

Other Studies:**L-754,030: Exploratory Sixteen-Day Oral Hepatotoxicity Study in Rats (TT #96-613-0)**

Methods: Male and female Sprague-Dawley rats (approximately 6 weeks old; 16 animals/sex/group) were administered L-754,030 (batch # 008; purity, 99%) at oral (gavage) doses of 5, 25 and 125 mg/kg/day for 17 days. Control animals were administered the vehicle (0.5% methylcellulose/0.02% sodium lauryl sulfate). The body weights of the animals were recorded pre-test, once in Week 1 and twice in Week 2. At the end of the treatment period, the animals were sacrificed and subjected to partial necropsy, limited to the liver. Samples of liver were collected for determination of hepatic 7-ethoxy-4-trifluoromethylcoumarin O-deethylase (EFCOD) activity. In addition, cytochrome P450 enzyme activity and peroxisomal fatty acyl COA-oxidase (FACO) activities of the liver samples were determined. Liver microsomes prepared from these rats were used to study the metabolism of L-754,030. Liver samples were examined microscopically after staining with hematoxylin and eosin.

For determination of the plasma drug concentrations, blood samples were collected from 4 animals/sex/time point on Day 1 and Day 14 at 0.5, 2, 4, 6, 8, 10 and 24 hours post-dosing.

Results:

Liver weights: There were significant increases in the mean liver weights of females receiving the 5 mg/kg/day dose, and in males and females receiving 25 and 125 mg/kg/day doses. The % changes in the mean liver weights of male and female animals are shown in the Table below.

Liver Weights as % Change from Controls

L-754,030	5 mg/kg/day	25 mg/kg/day	125 mg/kg/day
Males			
Liver weight (g)	0	+ 11.8 S	+ 14.8 S
Liver weight (% B.W.)	+ 1.9 NS	+ 11.7 S	+ 14.4 S
Females			
Liver weight (g)	+ 6.7 S	+ 7.5 S	+ 35 S
Liver weight (% B.W.)	+ 6.4 S	+ 10.5 S	+ 32 S

(g) = absolute organ weights

% B.W. = weights expressed as a percent of body weights

NS= trend not statistically significant through indicated dose (P > 0.05)

S = trend statistically significant through indicated dose (P ≤ 0.05)

Gross and microscopic changes: There were no treatment-related gross pathological changes in the liver. No treatment-related microscopic changes in the liver were observed in controls and in

females from the 5 mg/kg/day group. Centrilobular hypertrophy of hepatocytes (very slight to slight) was observed in all other groups treated with L-754,030. The incidences of centrilobular hepatocellular hypertrophy in male and female rats are shown in the Table below.

L-754,030	5 mg/kg/day	25 mg/kg/day	125 mg/kg/day
Hepatocyte, centrilobular hypertrophy	M (1/16) VS F (0/16)	M (5/16) VS F (1/16) VS	M (9/16) (7 VS ; 2SL) F (3/16) VS

VS: Very Slight

SL: Slight

Liver enzyme activities: EFCOD activity of the liver was significantly increased in both males and females at all dose levels. FACO activity of the liver was not affected by treatment of the rats with L-754,030. Administration of L-754,030 to male and female rats resulted in an induction of CYP3A, with greater effects occurring in females. In males, there were inductions of CYP2B.

Liver microsomal metabolism assays: The rate of metabolism of L-754,030 (10 μ M) was approximately 4.4 pmol/min/mg protein in liver microsomes from untreated male rats, and it was about 7-fold higher than that of females. The rate of metabolism was approximately 4-fold increased (20 pmol/min/mg protein) in male rats receiving the 5 mg/kg/day dose of L-754,030, with no further increase with increasing dose. In female rats, the rate of metabolism of L-754,030 was increased by about 23-fold (approximately 13 pmol/min/mg protein) at all three dose levels. Thus, repeated treatment of the rat with L-754,030 resulted in the induction of its own metabolism in liver.

Plasma concentrations of L-754,030 in male and female rats increased with increasing oral doses, and the maximum plasma concentrations were reached in 4 to 6 hours. The pharmacokinetic parameters for male and female rats are summarized in the Table below.

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Pharmacokinetic Parameters for L-754,030 in Plasma from Female Rats
Orally Dosed Daily at 5, 25, or 125 mg/kg for 15 days^a (#TT 96-613-0)

Parameters	Drug Day 1		
	5	25	125
Dose (mg/kg)	5	25	125
C _{max} (ng/mL)	857±62	2211±123	3764±383
T _{max} (hr)	6	6	6
AUC ^b (ng-hr/mL)	8588	21675	43618

Parameters	Drug Day 14		
	5	25	125
Dose (mg/kg)	5	25	125
C _{max} (ng/mL)	474±176	768±209	1631±269
T _{max} (hr)	4	4	6
AUC ^b (ng-hr/mL)	4231	6466	14657 ^b

^a See footnote "a" from Table 1.

^b See footnote "b" from Table 1.

Thus, treatment of rats with L-754,030 resulted in a significant increase in the liver weights and centrilobular hepatocellular hypertrophy in both males and females. There were increased EFCOD activities in the liver of male and female rats administered L-754,030. Metabolism of L-754,030 was increased in the liver fractions of rats treated with L-754,030 for 15 days, suggesting that L-754,030 induces its own metabolism in the liver.

