CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

21-881

PHARMACOLOGY REVIEW(S)

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: 21,881

SERIAL NUMBER: 000

DATE RECEIVED BY CENTER: July 10, 2005

DRUG NAME: Moviprep

INTENDED CLINICAL POPULATION: Patients undergoing colonoscopy,

SPONSOR: Norgine B.V.
United Kingdom

DOCUMENTS REVIEWED: Vol. 1-36

REVIEW DIVISION: Division of Gastroenterology Products (HFD-180)

PHARM/TOX REVIEWER: Ke Zhang, Ph.D.

PHARM/TOX SUPERVISOR: Jasti Choudary, B.V.Sc., Ph.D.

DIVISION DIRECTOR: Brian Harvey, M.D., Ph.D.

PROJECT MANAGER: Ms Tanya Clayton

Date of review submission to Division File System (DFS): February 24, 2006

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EXECUTIVE SUMMARY

I. Recommendations

A. Recommendation on approvability:

From a preclinical standpoint, approval of moviprep for bowel cleansing prior to colonoscopy is recommended.

B. Recommendation for nonclinical studies: None.

C. Recommendations on labeling:

Sponsor should be asked to revise the labeling as recommended.

II. Summary of nonclinical findings

A. Brief overview of nonclinical (toxicology) findings:

In the pre-NDA meeting with the sponsor held on July 28, 2004, the sponsor agreed to conduct 2-week repeated dose toxicity studies with moviprep in rodents and nonrodents. In addition, the Division agreed that the sponsor will submit the following studies with movicol: 90-day oral toxicity studies in rats and dogs, genotoxicity studies and Segment I fertility and reproductive toxicity study in rats, and Segment II teratology studies in rats and rabbits.

The toxicity profiles of moviprep were characterized in 2-week oral toxicity studies in rats and dogs. The toxicity profiles of movicol were characterized in 90-day oral toxicity studies in rats and dogs. The results indicated that the kidney was the target organ of toxicity in rats based on the changes of the clinical chemistry and the kidney weight. In dogs, the major treatment related toxicity was decreased terminal body weight gain, emesis, diarrhea, and salvation. The results suggested that the gastrointestinal tract was the target organ of toxicity in dogs based on the clinical signs of toxicity including emesis, diarrhea and salvation. The toxicity profiles of moviprep and movicol were similar.

Movicol was not teratogenic in the Segment II teratology studies in rats and rabbits.

Movicol was not genotoxic in the Ames test, the mouse lymphoma cell (L5178Y $TK^{+/-}$) forward mutation assay at tk locus, and the mouse micronucleus test.

Both sodium ascorbate and ascorbic acid were not mutagenic in the mouse lymphoma cell (L5178Y $TK^{+/-}$) forward mutation assay at tk locus (Cancer Letters, 14:151-158, 1981). Ascorbate induced a dose-dependent increase in sister-chromatid exchanges in Chinese hamster ovary cells and in human lymphocytes and increased the inhibition of DNA synthesis in Hela cells (Mutation Research, 60:321-327, 1979). Ascorbate induced mutation at HGPRT locus in Chinese hamster cells (Cancer Letters, 8:299-305, 1980).

A NTP study report (NTP-81-140) of carcinogenesis bioassays with L-ascorbic acid indicatd that L-ascorbic acid was not carcinogenic in mice and in rats.

B. Pharmacologic activity:

Moviprep contains polyethylene glycol 3350, sodium sulfate, sodium chloride, potassium chloride, sodium ascorbate, and ascorbic acid. All these ingredients contribute to the overall osmolarity of moviprep and thus are considered as the active ingredients. The sponsor did not conduct any in vivo pharmacology studies with moviprep. However, it is expected that PEG 3350 along with other ingredients in moviprep can increase the water content of the stool and produce a voluminous liquid stool when given orally.

C. Nonclinical safety issues relevant to clinical use: None.

PHARMACOLOGY/TOXICOLOGY REVIEW

2.6.1 INTRODUCTION AND DRUG HISTORY

NDA number: 21,881
Review number: 01
Sequence number/date/type of submission: July 10, 2005
Information to sponsor: Yes ( ) No (x)
Sponsor and/or agent: Norgine B.V.
United Kingdom

Manufacturer for drug substance:

Manufacturer for Pouch A and Pouch B

Norgine B.V.
United Kingdom

Reviewer name: Ke Zhang
Division name: Division of Gastrointestinal and Coagulation Drug Products
HFD #: 180
Review completion date: February 24, 2006

Drug: Moviprep for oral solution.

Moviprep contains polyethylene glycol 3350 (PEG 3350), sodium sulfate, sodium chloride, potassium chloride, aspartame, acesulfame potassium, lemon flavor, sodium ascorbate and ascorbic acid.

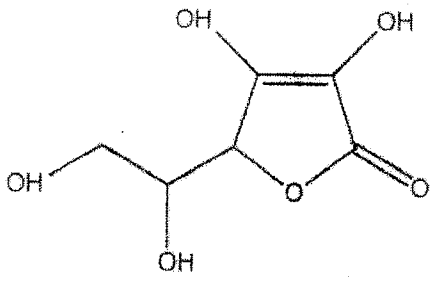
The structure, molecular formula and molecular weight of PEG 3350 are presented below.

Structural formula: H - (O - CH_2 - CH_2)$_n$ - OH

Where 'n' represents the average number of oxyethylene groups

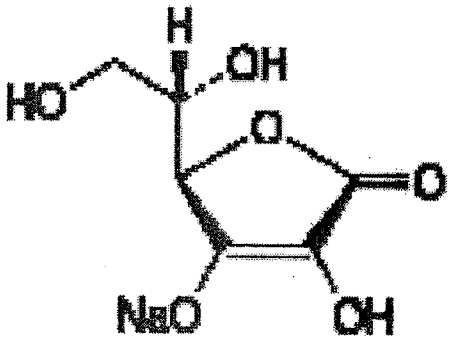
Molecular formula:	$H(C_2H_4O)_nOH$
Average molecular weight:	3350 (n=76)

The structure, molecular formula and molecular weight of ascorbic acid are presented below



Molecular formula:	$C_6H_8O_6$
Molecular weight:	176.12

The structure, molecular formula and molecular weight of sodium ascorbate are presented below



Molecular formula: $C_6H_7NaO_6$

Molecular weight: 198.12.

Relevant INDs/NDAs/DMFs: IND 63,268 and IND 67,947

Drug class: Bowel cleansing agent for use prior to colonoscopy,

Intended clinical population: Patients undergoing colonoscopy,

Indication: Moviprep is indicated for bowel cleansing prior to colonoscopy

Clinical formulation:

Table 1: Qualitative and Quantitative Composition of MOVIPREP Pouch A

Component	Reference to Quality Standard	Function	Quantity per Pouch (g)
Polyethylene Glycol 3350, NF	NF	Active Ingredient	100.00
Sodium Sulfate ——— , USP	USP	Active Ingredient	7.500
Sodium Chloride, USP	USP	Active Ingredient	2.691
Potassium Chloride, USP	USP	Active Ingredient	1.015
Aspartame, NF	NF	Sweetener	———
Acesulfame Potassium, NF	NF	Sweetener	———
Lemon Flavor ———	In-house standard	Flavor	———
Total			111.896

Table 1: Qualitative and Quantitative Composition of MOVIPREP Pouch B

Component	Reference to Quality Standard	Function	Quantity per Pouch (g)
Ascorbic Acid, USP	USP	Active Ingredient	4.700
Sodium Ascorbate, USP	USP	Active Ingredient	5.900
Total			**10.600**

Moviprep solution is prepared by emptying the contents of one pouch A and one pouch B into a container and adding one liter of lukewarm water into the container. After consuming the first liter, the procedure should be repeated with the second pouch A and pouch B. The recommended dose for adults is 2 liters of the moviprep solution.

Route of administration: Oral solution.

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

Data reliance: Except as specifically identified below, all data and information discussed below and necessary for approval of NDA 21,881 are owned by Norgine B.V.or are data for which Norgine B.V. has obtained a written right of reference. Any information or data necessary for approval of NDA 21,881 that Norgine B.V. does not own constitutes published literature.

Studies reviewed within this submission:

Type of Study	Study #	Lot #	lab	Page #
Pharmacology				9
Pharmacokinetics: None				12
Subacute/subchronic/chronic Toxicity:				
2-week oral toxicity study with moviprep in rats	18290/04	D02	1	15
90-day oral toxicity study with movicol in rats	17519/03	112803	1	18
2-week oral toxicity study with moviprep in dogs	18291/04	D02	1	31
90-day oral toxicity study with movicol in dogs	17520/03	112803	1	34
Genetic Toxicity:				
Movicol:				
Ames test	22722	27428	1	41
Mouse lymphoma cell (L5178Y/TK$^{+/-}$) forward mutation test at TK locus	22825	27428	1	49
Mouse micronucleus test with H376/95	22924	27428	1	52
Ascorbate:				
Mouse lymphoma mutation test in L5178Y TK+/- cells	published report	———		54
DNA synthesis inhibition and sister-chromatid exchange tests	published report	———		57
Mutation test at HGPRT locus in Chinese hamster ovary cells	published report	———		59
Reproductive Toxicity:				
Movicol:				
Oral segment I fertility and reproductive study in rats	17521/03	112903	1	78
Oral segment II teratology study in rats	17522/03	112903	1	86
Oral segment II teratology study in rabbits	17524/03	112903	1	103
Carcinogenicity:				
PEG 4000 A 2-year dietary carcinogenicity study in rats	published report	———		60
Ascorbate 2-year dietary carcinogenicity studies in mice and rats	published report	———		62

1 =

2.6.2 PHARMACOLOGY

2.6.2.1 Brief summary

Moviprep contains polyethylene glycol 3350, sodium sulfate, sodium chloride, potassium chloride, sodium ascorbate, and ascorbic acid. All these ingredients contribute to the overall

osmolarity of moviprep and thus are considered as the active ingredients. The sponsor did not conduct any in vivo pharmacology studies with moviprep. However, it is expected that PEG 3350 along with other ingredients in moviprep can increase the water content of the stool and produce a voluminous liquid stool when given orally.

2.6.2.2 Primary pharmacodynamics

Mechanism of action:

Moviprep contains polyethylene glycol 3350, sodium sulfate, sodium chloride, potassium chloride, sodium ascorbate, and ascorbic acid. The sponsor determined the osmolarity of each of these ingredients in moviprep. The results were summarized in Table 6 on page 14 in Volume 1.4. This table is attached below:

Table 6: Comparison of Theoretical and Measured Osmolarity of MOVIPREP® and its Components (Ref. 1)

	Theoretical Osmolarity mOsm/L	Measured Osmolarity (Norgine Ltd) mOsm/L	Measured Osmolarity (Pilot Data) mOsm/L
PEG 3350			
Sodium Sulphate			
Sodium Chloride			
Potassium Chloride			
Acesulfame K			
Aspartame			
Lemon Flavour			
Sum Sachet A			
Sachet A			
Ascorbic Acid			
Sodium Ascorbate			
Sum Sachet B			
Sachet B			
Sum of Sachet A and B combined			
Sachet A and B combined			

NP = Not performed

It appears that PEG 3350, sodium sulfate, sodium chloride, potassium chloride, sodium ascorbate, and ascorbic acid can contribute to the overall osmolarity of moviprep and thus are

considered as the active ingredients. [redacted] The results indicated that [redacted] The results were presented in Table 1 on page 11 in Volume 1.2 and this table is attached below.

TABLE 1
COMPARISON OF MEASURED VS. CALCULATED OSMOLARITY

Ingredients in Solution[1] (at MOVIPREP levels)	Measured Osmolarity (average mOsm/L)	Calculated Theoretical Osmolarity (mOsm/L)
PEG 3350	[redacted]	[redacted]
PEG 3350 and Na_2SO_4	[redacted]	[redacted]
PEG 3350, Na_2SO_4 and NaCl	[redacted]	[redacted]
PEG 3350, Na_2SO_4, NaCl and KCl	[redacted]	[redacted]

The sponsor did not conduct any in vivo pharmacology studies with moviprep.

2.6.2.3 **Secondary pharmacodynamics:** None

2.6.2.4 **Safety pharmacology: None**

2.6.2.5 **Pharmacodynamic drug interactions:** None

2.6.4 **PHARMACOKINETICS/TOXICOKINETICS**

No pharmacokinetic studies were conducted with moviprep. Toxicokinetic data with moviprep or movicol were presented with the toxicity studies.

The toxicokinetic results of the 2-week oral toxicity study in rats with moviprep indicated that the plasma AUC level of ascorbic acid was approximately 105, 113, and 200 µg.hr/ml (males) or 47, 48, and 107 µg hr/ml (females) on day 1 following oral doses of 5, 10, and 20 g/kg/day, respectively and the plasmal level of ascorbic acid was declined with a terminal half life ranging from approximately 5 to 34 hours.

The toxicokinetic results of the 2-week oral toxicity study in dogs with moviprep indicated that the plasma AUC level of ascorbic acid was approximately 161, 201, and 230 µg.hr/ml (males) or 168, 220, and 235 µg.hr/ml (females) on day 1 following oral doses of 5, 10, and 20 g/kg/day, respectively and the plasmal level of ascorbic acid was declined with a terminal half life ranging from approximately 4 to 20 hours.

2.6.6 TOXICOLOGY

2.6.6.1 Overall toxicology summary:

The toxicity profiles of moviprep were characterized in 2-week oral toxicity studies in rats and dogs. The toxicity profiles of movicol were characterized in 90-day oral toxicity studies in rats and dogs. The results indicated that the kidney was the target organ of toxicity in rats based on the changes of the clinical chemistry and the kidney weight. In dogs, the major treatment related toxicity was decreased terminal body weight gain, emesis, diarrhea, and salvation. The results suggested that the gastrointestinal tract was the target organ of toxicity in dogs based on the clinical signs of toxicity including emesis, diarrhea, and salvation. The toxicity profiles of moviprep and movicol were similar.

Movicol was not teratogenic in the Segment II teratology studies in rats and rabbits.

Movicol was not genotoxic in the Ames test, the mouse lymphoma cell (L5178Y $TK^{+/-}$) forward mutation assay at tk locus, and the mouse micronucleus test.

Both sodium ascorbate and ascorbic acid were not mutagenic in mouse lymphoma L5178Y TK+/- cell mutation assay (Cancer Letters, 14:151-158, 1981). Ascorbate induced a dose-dependent increase in sister-chromatid exchanges in Chinese hamster ovary cells and in human lymphocytes and increased the inhibition of DNA synthesis in Hela cells (Mutation Research, 60:321-327, 1979). Ascorbate induced mutation at HGPRT locus in Chinese hamster cells (Cancer Letters, 8:299-305, 1980).

A NTP study report (NTP-81-140) of carcinogenesis bioassays with L-ascorbic acid indicatd that L-ascorbic acid was not carcinogenic in mice and in rats.

2.6.6.2 **Single-dose toxicity: None.**

2.6.6.3 **Repeat-dose toxicity**

Some toxicity studies were conducted with movicol. Movicol does not contain ascorbic acid and sodium ascobate. The formulations of moviprep and movicol are listed below.

Table 1: Qualitative and Quantitative Composition of MOVIPREP Pouch A

Component	Reference to Quality Standard	Function	Quantity per Pouch (g)
Polyethylene Glycol 3350, NF	NF	Active Ingredient	100.00
Sodium Sulfate ——— USP	USP	Active Ingredient	7.500
Sodium Chloride, USP	USP	Active Ingredient	2.691
Potassium Chloride, USP	USP	Active Ingredient	1.015
Aspartame, NF	NF	Sweetener	———
Acesulfame Potassium, NF	NF	Sweetener	———
Lemon Flavor ———	In-house standard	Flavor	———
Total			111.896

Table 1: Qualitative and Quantitative Composition of MOVIPREP Pouch B

Component	Reference to Quality Standard	Function	Quantity per Pouch (g)
Ascorbic Acid, USP	USP	Active Ingredient	4.700
Sodium Ascorbate, USP	USP	Active Ingredient	5.900
Total			**10.600**

Appears This Way
On Original

Table 3.5.1 MOVICOL® Formulation

Ingredient	Amount
Polyethylene Glycol 3350 NF	[redacted]
Sodium Chloride USP	[redacted]
Sodium Bicarbonate USP	[redacted]
Potassium Chloride USP	[redacted]
Acesulfame potassium	[redacted]
Lemon-lime flavoring	[redacted]

RAT:

Two-Week Oral Toxicity Study of Moviprep in Rats
(18290/04)

Testing Laboratory: [redacted]

Study Start and Completion Dates: August 23, 2004 and April 12, 2005

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulations and a quality assurance statement.

Animals: Male (150-190 g, ~5 weeks old)
Female (121-162 g, ~5 weeks old)
CD/ [redacted] :CD/ [redacted] rats

Drug Batch No.: D02

Methods: To assess the repeated dose toxicity of moviprep in rats, moviprep was given by oral gavage to rats (10/sex/group) at 0, 5, 10, and 20 g/kg/day (30 ml/kg). Moviprep was dissolved in the tap water. The dose selection was based on the results of the 90-day oral toxicity study with movicol (study #17519/03). In this study, the high dose of 60/50 g/kg/day was lethal. The terminal body weight gain was decreased by 18% and 26% in the mid (40 g/kg/day) and high (60/50 g/kg/day) dose

males, respectively. In the current study, clinical signs of toxicity and mortality were observed daily. Body weights were recorded weekly. Food consumption was recorded weekly. Ophthalmological examination was performed at termination. Hematology, clinical chemistry and urinalysis were performed at termination. The following organs were weighed at necropsy for all animals: adrenals, brain, heart, kidneys, liver, lungs, lymph nodes, ovaries, pituitary, spleen, testes, thyroids, and thymus. Histopathological examination was performed on the following tissues from all animals: adrenals, aorta, bone, bone marrow, brain, cecum, epididymides, eyes, heart, ileum, intestine, kidneys, liver, lungs, lymph nodes, mammary gland, esophagus, sciatic nerves, ovaries, pancreas, parathyroids, pituitary, prostate, salivary gland, seminal vesicles, skeletal muscle, skin, spinal cord, spleen, stomach, testes, thymus, thyroids, trachea, urinary bladder, uterus, uterus, vagina, and togue. Plasma levels of ascorbic acid were determined on days 1 and 14 in the satellite animals at pre-dose, 0.5, 1, 2, 4, and 8 hours after dosing.

Results:

1. Clinical Signs of Toxicity: Soft feces were noted in the high dose animals.

2. Mortality: Two high dose animals (one male and one female) were found dead on days 12 and 13. The deaths were considered as treatment related but the cause for the deaths was not known.

3. Body Weight: The average initial and final body weights of the control animals were 168 g and 255 g (males) or 146 g and 192 g (females), respectively. There were no clear treatment related changes.

4. Food Consumption: The average food consumption of the control animals were 91-101 g/kg/day (males) or 110-114 g/kg/day (females). The average food consumption of the control animals were 19-21.4 g/animal/day (males) or 18.6-19.3 g/animal/day (females) based on the average body weight of 0.212 kg for males and 0.169 kg for females. Food consumption was lower in the mid and high dose groups than that in the control. The results were presented in a table on page 48 in Volume 1.7 and this table is attached below.

Changes of food consumption compared to the Control in %				
Test week	Group 3 (10 g/kg)		Group 4 (20 g/kg)	
	males	females	males	females
1	-13**	none	-15**	-17**

** statistically significant at $p \leq 0.01$

5. Ophthalmology: There were no treatment-related eye lesions.

6. Hematology: There were no treatment related changes

7. Clinical Chemistry: Slight increase in bilirubin, urea, and alanine aminotransferase (ALAT) was noted in the high dose group. The results were summarized in a table on page 51 in Volume 1.7 and this table is attached below.

Changes in biochemical parameters compared to the Control group on Test Day 15				
Parameter	Difference to the Control in [%]			
	Group 3 (10 g/kg)		Group 4 (20 g/kg)	
	males	females	males	females
Bilirubin (total)	none	none	+15**	none
Chloride	none	-3**	none	-3**
Potassium	-6	-4	-9	-10
Urea	+18	none	+58**	+59**
ALAT	none	none	+41**	none

** = statistically significant at $p \leq 0.01$

8. Urinalysis: Slight increase in specific gravity was noted in the mid and high dose groups.

9. Organ Weights: Increase in the kidey weight was noted in the high dose group. The results were summarized in a table on page 54 in Volume 1.7 and this table is attached below.

Organ	Difference to the Control [%]	
	Group 4 (20 g/kg) males	
	rel.	abs.
Kidney, left	+ 10	+ 9
Kidney, right	+ 14**	+ 12

** = statistically significant at $p < 0.01$

10. Gross pathology: There were no treatment related changes

11. Histopathology: There were no treatment related changes

12. Toxicokinetics: The plasma level of ascorbic acid was summarized in a table on page 56 in Volume 1.7 and this table is attached below.

Ascorbic acid (mean values per group)						
Group	Test day	C_{max} [µg/mL]	T_{max} [h]	$AUC_{0-8\,h}$ [µg*h/mL]	$AUC_{0-8\,h}$/dose [µg*h/mL/(mg/kg/day)]	$t_{1/2}$ [h]
Males						
2	1	14.87	4.00	104.68	20.94	16.50
	14	14.10	4.00	96.86	19.37	20.96
3	1	15.63	4.00	112.58	11.26	33.75
	14	16.83	4.00	112.74	11.27	11.29
4	1	33.63	2.00	199.89	9.99	4.65
	14	24.93	0.50	156.60	7.83	12.95
Females						
2	1	6.92	4.00	46.81	9.36	11.17
	14	5.64	2.00	42.96	8.59	27.79
3	1	6.95	4.00	47.61	4.76	10.91
	14	6.45	4.00	46.45	4.64	24.40
4	1	18.70	2.00	106.46	5.32	6.83
	14	10.60	0.50	87.74	4.39	7.98

The results indicated that the plasma level of ascorbic acid was increased with the dose and not accumulated over time during the 2-weeks of treatment.

In summary, moviprep was tested orally in rats at 0, 5, 10, and 20 g/kg/day for 2 weeks. Major treatment related changes were observed mainly at the high dose of 20 g/kg/day. These included mortality, increase in bilirubin, urea, and kidney weight. The kidney was the target organ of toxicity based on the changes of the clinical chemistry and kidney weight (no treatment related histopathological changes in the kidney).

