal, minimal		1	2			
Lungs, fibrosis, pleural, focal, minimal			1			
macrophages, alveolar, foamy, minimal						1
Spleen, pigment deposition, brown, minimal		1	1			
Stomach, hemorrhage, focal, minimal			1			
Testes, multinucleated spermatids, focal, minimal		1	1			

Toxicokinetics:

Phase A:

Days 5 and 61

Phases B & C:

Days 5, (1 cycle), 61 (3 cycles)
and 145 (6 cycles)

at 0, 1, 2, 4, 8, 12 h, and 24 h

Summary Clofarabine Toxicokinetic Parameters Following Multiple Intravenous Infusions to Beagle Dogs

Parameter	Dose (mg/kg)							
	0.375		0.75		1.5		3.0	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Day 5								
n	8	-	12	-	12	-	6	-
C _{max} , ng/mL	300.9	40.1	621.0	64.6	1237.9	142.5	2348.5	205.7
AUC(0-12), ng·h/mL	701.2	129.3	1622.3	366.4	2941.6	510.3	5326.9	791.4
T _{1/2} ^a , h	1.7 ^d	0.1	1.8 ^b	0.4	1.8	0.3	1.8	0.2
Day 61								
n	8	-	12	-	8	-	-	-
C _{max} , ng/mL	239.9	17.2	588.6	55.4	1240.0	57.8	NC	NC
AUC(0-12), ng·h/mL	603.0	109.7	1456.7	259.8	2927.3	273.1	NC	NC
T _{1/2} ^a , h	1.7 ^e	0.1	1.8 ^b	0.5	2.0	0.2	NC	NC
Day 145								
n	8	-	8	-	8	-	-	-
C _{max} , ng/mL	298.8	44.9	562.9	84.5	1125.2	98.9	NC	NC
AUC(0-12), ng·h/mL	715.7	146.9	1338.1	320.9	2586.2	299.2	NC	NC
T _{1/2} ^a , h	1.6 ^e	0.1	1.9 ^c	0.3	1.8	0.3	NC	NC

^a Expressed as harmonic mean and pseudo SD based on jackknife variance

^b n=11

^c n=7

^d n=5

^e n=4

NC - Not calculated

(Excerpted from the sponsor's submission)

There were no apparent gender differences in the toxicokinetics.

Mean Clofarabine Pharmacokinetic Parameters by Dose
(from Supplemental Toxicokinetics Report: MC02111-A1)

Parameter	Dose	Units	Mean	Std Dev	n	Minimum	Maximum
C _{max}	0.375 mg/kg	ng/mL	279.9	45.0	24		
	0.75 mg/kg		594.3	68.7	32		
	1.5 mg/kg		1206.3	120.0	28		
	3.0 mg/kg		2348.5	205.7	6		
AUC(0-12)	0.375 mg/kg	ng·h/mL	769.0	138.4	12		
	0.75 mg/kg		1575.2	296.7	29		
	1.5 mg/kg		2874.5	437.0	28		
	3.0 mg/kg		5393.8	821.9	6		
CL _{ss}	0.375 mg/kg	mL/h/kg	502.7	92.5	12		
	0.75 mg/kg		491.9	89.6	29		
	1.5 mg/kg		534.9	91.9	28		
	3.0 mg/kg		567.3	87.9	6		
Vd _{ss}	0.375 mg/kg	mL/kg	1182.2	285.6	12		
	0.75 mg/kg		1112.2	166.1	29		
	1.5 mg/kg		1112.9	134.2	28		
	3.0 mg/kg		1127.6	82.6	6		
Half-life	0.375 mg/kg	hours	1.7	0.1	12		
	0.75 mg/kg		1.9	0.4	29		
	1.5 mg/kg		1.9	0.3	28		
	3.0 mg/kg		1.8	0.2	6		

(Excerpted from the sponsor's submission)

Histopathology inventory

Study	0203-01	
	rat	dog
Adrenals	X*	X*
Aorta	X	X
Bone Marrow smear	X	X
Bone (femur)	X	X
Brain	X*	X*
Cecum	X	X
Cervix	X	
Colon	X	X
Duodenum	X	X
Epididymis	X	X
Esophagus	X	X
Eye	X	X
Fallopian tube		
Gall bladder		X
Gross lesions	X	
Harderian gland		
Heart	X*	X*
Ileum	X	X
Injection site	X	X
Jejunum	X	X
Kidneys	X*	X*
Lachrymal gland		
Larynx		
Liver	X*	X*
Lungs	X	X
Lymph nodes, cervical		
Lymph nodes mandibular	X	X
Lymph nodes, mesenteric	X	X
Mammary Gland	X	X
Nasal cavity		
Optic nerves	X	X
Ovaries	X*	X*
Pancreas	X	X
Parathyroid	X	X
Peripheral nerve		
Pharynx		
Pituitary	X	X
Prostate	X	X
Rectum		
Salivary gland	X	X
Sciatic nerve	X	X
Seminal vesicles	X	
Skeletal muscle	X	X

Study	0203-01	
	rat	dog
Species	rat	dog
Skin	X	X
Spinal cord	X	X
Spleen	X*	X*
Sternum	X	X
Stomach	X	X
Testes	X*	X*
Thymus	X	X
Thyroid	X	X
Tongue	X	X
Trachea	X	X
Urinary bladder	X	X
Uterus	X	X
Vagina	X	X
Zymbal gland		

X, histopathology performed; *, organ weight obtained

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2.6.6.4 Genetic toxicology**Study title:** Bacterial reverse mutation report.**Key findings:** Negative under conditions of the assay.**Study no.:** Report AA53PR.503. - CLO.00.07.02**Volume #, and page #:** Module 5.4.12.1**Conducting laboratory and location:** ☐**Date of study initiation:** completed 1/7/02**GLP compliance:** Yes**QA reports:** yes (x), no ()**Drug, lot #, and % purity:** clofarabine; 010188; - % (HPLC, provided by the sponsor).**Methods****Strains/species/cell line:** S. typhimurium TA98, TA100, TA1535, TA1537; E. coli WP2 *uvrA*.**Doses used in definitive study:** 75, 200, 600, 1800, 5000 µg/plate.**Basis of dose selection:** toxicity in preliminary assay**Negative controls:** DMSO (vehicle)**Positive controls:**

Strain	Compound	S9	Concentration (µg/plate)
Salmonella strains	2-aminoanthracene	-	1
WP2 _{uvrA}	2-aminoanthracene	-	10
TA98	2-nitrofluorene	+	1
TA100, TA1535	Sodium azide	+	1
TA1537	9-aminoacridine	+	75
WP2 _{uvrA}	methylmethanesulfonat	+	1000

Incubation and sampling times: plate incorporation method; 48-72 h**Results****Study validity** (comment on replicates, counting method, criteria for positive results, etc.):**Replicates:** triplicate plates for confirmatory assay**Counting method:** automatic or manual

Valid assay requires at least 3 non-toxic dose levels for evaluation.

Criteria for a positive result: a dose-related increase in the mean revertants per plate over two increasing concentrations or an increase in mean revertants at the peak of the dose response relative to mean vehicle control.

TA1535 and TA1537; $\geq 3x$.

TA98, TA100, WP2 uvrA; $\geq 2x$.

Positive control: $\geq 3x$

Study outcome: Study met the criteria for an acceptable assay. Clofarabine was negative for mutagenicity under the conditions of the assay.

Study title: **In vitro mammalian chromosome aberration test.**

Key findings: Clofarabine was positive as a clastogen in the presence and absence of metabolic activation

Study no.: Report AA53PR.331. — CLO.00.07.03

Volume #, and page #: Module 5.4.12.2

Conducting laboratory and location: []

Date of study initiation: 1/7/02

GLP compliance: yes

QA reports: yes (x) no ()

Drug, lot #, and % purity: clofarabine; 010188; — % (by sponsor)

Methods

Strains/species/cell line: Chinese hamster ovary cells

Concentrations used in definitive study:

4 h: 0.1, 0.5, 1, 5, 10, 15 $\mu\text{g/mL}$

20 h: 0.05, 0.1, 0.5, 1, 5, 10 $\mu\text{g/mL}$

Basis of concentration selection: cell growth inhibition/reduction in mitotic index $\geq 50\%$ in preliminary assays

Negative controls: DMSO (vehicle solvent)

Positive controls: 0.1 and 0.2 $\mu\text{g/mL}$ Mitomycin C; 10 and 20 $\mu\text{g/mL}$ cyclophosphamide

Incubation and sampling times: \pm S9, 4 h; -S9, 20 h*; all cells harvested 20 h after treatment initiation (*, not analyzed in this assay)

Results

Study validity (comment on replicates, counting method, criteria for positive results, etc.):

Replicates: duplicate cultures

Concentrations selected for analysis: highest concentration with >50% ↓ in cell growth and next 2 lower concentrations.

