CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER: 21036

PHARMACOLOGY REVIEW(S)

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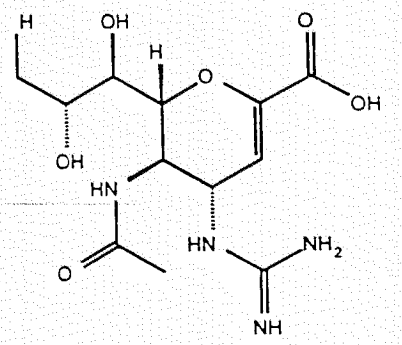
MAR 4 1999

PHARMACOLOGIST'S REVIEW



Zanamivir

NDA 21-036 Original NDA
Date Submitted: 10/19/98
Date Assigned: 10/21/98
Date Review Completed: 2/10/99
HFD-530

SPONSOR: GlaxoWellcome Inc.
5 Moore Drive, Research Triangle Park, NC

DRUG: Zanamivir, GG167; GR121167; 5-(Acetylamino)-4-[(aminoiminomethyl)amino]-2,6-anhydro-3,4,5-trideoxy-D-glycero-D-galacto-non-2-enonic acid; $C_{12}H_{20}N_4O_7$; MW: 332.3

FORMULATION: Oral Inhalation (solubility: 18 mg Zanamivir off-white powder/ml water); pK_a1 (guanidine group)=13; pK_a2 (carboxyl group)=2.4; administered to the respiratory tract by ROTADISK® which contains four regularly spaced double-foil blisters with each blister containing a powder mixture of 5 mg of Zanamivir and 20 mg of lactose. The contents of each blister are inhaled through the mouthpiece of DISKHALER, a breath-activated plastic device.

INDICATION: Anti-Influenza

INTRODUCTION

Zanamivir is a competitive inhibitor of influenza A and B virus neuraminidase (NA). It inhibits the growth of influenza A and influenza B viruses *in vitro*, and is active against animal models of influenza virus infection. This NDA is originated from IND [redacted] The preclinical drug development program of this NDA consisted of more than 50 in vitro and in vivo animal toxicity studies conducted in rodents, dogs, and rabbits in support of the intended human use. All toxicity studies have been submitted and reviewed under IND [redacted] and its amendments). This document summarizes and comments on the overall preclinical safety information and the risk assessment of zanamivir based on non-clinical toxicology studies.

PHARMACOLOGY OF ZANAMIVIR

IC_{50} AND K_i. Zanamivir is an analogue of neuraminidase (NA) and is a product of rational drug design based on the x-ray determination of the crystal structure of viral lipid envelope hemagglutinin and NA. It is a slow-binding and high affinity competitive inhibitor for the influenza virus NA. K_i values determined for the three strains of influenza virus were 0.46-1.0x10^{-10} M (A/Brazil/11/78), 0.38-2.3x10^{-10} M (reassortant X31) and 5.0-10x10^{-10} M (B/Hong Kong/3/91). IC_{50} values for the inhibition of NA by zanamivir from a range of influenza A and B viruses lay in the range of 0.64 nM to 7.9 nM.

ANIMAL MODELS. In animal models, zanamivir inhibited influenza A and B virus replication in mouse and ferret models of infection when administered intranasally or by aerosol, but was substantially less active when administered intraperitoneally. In addition to inhibiting viral replication in vivo, the compound reduced the pyrexia in infected ferrets. In all in vitro and in vivo tests, zanamivir showed inhibition of influenza viruses that are resistant to amantadine or rimantadine.

NONSPECIFICITY AND CYTOTOXICITY. Zanamivir has low inhibitory activity against mammalian cell neuraminidase (e.g., IC_{50}=0.93-1.0 mM for human lysosomal sialidase isolated from placenta), and has lower cytotoxic potential when compared with other NA inhibitors (rimantadine, amantadine and ribavirin), as tested in the following in vitro cytotoxicity assays: MRC-5 (a fibroblast cell line established from normal human lung tissue of a 14-week old fetus); PANC-1 (a human epithelial cell line derived from male pancreatic carcinoma); 161-BR (a human skin fibroblast cell line from normal male skin); and MDCK (a canine epithelial cell line).

RESISTANCE. Zanamivir is a weak inhibitor of the NA of influenza viruses that are resistant to zanamivir in vitro and selected by passage of viruses in the presence of the compound. Characterization of these viruses has shown that viral resistance to zanamivir in vitro can result from amino acid sequence changes either to the viral hemagglutinin, or to the viral neuraminidase, or both. The clinical importance of the resistant genotypes which have been selected from in vitro experiments has not been established.