90-day Oral Toxicity Study of Movicol in Rats
(17519/03)

Testing Laboratory: [redacted]

Study Start and Completion Dates: February 10, 2004 and April 29, 2005

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulations and a quality assurance statement.

Animals: Male (136-171 g, ~5 weeks old)
Female (125-155 g, ~5 weeks old)
CD/[redacted]:CD/[redacted] rats

Drug Batch No.: 112803

Methods: To assess the repeated dose toxicity of movicol in rats, movicol was given by oral gavage to rats (15/sex/group) at 0, 5, 20, and 30/25 g/kg twice a day (10, 40, and 60/50 g/kg/day) for 90 days. The high dose of 60 g/kg/day was reduced to 50 g/kg/day (25 g/kg twice a day) from day 22 onwards due to increased mortality in this group. Movicol was dissolved in tap water and 40 ml/kg was given per administration (80 ml/kg/day). The dose selection was based on the results of a 7-day dose-ranging study (study #17198/03). In this study, the high dose of 60 g/kg/day was maximum feasible dose In the current study, clinical signs of toxicity and mortality were observed daily.

Body weights were recorded weekly. Food consumption was recorded weekly. Ophthalmological examination was performed at the end of week 4. Hematology, clinical chemistry, and urinalysis were performed at the end of week 4 and at termination. The following organs were weighed at necropsy for all animals in the control and high dose group and all prematurely deceased animals: adrenals, brain, heart, kidneys, liver, lungs, lymph nodes, ovaries, pituitary, spleen, testes, thyroids, and thymus. Histopathological examination was performed on the following tissues for all animals: adrenals, aorta, bone, bone marrow, brain, cecum, epididymides, eyes, heart, ileum, intestine, kidneys, liver, lungs, lymph nodes, mammary gland, esophagus, sciatic nerves, ovaries, pancreas, parathyroids, pituitary, prostate, salivary gland, seminal vesicles, skeletal muscle, skin, spinal cord, spleen, stomach, testes, thymus, thyroids, trachea, urinary bladder, uterus, uterus, vagina, and togue. Some organs or tissues were also examined histopathologically in all animals in the low and mid dose groups. Plasma levels of low molecular weight PEG were determined on days 1, 28, and 90 in the satellite animals at pre-dose, 0.5, 1, 2, 4, and 6 hours after dosing.

Results:

1. Clinical Signs of Toxicity: Soft or liquid feces were noted in the treated animals. Enlarged abdomen and reduced motility were noted in the mid and high dose groups. Pilo-erection was also ntoed in the high dose animals.

2. Mortality: Nine males and eight females in the high dose group were found dead during the study. The deaths were considered as treatment. Most of the prematurely terminated animals had dilated stomach and intestinal tract filled with yellowish liquid. The sponsor stated that the deaths were most likely due to the dilation of the lumen of gastrointestinal tract and the stress associated with dosing.

3. Body Weight: The average initial and final body weights of the control animals were 154.6 g and 441.8 g (males) or 139.6 g and 282.5 g (females), respectively. The terminal body weight gain was decreased by ~18% and 26% in the mid and high dose males as compared to the control.

4. Food Consumption: The average food consumption of the control animals were 50-103 g/kg/day (males) or 72-126 g/kg/day (females). The average food consumption of the control animals were 15-31 g/animal/day (males) or 15-26.5 g/animal/day

(females) based on the average body weight of 0.3 kg for males and 0.21 kg for females. Food consumption was lower in the mid and high dose groups than that in the control during the first week of the study. The results were presented in a table on page 50 in Volume 1.14 and this table is attached below.

Changes in food consumption compared to Control in %				
Test week	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
1	13**	17**	-31**	-38**

** statistically significant at $p \leq 0.01$

5. Ophthalmology: There were no treatment related changes.

6. Hematology: Treatment with movicol increased erythrocytes and hemoglobin levels and hematocrit, decreased levels of leucocytes and lymphocytes. The results were presented in tables on pages 53 and 54 in Volume 1.14. These tables are attached below.

Changes in haematological parameters compared to the Control group in test week 4				
Parameter	Difference to the Control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Haemoglobin content	+9**	none	+14**	none
Erythrocytes	+9**	none	+11**	none
Haematocrit value	+8**	none	+15**	none
Leucocytes	-27	-10	-36	-7
Neutrophilic granulocytes	+211	+57	+269	+61
Lymphocytes	-22	-11	-28	12
Monocytes	+58	+24	+89	+51

** = statistically significant at $p \leq 0.01$

Changes in haematological parameters compared to the Control group in test week 13				
Parameter	Difference to the Control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Haemoglobin content	+11**	+9**	+19**	+18**
Erythrocytes	+9**	+8**	+14**	+15**
Haematocrit value	+10**	+7**	+21**	+19**
Leucocytes	-31**	none	-58**	-22
Neutrophilic granulocytes	+53	+92	+165	+181
Lymphocytes	-12	-17	-39	-34
Monocytes	+35	none	+118	+45

** = statistically significant at $p \leq 0.01$

7. Clinical Chemistry: Increase in bilirubin, creatinin, urea, glucose, total protein, calcium, chloride, potassium, sodium,

and alanine aminotransferase (ALAT) was noted in the mid and high dose groups. Some of these changes were observed at 4 week but not at 13 week. The results were summarized in tables on pages 56 and 57 in Volume 1.14 and these tables are attached below.

Changes in biochemical parameters compared to the Control group in test week 4

Parameter	Difference to the Control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Bilirubin (total)	+26**	none	+36**	none
Creatinine	+7	none	+11**	none
Glucose	+18	none	+30**	none
Protein (total)	+11**	+11**	+16**	+6**
Urea	+115**	+59**	+153**	+74**
Calcium	none	+5**	+4**	+7**
Chloride	+6**	+4	+7**	+4**
Potassium	-10	-14**	-14**	-11**
Sodium	+4**	+3**	+6**	+4**
ALAT	+18	none	+31**	none
ASAT	+19**	none	+15	none

** = statistically significant at $p \leq 0.01$

Changes in biochemical parameters compared to the Control group in test week 13

Parameter	Difference to the Control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Glucose	none	none	+19	+16
Protein (total)	+8**	+11**	+16**	+16**
Urea	+80**	+51**	+157**	+89**
Calcium	none	-5**	+7**	+10**
Chloride	+4	-3**	+13**	+10**
Potassium	none	-13**	none	-17**
Sodium	+3	+3**	+10**	+9**
ASAT	+28**	none	+26**	none

** = statistically significant at $p \leq 0.01$

8. Urinalysis: Increase in specific gravity, urine pH and volume was noted in the mid and high dose groups. The results were presented in tables on page 58 in Volume 1.14. These tables are attached below.

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Changes in urinary parameters compared to the Control group in test week 4				
Parameter	Difference to the Control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Specific gravity	+5**	+4**	+5**	+4**
pH	+8**	+16**	+9**	+16**
Urine volume	+199**	+138**	+195**	+186**

** = statistically significant at $p \leq 0.01$

Changes in urinary parameters compared to the Control group in test week 13				
Parameter	Difference to the Control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Specific gravity	+4**	+4**	+3**	+4**
pH	+6**	+18**	+6**	+17**
Urine volume	+241**	+386**	+302**	+379**

** = statistically significant at $p \leq 0.01$

9. Organ Weights: Increase in the kidey and adrenal weights and decrese in the spleen and thymus weights were noted in the mid and high dose groups. The results were summarized in a table on page 61 in Volume 1.14 and this table is attached below.

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Organ	Difference to the Control [%]							
	Group 3 (40 g/kg)				Group 4 (60/50 g/kg)			
	males		females		males		females	
	rel.	abs.	rel.	abs.	rel.	abs.	rel.	abs.
Adrenal, left	+17	none	none	none	+35**	+12	+22	+25
Adrenal, right	+25	none	none	none	+39**	+14	+7	+8
Kidney, left	+17**	none	+14**	+18	+21**	none	+16**	+18
Kidney, right	+18**	none	+11**	+15	+19**	none	+15**	+17
Spleen	-19**	-30	-13	-10	-30**	-43	-30**	-28
Thymus	-27	-37	-25**	-24	-25	-38	-39**	-38

** = statistically significant at $p \leq 0.01$

10. Gross pathology: Enlarged cecum and dilated intestal tract were noted in all treatment groups and the incidence was dose dependent. Small spleen and thymus were found in the high dose animals. Most of the prematurely terminated animals had dilated stomach and intestinal tract filled with yellowish liquid.

11. Histopathology: A number of histopathological changes were identified in the control and treatment groups. The results were presented in the following table.

Histopathological changes (n=15 per group)

	Control		10 g/kg		40 g/kg		60/50 g/kg	
	Male	Female	male	female	Male	female	Male	Female
Adrenal: contical hypertrophy/hyperplasia	2	3	2	0	0	0	4	7
Brain stem: congension	0	0	0	0	0	0	0	0
Kidney: fatty infiltration (tubular epithelium)	3	11	3	9	10	12	9	9
dilatation of tubule	0	0	0	0	2	4	0	0
Cecum: Dilation	0	0	0	1	10	12	12	11
Thin layer of mucosa	0	0	0	0	0	1	7	7
Liver: Hepatocellular atrophy	0	0	0	0	0	0	4	4
Lympho-histoc. infiltr	8	4	5	3	7	3	2	2
Lung: Granuloma	4	2	0	0	0	0	3	1
Peribronch./perivasc. infla	1	3	0	0	0	0	4	2
Lymph node (cervical): Reduction of cellularity	0	0	0	2	1	1	2	5
Lymphoid hyperplasia	10	11	11	9	12	10	5	4
Increase of macrophages	0	1	0	1	3	3	8	5

Seminal vesicles: inactivation	1	-	0	-	0	-	1	-
Ileum:								
Thin layer of mucosa	0	0	0	0	0	0	3	2
Lymphoid hyperplasia	14	14	8	11	5	5	0	5
Spleen:								
Reduction of cellularity	2	0	0	0	1	4	10	5
Stomach:								
Thin layer of mucosa	0	0	0	0	0	0	1	2
Thymus:								
Hemorrhage	0	0	0	1	1	1	1	2
Increase of macrophages	0	0	1	0	0	0	4	5

Increased incidences of thin layer of mucosa in the gastrointestinal tract, reduced cellularity in the lymph node and spleen, and increased macrophages in the lymph nodes and thymus were noted in the high dose group. A few cases (1 or 2) of hemorrhages in the thymus were found in the treatment groups (none in the control group). The incidences of all other changes were not clearly dose dependent. All prematurely terminated animals had a mild to moderate congestion with hemorrhage in the cerebrum, cerebellum, and brain stem (none in the control animals and the terminal sacrificed animals in the treatment groups).

12. Toxicokinetics: PEG 3350 is a heterogenous group of PEG oligomers. Sponsor determined the plasma levels of lower molecular weight PEGs including oligomers 11, 13, 16, and 18 since these oligomers are more likely absorbed. Oligomers 11, 13, 16, and 18 represent PEGs with molecular weight ranging approximately 500 to 800. The results were presented in tables on pages 66-68 in Volume 1.14. These tables are attached below.

Oligomer 11
(mean values per group)

Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{8-12}#3 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{8-12}#3 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
3	1	0.550	2.0	3.09#2	-#4	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
4	1	0.610	0.5	3.15	12.20	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	0.860	2.0	3.03	5.55	-	-	-	-

#: AUC_{0-4} calculated for test day 90 only for the high dose group
#2: AUC_{0-6} calculated for test day 1 #3: AUC_{8-12} calculated for test days 28 and 90
#4: $t_{1/2}$-value not detectable; the calculated AUC-value is equivalent to the AUC_{0-4}-value given in the table

Oligomer 13
(mean values per group)

Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{8-12}#3 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{8-12}#3 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
3	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
4	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-

#: AUC_{0-4} calculated for test day 90 only for the high dose group
#2: AUC_{0-6} calculated for test day 1
#3: AUC_{8-12} calculated for test days 28 and 90

Oligomer 16
(mean values per group)

Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{0-12}#3 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{0-12}#3 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
3	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
4	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	0.720	4.0	2.71#4	#4	-	-	-	-

#: AUC_{0-4} calculated for test day 90 only for the high dose group
#2: AUC_{0-6} calculated for test day 1
#3: AUC_{0-12} calculated for test days 28 and 90
#4: $t_{1/2}$-value not detectable; the calculated AUC-value is equivalent to the AUC-value given in the table

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Oligomer 18
(mean values per group)

Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{0-12}#3 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-4}#1 / AUC_{0-6}#2 / AUC_{0-12}#3 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	-	-	-	-	-	-	-	-
3	1	1.010	0.5	3.76	2.90	-	-	-	-
	28	-	-	-	-	-	-	-	-
	90	1.110	2.0	7.61	1.74	-	-	-	-
4	1	4.510	0.5	6.03	1.33	4.000	0.5	5.44	1.54
	28	1.070	2.0	7.30	3.95	-	-	-	-
	90	2.100	2.0	7.91#4	-#4	0.560	2.0	2.13#4	-#4

#1: AUC_{0-4} calculated for test day 90 only for the high dose group
#2: AUC_{0-6} calculated for test day 1
#3: AUC_{0-12} calculated for test days 28 and 90
#4: $t_{1/2}$-value not detectable; the calculated AUD value is equivalent to the AUC-value given in the table

In summary, movicol was tested orally in rats at 0, 5, 20, and 30/25 g/kg twice a day (10, 40, and 60/50 g/kg/day) for 90 days. The high dose was lethal. Decreased body weight gain was noted in the mid and high dose males. Serum levels of bilirubin and creatinine were increased in the mid and high dose males in week 4 but not in week 13. The kidney weight was increased in the mid and high dose groups. Increased incidences of thin layer of mucosa in the gastrointestinal tract, reduced cellularity in the lymph node and spleen, and increased macrophages in the lymph nodes and thymus were noted in the high dose group. A few cases (1 or 2) of hemorrhages in the thymus were found in the treatment groups (none in the control group). These results indicated that these animals had a marked stress due to receiving a large volume of test drug for a long period time (90 days).

DOG:

Two-Week Oral Toxicity Study of Moviprep in Dogs
(18291/04)

Testing Laboratory: [redacted]

Study Start and Completion Dates: September 15, 2004 and April 13, 2005

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulations and a quality assurance statement.

Animals: Male (6.5-8.9 kg, 5.5-6 months old)
Female (5-6.6 kg, 5.5-6 months old)
Beagle dogs

Drug Batch No.: D02

Methods: To assess the repeated dose toxicity of moviprep in dogs, moviprep was given by oral gavage to dogs (3/sex/group) at 0, 5, 10, and 20 g/kg/day (40 ml/kg/day). Moviprep was dissolved in tap water. The dose selection was based on the results of 90-day oral toxicity study with movicol in dogs. In this study, emesis, diarrhea, and salvation were noted at doses of 10 g/kg/day or higher and decreased terminal body weight gain at high dose of 60/50 g/kg/day. The dose of 60 g/kg/day was maximum feasible dose. In the current study, clinical signs of toxicity and mortality were observed daily. Body weights were recorded weekly. Food consumption was recorded weekly. Ophthalmological examination was performed at termination. ECG and blood pressure were determined on day 1 and at termination. Hematology, clinical chemistry and urinalysis were performed at termination. The following organs were weighed at necropsy for all animals: adrenals, brain, heart, kidneys, liver, lungs, lymph nodes, ovaries, pituitary, spleen, testes, thyroids, and thymus. Histopathological examination was performed on the following tissues for all animals: adrenals, aorta, bone, bone marrow, brain, cecum, epididymides, eyes, heart, ileum, intestine, kidneys, liver, lungs, lymph nodes, mammary gland, esophagus, sciatic nerves, ovaries, pancreas, parathyroids, pituitary, prostate, salivary gland, seminal vesicles, skeletal muscle, skin, spinal cord, spleen, stomach, testes, thymus,

thyroids, trachea, urinary bladder, uterus, uterus, vagina, and togue. Plasma levels of ascorbic acid were determined on days 1 and 14 in the satellite animals at pre-dose, 0.5, 1, 2, 4, and 8 hours after dosing.

Results:

1. Clinical Signs of Toxicity: Emesis and soft or liquid feces were noted in the treated animals. Salvation was also noted in the high dose group.

2. Mortality: There were no deaths.

3. Body Weight: The average initial and final body weights of the control animals were 7.46 kg and 7.42 kg (males) or 5.62 kg and 5.68 kg (females), respectively. There were no clear treatment related changes.

4. Food Consumption: The average food consumption of the control animals were 33.6-43.6 g/kg/day (males) or 34.8-43 g/kg/day (females). The average food consumption of the control animals were 250-324 g/animal/day (males) or 197-243 g/animals/day (females) based on the average body weight of 7.44 kg for males and 5.65 kg for females. There were no clear treatment related changes.

5. Ophthalmology: There were no treatment-related eye lesions.

6. ECG and Blood Pressure: There were no clear treatment related changes.

7. Hematology: There were no treatment related changes.

8. Clinical Chemistry: Slight decrease (1-2%) in plasma sodium level was noted in the high dose group.

9. Urinalysis: There were no clear treatment related changes.

10. Organ Weights: There were no clear treatment related changes.

11. Gross pathology: There were no treatment related changes

12. Histopathology: There were no treatment related changes

13. Toxicokinetics: The plasma level of ascorbic acid was summarized in a table on page 56 in Volume 1.9 and this table is attached below.

Ascorbic acid (mean values per group)						
Group	Test day	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-8h} [µg*h/mL]	AUC_{0-8h}/dose [µg*h/mL/(mg/kg/day)]	$t_{1/2}$ [h]
Males						
2	1	27.5	4.0	161.09	32.22	4.14
	14	27.6	4.7	152.97	30.59	8.09
3	1	36.1	4.0	200.60	20.06	7.99
	14	32.6	5.3	176.75	17.68	4.24
4	1	39.0	4.0	230.15	11.51	5.43
	14	42.4	2.7	236.40	11.82	5.14
Females						
2	1	29.3	4.0	168.92	33.78	5.47
	14	26.6	5.3	152.99	30.60	20.42
3	1	42.3	4.0	219.56	21.96	4.70
	14	40.7	4.0	206.53	20.65	5.13
4	1	39.7	3.3	234.87	11.74	3.88
	14	49.7	3.3	295.68	14.78	7.10

The results indicated that the plasma level of ascorbic acid was increased with the dose and not accumulated over time during the 2-weeks of treatment.

In summary, moviprep was tested orally in dog at 0, 5, 10, and 20 g/kg/day for 2 weeks. Major treatment related changes were clinical signs of toxicity including emesis and soft or liquid feces noted in all treatment groups. Salvation was also noted in the high dose group.

90-day Oral Toxicity Study of Movicol in Dogs
(17520/03)

Testing Laboratory: [redacted]

Study Start and Completion Dates: February 10, 2004 and April 29, 2005

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulations and a quality assurance statement.

Animals: Male (6.8-9.1 kg, 6 months old)
Female (5.9-8.6 kg, 6 months old)
Beagle dogs

Drug Batch No.: 112803

Methods: To assess the repeated dose toxicity of movicol in dogs, movicol was given by oral gavage to dogs (4/sex/group) at 0, 5, 20, and 30/25 g/kg twice a day (10, 40, and 60/50 g/kg/day) for 90 days (40 ml/kg/administration). The high dose of 60 g/kg/day was reduced to 50 g/kg/day (25 g/kg twice a day) on day 29 onwards due to the severe toxicity. Movicol was dissolved in tap water. The dose selection was based on a maximum tolerated dose study in dogs (17197/03). The high dose of 60 g/kg/day was the maximum feasible dose. In the current study, clinical signs of toxicity and mortality were observed daily. Body weights were recorded weekly. Food consumption was recorded weekly. Ophthalmological examination was performed at the end of weeks 4 and 13. ECG and blood pressure were determined on day 1, during weeks 4 and 13. Hematology, clinical chemistry and urinalysis were performed at the end of weeks 4 and 13. The following organs were weighed at necropsy for all animals: adrenals, brain, heart, kidneys, liver, lungs, lymph nodes, ovaries, pituitary, spleen, testes, thyroids, and thymus. Histopathological examination was performed on the following tissues for all animals: adrenals, aorta, bone, bone marrow, brain, cecum, epididymides, eyes, heart, ileum, intestine, kidneys, liver, lungs, lymph nodes, mammary gland, esophagus, sciatic nerves, ovaries, pancreas, parathyroids, pituitary, prostate, salivary gland, seminal vesicles, skeletal muscle, skin, spinal cord, spleen, stomach, testes, thymus, thyroids, trachea, urinary bladder, uterus, uterus, vagina, and togue. Plasma levels of ascorbic acid were determined on days

1, 28, and 90 in the satellite animals at pre-dose, 0.5, 1, 2, 4, 6, 8, and 12 hours after dosing (second dose).

Results:

1. Clinical Signs of Toxicity: Emesis, diarrhea, and salvation were noted in all treatment groups and the severity and incidence were treatment related.

2. Mortality: One high dose male was found dead on day 10. One high dose female was sacrificed due to moribund condition. The sponsor stated that the deaths of these two dogs were due to repeated emesis with inhalation of foreign bodies into the lungs and thus a purulent pneumonia.

3. Body Weight: The average initial and final body weights of the control animals were 8 kg and 10 kg (males) or 7.1 kg and 8.55 kg (females), respectively. Treatment with high dose decreased terminal body weight gain in females (7.15 kg at week 0 and 7.06 kg at week 13). The high dose male also gained less weight as compared to the control (8 kg at week 0 and 10.0 kg at week 13 for control and 8.08 kg at week 0 and 8.14 kg at week 13).

4. Food Consumption: The average food consumption of the control animals were 34-40 g/kg/day (males) or 32.3-40 g/kg/day (females). The average food consumption of the control animals were 306-360 g/animal/day (males) or 252-312 g/animal/day (females) based on the average body weight of 9 kg for males and 7.8 kg for females. Reduced food consumption was noted in the high dose males (26 g/kg/day) as compared to the control (34 g/kg/day) during first week.