Criteria for valid study: frequency of cells with structural chromosome aberrations in the solvent control must be within the range of the historical solvent control. The percentage of cells with chromosome aberrations in the positive control must be statistically increased relative to the solvent control.

Criteria for positive result: % cells with aberrations ↑ in a concentration-responsive manner with one or more concentrations statistically significant. Values that were statistically significant but within range of historic solvent controls were judged as not biologically significant (sponsor's criteria).

Study outcome: Sponsor judged the study positive for structural and numerical aberrations in the non activated system. The reviewer concurs with this conclusion. The sponsor judged the assay positive for statistically significant increases in structural and numerical aberrations in the activated system. The sponsor concludes that these findings are not biologically relevant as they fall within the range of historic control values. The reviewer disagrees and judges the assay positive because of the findings in the non activated arm and statistical significance of the findings. This is consistent with the interpretation of assays discussed in ICH S2A.

TABLE 9
SUMMARY

Treatment (µg/ml.)	S9 Activation	Treatment Time	Mean Mitotic Index	Cells Scored	Aberrations Per Cell (Mean ± SD)	Cells With Aberrations Numerical (%)	Aberrations Structural (%)
DMSO	-	4	8.0	200	0.000 ±0.000	3.3	0.0
Clofarabine							
1	-	4	7.0	200	0.000 ±0.000	0.0	0.0
5	-	4	7.8	200	0.100 ±0.117	0.0	9.5**
10	-	4	5.8	200	0.200 ±0.227	13.0**	13.0**
MMC, 0.2	-	4	6.1	200	0.265 ±1.073	0.0	15.5**
DMSO	+	4	8.5	200	0.000 ±0.000	0.5	0.0
Clofarabine							
1	+	4	7.5	200	0.000 ±0.000	1.0	0.0
5	+	4	7.5	200	0.005 ±0.071	1.0	0.5
10	+	4	6.1	200	0.035 ±0.253	4.5**	2.5*
CP, 10	+	4	6.2	200	0.125 ±0.361	2.0	11.5**

Treatment: Cells from all treatment conditions were harvested at 20 hours after the initiation of the treatments.

Aberrations per Cell: Severely damaged cells were counted as 10 aberrations.

Percent Aberrant Cells: *, p<0.05; **, p<0.01; using Fisher's exact test.

(Excerpted from the sponsor's submission)

Study title: Mammalian erythrocyte micronucleus test.

Key findings: Clofarabine induced micronuclei in rat bone marrow after iv administration.

Study no.: Report AA53PR.125. — CLO.00.48

Volume #, and page #: module 5.4.12.3

Conducting laboratory and location: E J

Date of study initiation: 2/25/02

GLP compliance: yes

QA reports: yes (x) no ()

Drug, lot #, and % purity: clofarabine; 010188; — (by sponsor)

Methods

Strains/species/cell line: male and female Sprague Dawley rats
5/sex/group; 10 mL/kg dose volume

Doses used in definitive study: 8.75, 17.5, 35 mg/kg iv

Basis of dose selection: mortality in preliminary assay:
50 mg/kg, 1/3 • ; 0/3 • •
100 mg/kg, 3/3 • ; 3/3 • •

Negative controls: 25% PEG 400 in 0.9% saline (vehicle) iv

Positive controls: cyclophosphamide 40 mg/mL iv

Incubation and sampling times:
test article: single dose; bone marrow collected at 24 (LD-HD), 48 h (HD only)
negative control: 24, 48 h
positive control: 24 h

Results

Study validity (comment on replicates, counting method, criteria for positive results, etc.):

Replicates: n= 5; 2000 PCE/animal scored

Counting: manual; slides coded by individual not involved in counting

Criteria for a Valid Test: mean MPCE must not exceed 3/1000 in the vehicle control; MPCE in the positive control group must be significantly increased relative to the vehicle control.

Criteria for positive result: a dose-responsive increase in MPCE and one or more doses were statistically elevated relative to the vehicle control. Values that were statistically significant but within range of historical negative or vehicle controls were judged as not biologically significant (sponsor's criteria).

Study outcome:

Positive for induction of micronucleated polychromatic erythrocytes under the conditions

Table 4: Summary of Bone Marrow Micronucleus Analysis After a Single Dose of Clofarabine in Sprague Dawley Rats

Treatment (10 mL/kg)	Sex	Time (hr)	Number of Rats	PCE/Total Erythrocytes (Mean ± SD)	Change from Control (%)	Micronucleated Polychromatic Erythrocytes	
						Number per 1000 PCEs (Mean ± SD)	Number per PCEs Scored ¹
25% PEG 400 in 0.9% saline	M	24	5	0.640 ± 0.02	---	0.6 ± 0.65	6 / 10000
	F	24	5	0.642 ± 0.04	---	0.6 ± 0.65	6 / 10000
Clofarabine 8.75 mg/kg	M	24	5	0.646 ± 0.05	1	0.0 ± 0.00	0 / 10000
	F	24	5	0.626 ± 0.03	-2	0.2 ± 0.45	2 / 10000
17.5 mg/kg	M	24	5	0.631 ± 0.06	-1	1.5 ± 1.27	*15 / 10000
	F	24	5	0.637 ± 0.03	-1	0.6 ± 0.82	6 / 10000
35 mg/kg	M	24	5	0.615 ± 0.05	-4	2.6 ± 0.89	*26 / 10000
	F	24	5	0.634 ± 0.01	-1	1.8 ± 0.76	*18 / 10000
CP 40 mg/kg	M	24	5	0.589 ± 0.05	-8	25.4 ± 8.76	*254 / 10000
	F	24	5	0.578 ± 0.03	-10	16.6 ± 3.55	*166 / 10000
25% PEG 400 in 0.9% saline	M	48	5	0.636 ± 0.05	---	0.4 ± 0.42	4 / 10000
	F	48	5	0.616 ± 0.02	---	0.3 ± 0.45	3 / 10000
Clofarabine 35 mg/kg	M	48	5	0.611 ± 0.03	-4	1.5 ± 0.50	*15 / 10000
	F	48	5	0.632 ± 0.07	3	2.2 ± 0.45	*22 / 10000

¹*Statistically significant, p<0.05 (Kastenbaum-Bowman Tables)

(Excerpted from the sponsor's submission)

2.6.6.5 Carcinogenicity: Studies not conducted

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2.6.6.6 Reproductive and developmental toxicology

Fertility and early embryonic development: Not done

Embryofetal development

1 Intravenous Dosage-Range Developmental Toxicity Study of Clofarabine in Rats. Sponsor's Study Number: 0202-01.

Key study findings: Clofarabine administration at 30 mg/kg/day via I.V. bolus to female rats once daily from DG 7 through 17 caused three mortalities (38%). Clinical observations included piloerection, decreased motor activity, impaired or loss of righting reflex, labored breathing, and disorientation. Dose dependent reductions in maternal body weight gains were observed at 3, 10 and 30 mg/kg/day. Increased postimplantation loss and decreased fetal body weights were evident in the 10 and 30 mg/kg/day dosage groups. Histopathological findings in dams (small thymus and spleen, enlarged thymus and stomach, black areas on lungs and red areas on the thymus) were limited to the high dose animals.

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2 Intravenous Developmental Toxicity Study of Clofarabine in Rats.
Sponsor's Study Number: 0204-01.

Key study findings: The intravenous administration of clofarabine at 9 mg/kg/day from DG 7 to 17 was associated with reduced maternal body weight gains, reduced fetal body weights, increased postimplantation loss, and increased incidences of malformations and variations (gross external, soft tissue, skeletal and retarded ossification). There was no maternal toxicity.

Study no.: Sponsor – 0204-01
 — 1209-001
Volume #, and page #: Electronic Module
Conducting laboratory and location: L

1

Date of study initiation: 16 April 2002
GLP compliance: Yes
QA reports: yes (X) no ()
Drug, lot #, and % purity: Clofarabine, lot # 010188, — purity
Formation/vehicle: PEG 400, lot # T42604 by . —
 0.9% NaCl, lot # JIP577 by —

Methods

Doses:

Dosage Group	Dosage* (mg/kg/day)	Concentration (mg/mL)	Dosage Volume (mL/kg)	Injection Rate	Number of Rats	Assigned Rat Numbers
I	0 (Vehicle)	0	10	Slow Bolus	25	19601-19625
II	1	0.1	10	Slow Bolus	25	19626-19650
III	3	0.3	10	Slow Bolus	25	19651-19675
IV	9	0.9	10	Slow Bolus	25	19676-19700

a. The test article was considered 100% active for the purpose of dosage calculations.

(Excerpted from the sponsor's submission)

Animals were dosed once daily on DGs 7 through 17.