2.6.6.9 Discussion and Conclusions

L-758, 298 caused a local irritation at the injection sites (discoloration, stiffness, sloughing of the skin, discharge) in rats when administered at i.v. doses up to 25 mg/kg/day for 5 days. Intravenous administration of single doses of 5.0 and 7.5 mg/kg doses to rats was associated with edema, hemorrhage and/or cellular infiltration in the subcutis at the injection sites of the tail. Topical application of L-754, 030 was not irritating to the skin of rabbits, and it was mildly irritating to the eyes of rabbits. L-758, 298 caused hemolysis of washed red blood cells from rats, dogs and humans. However, it did not cause a hemolysis in human whole blood. MK-0869 caused an increase in TSH levels and thyroxine clearance in male and female rats. Increased TSH levels may be related to increased live enzyme activity that caused increased metabolism of thyroid hormones. Fosaprepitant caused a moderate induction of cytochrome P₄₅₀ enzyme activity (7-ethoxy-4-trifluoromethylcoumarin-O-deethylase and fatty acyl-CoA oxidase activity) in the mouse liver. It also caused an increase in the liver enzyme activity (7-ethoxy-4-trifluorocoumarin-O-deethylase and fatty acyl-CoA-oxidase) in microsomes prepared from mouse liver. The liver enzyme inducing

capability of the drug may be responsible for the hepatocellular hypertrophy and thyroid follicular cell hyperplasia observed in rats during repeated administration of the drug.

2.6.6.10 Tables and Figures

Tables and Figures are incorporated in appropriate sections of the review.

2.6.7 TOXICOLOGY TABULATED SUMMARY

N/A

LABELING:

CARCINOGENESIS, MUTAGENESIS AND IMPAIRMENT OF FERTILITY

Sponsor's Version:

Carcinogenicity studies were conducted in SpragueDawley rats and CD-1 mice for 2 years. In the rat carcinogenicity studies, animals were treated with oral doses ranging from 0.05 to 1000 mg/kg twice daily. The highest dose which was the maximum feasible dose based on the physical properties of the dosing formulation, produced a systemic exposure to Aprepitant (plasma $AUC_{0-24 \text{ hr}}$) of 0.7 to 1.6 times the human exposure ($AUC_{0-24 \text{ hr}} = 19.6 \text{ mcg.hr/ml}$) at the recommended dose of 125 mg/day. Treatment with Aprepitant at doses of 5 to 1000 mg/kg twice daily produced thyroid follicular cell adenomas and carcinomas in male rats. In female rats, it produced hepatocellular adenomas at 5 to 1000 mg/kg twice daily and hepatocellular carcinomas and thyroid follicular cell adenomas at 125 to 1000 mg/kg twice daily. In mouse carcinogenicity studies, animals were treated with oral doses ranging from 2.5 to 2000 mg/kg/day. The highest dose, which was the maximum feasible dose, produced a systemic exposure of about 2.8 to 3.6 times the human exposure at the recommended dose. Treatment with Aprepitant produced hepatocellular adenomas and/or carcinomas at 500 to 2000 mg/kg in female mice, hepatocellular carcinomas at 1000 and 2000 mg/kg/day in male mice, and skin fibrosarcomas in male mice of 125 and 500 mg/kg/day groups. Carcinogenicity studies were not conducted with Fosaprepitant.

Aprepitant and Fosaprepitant were not genotoxic in the Ames test, the human lymphoblastoid cell (TK6) mutagenesis test, the rat hepatocyte DNA strand break test, the Chinese hamster ovary (CHO) cell chromosome aberration test and the mouse micronucleus test.

Fosaprepitant, when administered intravenously, is rapidly converted to Aprepitant. In the fertility studies conducted with Fosaprepitant and Aprepitant, the highest systemic exposures to Aprepitant were obtained following oral administration of Aprepitant. Oral Aprepitant did not affect the fertility or general reproductive performance of male or female rats at doses up to the

maximum feasible dose of 1000 mg/kg twice daily (providing exposure in male rats lower than the exposure at the recommended human dose and exposure in female rats at about 1.6 times the human exposure).

Evaluation: The carcinogenicity studies were conducted with aprepitant (MK-0869) and not with fosaprepitant. This should be mentioned in this section of the labeling. Since no studies were conducted with fosaprepitant, the labeling should conform to the existing labeling of aprepitant (EMEND). Studies on the fertility and general reproductive performance of male and female rats were conducted with intravenous fosaprepitant (L-758, 298). The findings of these studies should be included in this section of the labeling.

Proposed version:

Carcinogenesis, Mutagenesis and Impairment of Fertility

Carcinogenicity studies were conducted with aprepitant in SpragueDawley rats and CD-1 mice for 2 years. In the rat carcinogenicity studies, animals were treated with oral doses of ranging from 0.05 to 1000 mg/kg twice daily. The highest dose produced a systemic exposure to aprepitant (plasma $AUC_{0-24 \text{ hr}}$) of 0.7 to 1.6 times the human exposure ($AUC_{0-24 \text{ hr}} = 19.6 \text{ mcg.hr/ml}$) at the recommended dose of 125 mg/day. Treatment with aprepitant at doses of 5 to 1000 mg/kg twice daily caused an increase in the incidences of thyroid follicular cell adenomas and carcinomas in male rats. In female rats, it produced hepatocellular adenomas at 5 to 1000 mg/kg twice daily and hepatocellular carcinomas and thyroid follicular cell adenomas at 125 to 1000 mg/kg twice daily. In mouse carcinogenicity studies, animals were treated with oral doses ranging from 2.5 to 2000 mg/kg/day. The highest dose produced a systemic exposure of about 2.8 to 3.6 times the human exposure at the recommended dose. Treatment with aprepitant produced skin fibrosarcomas at 125 and 500 mg/kg/day doses in male mice. Carcinogenicity studies were not conducted with fosaprepitant.