5. Ophthalmology: There were no treatment-related eye lesions.

6. ECG and Blood Pressure: Prolongation of QTc was noted during week 4 (before second dose) in the treatment groups (234.2, 259.1, and 259.6 msec for low, mid, and high dose groups) as compared to the control (222 msec). It was noted that the mean QTc value of the control group during this period (222 msec) is much shorter than those during other period ranging from 234.3 msec to 253.8 msec. Therefore, the prolongation of QTc interval would be due to the shorter QTc value of the control (than average of QTc). There were no clear treatment related changes in the blood pressure.

7. Hematology: The treatment decreased hematacrit and hemoglobin values in the high dose males by 11-12% as compared to the control.

8. Clinical Chemistry: The treatment related changes were summarized in tables on page 54 in Volume 1.20 and these tables are attached below.

Changes in biochemical parameters compared to the Control group in Test Week 4				
Parameter	Difference to the control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Albumin	none	none	-12**	none
Globulin	none	none	+10	none
α1-globulin	-24	-18	-36**	-24**
α2-globulin	+18	+14	+19	+18
β-globulin	+11	none	+11	none
γ-globulin	+28	+35	+37	+31
A/G ratio	none	none	-19	none
ALAT	none	+31	+106**	+38
Chloride	-3	-6	-5**	-7**
Sodium	-3**	-3	-4**	-5**

** = statistically significant at $p \leq 0.01$

Changes in biochemical parameters compared to the Control group in Test Week13				
Parameter	Difference to the control in [%]			
	Group 3 (40 g/kg)		Group 4 (60/50 g/kg)	
	males	females	males	females
Albumin	none	none	-9**	none
Globulin	none	none	+9	none
α1-globulin	-20**	-6	-22**	-15
α2-globulin	+6	none	+8	+18
β-globulin	+6	none	+7	none
γ-globulin	+17	+50	+24	+28
A/G ratio	none	none	-16	none
Chloride	none	none	-2	-3**
Creatinine	-14	-19**	-17**	-19**
Sodium	-2	-2	-2**	-2**

** = statistically significant at $p \leq 0.01$

9. Urinalysis: There were no clear treatment related changes.

10. Organ Weights: There were no clear treatment related changes.

11. Gross pathology: Hemorrhagic gastric mucosa, bluish foci on the spleen, and hemorrhagic content in the intestinal tract were noted in the dead high dose male. The sacrificed high dose female had light-brown discolored spleen and lung. Reddish discolored gastrointestinal mucosa was also noted in another high dose female.

12. Histopathology: The histopathological examination revealed a reduction of cellularity and increase of foamy macrophages in lymphoid tissues including spleen, thymus, lymph nodes, and lymph follicle of the stomach and Peyer's patches of the intestine in high dose group. The retarded developments of the testis, epididymis, prostate, seminal duct, ovary, uterus, and cervix were noted in all groups including control. These changes were reversible at the end of the 4-week recovery period.

13. Toxicokinetics: PEG 3350 is a heterogenous group of PEG oligomers. The sponsor determined the plasma levels of lower molecular weight PEGs including oligomers 11, 13, 16, and 18 since these oligomers are more likely absorbed. Oligomers 11, 13, 16, and 18 represent PEGs with molecular weight ranging approximately 500 to 800. The results were presented in tables on pages 60-62 in Volume 1.20. These tables are attached below.

Oligomer 11 (mean values per group)									
Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}#1 / AUC_{0-12}#2 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}#1 / AUC_{0-12}#2 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	0.850	1.3	5.87	1.99	0.208	0.7	2.08	2.07
	90	1.413	4.0	9.05	2.80	-	-	-	-
3	1	1.482	4.0	5.44	2.50	0.525	3.3	2.67	1.20
	28	2.720	2.0	13.30	1.67	-	-	-	-
	90	1.963	2.0	13.48	2.93	1.620	2.0	9.53	2.00
4	1	0.277	1.3	1.47	0.00	0.788	2.7	3.36	1.99
	28	1.765	2.0	9.63	2.22	0.328	0.7	2.43	1.36
	90	2.053	2.0	11.18	2.45	1.913	2.0	10.38	1.97

#: AUC_{0-6} calculated for test day 1
#2: AUC_{0-12} calculated for test days 28 and 90

Oligomer 13 (mean values per group)									
Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}#1 / AUC_{0-12}#2 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}#1 / AUC_{0-12}#2 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	0.213	1.3	2.16	1.90	-	-	-	-
	90	-	-	-	-	-	-	-	-
3	1	-	-	-	-	-	-	-	-
	28	0.970	1.3	5.92	1.27	-	-	-	-
	90	0.294	0.7	2.72	2.46	0.208	1.3	2.16	2.09
4	1	-	-	-	-	-	-	-	-
	28	0.520	0.7	2.80	0.81	-	-	-	-
	90	-	-	-	-	-	-	-	-

#: AUC_{0-6} calculated for test day 1
#2: AUC_{0-12} calculated for test days 28 and 90

Oligomer 16 (mean values per group)									
Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}#1 / AUC_{0-12}#2 [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}#1 / AUC_{0-12}#2 [h*µg/mL]	$t_{1/2}$ [h]
2	1	-	-	-	-	-	-	-	-
	28	0.519	4.0	5.09	8.57	-	-	-	
	90	-	-	-	-	-	-	-	-
3	1	-	-	-	-	-	-	-	-
	28	1.397	6.0	9.38	0.54	-	-	-	
	90	0.245	1.3	2.49	2.41	0.298	1.3	2.30	0.79
4	1	0.316	0.7	1.26	0.72		-	-	-
	28	0.918	2.0	5.56	0.96	-	-	-	
	90	-	-	-	-	-	-	-	-

#1: AUC_{0-6} calculated for test day 1
#2: AUC_{0-12} calculated for test days 28 and 90

Oligomer 18 (mean values per group)									
Group	Test day	Males				Females			
		C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}[#] / AUC_{0-12}[#2] [h*µg/mL]	$t_{1/2}$ [h]	C_{max} [µg/mL]	T_{max} [h]	AUC_{0-6}[#] / AUC_{0-12}[#2] [h*µg/mL]	$t_{1/2}$ [h]
2	1	0.199	0.2	1.02	2.30	-	-	-	-
	28	1.439	6.7	11.73	5.08	-	-	-	-
	90	-	-	-	-	-	-	-	-
3	1	1.349	2.2	4.90	2.07	-	-	-	-
	28	2.947	4.7	15.90	0.32	0.315	0.7	2.31	1.45
	90	1.643	1.3	8.77	1.68	1.158	2.0	6.22	1.25
4	1	1.843	1.7	5.99	2.34	0.473	2.7	2.37	2.70
	28	2.253	2.7	11.27	2.01	1.177	1.3	5.91	1.49
	90	1.430	2.0	8.61	2.24	1.257	1.3	6.29	1.28

#: AUC_{0-6} calculated for test day 1
#2: AUC_{0-12} calculated for test days 28 and 90

In summary, movicol was given by oral gavage to dogs (4/sex/group, 3/sex/control group) at 0, 5, 20, and 30/25 g/kg twice a day (10, 40, and 60/50 g/kg/day) for 90 days. Major treatment related changes were clinical signs of toxicity including emesis, diarrhea, and salvation noted (all treatment groups), decreased terminal body weight gain (high dose group), and a reduction of cellularity and increase of foamy macrophages in lymphoid tissues (high dose)

2.6.6.4. Genetic toxicology

Movicol:

Testing for Mutagenic activity of movicol with Salmonella typhimurium and Escherichia coli
(study #22722)

Testing Laboratories: [ ]

Dates Started and Completed: October 25, 2002 and

October 8, 2003

GLP Requirement: A statement of compliance with GLP regulations and the quality assurance unit was included.

Drug Batch No.: 27428

Methods: To examine the potential mutagenic effects of movicol, the reverse mutation assay (Ames test) was conducted using both direct plate and pre-incubation methods in five strains Salmonella typhimurium (TA1535, TA100, TA98, TA1537) and one strain Escherichia coli (WP2uvrA) in the presence and absence of metabolic activation, S-9 mix from rat liver. The following concentrations were tested in the presence and absence of S-9: 17, 50, 167, 500, 1667, and 5000 µg/plate. Four positive controls (sodium azide, 2-nitrofluorene, 9-aminoacridine, and 2-aminoanthracene) were tested.

Results: Movicol did not significantly increase the colonies as compared to the solvent control in the presence and absence of S-9 mix. The positive controls, however, significantly increased in the colonies compared to the solvent controls. The results were presented in Tables 2-5 on pages 20-25 of this report in volume c1.24. These tables are attached below.

Table 2 ***First Mutation Assay***

Mean Number of Revertant Colonies Per Plate in the Presence of S9 Mix (FLI 097).

Item	Dose Level µg per plate	TA 1535	TA 1537	TA 98	TA 100	WP2uvrA
		Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
WATER	100 µl	16 ± 2	13 ± 4	33 ± 3	77 ± 13	4 ± 3
MOVICOL	17	12 ± 2	17 ± 3	25 ± 5	88 ± 12	3 ± 2
	50	12 ± 3	17 ± 9	34 ± 8	92 ± 19	3 ± 3
	167	9 ± 5	15 ± 4	32 ± 5	83 ± 2	2 ± 2
	500	14 ± 4	19 ± 5	37 ± 2	91 ± 3	5 ± 1
	1667	13 ± 4	11 ± 2	27 ± 7	90 ± 9	5 ± 2
	5000	14 ± 1	20 ± 6	31 ± 6	104 ± 15	3 ± 1
Positive controls	Compound	2AAN	2AAN	2AAN	2AAN	2AAN
	Dose Level µg per plate	2	2	0.5	0.5	20
	Mean ± SD	940 ± 58	521 ± 59	799 ± 105	1160 ± 86	420 ± 34

SD Standard Deviation
2AAN 2-Aminoanthracene

Table 2 First Mutation Assay (continued)

Mean Number of Revertant Colonies Per Plate in the Absence of S9 Mix.

Item	Dose Level µg per plate	TA 1535	TA 1537	TA 98	TA 100	WP2uvrA
		Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
WATER	100 µl	15 ± 4	10 ± 1	20 ± 5	97 ± 2	8 ± 1
MOVICOL	17	11 ± 3	4 ± 1	14 ± 5	90 ± 2	7 ± 1
	50	17 ± 5	9 ± 1	16 ± 6	92 ± 5	3 ± 2
	167	12 ± 2	11 ± 4	16 ± 6	84 ± 6	6 ± 2
	500	10 ± 5	13 ± 5	13 ± 5	95 ± 6	4 ± 2
	1667	8 ± 2	12 ± 3	12 ± 4	80 ± 7	4 ± 2
	5000	12 ± 5	10 ± 5	12 ± 0	91 ± 12	6 ± 0
Positive controls	Compound	NaN_3	9AA	2NF	NaN_3	ENNG
	Dose Level µg per plate	1	80	1	1	2
	Mean ± SD	634 ± 22	3752 ± 712	1532 ± 287	1143 ± 22	515 ± 32

SD Standard Deviation
NaN_3 Sodium azide
9AA 9-Aminoacridine
2NF 2-Nitrofluorene
ENNG N-Ethyl-N-nitro-N-nitrosoguanidine

Table 3 Second Mutation Assay

Mean Number of Revertant Colonies Per Plate in the Presence of S9 Mix (FLI 097).

Item	Dose Level µg per plate	TA 1535	TA 1537	TA 98	TA 100	WP2uvrA
		Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
WATER	100 µl	16 ± 3	13 ± 2	29 ± 8	89 ± 7	6 ± 4
MOVICOL	17	15 ± 2	8 ± 3	23 ± 8	94 ± 10	2 ± 2
	50	14 ± 4	14 ± 2	27 ± 3	96 ± 11	3 ± 1
	167	13 ± 2	5 ± 4	24 ± 6	96 ± 6	2 ± 2
	500	12 ± 2	14 ± 6	26 ± 4	95 ± 2	4 ± 1
	1667	13 ± 8	9 ± 7	23 ± 3	100 ± 12	4 ± 2
	5000	12 ± 4	11 ± 1	31 ± 1	90 ± 13	3 ± 2
Positive controls	Compound	2AAN	2AAN	2AAN	2AAN	2AAN
	Dose Level µg per plate	2	2	0.5	0.5	20
	Mean ± SD	255 ± 22	145 ± 17	194 ± 23	302 ± 9	695 ± 46

SD Standard Deviation
2AAN 2-Aminoanthracene

The counts obtained for 2AAN with the *Salmonella* strains were lower on this occasion than those obtained with the first mutation assay (although the counts were considered valid as they were within the acceptance criteria and the historical range). At the Sponsor's request, TA 1537, TA 98 and TA 100 were repeated due to the inconsistency of the results between tests.

Table 3 Second Mutation Assay
(continued)

Mean Number of Revertant Colonies Per Plate In the Absence of S9 Mix

Item	Dose Level µg per plate	TA 1535	TA 1537	TA 98	TA 100	WP2uvrA
		Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
WATER	100 µl	11 ± 3	6 ± 3	21 ± 5	112 ± 9	7 ± 3
MOVICOL	17	14 ± 3	7 ± 5	17 ± 6	96 ± 12	4 ± 2
	50	15 ± 5	5 ± 3	20 ± 3	99 ± 18	4 ± 2
	167	8 ± 6	7 ± 3	17 ± 1	102 ± 5	4 ± 2
	500	14 ± 3	6 ± 4	23 ± 3	110 ± 6	7 ± 2
	1667	17 ± 0	5 ± 2	28 ± 2	107 ± 17	3 ± 1
	5000	15 ± 1	8 ± 5	22 ± 2	114 ± 11	6 ± 1
Positive controls	Compound	NaN_3	9AA	2NF	NaN_3	ENNG
	Dose Level µg per plate	1	80	1	1	2
	Mean ± SD	636 ± 8	78 ± 9	10 ± 7	1239 ± 121	399 ± 49

SD Standard Deviation
NaN_3 Sodium azide
9AA 9-Aminoacridine
2NF 2-Nitrofluorene
ENNG *N*-Ethyl-N-nitro-N-nitrosoguanidine

TA 1537 and TA 98 were repeated due to the poor responses obtained with 9AA and 2NF

Table 4 ***Second Mutation Assay Retest with TA 1537 and TA 98 (-S9 Mix)***

Mean number of Revertant Colonies Obtained in the Absence of S9 Mix

Item	Dose Level µg per plate	TA 1537	TA 98
		Mean ± SD	Mean ± SD
WATER	100 µl	3 ± 2	11 ± 2
MOVICOL	17	6 ± 2	11 ± 3
	50	9 ± 4	11 ± 2
	167	9 ± 2	16 ± 4
	500	5 ± 2	11 ± 3
	1667	8 ± 1	13 ± 1
	5000	5 ± 2	15 ± 8
Positive controls	Compound	9AA	2NF
	Dose Level µg per plate	80	1
	Mean ± SD	1002 ± 113	644 ± 39

SD Standard Deviation
9AA 9-Aminoacridine
2NF 2-Nitrofluorene

Table 5 ***Second Mutation Assay Retest with TA 1537, TA 98 and TA 100 (+S9 Mix only)***

Mean number of Revertant Colonies Obtained in the Presence of S9 Mix

Item	Dose Level µg per plate	TA 1537	TA 98	TA 100
		Mean ± SD	Mean ± SD	Mean ± SD
WATER	100 µl	16 ± 6	33 ± 7	94 ± 17
MOVICOL	17	14 ± 1	29 ± 7	91 ± 8
	50	14 ± 2	31 ± 2	95 ± 6
	167	21 ± 4	31 ± 4	101 ± 3
	500	19 ± 1	27 ± 6	88 ± 7
	1667	18 ± 5	33 ± 1	95 ± 20
	5000	20 ± 5	29 ± 2	90 ± 14
Positive controls	Compound	2AAN	2AAN	2AAN
	Dose Level µg per plate	2	0.5	0.5
	Mean ± SD	298 ± 33	547 ± 32	530 ± 31

SD Standard Deviation
2AAN 2-Aminoanthracene

In conclusion, the results suggest that mivocol was not mutagenic in this test system.

Mouse lymphoma cell mutation assay with movicol
(study #22825)

Testing Laboratories: [ ]

Dates Started and Completed: October 25, 2002 and July 25, 2003

GLP Requirement: A statement of compliance with GLP regulations and the quality assurance unit was included.

Drug Batch No.: 27428

Methods: To examine the potential mutagenic effects of movicol, the L5178Y TK+/- mouse lymphoma cell assay was conducted in the presence and absence of metabolic activation, S-9 mix from rat liver. The following concentrations of movicol were used: 625, 1250, 2500, and 5000 µg/ml with and without S9. Positive controls (ethyl methanesulphonate and methyl methanesulphonate) were also tested. The TK+/- cell suspension was incubated with the test drug for 4 or 24 hours at 37° C and cells were then washed free of drug. The expression period was 2 days. Plates were incubated for 9-12 days at 37° C. The number of TK+/- mutant colonies were then determined. The result is considered positive if a dose dependent increase in mutation frequency is observed at one or more concentrations.

Results:

- **Study validation**: The positive controls significantly increased the colonies compared to the solvent controls.

- Study outcome: There were no significant increases in the mutant frequency in the cultures treated with movicol in the presence and absence metabolic activation. The results were summarized in Tables 4, 5, 6, and 7 on pages 35-38 of this report in volume 1.24. These tables are attached below.

MOVICOL
Mouse Lymphoma Mutation Assay
Table 4 *Mutation Test in the Absence of S9 Mix (4 h Exposure)*
Summary of Means of Data (Assay 1)

Chemical	Concentration ($\mu g.ml^{-1}$)	Day 0	Day 2			
		Relative Survival %	Relative Total Growth %	Mutant Fraction ($\times 10^{-6}$)	Ratio of Small to Large Colonies	Statistical Comparison
Water	(100µl added)	100	100	100	1.15	-
EMS	250	103	80	651	0.63	*
MMS	10	69	28	688	2.07	*
MOVICOL	625	102	105	86	1.36	-
	1250	86	116	116	1.30	-
	2500	94	112	102	1.11	-
	5000	103	107	83	1.27	-

* = Significant difference in log mutant fraction compared with vehicle control ($P < 0.05$)
Test for linear trend of mutant fraction with concentration of MOVICOL = not reported - slope negative

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MOVICOL
Mouse Lymphoma Mutation Assay
Table 5 *Mutation Test in the Presence of S9 Mix (4 h Exposure)*
Summary of Means of Data (Assay 2)

Chemical	Concentration ($\mu g.ml^{-1}$)	Day 0	Day 2			
		Relative Survival %	Relative Total Growth %	Mutant Fraction ($\times 10^{-6}$)	Ratio of Small to Large Colonies	Statistical Comparison
Water	(100µl added)	100	100	120	0.79	-
3-MC	2.5	65	44	2212	1.16	*
MOVICOL	625	109	100	110	0.55	-
	1250	99	93	112	0.60	-
	2500	109	137	102	1.32	-
	5000	106	127	106	0.78	-

* = Significant difference in log mutant fraction compared with vehicle control ($P < 0.05$)
Test for linear trend of mutant fraction with concentration of MOVICOL = not significant - slope negative

MOVICOL
Mouse Lymphoma Mutation Assay
Table 6 *Mutation Test in the Absence of S9 Mix (24 h Exposure)*
Summary of Means of Data (Assay 3)

Chemical	Concentration ($\mu g.ml^{-1}$)	Day 1	Day 3			
		Relative Survival %	Relative Total Growth %	Mutant Fraction ($\times 10^{-6}$)	Ratio of Small to Large Colonies	Statistical Comparison
Water	(200µl added)	100	100	80	0.63	-
EMS	150	42	34	1622	0.41	*
MMS	5	50	40	1038	2.22	*
MOVICOL	625	97	126	66	0.84	-
	1250	94	110	90	0.55	-
	2500	106	85	75	0.50	-
	5000	97	96	91	0.59	-

* = Significant difference in log mutant fraction compared with vehicle control ($P < 0.05$)
Test for linear trend of mutant fraction with concentration of MOVICOL = not significant ($P = 0.50$)

Table 7 ***MOVICOL***
Mouse Lymphoma Mutation Assay
Mutation Test in the Presence of S9 Mix (4 h Exposure)
Summary of Means of Data (Assay 4)

Chemical	Concentration ($\mu g.ml^{-1}$)	Day 0	Day 2			
		Relative Survival %	Relative Total Growth %	Mutant Fraction ($\times 10^{-6}$)	Ratio of Small to Large Colonies	Statistical Comparison
Water	(100µl added)	100	100	99	0.79	-
3-MC	2.5	72	39	1256	0.92	*
MOVICOL	625	103	104	84	1.06	-
	1250	116	101	85	0.49	-
	2500	106	114	95	0.80	-
	5000	109	107	97	0.35	-

* = Significant difference in log mutant fraction compared with vehicle control ($P < 0.05$)
Test for linear trend of mutant fraction with concentration of MOVICOL = not significant ($P = 0.84$)

Summary: There were no significant increases in the mutant frequency in the cultures treated with movicol with and without metabolic activation. The results suggest that movicol was not mutagenic in this testing system.

Micronucleus Test in bone marrow of CD-1 mice with movicol
(study #22924)

Testing Laboratories: [ ]

Dates Started and Completed: January 22, 2003 and July 27, 2003

GLP Requirement: A statement of compliance with GLP regulations and the quality assurance unit was included.

Animals: Male: 23-35 g, 6-7 weeks old
Female: 20-27 g, 6-7 weeks old
CD-1 Mice

Drug Batch No.: 27428

Methods: To examine the potential mutagenic effects of movicol, micronucleus test was conducted using mouse bone marrow cells. Movicol was given to mice by oral gavage at 2000 mg/kg/day on day 1 (0 hour) and day 2 (24 hours). The treated mice were then sacrificed 48 hours after the first dose and bone marrow was collected. There was only one sample collection of bone marrow in this study. Vehicle and positive controls (cyclophosphamide) were also tested. The frequency of micronucleated polychromatic erythrocytes was then determined.

Results: The results were presented in Table 3 on page 23 of this report in volume c1.24. This table is attached below.