Species/strain: — CD@ (SD) IGS BR VAF/Plus®
Number/sex/group: 25
Route, formulation, volume, and infusion rate: IV
Satellite groups used for toxicokinetics: No
Study design: Segment II (effects on embryo-fetal development, implantation to closure of the hard palate, stages C to D)

Parameters and endpoints evaluated:

Females: Clinical signs, body weights, and food consumption.

DG 22- combined weight of uterus, number and placements of implantation sites, live and dead fetuses, early and late resorptions, ovaries and oviducts, and corpora lutea.

Fetuses: Live fetuses were examined for weight, size, shape, color, and position of the head, palate, limb and digits, back tail, anus, and external genitalia.

Half of the fetuses were examined viscerally and the remaining fetuses were examined for skeletal abnormalities.

Results

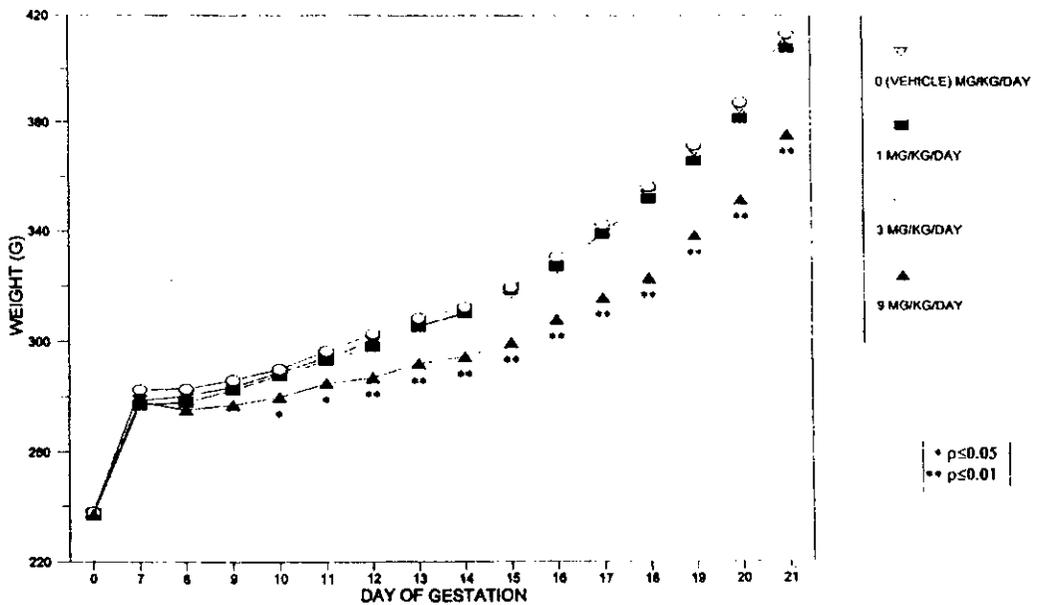
Mortality (dams):

3 mg/kg/day – 1 died on DG 11 and 1 delivered before caesarean sectioning on DG 21. These events are not related to test article because similar events were not observed in the high dosage group.

Clinical signs (dams):

Localized alopecia (limbs), urine-stained abdominal fur, red perivaginal substance, swollen hindlimbs and hindpaws, chromorhinorrhea, and discolored tail in all groups.

Body weight (dams):



(Excerpted from the sponsor's submission)

Reductions in body weight and body weight gain were observed at 9 mg/kg/day as shown below.

Maternal body weights changes vs. vehicle control group

Day of gestation	Absolute weight		% change
	Group 1	Group 4	
10	289±11	280±15	-3*
13	306±12	292±18	-5*
15	317±14	300±19	-5*
18	353±15	323±25	-8.5*
21	411±23	375±44	-8.8*

* Significantly different from the vehicle control group value (p≤0.01)

Food consumption (dams): Reduction in absolute (g/day) and relative (g/kg/day) in high dose animals.

Toxicokinetics: Not done

Terminal and necropsic evaluations:

Pregnancy: Increased post implantation loss in 9 mg/kg/day dosage group as compared with vehicle control group values as shown below.

Caesarean section observations

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 1	III 3	IV 9	
RATS TESTED		N	25	25	25	25
PREGNANT	N(%)	25 (100.0)	24 (96.0)	24 (96.0)	24 (96.0)	
FOUND DEAD	N(%)	0 (0.0)	0 (0.0)	1 (4.2)	0 (0.0)	
DELIVERED AND SACRIFICED	N(%)	0 (0.0)	0 (0.0)	1 (4.2)	0 (0.0)	
RATS PREGNANT AND CAESAREAN-SECTIONED ON DAY 21 OF GESTATION		N	25	24	22	24
CORPORA LUTEA	MEAN±S.D.	16.4 ± 2.9	16.6 ± 2.2	16.4 ± 2.3	18.2 ± 2.9	
IMPLANTATIONS	MEAN±S.D.	15.4 ± 2.1	15.2 ± 1.4	15.4 ± 1.7	15.6 ± 2.2	
LITTER SIZES	MEAN±S.D.	14.6 ± 2.0	13.9 ± 2.1	15.0 ± 1.7	13.1 ± 4.5	
LIVE FETUSES	N	364	333	330	315	
	MEAN±S.D.	14.6 ± 2.0	13.9 ± 2.1	15.0 ± 1.7	13.1 ± 4.5	
DEAD FETUSES	N	0	0	0	0	
RESORPTIONS	MEAN±S.D.	0.8 ± 1.1	1.3 ± 1.4	0.4 ± 0.6	2.4 ± 4.0	
EARLY RESORPTIONS	N	20	31	10	53	
	MEAN±S.D.	0.8 ± 1.1	1.3 ± 1.4	0.4 ± 0.6	2.2 ± 4.0	
LATE RESORPTIONS	N	0	0	0	6	
	MEAN±S.D.	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.2 ± 0.4*	
DAMS WITH ANY RESORPTIONS	N(%)	11 (44.0)	17 (70.8)	9 (40.9)	15 (62.5)	
DAMS WITH ALL CONCEPTUSES RESORBED	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	2 (8.3)	
DAMS WITH VIABLE FETUSES	N(%)	25 (100.0)	24 (100.0)	22 (100.0)	22 (91.7)	
PLACENTAE APPEARED NORMAL ^b	N(%)	25 (100.0)	24 (100.0)	22 (100.0)	22 (100.0)	

a. Dosage occurred on days 7 through 17 of gestation.
 b. Excludes dams with all early resorptions.
 * Significantly different from the vehicle control group value (p<0.05).

(Excerpted from the sponsor's submission)

Corpora lutea: Comparable among groups as expected since the dosing did not begin until after ovulation occurred.

Offspring:

Body weights: Reduced in the 9 mg/kg/day dosage group vs. control group values.
 Litter size and sex: Reduced litter size and number of live fetuses in high dose animals as shown below.

Litter Observations

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 1	III 3	IV 9
LITTERS WITH ONE OR MORE LIVE FETUSES	N	25	24	22	22
IMPLANTATIONS	MEAN ₂ S.D.	15.4 ± 2.1	15.2 ± 1.4	15.4 ± 1.7	15.7 ± 2.3
LIVE FETUSES	N	364	333	330	315
	MEAN ₂ S.D.	14.6 ± 2.0	13.9 ± 2.1	15.0 ± 1.7	14.3 ± 2.1
LIVE MALE FETUSES	N	187	173	165	144
† LIVE MALE FETUSES/LITTER	MEAN ₂ S.D.	50.9 ± 12.7	52.3 ± 15.0	50.1 ± 10.8	45.4 ± 13.4
LIVE FETAL BODY WEIGHTS (GRAMS)/LITTER	MEAN ₂ S.D.	5.45 ± 0.25	5.37 ± 0.24	5.28 ± 0.27	4.67 ± 0.54**
MALE FETUSES	MEAN ₂ S.D.	5.55 ± 0.30	5.51 ± 0.29	5.42 ± 0.28	4.73 ± 0.61**
FEMALE FETUSES	MEAN ₂ S.D.	5.34 ± 0.25	5.22 ± 0.27	5.13 ± 0.29	4.58 ± 0.52**
† RESORBED CONCEPTUSES/LITTER	MEAN ₂ S.D.	5.0 ± 6.6	8.7 ± 9.6	2.9 ± 3.8	8.2 ± 9.3

a. Dosage occurred on days 7 through 17 of gestation.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

External, visceral, and skeletal anomalies:

Fetal alterations

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 1	III 3	IV 9
LITTERS EVALUATED	N	25	24	22	22
FETUSES EVALUATED	N	364	333	330	315
LIVE	N	364	333	330	315
LITTERS WITH FETUSES WITH ANY ALTERATION OBSERVED	N(%)	5 (20.0)	6 (25.0)	6 (27.3)	13 (59.1)**
FETUSES WITH ANY ALTERATION OBSERVED	N(%)	6 (1.6)	8 (2.4)	6 (1.8)	62 (19.7)**
† FETUSES WITH ANY ALTERATION/LITTER	MEAN ₂ S.D.	1.7 ± 4.0	2.4 ± 4.7	1.9 ± 3.3	21.4 ± 30.5**

a. Dosage occurred on days 7 through 17 of gestation.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Summary of fetal gross external alterations: Group 4 — ↑fore and hindlimbs (medially rotated, flexed downward, digits short, digit absent, digit fused, litter & fetal), body (trunk short, dark area, thin and transparent skin), tail (short, absent) vs group 1 (vehicle).