Aprepitant and fosaprepitant were not genotoxic in the Ames test, the human lymphoblastoid cell (TK6) mutagenesis test, the rat hepatocyte DNA strand break test, the Chinese hamster ovary (CHO) cell chromosome aberration test and the mouse micronucleus test.

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Pregnancy. Teratogenic Effects:

Sponsor's version:

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✓

Nursing mothers

✓

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Evaluation: The sponsor conducted teratogenicity studies with fosaprepitant in rats and rabbits following intravenous administration of the drug. The findings of the intravenous teratogenicity studies should be included in this section of the labeling.

Proposed version:

**Teratogenic Effects:
Pregnancy Category B**

✓

b(4)

✓

Nursing mothers

✓

b(4)

✓

OVERALL CONCLUSIONS AND RECOMMENDATIONS

Aprepitant (MK-0869; L-754, 030) is a selective neurokinin₁ (NK₁) receptor antagonist which binds with high affinities to NK₁ receptors from different species. It has >3,000-fold higher affinities for NK₁ receptors than NK₃ receptors, and has higher affinities than other G-protein coupled receptors. Aprepitant is currently approved for the prevention of acute and delayed nausea and vomiting due to highly and moderately emetogenic chemotherapy, in combination with other anti-emetic agents. The approved 3-day regimen for orally administered aprepitant is 125 on Day 1, followed by 80 mg on Days 2 and 3. Fosaprepitant dimeglumine (MK-0517; L-758, 298) is a water soluble phosphoryl prodrug of aprepitant, and is rapidly converted to aprepitant following i.v. administration. The pharmacologic activity of fosaprepitant is derived from aprepitant. The sponsor submitted NDA 22-023 for use of intravenous fosaprepitant in the prevention of acute and delayed nausea and vomiting associated with initial and repeat courses of highly and moderately emetogenic chemotherapy. In the proposed 3-day dosing regimen, fosaprepitant will be administered at an i.v. dose of 115 mg (substituted for 125 mg EMEND) on Day 1 only of the chemotherapy induced nausea and vomiting (CINV) regimen.

Fosaprepitant (L-758,298) was rapidly converted to aprepitant following i.v. administration to experimental animals. After i.v. dosing of fosaprepitant to rhesus monkeys, a good correlation between plasma active drug (aprepitant) concentration and central receptor occupancy was observed. Aprepitant caused an inhibition of binding of [¹²⁵I]substance P to cloned human, rat, dog and guinea pig NK₁ receptors, and to NK₁ receptors from dog, monkey, mouse, rabbit and ferret cerebral cortex with IC₅₀ values in the nM range. Intravenously administered aprepitant (1.0 mg/kg) caused an inhibition of facilitation of the nociceptive spinal flexor reflex in a rabbit model. Oral or i.v. aprepitant caused dose-dependent increases in the latency of cisplatin-induced retching and vomiting and reduced the number of retches and vomits in ferrets. Aprepitant, at oral doses of 2 and 4 mg/kg, caused a complete inhibition of cisplatin-induced delayed retching and vomiting in ferrets. An additive anti-emetic effect was observed when the drug was administered with dexamethasone. At suboptimal doses, dexamethasone (20 mg/kg i.v.) or MK-0869 (0.1 mg/kg i.v.) was not effective in preventing cisplatin-induced emesis in ferrets, but the combination caused a significant inhibition of emesis in this animal model.

Pharmacokinetic/toxicokinetic studies were conducted with both fosaprepitant (L-758, 298; pro-drug) and the active drug (aprepitant; L-754, 030/MK-0869) following i.v. and oral administration to rats, mice, dogs and ferrets. Fosaprepitant (L-758,298) was rapidly converted to aprepitant in *in vitro* liver preparations from rat, dog and humans and by the S9 fractions from human lung, kidney and ileum, suggesting that the conversion of the pro-drug to active drug can take place in extrahepatic tissues. L-758,298 was rapidly (C_{max} was reached in 2 minutes) converted to its active metabolite, aprepitant following i.v. administration in rats and dogs, and in humans, it was converted to aprepitant within 30 minutes following the end of i.v. infusion. Following i.v. dosing of a 0.5 mg/kg dose to ferrets, the half-life, Cl and Vd_{ss} values were 10 hr, 1.5 ml/kg/min and 1.3 L/kg, respectively. The terminal half-life of intravenous aprepitant in rats was approximately 3 hours. In humans, the apparent terminal half-life of aprepitant ranged from approximately 9 to 13 hours. Aprepitant penetrated the blood brain barrier in rats and ferrets following i.v. and oral administration and it was excreted in the milk of lactating rats following

oral dosing. In human liver microsomes, CYP3A4 was the major CYP enzyme involved in the metabolism of MK-0869. Two other CYP enzymes, CYP1A2 and CYP2C19 were also involved in the metabolism of the compound. Fecal excretion was the major excretory pathway in rats. Fosaprepitant caused a moderate induction of cytochrome P₄₅₀ enzyme activity (7-ethoxy-4-trifluoromethylcoumarin-O-deethylase and fatty acyl-CoA oxidase activity) in the mouse liver. In rats, aprepitant caused an increase in the liver enzyme activity. The liver enzyme inducing capability of the drug may be responsible for the hepatocellular hypertrophy and thyroid follicular cell hyperplasia observed in rats during repeated administration of the drug.