MOVICOL
Micronucleus Test in Bone Marrow of CD-1 Mice
Table 3 *Summary of Assessment Data*

Treatment	Dose (h)	Sex	No. of Mice Scored	Erythrocytes				
				Normochromatic Cells (NCE)	Polychromatic Cells (PCE)			PCE/NCE
				No. of MN-NCE	PCE Analysed	No. of MN-PCE	% MN-PCE	Mean ± S.D.
10 ml water $kg^{-1}.day^{-1}$	0 + 24	♂	5	0	10012	3	0.03	0.89 ± 0.05
		♀	5	7	10013	2	0.02	0.92 ± 0.06
		♂♀	10	7	20025	5	0.02	0.91 ± 0.05
2000 mg MOVICOL kg^{-1} day^{-1}	0 + 24	♂	5	5	10009	9	0.09	0.94 ± 0.01
		♀	5	4	10016	9	0.09	0.87 ±0.08
		♂♀	10	9	20025	18	0.09	0.90 ± 0.07
50 mg Cyclophosphamide $kg^{-1}.day^{-1}$	0 + 24	♂	5	31 α	10012	185 Φ	1.85	0.81 ± 0.12

PCE = Polychromatic erythrocytes
MN-PCE = Micronucleated PCE
NCE = Normochromatic erythrocytes
MN-NCE = Micronucleated NCE
Φ = Positive response in PCE
α = Evident response in NCE

The results indicated that treatment with movicol did not significantly increase the frequency of micronucleated polychromatic erythrocytes. The frequency of micronucleated polychromatic erythrocytes in the treatment group was 0.09% which is within the range of the historical control data of 0.0%-0.5% provided by the sponsor. The positive control significantly increased it

In conclusion, movicol was not mutagenic in this test system.

Ascorbic acid: Following results were obtained from the literatures provided by the sponsor.

Ascorbate is not detectably mutagenic in the L5178Y TK+/- cell mutation assay
(Cancer Letters, 14:151-158, 1981)

Methods: To evaluate the potential mutagenicity of sodium ascorbate and ascoarbic acid, both sodium ascorbate and ascorbic acid were tested in the mouse lymphoma L5178Y TK+/- cell mutation assay at thymidine kinase locus in the presence and absence of bovine liver catalase. Sodium ascorbate and ascoarbic acid were tested up to a few mM concentrations. Positive control (ethyl methanesulphonate) was tested. The TK+/- cell suspension was incubated with the test drug for 3 hours at 37° C and cells were then washed free of drug. The number of TK+/- mutant colonies were then determined after incubation for 7 days at 37° C.

Results: The results were presented in Figures 1 and 2 in this report. These figures are attached below.

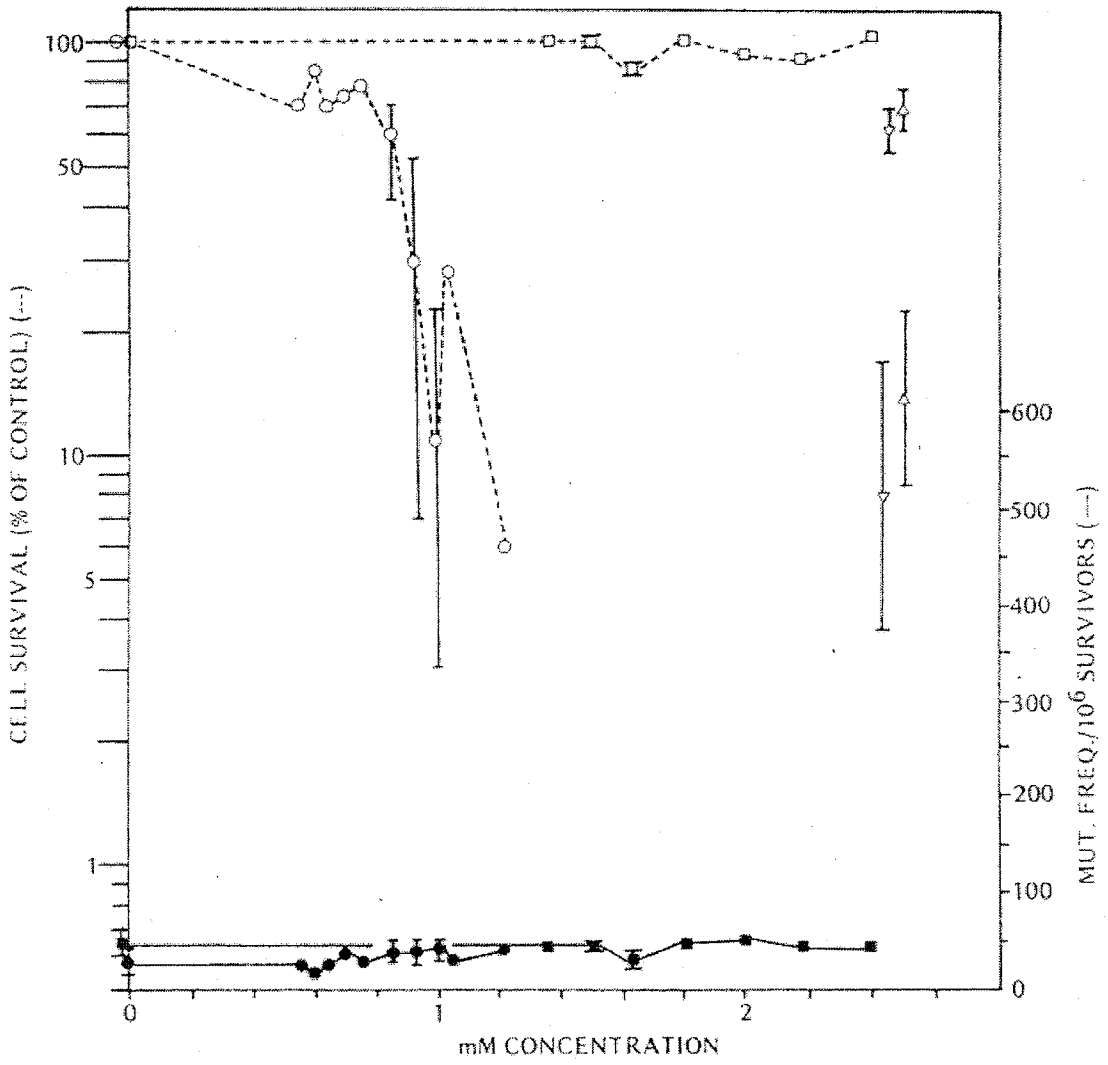

Fig. 1. Mutagenicity and cytotoxicity of sodium ascorbate in L5178Y/TK$^{+/-}$ cells. Solid lines indicate mutant frequency; broken lines indicate cell survival. Squares indicate the presence and circles indicate the absence of 0.1 mg/ml catalase in paired experiments. The mean plus or minus half the range is shown for solvent controls and replicate treated cultures from 2 different trials. Cell survival and mutant frequencies for concurrent positive controls (2.5×10^{3} M EMS) are indicated for experiments with (△) or without (▽) catalase.

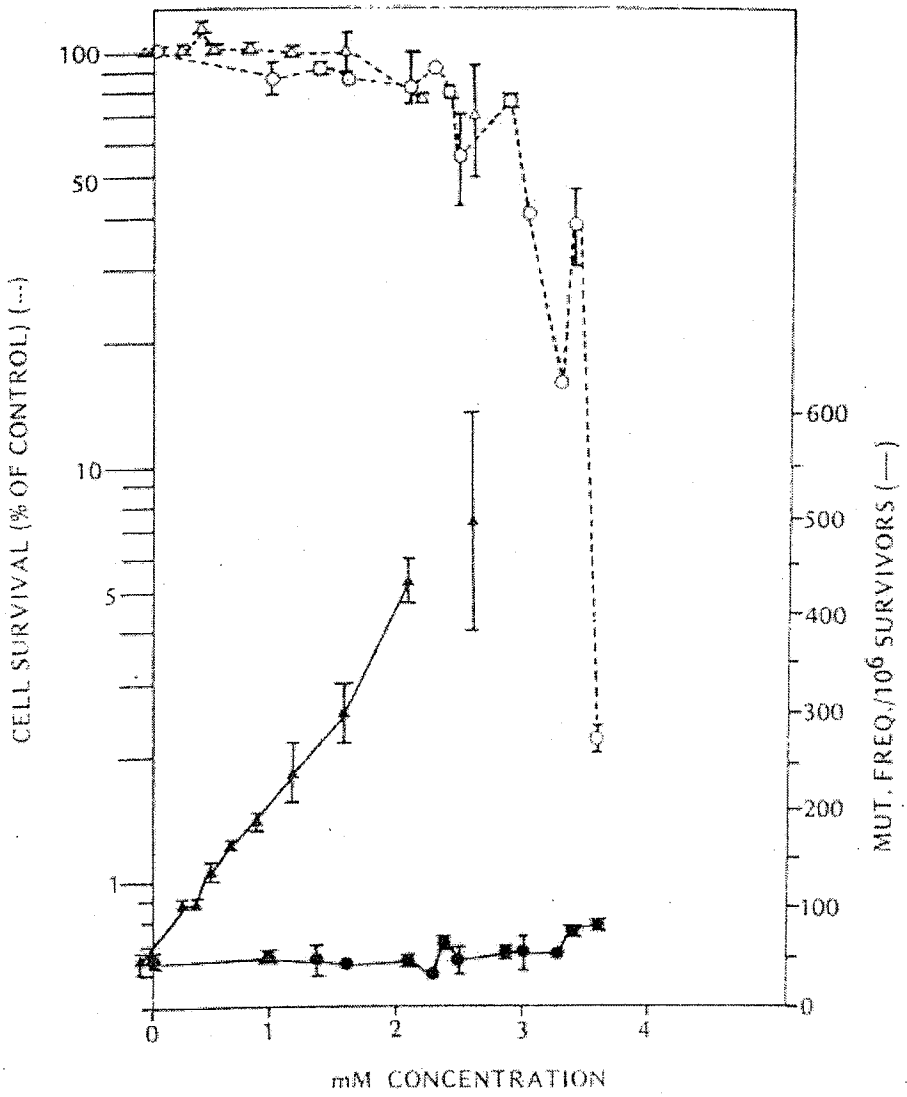

Fig. 2. Mutagenicity and cytotoxicity of ascorbic acid in L5178Y/TK$^{+/-}$ cells. Closed symbols indicate mutant frequency; open symbols indicate cell survival for 2 overlapping independent experiments. The mean plus or minus half the range is shown for all solvent controls and for some experimental data which included duplicate samples from different trials. The data from 2 independent trials with EMS (triangles) is derived from a previously published study [2]. Cell survival and mutant frequencies for concurrent positive controls (2.5×10^{3} M EMS) are shown.

The results indicated that treatment with sodium ascorbate and ascoarbic acid did not significantly increase the frequency of the mutant frequency. However, the positive control significantly increased it.

In conclusion, sodium ascorbate and ascoarbic acid were not mutagenic in this testing system.

Vitamin C is positive in the DNA synthesis inhibition and sister-chromatid exchange tests
(Mutation Research, 60:321-327, 1979)

Methods: To evaluate the potential mutagenicity of sodium ascorbate, sodium ascorbate was tested in the sister-chromatid exchange (SCE) test in Chinese hamster ovary cells and in human lymphocytes and DNA synthesis inhibition test in Hela cells.

SCE test:

The Chinese hamster ovary (CHO) cells were plated in 10 ml medium and 5-bromodeoxyuridine (BrdUrd) was added for two cell cycles (~26-28 hours). For the human lymphocyte study, whole blood samples were cultured for 72 hours with BrdUrd. Sodium ascorbate was added to the medium 1 hour before addition of BrdUrd. Colcemid was added for the final 2-2.5 hours. SCEs were then determined.

DNA synthesis inhibition test:

Hela cells were incubated with medium containing sodium ascorbate for 30 minute in the presence and absence catalase and the cells were washed with drug free medium. The cultures were then incubated with medium containing 3H-thymidine for 10 minutes. The 3H-DNA specific activity and the rate of DNA synthesis were deterimined using liquid scintillation spectrometer.

Results:

SCE test:

The results indicated that treatment with sodium ascobate increased the frequency of the SCE in a dose dependent manner in both CHO cells and human lymphocytes. The results were presented in Tables 1 in this report. This table is attached below.

TABLE 1

SISTER-CHROMATID EXCHANGES IN CHO CELLS AND HUMAN LYMPHOCYTES TREATED WITH SODIUM ASCORBATE

Concentration of sodium ascorbate [a] (M)	Number of SCEs per cell
CHO cells [b]	
Control	8.1 ± 0.5
1×10^{-4}	9.8 ± 0.5
3×10^{-4}	10.7 ± 0.6
1×10^{-3}	12.0 ± 0.5
3×10^{-3}	18.5 ± 0.9
1×10^{-2}	23.7 ± 1.1
Lymphocytes [c]	
Control	9.2 ± 0.6
5.4×10^{-4}	10.2 ± 0.9
1.8×10^{-3}	13.5 ± 0.7
5.4×10^{-3}	18.1 ± 0.9

[a] Ascorbate was added 1 h before BrdUrd and left in medium.
[b] Mean ± S.E. of 50 cells.
[c] Mean ± S.E. of 30 cells.

DNA synthesis inhibition test:

Sodium ascorbate also increased the inhibition of DNA synthesis in Hela cells. The results were presented in Figure 1 in this report and this figure is attached below.

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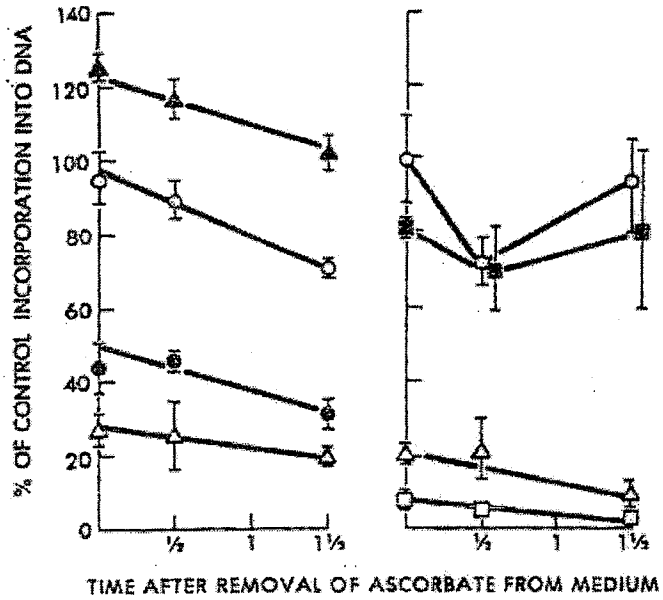


Fig. 1. Inhibition of HeLa DNA synthesis by ascorbate. Two experiments are shown. ○, 2 X 10^{-3} M ascorbate; ●, 3.5 X 10^{-3} M ascorbate; △, 5 X 10^{-3} M ascorbate; □, 2 X 10^{-2} M ascorbate; ▲, 5 X 10^{-3} M ascorbate + 20 μg/ml catalase; ■, 2 X 10^{-2} M ascorbate + 20 μg/ml catalase.

In conclusion, sodium ascorbate was mutagenic in these testing systems.

Ascorbate induced mutation at HGPRT locus in Chinese hamster cells

(Cancer Letters, 8:299-305, 1980)

Methods: To determine the mutagenic potential of ascorbate, a mutation assay at HGPRT locus was conducted with ascorbate in Chinese hamster ovary (CHO) cells. In this study, the CHO cells were exposed to sodium ascorbate at concentrations up to 10^{-3} M for 3 hours at 37° C with or without catalase from bovine liver. The cultures were then incubated for 7 days. The 6-thioguanine (6-TG) mutant colonies were determined.

Results: The result indicated that treatment with ascorbate induced the 6-TG resistant mutation in the presence and absence of catalase. The maximum induction occurred at 5 x 10^{-4} M concentration. The results were presented in Figure 2 in this report. This figure is attached below.

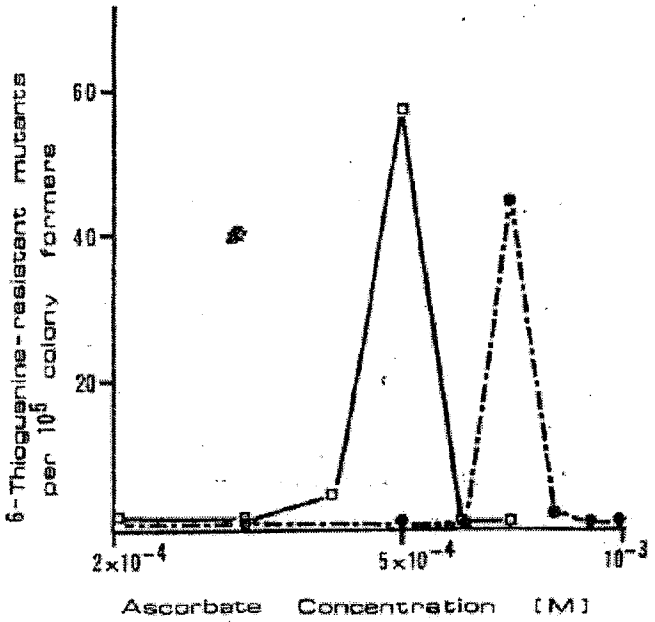


Fig. 2. Induction of 6-TG^r colonies by ascorbate in CHO cells. Treatment was: ▫, ascorbate; •, ascorbate + catalase.

There were no induction of mutation in the control culture and the positive control (N-methyl-N-nitro-N-nitrosoguanidine) induced it (no data were provided). In conclusion, sodium ascorbate was mutagenic in this tesing system.

2.6.6.5. Carcinogenicity

PEG:

Two years of repeated oral feeding of polyethylene glycol 4000 to rats

By [redacted]

Methods: To determine potential carcinogenicity of PEG 4000, a two-year oral (in diet) toxicity study with PEG 4000 was conducted in rats. In this study, Wistar rats (20/sex/group) were treated with PEG 4000 at 0, 0.5, 1, 2, 4 and 8% (approximately 0, 0.313, 0.625, 1.25, 2.5 and 5 g/kg/day) in diet. Following parameters were monitored: mortality, food consumption, body weight, hematology and organ weights (liver

and kidney at 1 and 2 years). Gross and histopathological examinations were conducted in the rats died before termination and at one year after treatment. The histopathological examination was conducted on adrenal, heart, small intestine, kidney, liver, lung, pancreas, spleen and testis. After 2 years, the survivors were sacrificed and liver and kidney weights were determined.

Results: The results indicated that there were no treatment related deaths in this study. The body weight was slightly but significantly lower in the high dose groups as compared to the control. The body weight data were presented in Table 17-144 in this report and this table is attached below.

Table 17-144

Summary of Weight Data

Concen., %	No. of Rats	Overall Mean Mean	Overall Mean p	No. of Twenty-Six Biweekly Means: Lower Than Control	Signi-ficantly Lower	Higher Than Control	Signi-ficantly Higher	Correl. between Overall Means & Conc. in Diet: Coef-ficient	p
				Male Rats					
8.0	17	198.6*	<0.0001	26	1	0	0		
4.0	15	211.7	0.34	4	0	22	0		
2.0	14	226.1*	<0.0001	0	0	26	8		
1.0	15	214.0	0.087	2	0	24	0		
0.5	17	208.2	0.76	12	0	14	0		
0.0	14	209.0	-	-	-	-	-		
								-0.494	0.30
				Female Rats					
8.0	16	163.5*	<0.0001	26	21	0	0		
4.0	19	174.2	0.052	25	1	1	0		
2.0	16	175.8	0.25	17	0	9	0		
1.0	18	176.1	0.32	17	1	9	0		
0.5	18	170.3*	<0.0001	26	4	0	0		
0.0	19	177.8	-	-	-	-	-		
								-0.777	0.06

* Deviation from control is statistically significant.

There were no other treatment related changes in this study. The treatment with PEG 4000 did not increase tumor incidences in this study.

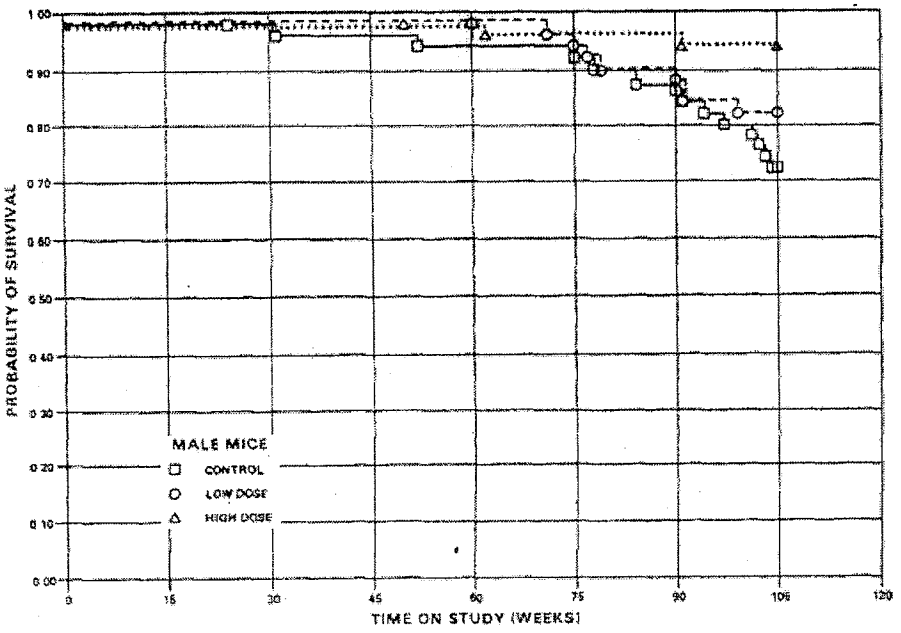
L-Ascorbic acid:

Carcinogenesis bioassay of L-Ascorbic acid (vitamin C) in F344/N rats and B6C3F1 mice (feed study)
(NTP-81-140)

Methods: To determine the potential carcinogenesis of L-ascorbic acid, 2-year carcinogenesis studies were conducted with L-ascobic acid in B6C3F1 mice and 50 F344/N rats. In these studies, both mice and rats were treated in diet with L-ascorbic acid at 25,000 and 50,000 ppm (2.5% and 5%) for 103 weeks. The doses of 2.5% and 5% in mice are approximatelty 6 g/kg/day and 12 g/kg/day, respectively. The doses of 2.5% and 5% in rats are approximatelty 1.2 g/kg/day and 2.5 g/kg/day, respectively. Body weight and food consumption were determined weekly. Examination for gross lesions were conducted on major tissues or organs. The following tissues or organs were examined microscropically: tissue masses, abnormal lymph nodes, skin, mandibular lymph nodes, mammary gland, salivary gland, thigh muscle, sciatic nerve, bone marrow, costochondral junctions, thymus, larynx, trachea, lungs and bronchi, heart, thyroid, parthyroid, esophagus, stomach, duodenum, jejunum, ileum, colon, mesenteric lymph nodes, liver, gallbladder (mice), pancreas, spleen, kidneys, adrenals, urinary bladder, seminal vesicles/prostate/testis or ovaries/uterus, nasal cavity, brain, pituitary, and spinal cord.