Summary of fetal soft tissue alterations: Group 4 - ↑brain (third and lateral ventricles dilation), vessels (umbilical artery descended), kidneys (absent or small), genitalia (testes undescended) vs group 1 (vehicle).

Summary of fetal skeletal alterations: Group 4 - ↑ cervical vertebrae (cervical rib present at the 7th cervical vertebra), interrelated vertebral/rib malformations, thoracic vertebrae (arch open, fused, irregularly shaped, centrum irregularly shaped, fused, unilateral ossification), lumber vertebrae (arches fused, open, centrum bifid, unilateral ossification), sacral vertebrae (centra fused), caudal vertebrae (irregular), ribs (fused, short, boned), sternal centra (fused, asymmetric), hindlimb (digit absent, not ossified) vs group 1 (vehicle).

See tables below for details.

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On Original

Fetal gross external alterations:

DOSAGE GROUP		DOSAGE (MG/KG/DAY) ^a			
		I	II	III	IV
		0 (VEHICLE)	1	3	9
LITTERS EVALUATED	N	25	24	22	22
FETUSES EVALUATED ^b	N	364	333	330	315
LIVE	N	364	333	330	315
HEAD					
EYE: LIDS OPEN					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.3)
EYE: BULGE DEPRESSED					
LITTER INCIDENCE	N(%)	0(0.0)	1(4.2)	0(0.0)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	1(0.3)	0(0.0)	0(0.0)
PALATE: CLEFT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.3)	0(0.0)
JAW: MICROGNATHIA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.3)	0(0.0)
FORE AND/OR HINDLIMBS: MEDIANLY ROTATED AND/OR FLEXED DOWNWARD					
LITTER INCIDENCE	N(%)	0(0.0)	1(4.2)	1(4.5)	6(27.3)**
FETAL INCIDENCE	N(%)	0(0.0)	1(0.3)	1(0.3)	17(5.4)**
FORE AND/OR HINDLIMBS: DIGITS SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(27.3)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	12(3.8)**
FORE AND/OR HINDLIMBS: DIGITS ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	18(5.7)**
FORE AND/OR HINDLIMBS: DIGITS FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(1.3)**
FORE AND/OR HINDLIMBS: SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(2.5)**
BODY					
TRUNK SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(31.8)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(6.3)**
DARK AREAS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(31.8)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	19(6.0)**
THIN AND TRANSPARENT SKIN					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(31.8)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(6.3)**
EDEMA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.3)	1(0.3)
UMBILICAL HERNIA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.3)
GASTROSCHEISIS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.3)
NO ANAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.3)
TAIL					
SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(22.7)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(2.2)**
PREUNCULATED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(1.3)**
ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(1.0)**
KINKED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(0.6)

a. Dosage occurred on days 7 through 17 of gestation.
 b. See the individual fetal alterations table (Table 21) for fetuses with multiple gross external alterations.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal soft tissue alterations:

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 1	III 3	IV 9
LITTERS EVALUATED	N	25	24	22	22
FETUSES EVALUATED ^b	N	177	162	160	154
LIVE	N	177	162	160	154
EYES: RETINA FOLDED					
LITTER INCIDENCE	N(%)	1(4.0)	1(4.2)	1(4.5)	0(0.0)
FETAL INCIDENCE	N(%)	1(0.6)	1(0.6)	1(0.6)	0(0.0)
EYES: MICROPTHALMIA					
LITTER INCIDENCE	N(%)	0(0.0)	1(4.2)	0(0.0)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	1(0.6)	0(0.0)	0(0.0)
BRAIN: THIRD VENTRICLE SLIGHT DILATION					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(22.7)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	13(8.4)**
BRAIN: LATERAL VENTRICLES SLIGHT DILATION					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(1.9)**
VESSELS: UMBILICAL ARTERY DESCENDED TO THE LEFT OF THE URINARY BLADDER					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(22.7)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(3.9)**
VESSELS: IMMINUTE ARTERY ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
KIDNEYS: PELVIS, DILATED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.3)
KIDNEYS: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	9(5.8)**
KIDNEYS: SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.6)**
INTESTINES: DIVERTICULUM					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
INTESTINES: PROTRUDED THROUGH UMBILICUS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
GENITALIA: TESTES UNDESCENDED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.2)**
GENITALIA: OVARIES DISPLACED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.3)
GENITALIA: UTERINE HORN REDUCED TO A LIGAMENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
GENITALIA: UTERINE HORN ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
GENITALIA: OVARIES AND TESTES PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
GENITALIA: TESTES ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
URETERS: DILATED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
MASS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)

a. Dosage occurred on days 7 through 17 of gestation.
 b. See the individual fetal alterations table (Table 21) for fetuses with multiple soft tissue alterations.

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP DOSAGE (MG/KG/DAY) a	0 (VEHICLE)	I 1	II 1	III 1	IV 1
FETUSES EVALUATED b	N	22	24	22	22
FETUSES EVALUATED b LIVE	N	197	171	170	161
	N	187	171	170	161
SKULL: PALATE, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (4.5)	0 (0.0)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (0.6)	0 (0.0)
SKULL: MANDIBLES, FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (4.5)	0 (0.0)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (0.6)	0 (0.0)
SKULL: MANDIBLES, SHORT					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (4.5)	0 (0.0)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (0.6)	0 (0.0)
SKULL: PARIETALS, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.6)
SKULL: INTERPARIETALS, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.6)
CERVICAL VERTEBRAE: CERVICAL RIB PRESENT AT 7TH CERVICAL VERTEBRA					
LITTER INCIDENCE	N(%)	2 (9.1)	2 (9.3)	0 (0.0)	6 (27.3)**
FETAL INCIDENCE	N(%)	2 (1.1)	2 (1.2)	0 (0.0)	9 (5.6)**
CERVICAL VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.6)
INTERRELATED VERTEBRAL/RIB MALFORMATIONS SUBSUMMATION (Includes Cervical, thoracic, lumbar, sacral, caudal and/or rib alterations, excluding absence of vertebrae and ribs)					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	8 (36.4)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	23 (14.3)**
THORACIC VERTEBRAE: CENTRUM, BIFID					
LITTER INCIDENCE	N(%)	2 (9.1)	1 (4.2)	3 (13.6)	9 (40.9)**
FETAL INCIDENCE	N(%)	3 (1.6)	2 (1.2)	3 (1.8)	24 (14.9)**
THORACIC VERTEBRAE: ARCH, OPEN					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	3 (13.6)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	5 (3.1)**
THORACIC VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	8 (36.4)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	22 (13.7)**
THORACIC VERTEBRAE: CENTRUM, UNILATERAL OSSIFICATION					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (4.5)	5 (22.7)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (0.6)	10 (6.2)**
THORACIC VERTEBRAE: ARCH, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	3 (13.6)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	5 (3.1)**
THORACIC VERTEBRAE: CENTRUM, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.6)
THORACIC VERTEBRAE: IRREGULAR NUMBER PRESENT					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	8 (36.4)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	16 (9.9)**
THORACIC VERTEBRAE: CENTRUM, FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	2 (9.1)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	2 (1.2)
THORACIC VERTEBRAE: ARCHES AND CENTRA FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	4 (18.2)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	6 (3.7)**
THORACIC VERTEBRAE: CENTRA, FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	6 (27.3)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	20 (12.4)**
THORACIC VERTEBRAE: CENTRUM, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (4.5)	2 (9.1)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	1 (0.6)	2 (1.2)
THORACIC VERTEBRAE: NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.6)
THORACIC VERTEBRAE: ARCH, SMALL					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	3 (13.6)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.6)
THORACIC VERTEBRAE: ARCH, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	2 (1.2)
LUMBAR VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	8 (36.4)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	17 (10.6)**
LUMBAR VERTEBRAE: CENTRUM, BIFID					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	7 (31.8)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	11 (6.8)**
LUMBAR VERTEBRAE: CENTRUM, UNILATERAL OSSIFICATION					
LITTER INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	2 (9.1)
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	3 (1.9)**
LUMBAR VERTEBRAE: ARCHES, OPEN					
LITTER INCIDENCE	N(%)	0 (4.0)	0 (0.0)	0 (0.0)	3 (13.6)**
FETAL INCIDENCE	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	4 (2.5)**