Acute toxicity studies of fosaprepitant (L-758, 298) were conducted in mice and rats following i.v. administration of 200 and 500 mg/kg doses, and an oral dose of 500 mg/kg. The minimal lethal dose (MLD) by the i.v. route was 500 mg/kg in both rats and mice. There were no deaths of rats and mice at the 500 mg/kg oral dose. The clinical signs observed in mice after i.v. dosing included gasping, convulsions, bradypnea and loss of righting reflex, which disappeared within 3 hours. In rats, the clinical signs included gasping and bradypnea.

In the 17-day i.v. toxicity study with the lyophilized i.v. formulation of fosaprepitant in rats, the drug was administered at doses of 0, 2.5, 5.0 and 7.5 mg/kg/day. The high dose (7.5 mg/kg/day) group was terminated early on Day 9, because dosing was not feasible due to treatment related changes at the injection sites of the tail. Treatment group females had decreased body weight gains when compared with the controls. High dose males and females had slightly decreased erythrocytes, hemoglobin and hematocrit levels, and increased reticulocytes, neutrophil and monocyte counts. Changes at the injection sites were observed in all groups, and consisted of cellular proliferation of venous intima, venous necrosis or thrombosis, skin necrosis, subcutaneous edema, cellular infiltration or fibroplasia and degeneration of muscle fibers. The target organ of toxicity was the injection site, and the NOAEL was not established.

In a 16-day intravenous toxicity study, rats received MK-0869 (— drug particle size) at doses of 0, 80, 160 and 240 µg/kg/day. The no effect dose appeared to be 240 µg/kg/day. Solubility of MK-0869 in the vehicle limited the amount of drug that could be administered by the intravenous route. Therefore, the doses appeared to be inadequate to assess the toxicity of MK-0869 when administered by the i.v. route. Body weight gains of female rats at 160 and 240 µg/kg/day were impaired by >10%; However, there were no effects on corresponding male treatment groups. A target organ of toxicity was not identified. **b(4)**

In a 4-week i.v. toxicity study with L-758, 298 (fosaprepitant) in rats, groups of animals were administered the drug at doses of 0, 0.25, 1 and 4 mg/kg/day. Treatment of the animals with L-758, 298 at i.v. doses up to 4 mg/kg/day was not associated with any adverse effects on the animals. Thus, the 4 mg/kg/day dose was the no effect dose, and no target organ of toxicity was identified.

In a 5-week i.v. toxicity study with L-758, 298 in rats, the drug was administered at 0, 2, 5 and 10 mg/kg/day doses. Dose-related increases in liver weights were observed in males and females receiving the drug. Hepatocellular hypertrophy was observed in both males and females at 5 and 10 mg/kg/day doses. The target organ of toxicity was the liver, and the 2 mg/kg/day dose was the no effect dose.

In a 5-week oral toxicity study with two MK-0869 formulations (formulation M, ~~particle~~ particle size; formulation NB, ~~particle~~ particle size) in rats, 0 and 125 mg/kg b.i.d. doses of formulation M and 0, 5, 125, 250, 500 and 750 mg/kg b.i.d. doses of formulation NB were used. A no effect dose was not observed with either formulation M or NB. Histopathological changes were observed in the liver and the thyroid gland. Hypertrophy and diffuse vacuolation of hepatocytes, and diffuse follicular cell hyperplasia of the thyroid gland were observed in males and females of all treatment groups. Benign parafollicular cell adenoma was observed in two animals, one receiving 125 mg/kg b.i.d of formulation M and the other receiving formulation NB at 250 mg/kg b.i.d. The target organs of toxicity were the liver and the thyroid gland. Plasma drug concentrations in females were higher than that of males. A plateau of plasma drug level was observed in both males and females receiving b.i.d. doses of formulation NB.

b(4)

In a 14-week oral dose-ranging study with L-754, 030 in rats, groups of animals were administered 0, 5, 25, 125 and 250 mg/kg b.i.d. doses of the drug. Very slight to slight hepatocellular hypertrophy was observed in all male and female treatment groups. Very slight to slight diffuse hepatocellular vacuolation, with increased incidences in males, was observed in animals receiving the high dose. Very slight to slight diffuse follicular cell hyperplasia of the thyroid gland was observed in all treatment group males and females. Vacuolation of individual cells of pars distalis of the pituitary gland was observed in males receiving 125 and 250 mg/kg b.i.d. doses. The change was characterized by enlargement of individual pituitary cells due to formation of large cytoplasmic vacuoles and occasional protein droplets. The target organs of toxicity were the liver, thyroid and the pituitary gland, and the no effect dose was not established.

In a second 14-week oral dose-ranging study with L-754030 in rats, the drug was administered at doses of 0, 125, 250, 500 and 1000 mg/kg/day. Males and females receiving 500 and 1000 mg/kg/day doses had increased neutrophil counts. Increased liver and thyroid weights were observed in male and female animals receiving the drug. Centrilobular hypertrophy of the liver and thyroid follicular cell hyperplasia were observed in male and female rats at all doses. High dose males had an increased incidence of pancreatic acinar cell atrophy. The target organs of toxicity were the liver, thyroid gland and pancreas, and the NOAEL was not established.

In a 14-week oral toxicity study with L-754030 in rats, the drug was administered at doses of 0, 0.2, 1.0 and 5.0 mg/kg/day. This was not a complete toxicology study, because hematology, clinical chemistry and urinalysis were not performed and gross pathology, organ weight and histopathology examinations were limited to only the liver. Slight increases in liver weights were observed in female animals receiving the drug, and a slight centrilobular hypertrophy of the hepatocytes was observed in two (of 15) high dose (5 mg/kg/day) male rats.