Results:

Mice: The results indicated that the survivals of the female mice were comparable between control and treatment groups. The survivals of the high dosed male mice were greater than that in the controls. The survival data were presented in Figure 4 in this report and this figure is attached below.



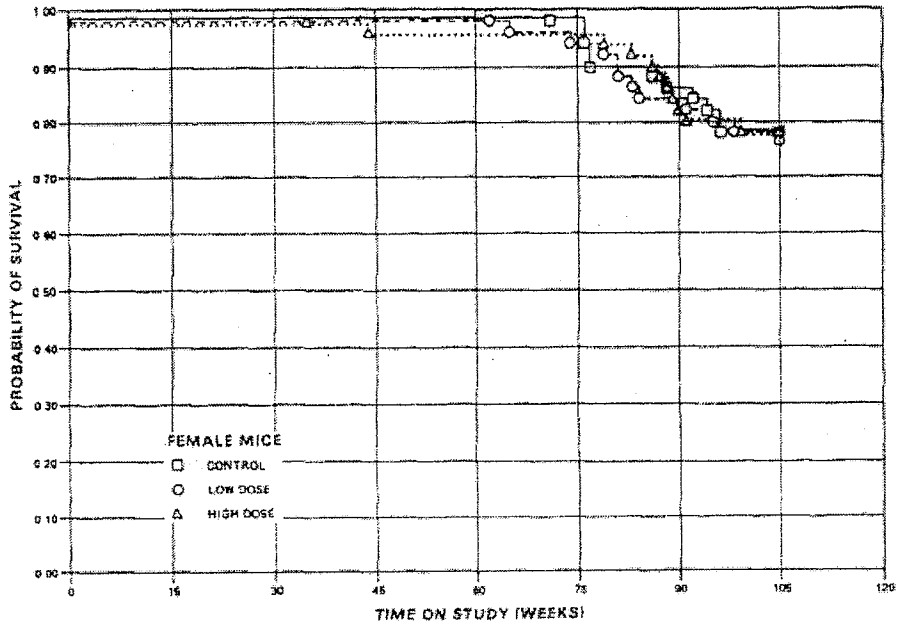


Figure 4. Survival Curves for Mice Fed Diets Containing L-Ascorbic Acid

The final body weight was lower by 8-11% in the treated mice as compared to the control. The body weight data were summarized in Table 13 in this report and this table is attached below.

TABLE 13. CUMULATIVE MEAN BODY WEIGHT CHANGE (RELATIVE TO CONTROLS) OF MICE FED DIETS CONTAINING L-ASCORBIC ACID IN THE 2-YEAR STUDY

		Cumulative Mean Body Weight Change (grams)			Weight Differential Relative to Controls *(a)* (percent)	
	Week No.	Control	Low Dose	High Dose	Low Dose	High Dose
Males	0	22 *(b)*	22 *(b)*	22 *(b)*		
	1	1	2	1	+100	0
	21	9	8	9	- 11	0
	42	11	12	13	+ 9	+18
	63	15	14	14	- 7	-7
	80	14	13	13	- 7	-7
	101	13	13	12	0	-8
	103	35 *(c)*	34 *(c)*	34 *(c)*	- 3 *(d)*	-3 *(d)*
Females	0	18 *(b)*	18 *(b)*	18*(b)*		
	1	2	2	1	0	-50
	21	9	6	6	- 33	-33
	42	15	13	13	- 13	-13
	63	19	15	16	- 21	-16
	80	18	15	16	- 17	-11
	101	19	16	15	- 16	-21
	103	36 *(c)*	33 *(c)*	32 *(c)*	- 8 *(d)*	-11 *(d)*

(a) Weight Differential Relative to Controls =

$$\frac{\text{Weight Change (Dosed Group)} - \text{Weight Change (Control Group)}}{\text{Weight Change (Control Group)}} \times 100$$

(b) Initial weight

(c) Mean body weight at week 103

(d) Weight at week 103 relative to controls

The tumor data were summarized in Tables 16 and 17 in this report and these tables are attached below.

TABLE 16. ANALYSIS OF PRIMARY TUMORS IN MALE MICE *(a)*

	Control	Low Dose	High Dose
Lung: Alveolar/Bronchiolar Adenoma			
Tumor Rates			
Overall *(b)*	3/49 (6%)	3/49 (6%)	3/49 (6%)
Adjusted *(c)*	8.3%	7.3%	6.4%
Terminal *(d)*	3/36 (8%)	3/41 (7%)	3/47 (6%)
Statistical Tests *(e)*			
Life Table	P=0.450N	P=0.602N	P=0.535N
Incidental Tumor Test	P=0.450N	P=0.602N	P=0.535N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.583	P=0.661	P=0.661
Lung: Alveolar/Bronchiolar Carcinoma			
Tumor Rates			
Overall *(b)*	2/49 (4%)	1/49 (2%)	5/49 (10%)
Adjusted *(c)*	5.0%	2.4%	10.4%
Terminal *(d)*	1/36 (3%)	1/41 (2%)	4/47 (9%)
Statistical Tests *(e)*			
Life Table	P=0.201	P=0.467N	P=0.316
Incidental Tumor Test	P=0.119	P=0.470N	P=0.163
Cochran-Armitage Trend, Fisher Exact Tests	P=0.133	P=0.500N	P=0.218
Lung: Alveolar/Bronchiolar Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	5/49 (10%)	4/49 (8%)	8/49 (16%)
Adjusted *(c)*	13.1%	9.8%	16.7%
Terminal *(d)*	4/36 (11%)	4/41 (10%)	7/47 (15%)
Statistical Tests *(e)*			
Life Table	P=0.365	P=0.427N	P=0.448
Incidental Tumor Test	P=0.287	P=0.428N	P=0.317
Cochran-Armitage Trend, Fisher Exact Tests	P=0.215	P=0.500N	P=0.276
Hematopoietic System: Malignant Lymphoma, Histiocytic Type			
Tumor Rates			
Overall *(b)*	3/50 (6%)	5/50 (10%)	3/50 (6%)
Adjusted *(c)*	7.3%	11.8%	6.4%
Terminal *(d)*	0/36 (0%)	4/41 (10%)	3/47 (6%)
Statistical Tests *(e)*			
Life Table	P=0.452N	P=0.407	P=0.559N
Incidental Tumor Test	P=0.318	P=0.226	P=0.281
Cochran-Armitage Trend, Fisher Exact Tests	P=0.576	P=0.357	P=0.661
Hematopoietic System: Malignant Lymphoma, Lymphocytic Type			
Tumor Rates			
Overall *(b)*	3/50 (6%)	1/50 (2%)	0/50 (0%)
Adjusted *(c)*	7.7%	2.4%	0.0%
Terminal *(d)*	2/36 (6%)	0/41 (0%)	0/47 (0%)
Statistical Tests *(e)*			
Life Table	P=0.045N	P=0.279N	P=0.089N
Incidental Tumor Test	P=0.126N	P=0.382N	P=0.141N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.060N	P=0.309N	P=0.121N

TABLE 16. ANALYSIS OF PRIMARY TUMORS IN MALE MICE *(a)* (Continued)

	Control	Low Dose	High Dose
Hematopoietic System: All Malignant Lymphoma			
Tumor Rates			
Overall *(b)*	8/50 (16%)	7/50 (14%)	3/50 (6%)
Adjusted *(c)*	18.7%	16.2%	6.4%
Terminal *(d)*	3/36 (8%)	5/41 (12%)	3/47 (6%)
Statistical Tests *(e)*			
Life Table	P=0.044N	P=0.431N	P=0.058N
Incidental Tumor Test	P=0.242N	P=0.602N	P=0.296N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.083N	P=0.500N	P=0.100N
Hematopoietic System: Lymphoma or Leukemia			
Tumor Rates			
Overall *(b)*	9/50 (18%)	8/50 (16%)	3/50 (6%)
Adjusted *(c)*	20.6%	17.9%	6.4%
Terminal *(d)*	3/36 (8%)	5/41 (12%)	3/47 (6%)
Statistical Tests *(e)*			
Life Table	P=0.028N	P=0.434N	P=0.035N
Incidental Tumor Test	P=0.246N	P=0.588	P=0.296N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.053N	P=0.500N	P=0.061N
Circulatory System: Hemangiosarcoma			
Tumor Rates			
Overall *(b)*	1/50 (2%)	4/50 (8%)	0/50 (0%)
Adjusted *(c)*	2.5%	9.5%	0.0%
Terminal *(d)*	0/36 (0%)	3/41 (7%)	0/47 (0%)
Statistical Tests *(e)*			
Life Table	P=0.315N	P=0.212	P=0.468N
Incidental Tumor Test	P=0.514	P=0.047	P=0.824N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.390N	P=0.181	P=0.500N
Liver: Adenoma			
Tumor Rates			
Overall *(b)*	6/50 (12%)	4/49 (8%)	9/50 (18%)
Adjusted *(c)*	16.7%	9.8%	19.1%
Terminal *(d)*	6/36 (17%)	4/41 (10%)	9/47 (19%)
Statistical Tests *(e)*			
Life Table	P=0.402	P=0.289N	P=0.499
Incidental Tumor Test	P=0.402	P=0.289N	P=0.499
Cochran-Armitage Trend, Fisher Exact Tests	P=0.227	P=0.383N	P=0.288
Liver: Carcinoma			
Tumor Rates			
Overall *(b)*	10/50 (20%)	12/49 (24%)	4/50 (8%)
Adjusted *(c)*	24.6%	26.4%	8.5%
Terminal *(d)*	6/36 (17%)	8/41 (20%)	4/47 (9%)
Statistical Tests *(e)*			
Life Table	P=0.031N	P=0.502	P=0.032N
Incidental Tumor Test	P=0.166N	P=0.347	P=0.168N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.074N	P=0.384	P=0.074N

TABLE 16. ANALYSIS OF PRIMARY TUMORS IN MALE MICE *(a)* (Continued)

	Control	Low Dose	High Dose
Liver: Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	16/50 (32%)	16/49 (33%)	13/50 (26%)
Adjusted *(c)*	39.7%	35.3%	27.7%
Terminal *(d)*	12/36 (33%)	12/41 (29%)	13/47 (28%)
Statistical Tests *(e)*			
Life Table	P=0.101N	P=0.447N	P=0.112N
Incidental Tumor Test	P=0.319N	P=0.580N	P=0.322N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.293N	P=0.558	P=0.330N

(a) Dosed groups received doses of 25,000 or 50,000 ppm of ascorbic acid in the diet.
(b) Number of tumor bearing animals/number of animals examined at the site.
(c) Kaplan-Meier estimated lifetime tumor incidence after adjusting for intercurrent mortality.
(d) Observed tumor incidence at terminal kill.
(e) Beneath the control incidence are the P-values associated with the trend test. Beneath the dosed group incidence are the P-values corresponding to pairwise comparisons between that dosed group and the controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as non-fatal. The Cochran-Armitage and Fisher's exact tests compare directly the overall incidence rates. A negative trend or lower incidence is indicated by (N).

TABLE 17. ANALYSIS OF PRIMARY TUMORS IN FEMALE MICE *(a)*

	Control	Low Dose	High Dose
Lung: Alveolar/Bronchiolar Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	1/49 (2%)	4/49 (8%)	1/50 (2%)
Adjusted *(c)*	2.6%	10.3%	2.6%
Terminal *(d)*	1/38 (3%)	4/39 (10%)	1/39 (3%)
Statistical Tests *(e)*			
Life Table	P=0.591N	P=0.187	P=0.756N
Incidental Tumor Test	P=0.591N	P=0.187	P=0.756N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.593N	P=0.181	P=0.747N
Hematopoietic System: Malignant Lymphoma, Lymphocytic Type			
Tumor Rates			
Overall *(b)*	5/50 (10%)	4/50 (8%)	6/50 (12%)
Adjusted *(c)*	11.4%	9.8%	15.0%
Terminal *(d)*	2/39 (5%)	3/39 (8%)	5/39 (13%)
Statistical Tests *(e)*			
Life Table	P=0.438	P=0.509N	P=0.503
Incidental Tumor Test	P=0.338	P=0.470N	P=0.295
Cochran-Armitage Trend, Fisher Exact Tests	P=0.434	P=0.500N	P=0.500
Hematopoietic System: Malignant Lymphoma, Histiocytic Type			
Tumor Rates			
Overall *(b)*	5/50 (10%)	6/50 (12%)	3/50 (6%)
Adjusted *(c)*	12.4%	14.9%	6.9%
Terminal *(d)*	4/39 (10%)	5/39 (13%)	1/39 (3%)
Statistical Tests *(e)*			
Life Table	P=0.310N	P=0.497	P=0.361N
Incidental Tumor Test	P=0.237N	P=0.517	P=0.296N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.303N	P=0.500	P=0.357N
Hematopoietic System: All Malignant Lymphoma			
Tumor Rates			
Overall *(b)*	11/50 (22%)	13/50 (26%)	16/50 (32%)
Adjusted *(c)*	25.2%	30.8%	36.9%
Terminal *(d)*	7/39 (18%)	10/39 (26%)	12/39 (31%)
Statistical Tests *(e)*			
Life Table	P=0.169	P=0.405	P=0.202
Incidental Tumor Test	P=0.135	P=0.420	P=0.132
Cochran-Armitage Trend, Fisher Exact Tests	P=0.154	P=0.408	P=0.184
Hematopoietic System: Lymphocytic Leukemia			
Tumor Rates			
Overall *(b)*	3/50 (6%)	0/50 (0%)	0/50 (0%)
Adjusted *(c)*	7.7%	0.0%	0.0%
Terminal *(d)*	3/39 (8%)	0/39 (0%)	0/39 (0%)
Statistical Tests *(e)*			
Life Table	P=0.037N	P=0.121N	P=0.121N
Incidental Tumor Test	P=0.037N	P=0.121N	P=0.121N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.037N	P=0.121N	P=0.121N

TABLE 17. ANALYSIS OF PRIMARY TUMORS IN FEMALE MICE *(a)* (Continued)

	Control	Low Dose	High Dose
Hematopoietic System: Leukemia			
Tumor Rates			
Overall *(b)*	3/50 (6%)	0/50 (0%)	1/50 (2%)
Adjusted *(c)*	7.7%	0.0%	2.1%
Terminal *(d)*	3/39 (8%)	0/39 (0%)	0/39 (0%)
Statistical Tests *(e)*			
Life Table	P=0.174N	P=0.121N	P=0.301N
Incidental Tumor Test	P=0.129N	P=0.121N	P=0.225N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.176N	P=0.121N	P=0.309N
Hematopoietic System: Malignant Lymphoma or Leukemia			
Tumor Rates			
Overall *(b)*	14/50 (28%)	13/50 (26%)	17/50 (34%)
Adjusted *(c)*	32.2%	30.8%	38.2%
Terminal *(d)*	10/39 (26%)	10/39 (26%)	12/39 (31%)
Statistical Tests *(e)*			
Life Table	P=0.306	P=0.508N	P=0.349
Incidental Tumor Test	P=0.292	P=0.486N	P=0.305
Cochran-Armitage Trend, Fisher Exact Tests	P=0.291	P=0.500N	P=0.333
Circulatory System: Hemangiosarcoma			
Tumor Rates			
Overall *(b)*	2/50 (4%)	1/50 (2%)	5/50 (10%)
Adjusted *(c)*	5.1%	2.6%	12.5%
Terminal *(d)*	2/39 (5%)	1/39 (3%)	4/39 (10%)
Statistical Tests *(e)*			
Life Table	P=0.135	P=0.500N	P=0.220
Incidental Tumor Test	P=0.102	P=0.500N	P=0.161
Cochran-Armitage Trend, Fisher Exact Tests	P=0.133	P=0.500N	P=0.218
Liver: Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	3/50 (6%)	1/49 (2%)	3/50 (6%)
Adjusted *(c)*	7.7%	2.6%	7.2%
Terminal *(d)*	3/39 (8%)	1/39 (3%)	2/39 (5%)
Statistical Tests *(e)*			
Life Table	P=0.592N	P=0.305N	P=0.660N
Incidental Tumor Test	P=0.539N	P=0.305N	P=0.592N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.593	P=0.316N	P=0.661
Pituitary: Adenoma, Chromophobe Adenoma, or Carcinoma			
Tumor Rates			
Overall *(b)*	3/43 (7%)	2/42 (5%)	1/47 (2%)
Adjusted *(c)*	8.4%	4.2%	2.6%
Terminal *(d)*	2/33 (6%)	0/33 (0%)	1/38 (3%)
Statistical Tests *(e)*			
Life Table	P=0.206N	P=0.502N	P=0.272N
Incidental Tumor Test	P=0.282N	P=0.561N	P=0.326N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.197N	P=0.511N	P=0.275N

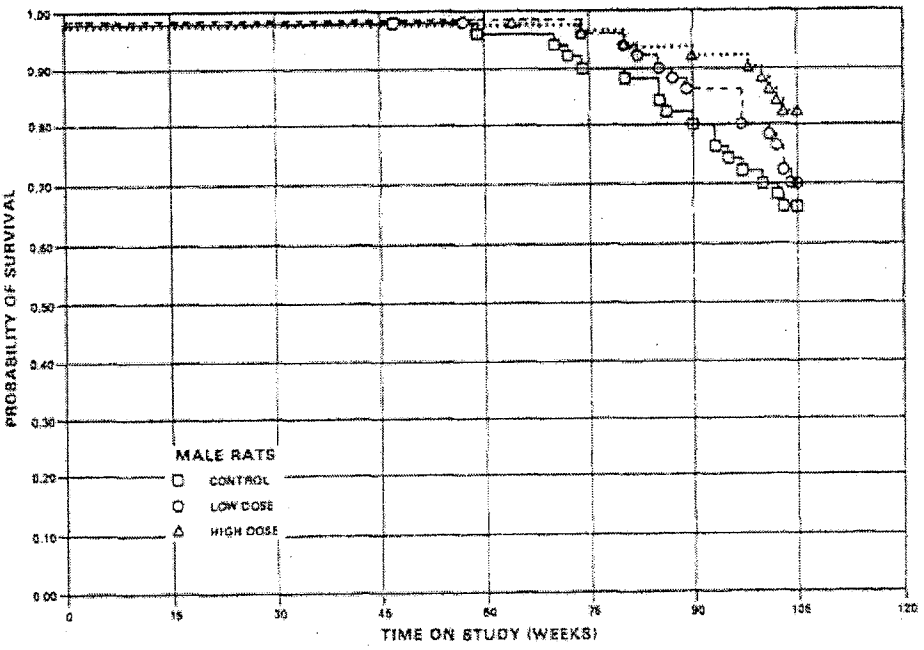
TABLE 17. ANALYSIS OF PRIMARY TUMORS IN FEMALE MICE *(a)* (Continued)

	Control	Low Dose	High Dose
Uterus: Endometrial Stromal Polyp			
Tumor Rates			
Overall *(b)*	3/50 (6%)	2/48 (4%)	0/50 (0%)
Adjusted *(c)*	7.3%	5.1%	0.0%
Terminal *(d)*	2/39 (5%)	2/39 (5%)	0/39 (0%)
Statistical Tests *(e)*			
Life Table	P=0.085N	P=0.504N	P=0.127N
Incidental Tumor Test	P=0.058N	P=0.454N	P=0.070N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.083N	P=0.520N	P=0.121N

(a) Dosed groups received doses of 25,000 or 50,000 ppm of ascorbic acid in the diet.
(b) Number of tumor bearing animals/number of animals examined at the site.
(c) Kaplan-Meier estimated lifetime tumor incidence after adjusting for intercurrent mortality.
(d) Observed tumor incidence at terminal kill.
(e) Beneath the control incidence are the P-values associated with the trend test. Beneath the dosed group incidence are the P-values corresponding to pairwise comparisons between that dosed group and the controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as non-fatal. The Cochran-Armitage and Fisher's exact tests compare directly the overall incidence rates. A negative trend or lower incidence is indicated by (N).

There were no treatment related effects on the tumor incidence. L-ascorbic acid was not carcinogenic in this study.

Rats: The results indicated that the survivals of the female rats were comparable between control and treatment groups. The survivals of the high dosed male rats were greater than that in the controls. The survival data were presented in Figure 2 in this report and this figure is attached below.



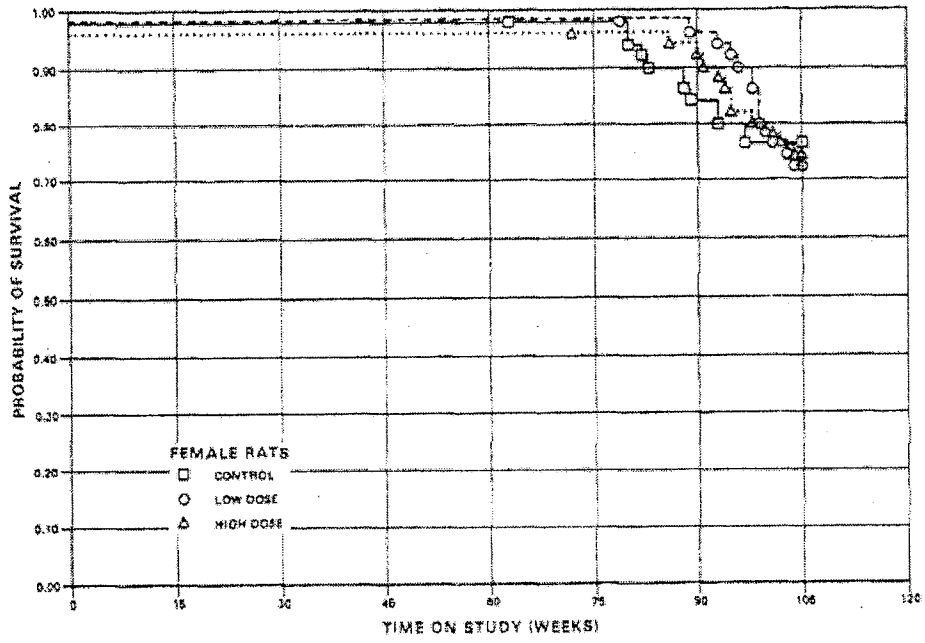


Figure 2. Survival Curves for Rats Fed Diets Containing L-Ascorbic Acid

The final body weight was lower by 13% in the high dose rats as compared to the control. The body weight data were summarized in Table 6 in this report and this table is attached below.