a. Dosage occurred on days 7 through 17 of gestation.
 b. See the individual fetal alterations table (Table 21) for fetuses with multiple skeletal alterations.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP (NO./EG/DAY) ^a		I 0 (VEHICLE)	II 1	III 3	IV 9
LETTERS EVALUATED ^b	N	25	26	22	22
FETUSES EVALUATED ^b	N	187	171	170	161
LIVE	N	187	171	170	161
<hr/>					
LUMBAR VERTEBRAE: ARCH, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(1.9)**
<hr/>					
LUMBAR VERTEBRAE: CENTRUM, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
LUMBAR VERTEBRAE: CENTRA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(27.3)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	10(6.2)**
<hr/>					
LUMBAR VERTEBRAE: ARCHES AND CENTRA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.5)**
<hr/>					
LUMBAR VERTEBRAE: ARCH, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.2)
<hr/>					
LUMBAR VERTEBRAE: ARCH, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
LUMBAR VERTEBRAE: IRREGULAR NUMBER PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(31.8)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	13(8.1)**
<hr/>					
LUMBAR VERTEBRAE: HEMI-VERTEBRA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
LUMBAR VERTEBRAE: NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
LUMBAR VERTEBRAE: (UNABLE TO LOCATE WITH RIBS NOT OSSIFIED)					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
SACRAL VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.1)**
<hr/>					
SACRAL VERTEBRAE: ARCH, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.2)
<hr/>					
SACRAL VERTEBRAE: CENTRA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.1)**
<hr/>					
SACRAL VERTEBRAE: CENTRUM, UNILATERAL OSSIFICATION					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
CAUDAL VERTEBRAE: HEMI-VERTEBRA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.2)
<hr/>					
CAUDAL VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.2)
<hr/>					
CAUDAL VERTEBRAE: CENTRA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
CAUDAL VERTEBRAE: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
<hr/>					
CAUDAL VERTEBRAE: IRREGULAR NUMBER PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(18.2)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.5)**
<hr/>					
RIBS: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(36.4)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	21(14.1)**
<hr/>					
RIBS: NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(31.8)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	9(5.6)**
<hr/>					
RIBS: SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.2)
<hr/>					
RIBS: BROAD					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.5)**
<hr/>					
RIBS: SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(31.8)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	10(6.2)**
<hr/>					
RIBS: BOWED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(22.7)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.1)**
<hr/>					
RIBS: WAVY					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	0(0.0)
<hr/>					
RIBS: ASYMMETRIC RIB PAIR NUMBERS OR IRREGULAR NUMBER PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(36.4)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	18(11.2)**

a. Dosages occurred on days 7 through 17 of gestation.

b. See the individual fetal alterations table (Table 21) for fetuses with multiple skeletal alterations.

** Significantly different from the vehicle control group value (p<0.01).

Fetal skeletal alterations:

DOSE GROUP		I	II	III	IV
DOSE (MG/50/DAY) ^a		0 (VEHICLE)	1	3	9
LITTER EVALUATED	N	25	24	22	22
FETUSES EVALUATED ^b	N	187	171	170	161
LIVE	N	187	171	170	161
RIBS: TWO SEGMENTS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
RIBS: THIN					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
RIBS: IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
MANUBRIUM: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.1)**
STERNAL CESTA: SUMMARISATION (Includes fused, incompletely ossified and asymmetric)					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	8(36.4)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	10(11.9)**
STERNAL CESTA: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(27.3)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	14(8.7)**
STERNAL CESTA: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	2(13.6)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	1(1.9)
STERNAL CESTA: ASYMMETRIC					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(1.9)**
XIPHOID: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(5.0)**
FORELIMB: METACARPALS, EXTRA OSSIFICATION					
LITTER INCIDENCE	N(%)	0(0.0)	1(4.2)	0(0.0)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	1(0.6)	0(0.0)	0(0.0)
FORELIMB: PHALANGES, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
PELVIS: SUMMARISATION (Includes ischium and pubis, incompletely ossified and ilium, irregularly shape)					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(9.1)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(1.9)**
PELVIS: ISCHIUM, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
PELVIS: PUBIS, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
PELVIS: ILIUM, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.2)
HINDLIMB: PHALANGES, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(22.7)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	11(6.8)**
HINDLIMB: METATARSALS, EXTRA OSSIFICATION					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	1(0.6)
HINDLIMB: TIBIA, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	0(0.0)
HINDLIMB: FIBULA, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(4.5)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	0(0.0)
HINDLIMB: DIGIT, ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(5.0)**
HINDLIMB: DIGITS, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
HINDLIMB: METATARSALS, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(13.6)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.5)**
HINDLIMB: FIBULA, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)
HINDLIMB: FEMUR, MISALIGNED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(4.5)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.6)

a. Dosage occurred on days 7 through 17 of gestation.
 b. See the individual fetal alterations table (Table 21) for fetuses with multiple skeletal alterations.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

3 Intravenous Dosage-Range Developmental Toxicity Study of Clofarabine in Rabbits. Sponsor's Study Number: 0202-02.

Key study findings: Clofarabine treatment at 20 mg/kg from DGs 6 through 18 killed all the rabbits. All conceptuses were resorbed and there were no live fetuses in the 6 mg/kg/day dosage group. Gross external fetal examination revealed multiple malformations in the 0.6 and 2 mg/kg/day dosage groups. Clinical observations included tachypnea, ungroomed coat and scant feces in the 6 and 20 mg/kg dosages groups.

4 Intravenous Developmental Toxicity Study of Clofarabine in Rabbits. Sponsor's Study Number: 0204-02.

Key study findings: The intravenous administration of clofarabine at 1 mg/kg/day from DG 6 to 18 did not affect the maternal body weight and food consumption. Decreased fetal body weight and increased postimplantation loss were attributed to 1 mg/kg/day clofarabine administration. Increased incidences of malformation and variations (gross external, soft tissue, skeletal and retarded ossifications) were also observed in high dose animals (1 mg/kg/day). The NOAEL dose of clofarabine for developmental toxicity in rabbit was 0.1 mg/kg/day.

Study no.: Sponsor – 0204-02, — — 1209-002
Volume #, and page #: Electronic module
Conducting laboratory and location: []

Date of study initiation:]

GLP compliance: Yes

QA reports: yes (X) no ()

Drug, lot #, and % purity: Clofarabine, lot # 010188, — purity

Methods

Doses:

Dosage Group	Dosage ^a (mg/kg/day)	Concentration (mg/mL)	Volume (mL/kg)	Infusion Rate (mL/hr)	Number of Rabbits	Assigned Rabbit Numbers
I	0 (Vehicle)	0	10	420	20	9901-9919 ^b , 4499 ^c
II	0.1	0.01	10	420	20	9921-9940
III	0.3	0.03	10	420	20	9941-9960
IV	1.0	0.1	10	420	20	9961-9980

- a. The test article was considered 100% active/pure for the purpose of dosage calculations.
 b. Rabbit 9919 was administered 7 mL of the 0.01 mg/mL concentration after the correct administration of the vehicle. This deviation did not adversely affect the outcome or interpretation of the study because the amount of 0.01 mg/mL test article that was administered was minimal (a resultant dose of 0.0194 mg/kg/day).
 c. Rabbit 9920 lost body weight between DGs 0 and 6. This rabbit was replaced by rabbit 4499 on DG 6.

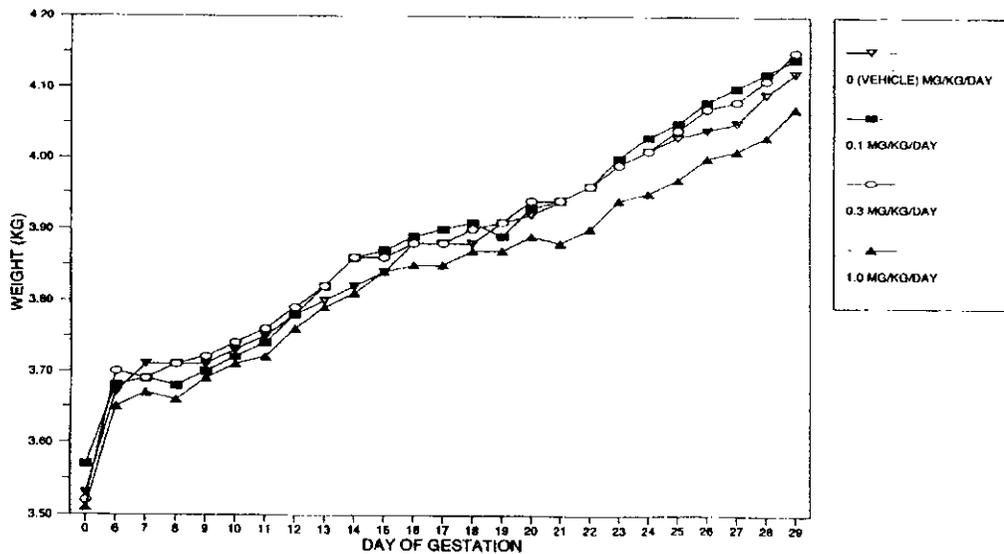
(Excerpted from the sponsor's submission)

Dosages were selected based on the dosage-range study (0202-02)
 Animals were dosed from DGs 6 through 18

Species/strain: Rabbit / Hra:(NZW)SPF
 Number/sex/group: 20
 Route, formulation, volume, and infusion rate: Intravenous, 10 mL/kg,
 Satellite groups used for toxicokinetics: No
 Study design: Segment II (effects on embryo-fetal development,
 implantation to closure of the hard palate, stages C to D)
 Parameters and endpoints evaluated: Clinical observations, mortality,
 moribundity, body weights, food consumption, corpora
 lutea/ovary, weight of uterus + contents, # and position of live and
 dead fetuses, early and late embryo/fetal losses, external
 observations of live fetuses, viscera and skeletons examinations.