In a 27-week oral toxicity study with MK-0869 (— particle size) in Sprague-Dawley rats, groups of animals received 125, 500 and 1000 mg/kg b.i.d. (250, 1000 and 2000 mg/kg/day) doses of the drug. Treatment-related changes in the hematology (increased platelet levels in males, and increased platelets, decreased hemoglobin and hematocrit values in females) and clinical chemistry (increased protein and cholesterol and decreased triglycerides in both sexes) parameters were observed in all treatment group animals. Hepatocellular hypertrophy and thyroid follicular cell hyperplasia were observed in males and females from all treatment groups. Thus, the target organs of toxicity were the liver and the thyroid gland, and the no effect dose was not established.

In the 53-week oral toxicity study with a 27-week interim necropsy in rats, L-754, 030 (— ug particle size) doses of 0, 0.25, 25 and 250 mg/kg/day were used. The target organs of toxicity were the liver and the thyroid gland following treatment for either 26 or 52 weeks. Slight centrilobular hypertrophy in the liver and slight follicular cell hyperplasia of the thyroid gland were observed in both males and females receiving 25 and 250 mg/kg/day doses for 26 or 52 weeks. The histopathological changes in the liver and the thyroid gland appeared to be associated with an induction of cytochrome P-450. The no effect dose was 0.25 mg/kg/day.

In a 17-day intravenous toxicity study with the lyophilized formulation of L-000758298 in dogs, the drug was administered at doses of 0, 2, 4 and 6 mg/kg/day. Treatment-related changes at the injection sites, consisting of swelling and/or cutaneous discoloration and the presence of hardening above and/or at the site of injection were observed at the mid and high doses. Venous thrombosis, fibroplasia and necrosis, as well as subcutaneous exudation and fibroplasia were observed in male and female animals receiving the mid and high doses. Thus, the injection site was the target organ of toxicity, and the 2 mg/kg/day dose was the no effect dose.

In a 4-week i.v. toxicity study with L-758, 298 in dogs, the drug was administered to groups of animals at doses of 0, 0.5, 2, 8 and 32 mg/kg/day. The 8 and 32 mg/kg/day doses produced slight to marked swelling and firmness of the forelimb, and a reddish discoloration of the skin. The 32 mg/kg/day produced venous thrombosis and vascularization. The no effect dose was 2 mg/kg/day, and no target organs of toxicity were identified.

In the 5-week intravenous toxicity study with L-758298 in dogs, the drug was administered at a dose of 8.0 mg/kg/day. Animals receiving L-758298 had decreased body weight throughout the dosing period. Injection site reactions consisting of perivascular cellular infiltration, fibrosis, hemorrhage and thrombosis, were observed in both control and treatment group animals, with higher incidences and more severity in the treatment group. Thus, the injection site was the target organ of toxicity. L-758298 was rapidly converted to its active metabolite (L-754030) following i.v. administration in dogs, and the maximum plasma concentrations of L-754030 were reached at approximately 2.0 minutes after administration.

In a 5-week oral dose range-finding toxicity study with MK-0869 (Formulation NB) in beagle dogs, groups of animals were administered 5, 25, 125, 250, 500 and 750 mg/kg b.i.d. (10, 50, 250, 500, 1000 and 1500 mg/kg/day) doses of formulation NB (µm particle size). A plateau of the plasma drug levels was achieved at the 500 mg/kg b.i.d. dose. The no effect dose was approximately 125 mg/kg b.i.d., and the target organs of toxicity were the testis, prostate and thymus. Testicular degeneration and prostatic atrophy were observed in male dogs at 125 mg/kg b.i.d. and higher doses. An increased incidence of thymic atrophy was observed in animals receiving ≥ 125 mg/kg b.i.d.; although a dose-response relationship was not evident. b(4)

In the 14-week oral toxicity study with L-754030 in dogs, the drug was administered at doses of 2, 8 and 32 mg/kg/day. No treatment related effects were observed in any groups at doses up to 32 mg/kg, suggesting that L-754030 was not examined at sufficiently high doses to show any toxic effects. No target organ of toxicity was identified in this study.

In a second 14-week oral toxicity study with MK-0869 in dogs, the drug was administered at 4, 32 and 128 mg/kg b.i.d. (8, 64 and 256 mg/kg/day) doses. No treatment related effects were observed in any group at doses up to 128 mg/kg b.i.d., suggesting that L-754030 was not examined at sufficiently high doses to show any toxic effects on the animals. No target organ of toxicity was identified.

In a 39-week oral toxicity study with MK-0869 in beagle dogs, groups of animals were administered 5, 25, 125 and 500 mg/kg b.i.d. (10, 50, 250 and 1000 mg/kg/day) doses of the drug. Suppression of body weight gains (15.4% to 69.2%) was observed at all doses. Increased plasma alkaline phosphatase and cholesterol levels were observed in all treatment group animals. Testicular degeneration and prostatic atrophy were observed in males receiving the 25 mg/kg b.i.d. (50 mg/kg/day) and higher doses. The target organs of toxicity were the testis and prostate, and the no effect dose was not established.

In a 53-week oral toxicity study of L-754,030 (µm drug particle size) in dogs, with a 27-week interim sacrifice, groups of animals were administered 0, 4, 16 and 32 mg/kg/day doses of the drug. No treatment-related toxic effects were observed in any group. The no effect dose was 32 mg/kg/day, and no target organs of toxicity were identified. b(4)

In a 17-day intravenous toxicity study with MK-0869 in rhesus monkeys, groups of animals were administered 0, 80, 160 and 240 µg/kg/day doses of the drug. Solubility of MK-0869 in the vehicle limited the amount of drug that could be administered by the i.v. route. Therefore, the doses appeared to be inadequate to assess the toxicity of MK-0869. The 240 µg/kg/day dose was the no effect dose, and no target organs of toxicity were identified.