TABLE 6. CUMULATIVE MEAN BODY WEIGHT CHANGE (RELATIVE TO CONTROLS) OF RATS FED DIETS CONTAINING L-ASCORBIC ACID IN THE 2-YEAR STUDY

		Cumulative Mean Body Weight Change (grams)			Weight Differential Relative to Controls *(a)* (percent)	
	Week No.	Control	Low Dose	High Dose	Low Dose	High Dose
Males	0	99 *(b)*	97 *(b)*	99 *(b)*		
	1	36	35	35	- 3	- 3
	22	246	241	250	- 2	+ 2
	39	294	282	292	- 4	- 1
	61	321	315	321	- 2	0
	82	298	296	301	- 1	+ 1
	100	299	283	288	- 5	- 4
		398 *(c)*	380 *(c)*	387 *(c)*	- 5 *(d)*	- 3 *(d)*
Females	0	87 *(b)*	88 *(b)*	88 *(b)*		
	1	18	15	16	-17	-11
	22	106	100	98	- 6	- 8
	39	126	121	114	- 4	-10
	61	163	151	142	- 7	-13
	82	173	162	149	- 6	-14
	100	193	169	157	-12	-19
		280 *(c)*	257 *(c)*	245 *(c)*	- 8 *(d)*	-13 *(d)*

(a) Weight Differential Relative to Controls =

$$\frac{\text{Weight Change (Dosed Group)} - \text{Weight Change (Control Group)}}{\text{Weight Change (Control Group)}} \times 100$$

(b) Initial weight
(c) Mean body weight at week 100
(d) Mean body weight relative to controls

The tumor data were summarized in Tables 9 and 10 in this report and these tables are attached below..

TABLE 9. ANALYSIS OF PRIMARY TUMORS IN MALE RATS *(a)*

	Control	Low Dose	High Dose
Hematopoietic System: Undifferentiated Leukemia			
Tumor Rates			
Overall *(b)*	17/50 (34%)	16/50 (32%)	14/50 (28%)
Adjusted *(c)*	39.5%	36.3%	29.6%
Terminal *(d)*	8/33 (24%)	8/35 (23%)	8/41 (20%)
Statistical Tests *(e)*			
Life Table	P=0.152N	P=0.415N	P=0.176N
Incidental Tumor Test	P=0.513N	P=0.577N	P=0.568N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.295N	P=0.500N	P=0.333N
Pituitary: Adenoma or Chromophobe Adenoma			
Tumor Rates			
Overall *(b)*	10/47 (21%)	9/45 (20%)	14/50 (28%)
Adjusted *(c)*	28.4%	26.6%	31.5%
Terminal *(d)*	8/32 (25%)	8/32 (25%)	11/41 (27%)
Statistical Tests *(e)*			
Life Table	P=0.415	P=0.490N	P=0.474
Incidental Tumor Test	P=0.297	P=0.564N	P=0.333
Cochran-Armitage Trend, Fisher Exact Tests	P=0.250	P=0.543	P=0.298
Pituitary: Adenoma, Adenocarcinoma, or Carcinoma			
Tumor Rates			
Overall *(b)*	12/47 (26%)	9/45 (20%)	15/50 (30%)
Adjusted *(c)*	33.0%	26.6%	33.2%
Terminal *(d)*	9/32 (28%)	8/32 (25%)	11/41 (27%)
Statistical Tests *(e)*			
Life Table	P=0.524	P=0.303N	P=0.583
Incidental Tumor Test	P=0.371	P=0.377N	P=0.398
Cochran-Armitage Trend, Fisher Exact Tests	P=0.342	P=0.351N	P=0.396
Adrenal: Pheochromocytoma			
Tumor Rates			
Overall *(b)*	8/49 (16%)	10/50 (20%)	14/50 (28%)
Adjusted *(c)*	21.9%	26.7%	32.3%
Terminal *(d)*	5/33 (15%)	8/35 (23%)	12/41 (29%)
Statistical Tests *(e)*			
Life Table	P=0.224	P=0.461	P=0.267
Incidental Tumor Test	P=0.135	P=0.475	P=0.161
Cochran-Armitage Trend, Fisher Exact Tests	P=0.098	P=0.416	P=0.124
Thyroid: C-Cell Adenoma			
Tumor Rates			
Overall *(b)*	2/49 (4%)	4/50 (8%)	6/50 (12%)
Adjusted *(c)*	6.1%	11.0%	14.6%
Terminal *(d)*	2/33 (6%)	3/35 (9%)	6/41 (15%)
Statistical Tests *(e)*			
Life Table	P=0.167	P=0.369	P=0.212
Incidental Tumor Test	P=0.151	P=0.371	P=0.212
Cochran-Armitage Trend, Fisher Exact Tests	P=0.103	P=0.349	P=0.141

TABLE 9. ANALYSIS OF PRIMARY TUMORS IN MALE RATS *(a)* (Continued)

	Control	Low Dose	High Dose
Thyroid: C-Cell Carcinoma			
Tumor Rates			
Overall *(b)*	4/49 (8%)	2/50 (4%)	2/50 (4%)
Adjusted *(c)*	12.1%	5.3%	4.6%
Terminal *(d)*	4/33 (12%)	1/35 (3%)	1/41 (2%)
Statistical Tests *(e)*			
Life Table	P=0.179N	P=0.305N	P=0.244N
Incidental Tumor Test	P=0.218N	P=0.305N	P=0.282N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.244N	P=0.329N	P=0.329N
Thyroid: C-Cell Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	5/49 (10%)	5/50 (10%)	8/50 (16%)
Adjusted *(c)*	15.2%	13.2%	18.9%
Terminal *(d)*	5/33 (15%)	3/35 (9%)	7/41 (17%)
Statistical Tests *(e)*			
Life Table	P=0.360	P=0.584N	P=0.429
Incidental Tumor Test	P=0.299	P=0.583N	P=0.397
Cochran-Armitage Trend, Fisher Exact Tests	P=0.232	P=0.617N	P=0.290
Preputial Gland: Adenocarcinoma			
Tumor Rates			
Overall *(b)*	3/50 (6%)	1/50 (2%)	0/50 (0%)
Adjusted *(c)*	8.4%	2.9%	0.0%
Terminal *(d)*	2/33 (6%)	1/35 (3%)	0/41 (0%)
Statistical Tests *(e)*			
Life Table	P=0.045N	P=0.287N	P=0.092N
Incidental Tumor Test	P=0.059N	P=0.291N	P=0.141N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.060N	P=0.309N	P=0.121N
Testis: Interstitial-Cell Tumor			
Tumor Rates			
Overall *(b)*	48/50 (96%)	49/50 (98%)	46/49 (94%)
Adjusted *(c)*	100.0%	100.0%	100.0%
Terminal *(d)*	33/33 (100%)	35/35 (100%)	40/40 (100%)
Statistical Tests *(e)*			
Life Table	P=0.016N	P=0.406N	P=0.018N
Incidental Tumor Test	P=0.029N	P=0.610N	P=0.059N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.391N	P=0.500	P=0.490N

(a) Dosed groups received doses of 25,000 or 50,000 ppm of ascorbic acid in the diet.
(b) Number of tumor bearing animals/number of animals examined at the site.
(c) Kaplan-Meier estimated lifetime tumor incidence after adjusting for intercurrent mortality.
(d) Observed tumor incidence at terminal kill.
(e) Beneath the control incidence are the P-values associated with the trend test. Beneath the dosed group incidence are the P-values corresponding to pairwise comparisons between that dosed group and the controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as non-fatal. The Cochran-Armitage and Fisher's exact tests compare directly the overall incidence rates. A negative trend or lower incidence is indicated by (N).

TABLE 10. ANALYSIS OF PRIMARY TUMORS IN FEMALE RATS *(a)*

	Control	Low Dose	High Dose
Hematopoietic System: Undifferentiated Leukemia			
Tumor Rates			
Overall *(b)*	6/50 (12%)	17/50 (34%)	12/50 (24%)
Adjusted *(c)*	13.9%	36.9%	27.8%
Terminal *(d)*	3/38 (8%)	8/36 (22%)	7/37 (19%)
Statistical Tests *(e)*			
Life Table	P=0.121	P=0.017	P=0.114
Incidental Tumor Test	P=0.070	P=0.012	P=0.072
Cochran-Armitage Trend, Fisher Exact Tests	P=0.097	P=0.008	P=0.096
Hematopoietic System: Lymphoma			
Tumor Rates			
Overall *(b)*	3/50 (6%)	2/50 (4%)	0/50 (0%)
Adjusted *(c)*	7.2%	4.4%	0.0%
Terminal *(d)*	1/38 (3%)	0/36 (0%)	0/37 (0%)
Statistical Tests *(e)*			
Life Table	P=0.078N	P=0.461N	P=0.122N
Incidental Tumor Test	P=0.053N	P=0.315N	P=0.123N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.082N	P=0.500N	P=0.121N
Pituitary: Adenoma or Chromophobe Adenoma			
Tumor Rates			
Overall *(b)*	25/50 (50%)	19/50 (38%)	15/50 (30%)
Adjusted *(c)*	57.9%	47.2%	38.4%
Terminal *(d)*	20/38 (53%)	15/36 (42%)	13/37 (35%)
Statistical Tests *(e)*			
Life Table	P=0.035N	P=0.197N	P=0.043N
Incidental Tumor Test	P=0.019N	P=0.090N	P=0.025N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.026N	P=0.157N	P=0.033N
Pituitary: Carcinoma			
Tumor Rates			
Overall *(b)*	1/50 (2%)	2/50 (4%)	3/50 (6%)
Adjusted *(c)*	2.6%	5.6%	7.9%
Terminal *(d)*	1/38 (3%)	2/36 (6%)	2/37 (5%)
Statistical Tests *(e)*			
Life Table	P=0.218	P=0.481	P=0.300
Incidental Tumor Test	P=0.238	P=0.481	P=0.359
Cochran-Armitage Trend, Fisher Exact Tests	P=0.222	P=0.500	P=0.309
Pituitary: Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	26/50 (52%)	20/50 (40%)	18/50 (36%)
Adjusted *(c)*	60.2%	49.7%	45.0%
Terminal *(d)*	21/38 (55%)	16/36 (44%)	15/37 (41%)
Statistical Tests *(e)*			
Life Table	P=0.083N	P=0.200N	P=0.100N
Incidental Tumor Test	P=0.047N	P=0.092N	P=0.055N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.065N	P=0.158N	P=0.079N

TABLE 10. ANALYSIS OF PRIMARY TUMORS IN FEMALE RATS *(a)* (Continued)

	Control	Low Dose	High Dose
Adrenal: Cortical Adenoma			
Tumor Rates			
Overall *(b)*	3/50 (6%) *(f)*	2/50 (4%)	1/49 (2%)
Adjusted *(c)*	7.9%	5.6%	2.7%
Terminal *(d)*	3/38 (8%)	2/36 (6%)	1/37 (3%)
Statistical Tests *(e)*			
Life Table	P=0.231N	P=0.525N	P=0.314N
Incidental Tumor Test	P=0.231N	P=0.525N	P=0.314N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.228N	P=0.500N	P=0.316N
Adrenal: Pheochromocytoma			
Tumor Rates			
Overall *(b)*	4/50 (8%)	6/50 (12%)	7/49 (14%)
Adjusted *(c)*	9.7%	15.0%	18.3%
Terminal *(d)*	3/38 (8%)	4/36 (11%)	6/37 (16%)
Statistical Tests *(e)*			
Life Table	P=0.213	P=0.368	P=0.255
Incidental Tumor Test	P=0.274	P=0.335	P=0.315
Cochran-Armitage Trend, Fisher Exact Tests	P=0.204	P=0.370	P=0.251
Thyroid: C-Cell Adenoma			
Tumor Rates			
Overall *(b)*	2/49 (4%)	6/50 (12%)	4/49 (8%)
Adjusted *(c)*	5.4%	16.7%	10.1%
Terminal *(d)*	2/37 (5%)	6/36 (17%)	3/37 (8%)
Statistical Tests *(e)*			
Life Table	P=0.294	P=0.124	P=0.345
Incidental Tumor Test	P=0.251	P=0.124	P=0.276
Cochran-Armitage Trend, Fisher Exact Tests	P=0.289	P=0.141	P=0.339
Thyroid: C-Cell Adenoma or Carcinoma			
Tumor Rates			
Overall *(b)*	2/49 (4%)	7/50 (14%)	5/49 (10%)
Adjusted *(c)*	5.4%	19.4%	12.0%
Terminal *(d)*	2/37 (5%)	7/36 (19%)	3/37 (8%)
Statistical Tests *(e)*			
Life Table	P=0.203	P=0.072	P=0.232
Incidental Tumor Test	P=0.140	P=0.072	P=0.131
Cochran-Armitage Trend, Fisher Exact Tests	P=0.194	P=0.085	P=0.218
Mammary Gland: Fibroadenoma			
Tumor Rates			
Overall *(b)*	5/50 (10%)	6/50 (12%)	8/50 (16%)
Adjusted *(c)*	12.3%	15.8%	18.9%
Terminal *(d)*	3/38 (8%)	5/36 (14%)	4/37 (11%)
Statistical Tests *(e)*			
Life Table	P=0.235	P=0.499	P=0.290
Incidental Tumor Test	P=0.295	P=0.530	P=0.400
Cochran-Armitage Trend, Fisher Exact Tests	P=0.226	P=0.500	P=0.277

TABLE 10. ANALYSIS OF PRIMARY TUMORS IN FEMALE RATS *(a)* (Continued)

	Control	Low Dose	High Dose
Clitoral Gland: Adenocarcinoma			
Tumor Rates			
Overall *(b)*	3/50 (6%)	0/50 (0%)	0/50 (0%)
Adjusted *(c)*	7.0%	0.0%	0.0%
Terminal *(d)*	1/38 (3%)	0/36 (0%)	0/37 (0%)
Statistical Tests *(e)*			
Life Table	P=0.038N	P=0.120N	P=0.125N
Incidental Tumor Test	P=0.045N	P=0.110N	P=0.123N
Cochran-Armitage Trend, Fisher Exact Tests	P=0.037N	P=0.121N	P=0.121N
Uterus: Endometrial Stromal Polyp			
Tumor Rates			
Overall *(b)*	13/50 (26%)	9/50 (18%)	13/50 (26%)
Adjusted *(c)*	33.1%	21.9%	32.1%
Terminal *(d)*	12/38 (32%)	5/36 (14%)	10/37 (27%)
Statistical Tests *(e)*			
Life Table	P=0.534	P=0.262N	P=0.572
Incidental Tumor Test	P=0.539N	P=0.162N	P=0.553
Cochran-Armitage Trend, Fisher Exact Tests	P=0.547	P=0.235N	P=0.590
Uterus: Endometrial Stromal Polyp or Sarcoma			
Tumor Rates			
Overall *(b)*	13/50 (26%)	10/50 (20%)	14/50 (28%)
Adjusted *(c)*	33.1%	24.4%	34.6%
Terminal *(d)*	12/38 (32%)	6/36 (17%)	11/37 (30%)
Statistical Tests *(e)*			
Life Table	P=0.442	P=0.348N	P=0.482
Incidental Tumor Test	P=0.460	P=0.236N	P=0.460
Cochran-Armitage Trend, Fisher Exact Tests	P=0.454	P=0.318N	P=0.500

(a) Dosed groups received doses of 25,000 or 50,000 ppm of ascorbic acid in the diet.
(b) Number of tumor bearing animals/number of animals examined at the site.
(c) Kaplan-Meier estimated lifetime tumor incidence after adjusting for intercurrent mortality.
(d) Observed tumor incidence at terminal kill.
(e) Beneath the control incidence are the P-values associated with the trend test. Beneath the dosed group incidence are the P-values corresponding to pairwise comparisons between that dosed group and the controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as non-fatal. The Cochran-Armitage and Fisher's exact tests compare directly the overall incidence rates. A negative trend or lower incidence is indicated by (N).
(f) One carcinoma was also seen in a control.

The incidence of undifferentiated leukemias was significantly higher in the low dose females (17/50, 34%) as compared to the control (6/50, 12%) with $p<0.02$. However, the incidence of this tumor was not significantly increased in the high dose group (12/50, 24% with $p>0.07$). The trend test was not significantly ($p\geq0.07$). Therefore, L-ascorbic acid was not carcinogenic in this study.

2.6.6.6. Reproductive and developmental toxicology

Oral Segment I Fertility and Reproductive Performance Study in Rat with Movicol
(17521/03)

Testing Laboratories: [redacted]

Study Start and Completion Dates: February 24, 2004 and April 28, 2005

GLP Requirement: A statement of compliance with GLP regulations and the quality assurance unit was included.

Animals: Males: 232-252 g, 8-9 weeks old
Females: 155-175 g, 8-9 weeks old
CD/[redacted] Rats

Drug Batch Nos.: 112903

Methods: To study the potential effects of movicol on fertility and reproductive functions in rats, movicol was given by oral gavage to male rats (20/group) for 4 weeks before mating, during mating until sacrifice (for a total of 49 days) and to female rats (20/group) for 2 weeks before mating, during mating, and until day 7 of gestation. The doses tested were 0, 10, 20, and 40 g/kg/day given by two divided doses daily. Clinical signs were observed daily. Body weights and food consumption were recorded. All females were sacrificed on day 13 of gestation, and the uterine contents and ovaries were examined for: number of fetuses and placenta, number and size of resorptions, number of corpora lutea, implantations, and location of fetuses. All animals were examined macroscopically. Sperm count, viability, and morphology were conducted.

Results: There were no treatment related deaths. Soft feces and/or diarrhea were noted in all treatment groups. Terminal body weight gain was decreased by 5.5%, 14%, and 58% in the low, mid, and high dose male groups, respectively. There were no clear treatment effects on the body weight in females. The food consumption was lower in the mid (62-68 g/kg/day) and high (50-65 g/kg/day) dose males than that in the control (66-75 g/kg/day). Treatment with movicol did not produce any effects on fertility index, sperm count, viability, and morphorlogy, estrous cycte, pre-coital time, number corpora lutea, implantation loss, resorption, live fetuses, and organ weights of testes, epididymides, and uterus. No macroscopic changes were noted. The results were summarized in Table 5.1 on pages 194-199 in volume c1.26. This table is attached below.

MOVICOL

TABLE 5-1 Fertility and Reproduction Parameters - Summary Rat

Parameter	Group 1 Control	Group 2 10 g/kg	Group 3 20 g/kg	Group 4 40 g/kg
		male animals		
Number of male animals at start of test	20	20	20	20
Number of male animals died before mating	0 of 20	0 of 20	0 of 20	0 of 20
Number of male animals evaluated for sperm parameters	20	19	20	20
Number of ultrasound-resistant spermatids per g testicular tissue x 10^6				
Mean:	68.47	72.59	64.66	71.97
SD:	8.58	21.67	19.98	15.49
t:		ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test				
Motile spermatozoa in the epididymal cauda				
Mean %:	63.76	68.93	66.29	70.56
SD:	16.05	13.83	18.62	10.23
*: ($p \leq 0.05$) / Fisher test				

MOVICOL

TABLE 5-1 Fertility and Reproduction Parameters - Summary Rat

Parameter	Group 1 Control	Group 2 10 g/kg	Group 3 20 g/kg	Group 4 40 g/kg
		male animals		
Morphologically normal spermatids in the cauda epididymis				
Mean %:	99.9	99.9	99.9	99.9
*: ($p \leq 0.05$) / Fisher test				
Number of males that mated	20	20	20	20
Number of fertile males	19	20	18	17
FERTILITY INDEX %:	95	100	90	85
*: ($p \leq 0.05$) / Fisher test				

MOVICOL

TABLE 5-1 Fertility and Reproduction Parameters - Summary Rat

Parameter		Group 1 Control	Group 2 10 g/kg	Group 3 20 g/kg	Group 4 40 g/kg
		female animals			
Number of female animals evaluated for oestrus cycle		20	20	20	20
Number of oestrus cycle					
Pre-treatment period:	Mean:	2.1	2.5	2.4	2.4
	SD:	1.0	0.7	0.7	0.6
	t:	-	ns	ns	ns
Treatment period:	Mean:	2.7	2.8	2.8	2.4
	SD:	0.6	0.4	0.4	0.7
	t:	-	ns	ns	ns
Mating period:	Mean:	0.5	0.6	0.7	0.6
	SD:	0.8	0.5	0.7	0.7
**: $p \leq 0.01$, Dunnett test or Student's t-test					
Number of female animals evaluated for pre-coital time		20	20	18	17
Pre-coital time [days]	Mean:	3.0	3.0	3.5	4.1
	SD:	2.6	1.8	2.4	3.0
	t:	-	ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test					
Females assigned to laparotomy	n=	20	20	20	20
pregnant	n=	19	20	18	18
aborted	n=	0 of 19	0 of 20	0 of 18	0 of 18

MOVICOL

TABLE 5-1 Fertility and Reproduction Parameters - Summary Rat

Parameter		Group 1 Control	Group 2 10 g/kg	Group 3 20 g/kg	Group 4 40 g/kg
		female animals			
Dams with total implantation loss	n=	0 of 19	0 of 20	0 of 18	1 of 18
Dams with viable fetuses	n=	19 of 19	20 of 20	18 of 18	17 of 17
FERTILITY INDEX	%:	95	100	90	90
*: ($p \leq 0.05$) / Fisher test					
Dams evaluated at Caesarian section	n=	19	20	16	15
Corpora lutea	total:	260	275	245	221
	Mean:	13.7	13.8	15.3	14.7
	SD:	3.4	3.1	2.4	3.2
	t:	·	ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test					
Implantation sites	total:	255	271	242	220
	Mean:	13.4	13.6	15.1	14.7
	SD:	4.1	3.6	2.9	3.5
	t:	·	ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test					
Pre-implantation loss					
	mean %:	4.4	2.9	1.9	1.7
	SD:	19.1	12.8	7.5	6.5
*: ($p \leq 0.05$) / Fisher test					

MOVICOL

TABLE 5-1 Fertility and Reproduction Parameters - Summary Rat

Parameter	Group 1 Control	Group 2 10 g/kg	Group 3 20 g/kg	Group 4 40 g/kg
female animals				
Post-implantation loss				
mean %:	6.7	3.2	8.7	12.9
SD:	11.4	3.9	10.3	26.4
*: ($p \leq 0.05$) / Fisher test				
Resorptions, total				
total:	17	10	18	17
Mean:	0.9	0.5	1.1	1.1
SD:	1.4	0.6	0.9	1.6
t:	-	ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test				
Resorptions, early				
total:	3	4	8	8
Mean:	0.2	0.2	0.5	0.5
SD:	0.4	0.4	0.8	0.7
t:	-	ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test				
Resorptions, late				
total:	14	6	10	9
Mean:	0.7	0.3	0.6	0.6
SD:	1.4	0.5	0.6	1.2
t:	-	ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test				
Resorptions, total in % of implants				
Mean %:	6.7	3.2	8.7	12.9
SD:	11.4	3.9	10.3	26.4
*: ($p \leq 0.05$) / Fisher test				

MOVICOL

TABLE 5-1 Fertility and Reproduction Parameters - Summary Rat

Parameter		Group 1 Control	Group 2 10 g/kg	Group 3 20 g/kg	Group 4 40 g/kg
		female animals			
Resorptions, early in % of implants					
	Mean %:	1.2	1.2	4.5	3.5
	SD:	2.9	2.5	10.6	5.2
*: ($p \leq 0.05$) / Fisher test					
Resorptions, late in % of implants					
	Mean %:	5.4	2.0	4.1	9.4
	SD:	11.3	3.1	4.4	26.1
*: ($p \leq 0.05$) / Fisher test					
Dead fetuses	n =	0	0	0	0
Malformed fetuses	n =	0	0	0	0
Live fetuses	total:	238	261	224	203
	Mean %:	12.5	13.1	14.0	13.5
	SD:	4.3	3.3	3.3	4.4
	t:		ns	ns	ns
**: $p \leq 0.01$, Dunnett test or Student's t-test					
Live fetuses in % of implants					
	Mean %:	93.3	96.8	91.3	87.1
	SD:	11.4	3.9	10.3	26.4
*: ($p \leq 0.05$) / Fisher test					

In summary, movicol was given by oral gavage to male rats for 4 weeks before mating, during mating until sacrifice and to female rats for 2 weeks before mating, during mating, and until day 7 of gestation. The doses tested were 0, 10, 20, and 40 g/kg/day. The mating performance and fertility were not affected.