Results

Mortality (dams): One control group doe aborted on DG27 and sacrificed.
 Clinical signs (dams): Soft/liquid feces, scant feces, localized alopecia (limbs and
 underside), head tilt, and ungroomed coat in a dose-
 unrelated manner.
 Body weight (dams): No statistically significant differences as shown below.



(Excerpted from the sponsor's submission)

Food consumption (dams): No differences
 Toxicokinetics: Not done

Terminal and necropsic evaluations: C-section data

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
RABBITS TESTED		N	20	20	20
PREGNANT	N(%)	18 (90.0)	18 (90.0)	19 (95.0)	20 (100.0)
ABORTED AND SACRIFICED	N(%)	1 (5.6)	0 (0.0)	0 (0.0)	0 (0.0)
RABBITS PREGNANT AND CAESAREAN-SECTIONED ON DAY 25 OF GESTATION		N	17	18	19
INCLUDED IN ANALYSES		N	16 ^b	18	19
CORPORA LUTEA	MEAN±S.D.	10.2 ± 3.5	10.9 ± 2.1	10.8 ± 2.6	10.9 ± 2.0
IMPLANTATIONS	MEAN±S.D.	8.2 ± 3.4	9.4 ± 2.1	9.5 ± 2.1	9.1 ± 2.2
LITTER SIZES	MEAN±S.D.	7.7 ± 3.2	8.8 ± 1.9	8.6 ± 2.4	7.0 ± 3.1
LIVE FETUSES	N	123	158	163	139
	MEAN±S.D.	7.7 ± 3.2	8.8 ± 1.9	8.6 ± 2.4	7.9 ± 3.1
DEAD FETUSES	N	0	0	0	1
	MEAN±S.D.	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.2
RESORPTIONS	MEAN±S.D.	0.6 ± 0.8	0.7 ± 1.0	0.9 ± 1.1	2.1 ± 2.2
EARLY RESORPTIONS	N	7	11	8	35
	MEAN±S.D.	0.4 ± 0.6	0.6 ± 1.0	0.4 ± 0.8	1.8 ± 2.1
LATE RESORPTIONS	N	2	1	10	7
	MEAN±S.D.	0.1 ± 0.3	0.0 ± 0.2	0.5 ± 0.8	0.4 ± 0.6
DOES WITH ANY RESORPTIONS	N(%)	6 (37.5)	8 (44.4)	11 (57.9)	15 (75.0)
DOES WITH ALL CONCEPTUSES DEAD OR RESORBED	N(%)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
DOES WITH VIABLE FETUSES	N(%)	16 (100.0)	18 (100.0)	19 (100.0)	20 (100.0)
PLACENTAE APPEARED NORMAL	N(%)	16 (100.0)	18 (100.0)	19 (100.0)	20 (100.0)

a. Dosage occurred on days 6 through 18 of gestation.
b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.

(Excerpted from the sponsor's submission)

Corpora lutea: Comparable among groups as expected since the dosing did not begin until after ovulation occurred.

Offspring:

Fetal alterations

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED		N	16 ^b	18	19
FETUSES EVALUATED		N	123	158	140
LIVE		N	123	158	139
DEAD		N	0	0	1 ^c
LITTERS WITH FETUSES WITH ANY ALTERATION OBSERVED	N(%)	9 (56.2)	13 (72.2)	13 (68.4)	20 (100.0)**
FETUSES WITH ANY ALTERATION OBSERVED	N(%)	13 (10.6)	24 (15.2)	26 (16.0)	139 (100.0)**
† FETUSES WITH ANY ALTERATION/LITTER	MEAN±S.D.	12.1 ± 14.7	15.7 ± 14.8	14.5 ± 14.0	100.0 ± 0.0**

a. Dosage occurred on days 6 through 18 of gestation.
b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
c. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Litter observations

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY) a		0 (VEHICLE)	0.1	0.3	1.0
LITTERS WITH ONE OR MORE LIVE FETUSES	N	17	18	19	20
INCLUDED IN ANALYSES	N	16b	18	19	20
IMPLANTATIONS	MEAN±S.D.	8.2 ± 3.4	9.4 ± 2.1	9.5 ± 2.1	9.1 ± 2.2
LIVE FETUSES	N	123	158	163	139
	MEAN±S.D.	7.7 ± 3.2	8.8 ± 1.9	8.6 ± 2.4	7.0 ± 3.1
LIVE MALE FETUSES	N	69	78	91	65
† LIVE MALE FETUSES/LITTER	MEAN±S.D.	57.2 ± 20.2	49.2 ± 15.2	56.5 ± 18.8	43.4 ± 20.6
LIVE FETAL BODY WEIGHTS (GRAMS)/LITTER	MEAN±S.D.	45.23 ± 4.58	44.73 ± 4.03	43.45 ± 4.06	30.27 ± 7.04**
MALE FETUSES	MEAN±S.D.	45.85 ± 5.04 [15]c	45.54 ± 3.92	43.78 ± 3.86	32.82 ± 5.32** [18]e
FEMALE FETUSES	MEAN±S.D.	44.15 ± 5.25	43.98 ± 4.75	42.56 ± 4.55 [18]d	29.21 ± 7.92**
‡ DEAD OR RESORBED CONCEPTUSES/LITTER	MEAN±S.D.	5.9 ± 8.7	6.4 ± 9.3	10.4 ± 13.0	24.2 ± 26.3

[] = NUMBER OF VALUES AVERAGED

a. Dosage occurred on days 6 through 10 of gestation.

b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.

c. Litter 4499 had no male fetuses.

d. Litter 9950 had no female fetuses.

e. Litter 9961 and 9972 had no male fetuses.

** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Summary of external, visceral, and skeletal anomalies:

Fetal gross external alterations: Group 4 (1 mg/kg/day) - ↑ Cleft snout, short snout, cleft palate, micrognathia jaw, absent teeth, open eyes, fore/hindlimbs (short, digit absent), trunk short body, and absent/short tail vs. control animals.

Fetal soft tissue alterations: Group 4 - ↑ Brain (dilated ventricles), heart (protruded through ventral opening), lungs (intermediate lobe absent, fused, small), vessels (protruded through ventral opening), liver (thick, edges rounded), kidneys (absent, small), pancreas, uterus, ovaries, and spleen (protruded through ventral opening) vs. control group animals.

Fetal skeletal alterations: Group 4 - ↑ Skull (irregularly shaped, small, fused, incompletely ossified, nasals short, nasal fused), hyoid (ala not ossified, small), thoracic vertebrae (arched, fused), lumbar vertebrae (fused), sacral vertebrae (fused, misaligned), ribs (fused, short), clavicular (not ossified, short), forelimb (fused, short, irregularly shaped), pelvis (not ossified), and hindlimb (short, not ossified) vs. control.