In a 5-week intravenous toxicity of L-758, 298 in monkeys, groups of animals were administered 2, 5 and 10 mg/kg/day doses of the drug. No treatment-related toxic effects were observed in any group. The 10 mg/kg/day dose was the no effect dose, and no target organs of toxicity were identified.

The genotoxic potential for L-758, 298 (fosaprepitant) was examined in the bacterial reverse mutation assay (Ames assay), the rat hepatocyte DNA damage assay, the mutagenicity assay in TK6 human lymphoblastoid cells, the chromosomal aberrations assay in the Chinese hamster ovary cells, and the *in vivo* mouse bone marrow micronucleus assay. L-758, 298 was not genotoxic in any of the assays.

The genotoxic potential for L-754, 030 (aprepitant; MK-0869) was examined in the bacterial reverse mutation assay (Ames assay), the *in vivo* mouse bone marrow micronucleus assay and the mutagenesis assay in TK6 human lymphoblastoid cells. It had no genotoxic potential in any of these assays.

The sponsor conducted three 2-year oral carcinogenicity studies in Sprague-Dawley rats and two 2-year oral carcinogenicity studies in mice with aprepitant (MK-0869). In the rat carcinogenicity studies, animals were treated with oral doses ranging from 0.05 to 1000 mg/kg twice daily. The highest dose produced systemic exposure to aprepitant (plasma $AUC_{0-24 \text{ hr}}$) of 0.7 to 1.6 times the human exposure ($AUC_{0-24 \text{ hr}} = 19.6 \text{ mcg}\cdot\text{hr}/\text{ml}$) at the recommended dose of 125 mg/day. Treatment with aprepitant at a dose of 1000 mg/kg twice daily caused an increase in thyroid parafollicular cell carcinoma, and at doses of 5 to 1000 mg/kg twice daily it caused an increase in the incidences of thyroid follicular cell adenomas and carcinomas in male rats. In female rats, it produced hepatocellular adenomas at 5 to 1000 mg twice daily and hepatocellular carcinomas and thyroid follicular adenomas at 125 to 1000 mg/kg twice daily. In the mouse carcinogenicity studies, the animals were treated with oral doses ranging from 2.5 to 2000 mg/kg/day. The highest dose produced a systemic exposure of about 2.8 to 3.6 times the human exposure at the recommended dose. Treatment with aprepitant produced skin fibrosarcomas at 125 and 500 mg/kg/day doses in male mice.

Reproductive and developmental toxicology studies were conducted with both fosaprepitant (pro-drug; L-758,298) and aprepitant (active drug; L-754,030; MK-0869) by the intravenous and oral routes of administration. In the i.v. Segment I fertility and general reproductive performance study in male rats, fosaprepitant doses of 2, 5 and 10 mg/kg/day were used. It had no treatment-related effects on the fertility and reproductive performance of male rats at i.v. doses up to 10 mg/kg/day. In the oral Segment I fertility and reproductive performance study with MK-0869 in male rats, it had no effects on the fertility and general reproductive performance at doses up to 2000 mg/kg/day (1000 mg/kg b.i.d). In the i.v. Segment I fertility and general reproductive performance study with L-758,298 in female rats, it had no treatment-related effects at i.v. doses up to 4 mg/kg/day. In the oral Segment I fertility and general reproductive performance study with MK-0869 in female rats, it had no treatment-related effects at doses up to 2000 mg/kg/day (1000 mg/kg, b.i.d.).

In the i.v. Segment II teratogenicity study with L-758, 298 in rats, the drug was administered at doses of 1, 2 and 4 mg/kg/day from gestation day 6 through 20. L-758, 298 had no teratogenic

effects in rats at i.v. doses up to 4 mg/kg/day. In the i.v. Segment II teratogenicity study with L-758, 298 in rabbits, the drug was administered to pregnant animals at doses of 1, 2 and 4 mg/kg/day. It was not teratogenic in rabbits at i.v. doses up to 4 mg/kg/day.

In a modified i.v. Segment II/III reproductive toxicity study with L-758, 298 in female rats, the drug was administered to pregnant females at doses of 1, 2 and 4 mg/kg from gestation day 6 through lactation day 20. There were no treatment-related effects on pregnancies of F0 animals, and no effects of the peri- and post- natal development and reproductive performance of the F1 generation were observed. In an oral Segment II/III reproductive toxicity study with MK-0869 in rats, it had no effects on the peri- and post- natal development or reproductive performance of the F1 animals.

L-758, 298 caused a local irritation at the injection sites (discoloration, stiffness, sloughing of the skin, discharge) in rats when administered at i.v. doses up to 25 mg/kg/day for 5 days. Intravenous administration of single i.v. doses of 5.0 and 7.5 mg/kg doses to rats was associated with edema, hemorrhage and/or cellular infiltration in the subcutis at the injection sites of the tail. L-758, 298 caused hemolysis of washed red blood cells from rats, dogs and humans. However, it did not cause a hemolysis in human whole blood. MK-0869 caused an increase in TSH levels and thyroxine clearance in male and female rats. Increased TSH levels may be related to increased liver enzyme activity that caused increased metabolism of thyroid hormones. Fosaprepitant caused a moderate induction of cytochrome P₄₅₀ enzyme activity (7-ethoxy-4-trifluoromethylcoumarin-O-deethylase and fatty acyl-CoA oxidase activity) in the mouse liver. It also caused an increase in the liver enzyme activity (7-ethoxy-4-trifluorocoumarin-O-deethylase and fatty acyl-CoA-oxidase) in microsomes prepared from mouse liver. Aprepitant caused an increase in the drug metabolizing enzyme activities in rats. The liver enzyme inducing capability of the drug may be responsible for the hepatocellular hypertrophy and thyroid follicular cell hyperplasia observed in rats during repeated administration of the drug.