Oral Segment II Embryo-Fetal Development Study in Rats (17522/03)

Testing Laboratories: [redacted]

Study Start and Completion Dates: February 24, 2004 and April 18, 2005

GLP Requirement: A statement of compliance with GLP regulations and the quality assurance unit was included.

Animals: Female rats: 206-280 g, ~8-9 weeks old
CD [redacted] :CD [redacted] rats

Drug Batch Nos.: 112903

Methods: To investigate the teratological potential of movicol, movicol was given by oral gavage to pregnant females from days 6 to 17 of gestation at 0, 10, 20, and 40 g/kg/day. The doses were given in two divided doses daily. In the current study, clinical condition was recorded daily. Body weights and food consumption were recorded. All pregnant females were sacrificed on day 20 of gestation for examination of their uterine contents. At necropsy, the females were examined macroscopically and live fetuses were weighed, sexed and examined for external abnormalities. Half of the fetuses were examined for visceral and the other half for skeletal abnormalities.

Results: One high dose female was found dead on test day 12. Necropsy revealed reddened lungs and the intestine filled with liquid. Diarrhea and/or soft feces were noted in the mid and high dose groups. Pilo-erection was noted in the high dose animals. Body weight gain during gestation days 7-18 were decreased by 12.6%, 25%, and 76% in the low, mid, and high dose groups, respectively. The food consumption was lower in the mid

(64-84 g/kg) and high (43-68 g/kg) dose groups as compared to the control (79-92 g/kg).

The number of resorption was increased and the number of viable fetuses was decreased due to the post-implantation loss in the high dose group. The results were presented in a table on page 48 in volume c1.27 and this table is attached below:

The following table presents the relevant reproduction data:

Parameter		Group 1 Control (n = 21)	Group 2 10 g/kg /day (n = 20)	Group 3 20 g/kg /day (n = 20)	Group 4 40 g/kg /day (n = 18)
Corpora lutea	total	317	305	279	281
	per dam	15.1	15.3	14.0	15.6
Implantation sites	total	295##	288###	265	270
	per dam	14.0	14.4	13.3	15.0
Resorptions	total	21	25	13	31
	per dam	1.0	1.3	0.7	1.7
Early resorptions	total	13	20	8	16
	per dam	0.6	1.0	0.4	0.9
Late resorptions	total	8	5	5	15
	per dam	0.4	0.3	0.3	0.8
Live fetuses	total	275	263	252	239*
	per dam	13.8 (n = 20 dams with viable fetuses	13.2	12.6	14.1 (n = 17 dams with viable fetuses
Fetuses	total	275	263	252	239*
	per dam	13.1 (n = 21 dams with both viable fetuses and resorbed implants)	13.2	12.6	13.3 (n = 18 dams with both viable fetuses and resorbed implants)
Dead fetuses at laparotomy	total	0	1	0	0
Pre-implantation loss	mean %	8.9	5.7	7.9	4.5
Post-implantation loss	mean %	10.9	10.0	5.5	13.3

The total number of live fetuses plus resorptions is 296 (275 total live fetuses plus 21 resorptions). As litter #17 had twin fetuses attached to one placenta, the number of implantation sites is reduced by one to 295.

The total number of live fetuses, resorptions and dead fetuses is 289 (263 total live fetuses plus 25 resorptions plus 1 dead fetus). As litter #28 had twin fetuses attached to one placenta, the number of implantation sites is reduced by one to 288.

* Significantly different from the controls at $p \leq 0.05$

There were no treatment related external, soft tissue, and skeletal malformations. Treatment with movicol increased the incidence of skeletal variation such as wavy ribs in the mid (59 of 126) and high (62 of 120) dose groups as compared to the control (28 of 138). The results were summarized in Tables 9, 10, and 11 on pages 74-95 in volume c1.27. These tables are attached below.

TABLE 9 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF ALL CLASSIFIED FETAL EXTERNAL OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	275	264	252	239
Live	N	275	263	252	239
Dead	N	0	1	0	0
TOTAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0
TOTAL VARIATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

Appears This Way
On Original

TABLE 9 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	275	264	252	239
Live	N	275	263	252	239
Dead	N	0	1	0	0
FOUND DEAD AT LAPAROTOMY					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.4	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	5.0	0.0	0.0
TOTAL FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.4	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	5.0	0.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF ALL CLASSIFIED FETAL SKELETAL OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	138	132	126	120
Live	N	138	131	126	120
Dead	N	0	1	0	0
TOTAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0
TOTAL VARIATIONS					
Fetal Incidence##	N	29	34	60**	63**
	%	21.0	25.8	47.6	52.5
Litter Incidence	N	13	13	17	15
	%	65.0	65.0	85.0	88.2
Affected Fetuses/Litter	MEAN%	20.6	26.6	53.6	54.2
	S.D.	24.0	31.3	38.3	32.7

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

MOVICOL - Embryo-Fetal Development in Rats
TABLE 10 SUMMARY OF ALL CLASSIFIED FETAL SKELETAL OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
TOTAL RETARDATIONS					
Fetal Incidence##	N	135	124	119	117
	%	97.8	93.9	94.4	97.5
Litter Incidence	N	20	20	20	17
	%	100.0	100.0	100.0	100.0
Affected Fetuses/Litter	MEAN%	98.0	94.3	95.5	98.2
	S.D.	6.3	8.7	14.3	7.3

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

MOVICOL - Embryo-Fetal Development in Rats
TABLE 10 SUMMARY OF FETAL SKELETAL MALFORMATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	138	132	126	120
Live	N	138	131	126	120
Dead	N	0	1	0	0
TOTAL FETAL SKELETAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

Appears This Way
On Original

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	138	132	125	120
Live	N	138	131	126	120
Dead	N	0	1	0	0
LESS THAN 13 RIB(S)					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.8	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	5.0	0.0	0.0
RIB(S) SHORTENED					
Fetal Incidence	N	0	1	2	3 *
	%	0.0	0.8	1.6	2.5
Litter Incidence	N	0	1	1	1
	%	0.0	5.0	5.0	5.9
RIB(S) WAVY					
Fetal Incidence	N	28	33	59**	62**
	%	20.3	25.0	46.8	51.7
Litter Incidence	N	12	13	17	14
	%	60.0	65.0	85.0	82.4

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
STERNEBRA(E) BIPARTITE					
Fetal Incidence	N	1	0	1	0
	%	0.7	0.0	0.8	0.0
Litter Incidence	N	1	0	1	0
	%	5.0	0.0	5.0	0.0
STERNEBRA(E) MISALIGNED (SEVERITY: SLIGHT)					
Fetal Incidence	N	1	0	1	1
	%	0.7	0.0	0.8	0.8
Litter Incidence	N	1	0	1	1
	%	5.0	0.0	5.0	5.9
TOTAL FETAL SKELETAL VARIATIONS					
Fetal Incidence##	N	29	34	60**	63**
	%	21.0	25.8	47.6	52.5
Litter Incidence	N	13	13	17	15
	%	65.0	65.0	85.0	88.2

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 10 MOVICOL - Embryo-fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	138	132	126	120
Live	N	138	131	126	120
Dead	N	0	1	0	0
5TH METACARPALIA NOT OSSIFIED					
Fetal Incidence	N	23	19	26	37**
	%	16.7	14.4	20.6	30.8
Litter Incidence	N	9	7	9	7
	%	45.0	35.0	45.0	41.2
5TH METATARSALIA NOT OSSIFIED					
Fetal Incidence	N	1	0	0	10**
	%	0.7	0.0	0.0	8.3
Litter Incidence	N	1	0	0	4
	%	5.0	0.0	0.0	23.5
CAUDAL VERTEBRAL ARCH(ES) NOT OSSIFIED					
Fetal Incidence	N	0	0	0	3 *
	%	0.0	0.0	0.0	2.5
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
CAUDAL VERTEBRAL BODIES, NO BODY OSSIFIED					
Fetal Incidence	N	3	0	0	5
	%	2.2	0.0	0.0	4.2
Litter Incidence	N	2	0	0	2
	%	10.0	0.0	0.0	11.8
CAUDAL VERTEBRAL BODIES, ONLY ONE BODY OSSIFIED					
Fetal Incidence	N	6	6	3	10
	%	4.3	4.5	2.4	8.3
Litter Incidence	N	3	3	3	5
	%	15.0	15.0	15.0	29.4
CERVICAL VERTEBRAL BODY/BODIES BIPARTITE					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.8	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	5.0	0.0	0.0
CERVICAL VERTEBRAL BODY/BODIES NOT OSSIFIED					
Fetal Incidence	N	0	0	0	3 *
	%	0.0	0.0	0.0	2.5
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
HYOID NOT OSSIFIED					
Fetal Incidence	N	35	23	32	47 *
	%	25.4	17.4	25.4	39.2
Litter Incidence	N	16	10	11	16
	%	80.0	50.0	55.0	94.1
LUMBAR VERTEBRAL ARCH/ARCHES INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	2	1	10**
	%	0.0	1.5	0.8	8.3
Litter Incidence	N	0	1	1	4 *
	%	0.0	5.0	5.0	23.5
LUMBAR VERTEBRAL ARCH/ARCHES NOT OSSIFIED					
Fetal Incidence	N	0	0	0	3 *
	%	0.0	0.0	0.0	2.5
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	11.8
LUMBAR VERTEBRAL BODY/BODIES BIPARTITE					
Fetal Incidence	N	0	1	0	1
	%	0.0	0.8	0.0	0.8
Litter Incidence	N	0	1	0	1
	%	0.0	5.0	0.0	5.9

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
LUMBAR VERTEBRAL BODY/BODIES DUMBBELL-SHAPED					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	0.8	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	5.0	0.0
LUMBAR VERTEBRAL BODY/BODIES INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	7**
	%	0.0	0.0	0.0	5.8
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	11.8
LUMBAR VERTEBRAL BODY/BODIES NOT OSSIFIED					
Fetal Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	1.7
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
METACARPALIA INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	4 *
	%	0.0	0.0	0.0	3.3
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
METATARSALIA INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	6**
	%	0.0	0.0	0.0	5.0
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	11.8
OS FIBULA NOT OSSIFIED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.8
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
OS ISCHII NOT OSSIFIED					
Fetal Incidence	N	0	0	0	8**
	%	0.0	0.0	0.0	6.7
Litter Incidence	N	0	0	0	3
	%	0.0	0.0	0.0	17.6
OS PUBIS NOT OSSIFIED					
Fetal Incidence	N	6	1 *	0 *	19**
	%	4.3	0.8	0.0	15.8
Litter Incidence	N	3	1	0	6
	%	15.0	5.0	0.0	35.3

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
PELVIC VERTEBRAL ARCH(ES) INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	6**
	%	0.0	0.0	0.0	5.0
Litter Incidence	N	0	0	0	4 *
	%	0.0	0.0	0.0	23.5
PELVIC VERTEBRAL ARCH(ES) NOT OSSIFIED					
Fetal Incidence	N	3	1	0	4
	%	2.2	0.8	0.0	3.3
Litter Incidence	N	1	1	0	1
	%	5.0	5.0	0.0	5.9
PELVIC VERTEBRAL BODY/BODIES BIPARTITE					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.8
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
PELVIC VERTEBRAL BODY/BODIES INCOMPLETELY OSSIFIED					
Fetal Incidence	N	3	0	0	9 *
	%	2.2	0.0	0.0	7.5
Litter Incidence	N	1	0	0	2
	%	5.0	0.0	0.0	11.8

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
PELVIC VERTEBRAL BODY/BODIES NOT OSSIFIED					
Fetal Incidence	N	0	0	0	4 *
	%	0.0	0.0	0.0	3.3
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
RIB(S) INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.8
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
SKULL INCOMPLETE OSSIFICATION (NASAL, FRONTAL, PARIETAL, INTERPARIETAL, SUPRAOCCIPITAL)					
Fetal Incidence	N	64	59	60	68
	%	46.4	44.7	47.6	56.7
Litter Incidence	N	15	13	14	15
	%	75.0	65.0	70.0	88.2
STERNEBRA(E) INCOMPLETELY OSSIFIED					
Fetal Incidence	N	38	35	32	15**
	%	27.5	26.5	25.4	12.5
Litter Incidence	N	15	16	14	7 *
	%	75.0	80.0	70.0	41.2

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
STERNEBRA(E) NOT OSSIFIED					
Fetal Incidence	N	87	70	77	96**
	%	63.0	53.0	61.1	80.0
Litter Incidence	N	20	17	17	17
	%	100.0	85.0	85.0	100.0
STERNEBRA(E) REDUCED IN SIZE					
Fetal Incidence	N	53	57	50	27**
	%	38.4	43.2	39.7	22.5
Litter Incidence	N	18	15	19	13
	%	90.0	75.0	95.0	76.5
THORACIC VERTEBRAL ARCHES INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	1	1
	%	0.0	0.0	0.8	0.8
Litter Incidence	N	0	0	1	1
	%	0.0	0.0	5.0	5.9
THORACIC VERTEBRAL BODIES INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	8**
	%	0.0	0.0	0.0	6.7
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	11.8

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

MOVICOL - Embryo-Fetal Development in Rats
TABLE 10 SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
THORACIC VERTEBRAL BODY/BODIES BIPARTITE					
Fetal Incidence	N	3	5	0	6
	%	2.2	3.8	0.0	5.0
Litter Incidence	N	2	4	0	4
	%	10.0	20.0	0.0	23.5
THORACIC VERTEBRAL BODY/BODIES DUMBBELL-SHAPED					
Fetal Incidence	N	0	2	4 *	4 *
	%	0.0	1.5	3.2	3.3
Litter Incidence	N	0	2	2	4 *
	%	0.0	10.0	10.0	23.5
THORACIC VERTEBRAL BODY/BODIES NOT OSSIFIED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.8
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
THORACIC VERTEBRAL BODY/BODIES REDUCED IN SIZE					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	0.8	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	5.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

MOVICOL - Embryo-Fetal Development in Rats
TABLE 10 SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
TOTAL FETAL SKELETAL RETARDATIONS					
Fetal Incidence##	N	135	124	119	117
	%	97.8	93.9	94.4	97.5
Litter Incidence	N	20	20	20	17
	%	100.0	100.0	100.0	100.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 11 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF ALL CLASSIFIED FETAL SOFT TISSUE OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	137	132	126	119
Live	N	137	132	126	119
Dead	N	0	0	0	0
TOTAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0
TOTAL VARIATIONS					
Fetal Incidence##	N	14	17	19	12
	%	10.2	12.9	15.1	10.1
Litter Incidence	N	8	12	14	7
	%	40.0	60.0	70.0	41.2
Affected Fetuses/Litter	MEAN%	10.6	17.1	13.6	12.1
	S.D.	17.2	24.3	14.0	17.5

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 11 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SOFT TISSUE MALFORMATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	137	132	126	119
Live	N	137	132	126	119
Dead	N	0	0	0	0
TOTAL FETAL SOFT TISSUE MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 11 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SOFT TISSUE VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
Litters Evaluated	N	20	20	20	17
Fetuses Evaluated	N	137	132	126	119
Live	N	137	132	126	119
Dead	N	0	0	0	0
4TH CEREBRAL VENTRICLE DILATED					
Fetal Incidence	N	1	3	3	3
	%	0.7	2.3	2.4	2.5
Litter Incidence	N	1	2	3	2
	%	5.0	10.0	15.0	11.8
BRAIN STEM: HAEMATOMA					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.8
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	5.9
CEREBELLUM: DARK DISCOLOURED					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	0.8	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	5.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 11 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SOFT TISSUE VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
DILATATION OF RENAL PELVIS					
Fetal Incidence	N	9	10	7	6
	%	6.6	7.6	5.6	5.0
Litter Incidence	N	5	9	4	4
	%	25.0	45.0	20.0	23.5
LIVER: HAEMORRHAGIC FOCUS/FOCI					
Fetal Incidence	N	5	5	6	3
	%	3.6	3.8	4.8	2.5
Litter Incidence	N	5	5	6	2
	%	25.0	25.0	30.0	11.8
MISPLACED KIDNEY					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	0.8	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	5.0	0.0
THORACIC CAVITY: HAEMORRHAGE					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	0.8	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	5.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 11 MOVICOL - Embryo-Fetal Development in Rats
SUMMARY OF FETAL SOFT TISSUE VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 10 g/kg	TEST GROUP 3 20 g/kg	TEST GROUP 4 40 g/kg
TOTAL FETAL SOFT TISSUE VARIATIONS					
Fetal Incidence	N	14	17	19	12
	%	10.2	12.9	15.1	10.1
Litter Incidence	N	8	12	14	7
	%	40.0	60.0	70.0	41.2

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

Toxicokinetics: Only the lower molecular weight PEGs were detected in the plasma and these PEGs included oligomers 11, 13, 16, and 18. Toxicokinetic data were presented in a table on page 57 in volume 1.27. This table is attached below.

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Dose [g/kg/day]	C_{max}## [µg/mL]	T_{max} [h]	$t_{1/2\ elim}$### [h]	AUC_{0-8h}### [µg/mL·h]
Gestation day 6 [sampling after first administration], administered dosages: 5, 10 or 20 g/kg/administration				
Oligomer 11				
10	-	-	-	-
20	-	-	-	-
40	0.57	2	-	-
Oligomer 13				
10	-	-	-	-
20	-	-	-	-
40	-	-	-	-
Oligomer 16				
10	-	-	-	-
20	-	-	-	-
40	0.53	0.5	-	3.03 (AUD value)
Oligomer 18				
10	-	-	-	-
20	0.61	0.5	5.81	3.07
40	1.40	0.5	2.34	4.28

Dose [g/kg/day]	C_{max}## [µg/mL]	T_{max} [h]	$t_{1/2\ elim}$### [h]	AUC_{0-8h}### [µg/mL·h]
Gestation day 17 [sampling after second administration], administered dosages: 10, 20 or 40 g/kg/day				
Oligomer 11				
10	0.61	4	-	-
20	-	-	-	-
40	-	-	-	-
Oligomer 13				
10	-	-	-	-
20	-	-	-	-
40	-	-	-	-
Oligomer 16				
10	0.56	4	-	-
20	-	-	-	-
40	-	-	-	-
Oligomer 18				
10	0.82	4.0	2.80	3.70
20	-	-	-	-
40	0.91	4.0	2.31	4.58

Values (above 0.5 µg/mL) measured in plasma samples (for timepoints see section 9.4)

Values calculated by the toxicokinetic analysis

In summary, movicol was given by oral gavage to pregnant rats from days 6 to 17 of gestation at 0, 10, 20, and 40 g/kg/day. The doses were given in two divided doses daily. Treatment with movicol suppressed body weight during gestation but did not induce malformations. Therefore, movicol was not teretogenic in this study.

Study of Embryo-Fetal Development in Rabbits with movicol by oral administration
(17524/03)

Testing Laboratories: [redacted]

Study Start and Completion Dates: February 24, 2004 and April 28, 2005

GLP Requirement: A statement of compliance with GLP regulations and the quality assurance unit was included.

Animals: Female: 2.2-3.2 kg, ~4-5 months old Himalayan Rabbits

Drug Batch Nos.: 112903

Methods: To investigate the teratological potential of mivocol, movicol was given by oral gavage to pregnant rabbits from days 6 to 20 of gestation at 0, 0.6, 2, and 6 g/kg/day. The doses were given by two divided doses daily (40 ml/kg/dose). Movicol was dissolved in tap water. The dose selection was based on the results of a dose-ranging study in rabbits (17523/03). In this study, movicol was given to pregnant rabbits (2/group) at 2, 6, 20, and 40 g/kg/day from gestation days 6 to 20. The dose of 40 g/kg/day was lethal. The dose of 20 g/kg/day induced abortion. In the current study, clinical condition was observed daily. Body weights and food consumption were recorded. All pregnant females were sacrificed on day 29 of gestation for examination of their uterine contents. At necropsy, females were examined macroscopically and live fetuses were weighed and sexed. All fetuses were examined for external, visceral, and skeletal abnormalities.