See tables below for details

Fetal gross external alterations:

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY) a		0 (VEHICLE)	0.1	0.3	1.0
LITTERS EVALUATED	N	168	18	19	20
FETUSES EVALUATED	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1c
SNOUT: CLEFT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(5.8)**
SNOUT: SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(35.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	13(9.4)**
PALATE: CLEFT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	124(89.2)**
JAW: MICROGNATHIA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(40.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	21(15.0)**
JAW: AGNATHIA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
TEETH: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
TONGUE: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
EYE: OPEN					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	14(70.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	67(48.2)**
EARS: LOW SET					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
HEAD: DOMED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
HEAD: SMALL ORAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
HEAD: MENINGOCELE					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORE AND/OR HINDLIMBS: FLEXED FORWARD OR DOWNWARD					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
FORE AND/OR HINDLIMBS: JOINT BENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(5.0)**
FORE AND/OR HINDLIMBS: ROTATED Laterally or Medially					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(5.8)**
FORE AND/OR HINDLIMBS: SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	12(60.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	52(37.4)**
FORE AND/OR HINDLIMBS: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORE AND/OR HINDLIMBS: DIGITS ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	135(97.1)**
FORE AND/OR HINDLIMBS: DIGITS FLEXED DOWNWARD					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORE AND/OR HINDLIMBS: DIGITS SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	9(45.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	22(15.8)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal gross external alterations:

DOSAGE GROUP		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16b	18	19	20
FETUSES EVALUATED	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1c
FORE AND/OR HINDLIMBS: TOENAIL ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	10(50.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	26(18.7)**
BODY: TRUNK SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
BODY: UMBILICAL HERNIA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
BODY: SPINE CURVED Laterally					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
BODY: SPINE CURVED UPWARD IN UPPER THORAX					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
BODY: THORACOGASTROSCHISIS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
BODY: GASTROSCHISIS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(5.3)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	0(0.0)
TAIL: CURVED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
TAIL: BENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
TAIL: SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
TAIL: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**

- a. Dosage occurred on days 6 through 18 of gestation.
- b. Excludes doe 9919, which was dosed 7 ml. of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
- c. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
- ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

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Fetal soft tissue alterations:

DOSE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16b	18	19	20
FETUSES EVALUATED ^c	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1d
EYES: CIRCUMCORNEAL HEMORRHAGE					
LITTER INCIDENCE	N(%)	1(6.2)	7(38.9)**	4(21.0)	1(5.0)
FETAL INCIDENCE	N(%)	1(0.8)	9(5.7)	6(3.7)	2(1.4)
BRAIN: DILATED VENTRICLES					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
HEART: INTERVENTRICULAR SEPTAL DEFECT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
HEART: VENTRICULAR CHAMBER ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
HEART: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
VESSELS: PULMONARY ARTERY CONSTRICTED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
VESSELS: SITUS INVERSUS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
VESSELS: DISTENDED AORTA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
VESSELS: AORTA DESCENDED TO THE RIGHT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
VESSELS: PERSISTANT TRUNCUS ARTERIOSIS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
VESSELS: RIGHT SUBCLAVIAN ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
VESSELS: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
LUNGS: INTERMEDIATE LOBE ABSENT					
LITTER INCIDENCE	N(%)	2(12.5)	2(11.1)	1(5.3)	9(45.0)**
FETAL INCIDENCE	N(%)	3(2.4)	2(1.3)	1(0.6)	25(18.0)**
LUNGS: SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(40.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	14(10.1)**
LUNGS: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
LUNGS: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
LUNGS: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
LIVER: EDGES ROUNDED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
LIVER: THICK					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
LIVER: PORTION OF LIVER PROTRUDED THROUGH UMBILICUS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(5.3)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	0(0.0)
LIVER: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes Doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conception are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal soft tissue alterations:

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16 ^b	10	19	20
FETUSES EVALUATED ^c	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1 ^d
LIVER: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
STOMACH: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
KIDNEYS: ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
KIDNEYS: LOW SET					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
KIDNEYS: SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
KIDNEYS: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
ADRENALS: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
ADRENALS: DISPLACED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
INTESTINES: PROTRUDED THROUGH UMBILICUS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(5.3)	3(15.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	3(2.2)
INTESTINES: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
PANCREAS: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
URINARY BLADDER: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
URETERS: DILATED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
UTERUS: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
OVARIES: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
SPLEEN: PROTRUDED THROUGH VENTRAL OPENING					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY) a		0 (VEHICLE)	0.1	0.3	1.0
LITTERS EVALUATED	N	14b	18	19	20
FETUSES EVALUATED c	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1d
SKULL - COMMON IRREGULARITIES:					
(SUMMARIZATION OF INTERNASAL, INTERFRONTAL, MIDLINE SUTURE DISPLACED, ANTERIOR AND POSTERIOR FONTANELLES LARGE, AND IRREGULARLY SHAPED)					
LITTER INCIDENCE	N(%)	0(0.0)	4(22.2)	2(10.5)	18(90.0)**
FETAL INCIDENCE	N(%)	0(0.0)	5(3.2)	2(1.2)	37(26.6)**
SKULL: NASALS, CONTAINED AN INTERNASAL					
LITTER INCIDENCE	N(%)	0(0.0)	2(11.1)	1(5.3)	9(45.0)**
FETAL INCIDENCE	N(%)	0(0.0)	3(1.9)	1(0.6)	11(7.9)**
SKULL: FRONTALS, CONTAINED AN INTERFRONTAL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
SKULL: NASALS, MIDLINE SUTURE DISPLACED					
LITTER INCIDENCE	N(%)	0(0.0)	2(11.1)	1(5.3)	14(70.0)**
FETAL INCIDENCE	N(%)	0(0.0)	2(1.3)	1(0.6)	20(14.4)**
SKULL: ANTERIOR AND POSTERIOR FONTANELLES, LARGE					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SKULL: IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
SKULL: PALATE, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	122(87.8)**
SKULL: MANDIBLE, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(40.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(14.4)**
SKULL: MANDIBLE, TWO SEGMENTS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	11(7.9)**
SKULL: NASALS AND FRONTALS, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	12(7.9)**
SKULL: NASALS, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	17(12.2)**
SKULL: NASALS, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
SKULL: NASALS, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	11(55.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	23(16.5)**
SKULL: MAXILLA, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SKULL: MAXILLA, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SKULL: MAXILLAE, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(5.0)**
SKULL: NASAL AND MAXILLA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SKULL: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(5.8)**
SKULL: SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
SKULL: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	14(10.1)**
SKULL: PARIETAL, CONTAINED A HOLE					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
SKULL: NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(5.0)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16b	18	19	20
FETUSES EVALUATED c	N	12j	15h	16j	16i
LIVE	N	12j	15h	16j	15g
DEAD	N	0	0	0	1d
HYOID: ALA, AMPLIATED					
LITTER INCIDENCE	N(%)	3(18.8)	3(16.7)	7(36.8)	11(55.0)**
FETAL INCIDENCE	N(%)	3(2.4)	3(2.9)	11(6.7)	18(12.9)**
HYOID: ALA, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	12(60.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	43(30.9)**
HYOID: ALA, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(40.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	10(7.2)**
CERVICAL VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	1(5.6)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	1(0.6)	0(0.0)	1(0.7)
CERVICAL VERTEBRAE: CENTRUM, UNILATERAL OSSIFICATION					
LITTER INCIDENCE	N(%)	1(6.2)	1(5.6)	0(0.0)	3(15.0)
FETAL INCIDENCE	N(%)	1(0.8)	1(0.6)	0(0.0)	3(2.2)
CERVICAL VERTEBRAE: ARCH, INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	1(5.6)	0(0.0)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	1(0.6)	0(0.0)	0(0.0)
CERVICAL VERTEBRAE: ARCH, SMALL					
LITTER INCIDENCE	N(%)	1(6.2)	1(5.6)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	1(0.8)	1(0.6)	0(0.0)	2(1.4)
CERVICAL VERTEBRAE: CENTRUM, BIFID					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
CERVICAL VERTEBRAE: CERVICAL RIB PRESENT AT 7TH CERVICAL VERTEBRA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
CERVICAL VERTEBRAE: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
THORACIC VERTEBRAE: ARCH, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
THORACIC VERTEBRAE: CENTRUM, UNILATERAL OSSIFICATION					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
THORACIC VERTEBRAE: HEMIVERTEBRA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
THORACIC VERTEBRAE: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
THORACIC VERTEBRAE: ARCHES, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
THORACIC VERTEBRAE: CENTRUM, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
THORACIC VERTEBRAE: 10 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
LUMBAR VERTEBRAE: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
LUMBAR VERTEBRAE: ARCHES AND CENTRUM, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
LUMBAR VERTEBRAE: 9 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
LUMBAR VERTEBRAE: CENTRUM, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
LUMBAR VERTEBRAE: 11 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SACRAL VERTEBRAE: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 ml of Group II test article (a resultant dose of 0.019% mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16b	18	19	20
FETUSES EVALUATED ^c	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1d
SACRAL VERTEBRAE: ARCH, OPEN					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SACRAL VERTEBRAE: ARCHES AND CENTRA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
CAUDAL VERTEBRAE: MISALIGNED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(5.3)	6(30.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	9(6.5)**
CAUDAL VERTEBRAE: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	10(7.2)**
CAUDAL VERTEBRAE: BIFID					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
CAUDAL VERTEBRAE: ARCH, OPEN					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
CAUDAL VERTEBRAE: HEMIVERTEBRA					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
CAUDAL VERTEBRAE: 8 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(8.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(8.0)	0(0.0)	3(2.2)**
CAUDAL VERTEBRAE: 2 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
CAUDAL VERTEBRAE: 7 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(8.0)	0(0.0)	1(0.7)
CAUDAL VERTEBRAE: 13 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
CAUDAL VERTEBRAE: 14 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
CAUDAL VERTEBRAE: 15 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(2.2)**
RIBS: FLAT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	1(5.3)	0(0.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	1(0.6)	0(0.0)
RIBS: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
RIBS: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
RIBS: 10 PRESENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
RIBS: SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
RIBS: BENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
RIBS: NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
MANUBRIUM: FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
STERNAL CENTRA: FUSED					
LITTER INCIDENCE	N(%)	4(25.0)	2(11.1)	2(10.5)	11(55.0)*
FETAL INCIDENCE	N(%)	4(3.2)	3(1.9)	6(3.7)	31(23.7)**
STERNAL CENTRA: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	1(6.2)	0(0.0)	2(10.5)	7(35.0)**
FETAL INCIDENCE	N(%)	1(0.8)	0(0.0)	3(1.8)	8(5.8)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe #919, which was dosed 7 ml of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 * Significantly different from the vehicle control group value (p<0.05).
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16 ^b	18	19	20
FETUSES EVALUATED ^c	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1 ^d