Conclusions:

Substance P, a tachykinin, is present in the sensory nerve terminals and is most likely involved in nociception. Substance P-containing vagal afferent fibers innervate the brainstem nucleus tactus solitarius, a region of the CNS involved in emesis. Aprepitant (also known as MK-0869 and L-754030) is a selective and potent NK₁/substance P receptor antagonist, and is approved for the prevention of acute and delayed nausea and vomiting due to highly and moderately emetogenic cancer chemotherapy. Fosaprepitant (also known as MK-0517 and L-758, 298) is the water soluble phosphoryl pro-drug of aprepitant, and is rapidly converted to aprepitant following intravenous administration. The pharmacological activity of fosaprepitant is derived from aprepitant. Under NDA 22-023, the sponsor is seeking approval of intravenous fosaprepitant for use in the prevention of acute and delayed nausea and vomiting associated with initial and repeat courses of highly and moderately emetogenic chemotherapy. In the proposed 3-day dosing regimen, fosaprepitant will be administered at an i.v. dose of 115 mg (substituted for 125 mg EMEND) on Day 1 only of the CINV regimen.

Acute, subacute/subchronic and chronic toxicity studies were conducted with fosaprepitant or aprepitant in different animal species. In i.v. acute toxicity studies of fosaprepitant (L-758, 298), the minimal lethal dose (MLD) was 500 mg/kg in both rats and mice. Seventeen-day i.v. toxicity studies with the lyophilized i.v. formulation of fosaprepitant were conducted in rats and dogs. In both rats and dogs, the injection site was the only target organ of toxicity, and the injection site changes included cellular proliferation of venous intima, venous necrosis or thrombosis, skin necrosis, subcutaneous edema, cellular infiltration and degeneration of muscle fibers. In repeat dose oral toxicity studies with aprepitant in rats, the target organs of toxicity were the liver (centrilobular hepatocellular hypertrophy), pituitary (vacuolation) and thyroid gland (follicular cell hyperplasia), and these effects may be related to activation of hepatic drug metabolizing enzymes. In a 5-week i.v. toxicity study with L-758, 298 in dogs, the no effect dose was 2 mg/kg/day, and no target organs of toxicity were identified. In a 53-week oral toxicity study with a 27-week interim sacrifice, no target organ of toxicity was identified. In monkeys, intravenous dosing of L-758, 298 for up to 240 mg/kg/day for 17 days, and up to 10 mg/kg/day for 5 weeks was not associated with any adverse effects, and no target organs of toxicity were identified. Thus, the preclinical toxicology studies with fosaprepitant indicate that it has very low toxicity profiles in rodents and non-rodents. It was not genotoxic in a battery of genotoxicity assays. Fosaprepitant or aprepitant had no effects on the fertility and general reproductive performance in rats, was not teratogenic in rats and rabbits, and had no effects on peri- and post- natal development in rats. Thus, the preclinical studies with fosaprepitant suggest that **the sponsor's proposed dose of the drug appears to be safe for use** in patients with chemotherapy induced nausea and vomiting at the proposed dose.

Thus, the sponsor conducted adequate preclinical studies with fosaprepitant/aprepitant to determine the safety of the drug, and the toxicity of fosaprepitant has been adequately characterized for the intended dose and route of administration.

Unresolved toxicology issues (if any): None

Recommendations:

The sponsor conducted adequate preclinical studies with fosaprepitant/aprepitant to determine the **safety of the drug, and the sponsor's proposed dose** appears to be safe for the proposed indication. Thus, from a preclinical standpoint, the NDA application is approvable.

Suggested labeling: See the labeling part of the review.

Sushanta Chakder, Ph. D.
Pharmacologist, HFD-180

Date

Comments:

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

Date

cc.

NDA

HFD-180

HFD-181/CSO

HFD-180/Dr. Chakder

HFD-180/Dr. Choudary

HFD-102/Dr Jacobs

HFD- 048/Dr. Viswanathan

R/D Init.: J. Choudary 12/22/06

APPENDIX/ATTACHMENTS:

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/s/

Sushanta Chakder
1/17/2007 11:38:28 AM
PHARMACOLOGIST

Jasti Choudary
1/19/2007 01:43:55 PM
PHARMACOLOGIST

Addendum To Pharmacologist's Review Dated January 17, 2007 of NDA 22-023

Sponsor and Address: Merck & Co. Inc., West Point, PA.

Reviewer: Sushanta Chakder, Ph. D.
Pharmacologist, HFD-180

Drug: Fosaprepitant (Emend I.V) Injection

Category: Neurokinin 1 (Substance P) Receptor Antagonist

The sponsor submitted an *in vitro* alkaline elution/rat hepatocyte assay study report under the NDA that was not reviewed earlier. The study report is reviewed.

Study Title: L-754,030: In vitro alkaline elution/rat hepatocyte assay

Study Report No. TT#95-8426 and TT#95-8433.

Conducting Laboratory: Merck Sharp & Dohme Research Laboratories, Merck & Co., Inc., West Point, PA.

Date of Study Initiation/completion: June 27, 1995/December 11, 1995

GLP Compliance: Yes

Drug Lot Number: L-754,030; Lot # L-754,030-000Z008; purity, 99.4%.