Results: Two low dose animals, five mid dose animlas, and five high dose animals aborted and were sacrificed. Soft feces

and/or diarrhea were noted mainly in the mid and high dose groups. Slight reduction of body weight (6-7%) was noted in the high dose group as compared to the control. Animals consumed less food in the treatment groups (19-78%) as compared to the control. Treatment increased resorptions and decreased number of viable fetuses in the mid and high dose groups. The mean fetal weights were lower in the treatment groups as compared to the control. The results are summarized in a table on page 45 in volume c1.30 and this table is attached below.

The following table gives the relevant reproduction data.

Parameter		Group 1 Control (n = 21)	Group 2 0.6 g/kg/day (n = 18)	Group 3 2 g/kg/day (n = 17)	Group 4 6 g/kg/day (n = 16)
Corpora lutea	total	152	134	118	126
	per dam	7.2	7.4	6.9	7.9
Implantation sites	total	105	116**	88	108**
	per dam	5.0	6.4	5.2	6.8
Resorptions	total	9	6	17*	19*
	per dam	0.4	0.3	1.0	1.2
Early resorptions	total	8	6	10	17*
	per dam	0.4	0.3	0.6	1.1
Late resorptions	total	1	0	7**	2
	per dam	0.0	0.0	0.4	0.1
Live fetuses	total	96	110	71*	89*
	per dam	4.8 (n = 20# dams with viable fetuses)	6.1	4.4 (n = 16## dams with viable fetuses)	5.6
Fetuses	total	96	110	71*	89
	per dam	4.6 (n = 21 dams with both viable fetuses and resorbed implants)	6.1	4.2 (n = 17 dams with both viable fetuses and resorbed implants)	5.6
Dead fetuses at laparotomy	total	0	0	0	0
Pre-implantation loss	mean %	30.6	13.2	25.0	15.1
Post-implantation loss	mean %	13.1	5.2	21.4	16.2

* Significantly different from the controls at $p \leq 0.05$

** Significantly different from the controls at $p \leq 0.01$

External evaluation revealed three fetuses with malrotated fore paws and one fetus with malrotated hind limbs in the high dose group (none in other groups including control). The incidences of these malformations were within the background incidences in the testing laboratory. The results were presented in a table on page 48 in volume c1.30 and this table is attached below.

Parameter	Mean value observed in this study [fetal incidence in %]		LPT background data range of individual values [fetal incidence in mean %] (n = 24 control or n = 66 test item treated groups data taken from 2000 - 2003)*	
malrotated fore paws	Group 1: Group 4:	0 3.4	0.0 ± 3.5 0.0 ± 3.7	(Control) (test item groups)
malrotated limbs	Group 1: Group 4:	0 1.1	0.0 ± 3.4 0.0 ± 6.3	(Control) (test item groups)

*data not audited by QAU

There were no treatment related skeletal and soft tissue malformations. The overall results of external, skeletal, and soft tissue examinations were presented in Tables 9, 10, and 11 on pages 74-92 in volume c1.30 and these tables are attached below.

MOVICOL - Embryo-Fetal Development in Rabbits

TABLE 9 SUMMARY OF ALL CLASSIFIED FETAL EXTERNAL OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
TOTAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	4
	%	0.0	0.0	0.0	4.5
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	12.5
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	3.0
	S.D.	0.0	0.0	0.0	8.4
TOTAL VARIATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 9 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL EXTERNAL MALFORMATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
MALROTATED FORE PAW(S)					
Fetal Incidence	N	0	0	0	3
	%	0.0	0.0	0.0	3.4
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	12.5
MALROTATED HIND LIMB(S)					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	1.1
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
TOTAL FETAL EXTERNAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	4
	%	0.0	0.0	0.0	4.5
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	12.5

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 9 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL EXTERNAL VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
TOTAL FETAL EXTERNAL VARIATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 9 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
DIED WITHIN 6 HOURS AFTER LAPAROTOMY					
Fetal Incidence	N	3	11 *	11**	12 *
	%	3.1	10.0	15.5	13.5
Litter Incidence	N	3	6	7	7
	%	15.0	33.3	43.8	43.8
DIED WITHIN 6 TO 24 HOURS AFTER LAPAROTOMY					
Fetal Incidence	N	5	8	5	6
	%	5.2	7.3	7.0	6.7
Litter Incidence	N	4	6	5	3
	%	20.0	33.3	31.3	18.8
PLACENTA: PALE					
Fetal Incidence	N	0	0	3	0
	%	0.0	0.0	4.2	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	6.3	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 9 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
TOTAL FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS					
Fetal Incidence##	N	8	19 *	18**	18 *
	%	8.3	17.3	25.4	20.2
Litter Incidence	N	6	11	8	8
	%	30.0	61.1	50.0	50.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF ALL CLASSIFIED FETAL SKELETAL OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
TOTAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0
TOTAL VARIATIONS					
Fetal Incidence##	N	7	14	8	17 *
	%	7.3	12.7	11.3	19.1
Litter Incidence	N	5	9	6	10 *
	%	25.0	50.0	37.5	62.5
Affected Fetuses/Litter	MEAN%	9.4	19.2	10.6	19.9
	S.D.	23.3	31.6	15.5	20.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF ALL CLASSIFIED FETAL SKELETAL OBSERVATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
TOTAL RETARDATIONS					
Fetal Incidence##	N	53	75 *	44	45
	%	55.2	68.2	62.0	50.6
Litter Incidence	N	17	18	15	15
	%	85.0	100.0	93.8	93.8
Affected Fetuses/Litter	MEAN%	57.1	68.5	58.6	50.1
	S.D.	36.7	32.6	34.4	31.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL SKELETAL MALFORMATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
TOTAL FETAL SKELETAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL SKELETAL VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	89
Dead	N	0	0	0	0
LESS THAN 12 RIB(S)					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	1.1
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
LUMBAR VERTEBRAL ARCH/ARCHES FUSED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	1.1
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
RIB(S) FURCATED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	1.1
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL SKELETAL VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
RIB(S) FUSED (SEVERITY: SLIGHT)					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	1.1
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
RIB(S) SHORTENED					
Fetal Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	2.2
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
STERNEBRA(E) BIPARTITE					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.9	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	5.6	0.0	0.0
STERNEBRA(E) FUSED (SEVERITY: SLIGHT)					
Fetal Incidence	N	7	11	7	12
	%	7.3	10.0	9.9	13.5
Litter Incidence	N	5	7	5	8
	%	25.0	38.9	31.3	50.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

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MOVICOL - Embryo-Fetal Development in Rabbits
TABLE 10 SUMMARY OF FETAL SKELETAL VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
STERNEBRA(E) MISALIGNED (SEVERITY: SLIGHT)					
Fetal Incidence	N	0	2	2	2
	%	0.0	1.8	2.8	2.2
Litter Incidence	N	0	2	2	2
	%	0.0	11.1	12.5	12.5
STERNEBRA(E) UPPER PART PROLONGED					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	1.4	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	6.3	0.0
TOTAL FETAL SKELETAL VARIATIONS					
Fetal Incidence##	N	7	14	8	17 *
	%	7.3	12.7	11.3	19.1
Litter Incidence	N	5	9	6	10 *
	%	25.0	50.0	37.5	62.5

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

MOVICOL - Embryo-Fetal Development in Rabbits
TABLE 10 SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	96	110	71	89
Live	N	96	110	71	99
Dead	N	0	0	0	0
HEEL BONE NOT OSSIFIED					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.9	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	5.6	0.0	0.0
HYOID NOT OSSIFIED					
Fetal Incidence	N	0	3	1	0
	%	0.0	2.7	1.4	0.0
Litter Incidence	N	0	2	1	0
	%	0.0	11.1	6.3	0.0
LESS THAN 7 LUMBAR VERTEBRAL BODIES OSSIFIED					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	1.4	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	6.3	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
OS PUBIS NOT OSSIFIED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	1.1
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
SKULL INCOMPLETE OSSIFICATION (FRONTAL, PARIETAL)					
Fetal Incidence	N	0	5 *	3*	0
	%	0.0	9.1	8.6	0.0
Litter Incidence	N	0	2	3	0
	%	0.0	11.1	18.8	0.0
STERNEBRA(E) NOT OSSIFIED					
Fetal Incidence	N	18	37 *	15	13
	%	18.8	33.6	21.1	14.6
Litter Incidence	N	10	10	6	5
	%	50.0	55.6	37.5	31.3
STERNEBRA(E) REDUCED IN SIZE					
Fetal Incidence	N	35	37	28	31
	%	36.5	33.6	39.4	34.8
Litter Incidence	N	17	15	14	14
	%	85.0	83.3	87.5	87.5

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 10 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL SKELETAL RETARDATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
TALUS NOT OSSIFIED					
Fetal Incidence	N	0	4 *	0	2
	%	0.0	3.6	0.0	2.2
Litter Incidence	N	0	1	0	1
	%	0.0	5.6	0.0	6.3
TOTAL FETAL SKELETAL RETARDATIONS					
Fetal Incidence##	N	53	75 *	44	45
	%	55.2	68.2	62.0	50.6
Litter Incidence	N	17	18	15	15
	%	85.0	100.0	93.8	93.8

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

MOVICOL - Embryo-Fetal Development in Rabbits

TABLE 11 SUMMARY OF ALL CLASSIFIED SOFT TISSUE OBSERVATIONS OF THE FETAL HEAD

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	48	55	36	44
Live	N	48	55	36	44
Dead	N	0	0	0	0
TOTAL MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN%	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0
TOTAL VARIATIONS					
Fetal Incidence##	N	9	4	1 *	6
	%	18.8	7.3	2.8	13.6
Litter Incidence	N	6	4	1	4
	%	30.0	22.2	6.3	25.0
Affected Fetuses/Litter	MEAN%	25.0	6.9	3.1	9.9
	S.D.	41.7	14.4	12.5	18.6

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

MOVICOL - Embryo-Fetal Development in Rabbits

TABLE 11 SUMMARY OF SOFT TISSUE MALFORMATIONS OF THE FETAL HEAD

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	48	55	36	44
Live	N	48	55	36	44
Dead	N	0	0	0	0
TOTAL FETAL SOFT TISSUE MALFORMATIONS					
Fetal Incidence##	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE II — MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF FETAL SOFT TISSUE VARIATIONS

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	48	55	36	44
Live	N	48	55	36	44
Dead	N	0	0	0	0
4TH CEREBRAL VENTRICLE DILATED					
Fetal Incidence	N	3	2	0	3
	%	6.3	3.6	0.0	6.8
Litter Incidence	N	2	2	0	2
	%	10.0	11.1	0.0	12.5
BRAIN STEM: DILATATION					
Fetal Incidence	N	1	0	0	0
	%	2.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	5.0	0.0	0.0	0.0
BRAIN STEM: GLOBULAR AREA CYSTIC					
Fetal Incidence	N	3	2	1	1
	%	6.3	3.6	2.8	2.3
Litter Incidence	N	2	2	1	1
	%	10.0	11.1	6.3	6.3

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)

TABLE 11 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF SOFT TISSUE VARIATIONS OF THE FETAL HEAD

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
BRAIN, MENINGES: SUBDURAL HAEMORRHAGE					
Fetal Incidence	N	3	1	0	2
	%	6.3	1.8	0.0	4.5
Litter Incidence	N	2	1	0	1
	%	10.0	5.6	0.0	6.3
MIDBRAIN: DISTINCT AREA CYSTIC					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	2.3
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	6.3
TOTAL FETAL SOFT TISSUE VARIATIONS					
Fetal Incidence##	N	9	4	1 *	6
	%	18.8	7.3	2.8	13.6
Litter Incidence	N	6	4	1	4
	%	30.0	22.2	6.3	25.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

TABLE 11 MOVICOL - Embryo-Fetal Development in Rabbits
SUMMARY OF SOFT TISSUE UNCLASSIFIED OBSERVATIONS OF THE FETAL HEAD

		TEST GROUP 1 Control	TEST GROUP 2 0.6 g/kg	TEST GROUP 3 2 g/kg	TEST GROUP 4 6 g/kg
Litters Evaluated	N	20	18	16	16
Fetuses Evaluated	N	48	55	36	44
Live	N	48	55	36	44
Dead	N	0	0	0	0
BRAIN: AUTOLYSIS					
Fetal Incidence	N	4	6	9 *	9
	%	8.3	10.9	25.0	20.5
Litter Incidence	N	4	5	6	7
	%	20.0	27.8	37.5	43.8
TOTAL FETAL SOFT TISSUE UNCLASSIFIED OBSERVATIONS					
Fetal Incidence##	N	4	6	9 *	9
	%	8.3	10.9	25.0	20.5
Litter Incidence	N	4	5	6	7
	%	20.0	27.8	37.5	43.8

SIGNIFICANTLY DIFFERENT FROM CONTROL: * = P≤0.05 ** = P≤0.01 (Fisher or Chi-square test)
FETUSES AFFECTED BY SEVERAL CHANGES WILL BE COUNTED AS ONE FETAL INCIDENCE

The results of toxicokinetic analysis indicated that only the lower molecular weight PEG oligomer 18 was detected in a few samples at concentration of 0.5 µg/ml or higher.

In summary, movicol was given to pregnant rabbits by oral gavage from days 6 to 20 of gestation at 0, 0.6, 2, and 6 g/kg/day. Treatment with movicol induced abortion in all treatment groups, increased resorptions and decreased number of viable fetuses in the mid and high dose groups. External evaluation revealed three fetuses with malrotated fore paws and one fetus with malrotated hind limbs in the high dose group (none in other groups including control). The incidences of these malformations were within the background incidences in the testing laboratory. There were no treatment related skeletal and soft tissue malformations. In conclusion, movicol was not teratologic in this study.

2.6.6.7 Local tolerance: None.

2.6.6.8 Special toxicology studies: None.

LABELING:

The labeling is according to 21 CFR, Subpart B. The following revisions in the labeling are recommended.

1. CARCINOGENESIS, MUTAGENESIS, IMPAIRMENT OF FERTILITY

Sponsor's Version:

[-]

Evaluation: Some editorial changes are recommended.

Suggested Version:

CARCINOGENESIS, MUTAGENESIS, IMPAIRMENT OF FERTILITY

Long-term studies in animals to evaluate carcinogenic potential have not been performed with moviprep. Studies to evaluate

potential for impairment of fertility or mutagenic potential have not been performed with moviprep.

2. Pregnancy Category

Sponsor's Version:

Pregnancy: Teratogenic Effects: Pregnancy Category B:

Animal reproduction studies have not been performed with moviprep. It is also not known if moviprep can cause fetal harm when administered to a pregnant women or can affect reproductive capacity. Moviprep should be given to a pregnant woman only if clearly needed.

Evaluation: If there are no animal reproduction studies and no adequate and well-controlled studies in humans, the labeling shall state "Pregnancy Category C".

Suggested Version:

Pregnancy: Teratogenic Effects: Pregnancy Category C.

Animal reproduction studies have not been conducted with moviprep. It is also not known whether moviprep can cause fetal harm when administered to a pregnant woman or can affect reproduction capacity. Moviprep should be given to a pregnant woman only if clearly needed.

3.6 OVERALL CONCLUSIONS AND RECOMMENDATIONS

Moviprep contains polyethylene glycol 3350, sodium sulfate, sodium chloride, potassium chloride, sodium ascorbate, and ascorbic acid. All these ingredients contribute to the overall osmolarity of moviprep and thus are considered as the active ingredients. The sponsor did not conduct any in vivo pharmacology studies with moviprep. However, it is expected that PEG 3350 along with other ingredients in moviprep can increase the water content of the stool and produce a voluminous liquid stool when given orally.

In the present NDA, sponsor is seeking market approval of oral moviprep for bowel cleansing prior to colonoscopy, [redacted]. In

support of this NDA, sponsor submitted the following studies with moniprep: pharmacology and 2-week oral toxicity studies in rats and dogs. Following toxicity studies with movicol were also submitted: 90-day oral toxicity studies in rats and dogs (movicol), oral segment I, oral segment II reproductive toxicity studies of in rats and rabbits (movicol), Ames tests (movicol), mouse lymphoma cell (L5178Y $TK^{+/-}$) assays at tk locus (movicol), and mouse micronucleus test (movicol). Some published reports were also submitted. No in vivo pharmacology studies, safety pharmacology studies, and pharmacokinetic studies with moviprep were submitted.

In the 2-week oral toxicity study in rats, moviprep was given by oral gavage to rats at 0, 5, 10, and 20 g/kg/day. Major treatment related changes were observed mainly at the high dose of 20 g/kg/day. These included mortality, increase in bilirubin, urea, and kidney weight. The kidney was the target organ of toxicity based on the changes of the clinical chemistry and kidney weight (no treatment related histopathological changes in the kidney).

In the 90-day oral toxicity study in rats, movicol was given by oral gavage to rats at 0, 5, 20, and 30/25 g/kg twice a day (10, 40, and 60/50 g/kg/day) for 90 days. The high dose was lethal. Treatment decreased body weight gain in the mid and high dose groups. Serum levels of bilirubin and creatinine were increased in the mid and high dose males in week 4 but not in week 13. The kidney weight was increased in the mid and high dose groups. The kidney was the target organ of toxicity based on the changes of the clinical chemistry and kidney weight (no treatment related histopathological changes in the kidney). Increased incidences of thin layer of mucosa in the gastrointestinal tract, reduced cellularity in the lymph node and spleen, and increased macrophages in the lymph nodes and thymus were noted in the high dose group. A few cases (1 or 2) of hemorrhages in the thymus were found in the treatment groups (none in the control group). The results indicated that these animals had a marked stress due to receiving a large volume of test drug for a long period time (90 days).

In the 2-week oral toxicity study in dogs, moviprep was given by oral gavage to dogs at 0, 5, 10, and 20 g/kg/day. Major treatment related changes were clinical signs of toxicity including emesis and soft or liquid feces noted in all treatment groups. Salvation was also noted in the high dose group. The gastrointestinal tract was the target organ of toxicity based on

the clinical signs of toxicity including emesis, diarrhea, and salvation.

In the 90-day oral toxicity study in dogs, movicol was given by oral gavage to dogs (4/sex/group, 3/sex/control group) at 0, 5, 20, and 30/25 g/kg twice a day (10, 40, and 60/50 g/kg/day) for 90 days. Major treatment related changes were clinical signs of toxicity including emesis, diarrhea, and salvation noted (all treatment groups), decreased terminal body weight gain (high dose group), and a reduction of cellularity and increase of foamy macrophages in lymphoid tissues. The gastrointestinal tract was the target organ of toxicity based on the clinical signs of toxicity including emesis, diarrhea, and salvation.

In Segment I oral fertility and reproductive performance study in rats, movicol was given by oral gavage to male rats for 4 weeks before mating, during mating until sacrifice and to female rats for 2 weeks before mating, during mating, and until day 7 of gestation. The doses tested were 0, 10, 20, and 40 g/kg/day. The mating performance and fertility were not affected.

In the Segment II oral teratology reproductive toxicity study in rats, movicol was given by oral gavage to pregnant females from days 6 to 17 of gestation at 0, 10, 20, and 40 g/kg/day. The doses were given in two divided doses daily. Treatment with movicol suppressed body weight during gestation but did not induce malformations. Therefore, movicol was not teretogenic in this study.

In the Segment II oral teratology reproductive toxicity study in rabbits, movicol was given by oral gavage to pregnant rabbits from days 6 to 20 of gestation at 0, 0.6, 2, and 6 g/kg/day. The doses were given by two divided doses daily. Treatment with movicol induced abortion in all treatment groups, increased resorptions and decreased number of viable fetuses in the mid and high dose groups. External evaluation revealed three fetuses with malrotated fore paws and one fetus with malrotated hind limbs in the high dose group (none in other groups including control). The incidences of these malformations were within the background incidences in the testing laboratory. There were no treatment related skeletal and soft tissue malformations. In conclusion, movicol was not teratologic in this study.

The mutagenic potential of movicol was tested in the Ames test, the mouse lymphoma cell (L5178Y $TK^{+/-}$) forward mutation assay at tk locus, and the mouse micronucleus test. Movicol was negative in these tests.

Both sodium ascorbate and ascorbic acid were not mutagenic in mouse lymphoma L5178Y TK+/- cell mutation assay (Cancer Letters, 14:151-158, 1981). Ascorbate induced a dose-dependent increase in sister-chromatid exchanges in Chinese hamster ovary cells and in human lymphocytes and increased the inhibition of DNA synthesis in Hela cells (Mutation Research, 60:321-327, 1979). Ascorbate induced mutation at HGPRT locus in Chinese hamster cells (Cancer Letters, 8:299-305, 1980).

A NTP study report (NTP-81-140) of carcinogenesis bioassays with L-ascorbic acid indicatd that L-ascorbic acid was not carcinogenic in mice and in rats.

The toxicity profiles of moviprep and movicol were characterized in rats and dogs. The results indicated that the kidney was the target organ of toxicity in rats based on the changes of the clinical chemistry and the kidney weight. In dogs, the major treatment related toxicity was emesis, diarrhea, salvation, and decreased terminal body weight gain. The gastrointestinal tract was the target organ of toxicity based on the clinical signs of toxicity including emesis, diarrhea, and salvation. The toxicity profiles of moviprep and movicol were similar. From a preclinical standpoint, approval of oral moviprep for bowel cleansing prior to colonoscopy, intestinal surgery, and barium enema X-ray examination is recommended.

Recommendations:

1. From a preclinical standpoint, approval of oral moviprep for bowel cleansing prior to colonoscopy [redacted] is recommended.

2. Labeling should be revised as recommended.

Ke Zhang, Ph.D. Date
Pharmacologist, HFD-180

Comments:

Jasti B. Choudary, B.V.Sc., Ph.D. Date
Supervisory Pharmacologist, HFD-180

CC:
NDA
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HFD-181/CSO
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/s/

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2/24/2006 02:49:29 PM
PHARMACOLOGIST

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