STERNAL CENTRA: ASYMMETRIC					
LITTER INCIDENCE	N(N)	0(0.0)	1(5.6)	0(0.0)	3(15.0)
FETAL INCIDENCE	N(N)	0(0.0)	1(0.6)	0(0.0)	3(2.2)
CLAVICULAE: NOT OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	8(5.8)**
CLAVICULAE: SMALL					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
SCAPULAE: ALA, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	1(5.3)	5(25.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	1(0.6)	14(10.1)**
SCAPULAE: ALA, SHORT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	7(35.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	24(17.3)**
SCAPULAE: ALA, NOT OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
SCAPULAE: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
SCAPULAE: SMALL					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**

SCAPULAE: NOT OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
SCAPULAE: BENT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORELIMB: FUSED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
FORELIMB: HUMERUS, NOT OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	7(5.0)**
FORELIMB: HUMERUS, BENT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
FORELIMB: HUMERUS, SHORT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	8(5.8)**
FORELIMB: HUMERUS, SMALL					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
FORELIMB: HUMERUS, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	6(4.3)**
FORELIMB: HUMERUS AND ULNA, FUSED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
FORELIMB: RADIUS, NOT OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	10(50.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	36(25.9)**
FORELIMB: RADIUS, SMALL					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	7(35.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	14(10.1)**
FORELIMB: RADIUS, SHORT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	7(5.0)**
FORELIMB: RADIUS, BENT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORELIMB: ULNA, SMALL					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	8(40.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	17(12.2)**
FORELIMB: ULNA, NOT OSSIFIED					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	9(45.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	26(18.7)**
FORELIMB: ULNA, SHORT					
LITTER INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(N)	0(0.0)	0(0.0)	0(0.0)	8(5.8)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP DOSAGE (MG/KG/DAY) ^a		I 0 (VEHICLE)	II 0.1	III 0.3	IV 1.0
LITTERS EVALUATED	N	16b	18	19	20
FETUSES EVALUATED ^c	N	123	158	163	140
LIVE	N	123	158	163	139
DEAD	N	0	0	0	1d

FORELIMB: ULNA, BENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
FORELIMB: ULNA, TWO SEGMENTS					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORELIMB: CARPALS, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	17(85.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	72(53.8)**
FORELIMB: METACARPALS, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	130(99.3)**
FORELIMB: METACARPALS, SMALL OR FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
FORELIMB: DIGITS, ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	130(99.3)**
FORELIMB: PHALANXES, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	130(99.3)**

PELVIS: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
PELVIS: ILIUM AND ISCHIUM, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(20.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(3.6)**
PELVIS: ILIUM, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(10.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
PELVIS: ILIUM, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	2(1.4)
PELVIS: ISCHIUM, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	9(6.5)**
PELVIS: PUBES, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	12(8.6)**
PELVIS: PUBES, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
PELVIS: PUBES, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)

HINDLIMB: INCOMPLETELY OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
HINDLIMB: FEMUR, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
HINDLIMB: FEMUR, BENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(35.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	16(11.5)**
HINDLIMB: FEMUR, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
HINDLIMB: FEMUR, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(35.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	12(8.6)**
HINDLIMB: FEMUR, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	7(35.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	16(11.5)**
HINDLIMB: FEMUR AND TIBIA, FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	13(9.4)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 ml of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Fetal skeletal alterations:

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY) a		0 (VEHICLE)	0.1	0.3	1.0
LITTERS EVALUATED	N	16b	18	19	20
FETUSES EVALUATED c	N	12j	158	163	140
LIVE	N	12j	158	163	139
DEAD	N	0	0	0	1d

HINDLIMB: TIBIA, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	8(40.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	16(11.5)**
HINDLIMB: TIBIA, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	11(7.9)**
HINDLIMB: TIBIA, IRREGULARLY SHAPED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(5.0)
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	1(0.7)
HINDLIMB: TIBIA, SHORT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	3(15.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	4(2.9)**
HINDLIMB: FIBULA, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	15(75.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	59(42.4)**
HINDLIMB: FIBULA, SMALL					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	6(30.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	28(20.1)**

HINDLIMB: TARSALS, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	17(85.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	83(59.7)**
HINDLIMB: METATARSALS, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	118(84.9)**
HINDLIMB: METATARSALS, SMALL OR FUSED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	5(25.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	9(6.5)**
HINDLIMB: DIGITS, ABSENT					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	135(97.1)**
HINDLIMB: PHALANGES, NOT OSSIFIED					
LITTER INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	20(100.0)**
FETAL INCIDENCE	N(%)	0(0.0)	0(0.0)	0(0.0)	137(98.6)**

a. Dosage occurred on days 6 through 18 of gestation.
 b. Excludes doe 9919, which was dosed 7 mL of Group II test article (a resultant dose of 0.0194 mg/kg/day) on day 10 of gestation.
 c. See the individual fetal alterations table (Table 22) for fetuses with multiple soft tissue alterations.
 d. Dead fetus was excluded from group averages and statistical analyses. Observations for this conceptus are cited on Table 22.
 ** Significantly different from the vehicle control group value (p<0.01).

(Excerpted from the sponsor's submission)

Prenatal and postnatal development: Not done

2.6.6.7 Local tolerance: Not done

2.6.6.8 Special toxicology studies: None

2.6.6.9 Discussion and Conclusions: Clofarabine demonstrated teratogenic effects in rats and rabbits. Similar adverse embryotoxic and teratogenic effects have been reported for other nucleoside analogues, such as cladribine (Teratology 2002; 66:6-18), 5-Aza-2'-deoxycytidine (Teratology 2002; 65:180-90), and gemcitabine (Teratology 1993; 48:365-81).

2.6.6.10 Tables and Figures: See within text and below.

2.6.7 TOXICOLOGY TABULATED SUMMARY

Study	Route	MTD		Target organs of toxicity
		mg/kg/day	mg/m ² /day	
Daily x 5 Rat (6 courses of treatment)	IV Bolus	12.5	75	Heart, eye, kidney, liver, WBC
Daily x 5 Dog (6 courses of treatment)	IV Bolus	0.75	15	Bone marrow, GI tract, lymphoid tissue, eye, testes

Study	Dose	Result		
		Positive control	No metabolic (-S9)	Plus metabolic (+S9)
Bacterial reverse mutation assay	75-5000 µg / plate	Positive	Negative	Negative
In vitro mammalian chromosome aberration assay	1-10 µg/mL	Positive	Positive	Positive
Mammalian erythrocyte micronucleus test	8.75-35 mg/kg	Positive	Positive	

Study	Route	Duration	High dose	Result
Embryofetal development, rat	IV bolus	DG 7 to 17	54 mg/m ² /day	Increased external, visceral, and skeletal anomalies (teratogenic)
Embryofetal development, rabbit	IV bolus	DG 6 to 18	12 mg/m ² /day	Increased malformation and variations (teratogenic)

Reviewer: Anwar Goheer, Ph.D.

NDA No. 21-673

OVERALL CONCLUSIONS AND RECOMMENDATIONS

Conclusions: Product is considered approvable from the pharmacology/toxicology point of view.

Unresolved toxicology issues (if any): None

Recommendations: This NDA is approvable from a pharmacology perspective.

Suggested labeling: A separate review will be conducted.



Reviewer Signature _____

Supervisor Signature _____  Concurrency Yes ___ No ___

APPENDIX/ATTACHMENTS: None

**This is a representation of an electronic record that was signed electronically and
this page is the manifestation of the electronic signature.**

/s/

Anwar Goheer
9/16/04 03:01:51 PM
PHARMACOLOGIST

John Leighton
9/20/04 02:26:46 PM
PHARMACOLOGIST