Study Endpoint: DNA strand break.

METHODOLOGY:

Strains/Species/Cell line: Hepatocytes from _____ \ BR Sprague Dawley rats.

b(4)

Dose Selection Criteria: Doses were selected on the basis of cytotoxicity and precipitation of the test substance in the culture medium. In the 3 hr solubility test in the culture medium (Leibovitz L-15), at 37°C, L-754,030 was found to be soluble at a concentration up to 45 µM. It produced significant cytotoxicity at 50 and 75 µM. Thus, L-754,030 was tested at concentrations of 10, 20, 30 and 40 µM in the final study.

Test Agent Stability Considerations: The sponsor stated that determination of the concentration of L-754,030 solution in DMSO showed that it was within acceptable limits.

Metabolic Activation System: N/A

Controls: Dimethylsulfoxide (DMSO) was used as a negative control. Aflatoxin B1 and 3 Gy gamma radiation were used as positive controls.

Exposure conditions: Primary cultures of rat hepatocytes (2.2×10^6 cells/plate) were treated with L-754,030 or the negative and positive controls for 3 hours. Duplicate samples were used for all groups and the positive control, and quadruplicate samples were used for the negative control. The cells were harvested by gentle scrapping and suspended in the original medium. Cytotoxicity was determined by trypan blue exclusion.

Analysis: For each drug concentration, one 5 mL aliquot of cell suspension (1×10^6 total cells) was taken from each of two separately dosed plates and each was loaded onto a 2.0 µ polycarbonate filter and lysed. A solution of tetrapropyl ammonium hydroxide, pH 12.1 was added to the DNA held on the filter and three fractions of 3 hours elution time each were collected. The DNA from each fraction was

trapped on non-fluorescent 0.2 μ polycarbonate filters and the amount of DNA on each filter was measured as the fluorescent product of 3,5-diaminobenzoic acid and deoxyribose.

Criteria for Positive Results: The results were considered positive if the compound produced and induced elution slope (treatment slope – negative control slope) of greater than or equal to 0.034, and was not associated with any significant cytotoxicity.

RESULTS

Study Validity: The study was valid, as the positive controls produced an induced slope of 0.034 or greater, and L-754,030 did not produce significant cytotoxicity at the concentrations used.

Study Outcome: L-754,030 did not show any significant cytotoxicity at concentrations of 10 to 40 μ M. Alkaline elution results show that L-754,030 did not produce an induced slope of 0.034 or greater at any concentrations tested. The positive control, Aflatoxin B1, at a final concentration of 1 μ M, produced an induced elution slope of 0.219 with viability of 103% compared to DMSO negative control, and a relative cellular ATP content of 91% immediately after treatment, and relative viability of 89% after 3-hour recovery assay, indicating that the assay was working as expected. The 3 Gy gamma radiation (positive control) produced an induced elution slope of 0.134 with a 94% relative cell viability. The data is summarized in the Table below.

Table 2. L-754,030: *In Vitro* Alkaline Elution/Rat Hepatocyte Assay-GLP.
Summary of DNA Strand Breaks and Cytotoxicity of L-754,030 in Rat Hepatocytes.
TT #95-8433

Treatment	Ppt. at Harvest	Elution Slopes	Mean Slope	Induced Elution Slope ¹	0-hour Trypan Blue Exclusion ²			3-hour Trypan Blue Exclusion ²			Cellular ATP Content (Percent)
					Absolute Viability (Percent)	Mean Absolute Viability (Percent)	Relative Viability (Percent)	Absolute Viability (Percent)	Mean Absolute Viability (Percent)	Relative Viability (Percent)	
Negative Control											
DMSO 1%	-	0.001, 0.002 0.004, 0.002	0.002	0.000	84, 82 79, 76	80	100	75, 70 77, 84	77	100	100
Positive Control											
(1 µM Aflatoxin B ₁)	-	0.236, 0.201	0.219	0.217	81, 83	82	103	62, 64	68	89 ³	91
(Gamma Radiation 3 Gy)	-	0.152, 0.119	0.136	0.134	74, 76	75	94	68, 67	68	88	87
L-754,030											
10 µM	-	0.003, 0.002	0.003	0.001	88, 88	88	110	79, 71	75	98	100
20 µM	-	0.003, 0.006	0.005	0.003	87, 76	82	103	75, 76	76	99	94
30 µM	-	0.005, 0.006	0.006	0.004	75, 69	72	90	78, 57	68	88	90
40 µM	-	0.003, 0.006	0.005	0.003	66, 64	65	81	59, 54	57	74	77

¹ Induced Elution Slope = Mean Treatment Slope - Mean Negative Control Slope.

This assay is considered positive if the induced slope is greater than or equal to 0.034 at a dose level with 70% or greater relative cell viability and 50% or greater relative ATP content at a soluble dose level.

² Viabilities determined immediately after treatment and after a 3-hour recovery period in fresh medium.

³ Blebbing.

SUMMARY: Based on these negative findings in the alkaline elution/rat hepatocyte assay, L-754,030 did not induce DNA strand breaks in isolated rat hepatocytes, and was not mutagenic under the conditions of the study.

CONCLUSION: L-754,030 was not mutagenic in the alkaline elution/rat hepatocyte assay under the conditions of the study.

Sushanta Chakder, Ph.D.
Pharmacologist, HFD-180

Date

cc:
NDA
HFD- 180
HFD- 181/CSO

HFD- 180/Dr. Chakder

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this page is the manifestation of the electronic signature.**

/s/

Sushanta Chakder
1/18/2008 04:20:42 PM
PHARMACOLOGIST