SUMMARY OF PRECLINICAL SAFETY INFORMATION: ZANAMIVIR

Animal toxicity studies conducted with zanamivir include

- Single-dose toxicity studies in the B6C3F1 strain of mouse and the Charles River Wistar strain of rat. (oral, iv in all and inhalation in the rat only)
- Repeat-dose toxicity studies in Wistar rat and beagle dog (inhaled, iv and intranasal; up to 26 weeks duration in the rat, and 52 weeks duration in the beagle dog)
- Pre-carcinogenicity range-finding studies in rats and mice
- Carcinogenicity studies in the B6C3F1 strain of mouse and the Charles River Wistar strain of rat.
- Reproductive toxicity studies in the Sprague Dawley rat (Crl:CD[SD]BR VAF/Plus) and New Zealand White (NZW) rabbit.
- *In vitro* and *in vivo* mutagenicity assays
- Special toxicity studies on irritancy, antigenicity, and sensitization.

For a detailed evaluation on the animal toxicology studies, please refer to IND [redacted] Key preclinical safety information is recapitulated and issues discussed below.

(1) TARGET ORGAN/SYSTEM AND PROFILE OF TOXICITY OF ZANAMIVIR

EXPLORATION OF TOXICITY PROFILE: ***Key Issues.***

Toxicologically, the full toxicity profile and true target organ/system of toxicity of zanamivir have been difficult to identify because of the short half-life and comparably low clinical exposure of the drug. In addition to the inhalation route of administration used in the animal toxicity testings, the sponsor has employed iv bolus and iv infusion to increase drug exposure to explore the toxicity profile. However, the attempts had not been fruitful since none of the chronic, repeat-dose toxicity studies showed a steady-state exposure to zanamivir (i.e., Cmin, trough drug levels or pre-dose drug concentrations were often non-detectable). This is the case even for the iv infusion studies because of the limitation of methodology (duration of the iv infusion in the dog was limited and rather short.) With the issue of drug exposure in mind, systemic and local toxicity findings and key target organ/system of toxicity are highlighted below.

KIDNEY: ***Renal Tubule Necrosis in Rats.***

Continuous iv infusion of zanamivir in rats at dosages of 864 and 1728 mg/kg/day caused a dose-related, vacuolation of the proximal convoluted tubules in the renal cortex. There was no vacuolation in the renal cortex following the 7-day recovery period. The no-effect level was 432 mg/kg/day. In a higher iv (bolus) study in rats (912 and 13,824 mg/kg/day), similar renal toxicity findings were also reported (cortical tubular vacuolation/glomerular sclerosis with eosinophilic material or adhesions in the Bowman's space). This renal toxicity was not reported in any inhalational studies or iv bolus studies. At the no-effect dose of 432 mg/kg, the systemic exposure was 1000 times higher than proposed for the clinical use of zanamivir.

There has been no parallel evidence of zanamivir-related nephrotoxicity reported humans at lower doses studied.

RESPIRATORY SYSTEM: ***Epithelial Hyperplasia and Loss of Ciliated Cells in the Trachea.***

An increase in incidence and degree of epithelial hyperplasia at the carina (with or without loss of cilia) was seen in all zanamivir-treated groups in the 26-week dog inhalation study. In the 52-week dog inhalation study, there was an increase in loss of cilia at the carina in females in the intermediate and high dose groups. The incidence of this lesion in the 52-week dog inhalation study is shown in the Table below. An increased incidence of loss of ciliated cells at the carina was also recorded in rats following 104 weeks administration. There were statistically significant trends of hyperplasia (data row 2 [minimal]; p=0.0186) and loss of cilia hyperplasia (data row 3; p=0.0004) in the trachea and bifurcation of the female dogs. Following a 2-week withdrawal period, the incidence of this lesion in the treated group was not greater than the control groups (i.e., the toxicities were reversible). The sponsor indicated that the toxicities were not accompanied by any evidence of inflammation.

Effects of Lactose. The histopathologic changes produced by lactose vehicle alone are considered adaptive phenomena due to a prolonged inhalation exposure to high aerosol concentration and the high lung burden. The lactose-induced phenomena have been observed in many other inhalational drug testings.

Table 1.
Overall Incidence of Histopathological Changes in the Respiratory Epithelium (52-Week Dog Inhalational Study)

Histopathology Findings	Air (M)	Air (F)	Veh (M)	Veh (F)	Low dose (M)	Low dose (F)	Mid dose (M)	Mid dose (F)	High dose (M)	High dose (F)
Trachea and bifurcation, epithelial hyperplasia at carnia:										
Trace -	1/4	2/4	1/4	2/4	3/4	0/4	2/4	1/4	1/4	1/4
minimal -	1/4	0/4	0/4	1/4	0/4	0/4	0/4	2/4	1/4	3/4
Trachea and bifurcation, loss of cilia at carnia	1/4	0/4	1/4	0/4	1/4	0/4	1/4	3/4	2/4	4/4
Following Recovery										
Trachea and bifurcation, epithelial hyperplasia at carnia:										
Trace -		1/2		1/2						1/2
minimal -		1/2		1/2						0/2
Trachea and bifurcation, loss of cilia at carnia		2/2		2/2						0/2

M: Male; F: Female; Low, Mid and High Dose: 0.93, 4.31 or 11.2 mg/kg/day

Table 2.
Overall Incidence of Focal Loss of Ciliated Cells In the Respiratory and Olfactory Epithelium (Rat Carcinogenicity Study)

	Male					Female				
Group	1	2	3	4	5	1	2	3	4	5
Dosage level	0	Lac.	Low	Middle	High	0	Lac.	Low	Middle	High
Focal loss of ciliated cells (bifurcation)	4	10	16**	18**	14**	9	14	26** #.	14	18*
Number carina examined	55	55	55	55	55	55	55	55	55	55

*Fisher's Exact Test; Air Control (*) or Vehicle Control (#) compared with all other Groups ** p<0.01 one-sided; # or * p<0.05 one-sided*

Low, Middle and High Doses: 7.6, 15.1 and 30.2 mg/kg/day (Weeks 1-17); 14.2, 27.4 and 53.1 mg/kg/day (Weeks 17-104).

RESPIRATORY SYSTEM: *Increased Number in Enlarged and Foamy Alveolar Macrophages in the Lung.*

Increased numbers of enlarged, diffusely distributed (prominent) alveolar macrophages were seen in the alveoli of a small number of rats exposed chronically to zanamivir. In the 26 weeks study this was present at the higher dose of 44.5mg/kg/day. This finding was also observed in the 104-week rat carcinogenicity study. The incidence of this finding in both these studies is summarized in the Table below.

Increased numbers and size of alveolar macrophages are a reflection of the clearance of particulate matter from the lung. The clinical relevance of this finding is uncertain, however, the sponsor estimated the concentration of particulate matter in the lung (high dose) of the rat to be approximately 34mg/m^2, whereas the inhaled dose in man is approximately 0.22mg/m^2; with a 150 fold difference. At the present stage of the intended indication for zanamivir (short-term administration), there is little concern over this toxicity upon risk/assessment of this NDA.

Table 3.
Overall Incidence of Enlarged or Foamy Alveolar Macrophages
(Rat Carcinogenicity Study)

	Male					Female				
Group	1	2	3	4	5	1	2	3	4	5
Dosage level	0	Lac.	Low	Middle	High	0	Lac.	Low	Middle	High
Prominent numbers of alveolar macrophages	3	1	2	8*	8*	1	1	1	2	5
Foamy alveolar macrophages										
Total	16	8	5	8	20**	12	8	10	12	13
Trace	8	4	2	4	3	8	6	7	6	5
Minimal	8	3	3	4	13**	3	2	3	6	5
Moderate	0	0	0	0	4	1	0	0	0	3
Severe	0	1	0	0	0	0	0	0	0	0
Subpleural aggregations of foamy alveolar macrophages										
Total	23	22	22	16	25	19	27	22	19	26
Trace	14	16	12	9	6	8	16	8	8	6
Minimal	9	5	10	7	14*	11	10	13	11	15
Moderate	0	1	0	0	5#	0	1	1	0	4
Marked	0	0	0	0	0	0	0	0	0	1
Number lungs examined	55	55	55	55	55	55	55	55	55	55

*Fisher's Exact Test; Air Control (#) or Vehicle Control (*) compared with all other GG167 treated Groups ** $p<0.01$; # or * $p<0.05$ one-sided. Low, Middle and High Doses: 7.6, 15.1 and 30.2 mg/kg/day (Weeks 1-17); 14.2, 27.4 and 53.1 mg/kg/day (Weeks 17-104).*

RESPIRATORY SYSTEM: *Nasal Passage.*

Increased incidences of eosinophilic inclusions were noted in nasal and respiratory epithelium in both rat and mouse carcinogenicity (inhalational) studies. The sponsor indicated that increases in incidence and severity of these droplets has been described for a number of different and unrelated compounds administered by the

intranasal route to rodents. The eosinophilic material is contained within endoplasmic reticula (by electron microscopy) and again, was considered to be a non-specific defense response.

The increased incidence of goblet cell hyperplasia seen in the rat carcinogenicity study was statistically significant. The toxicity was dose-related in incidence and severity, and a no-effect level was not determined (see Table below). The change was not accompanied by any degeneration or inflammatory changes. The sponsor indicated that goblet cells produce mucus and its proliferation are adaptive response to high concentrations of particulate matter.

Table 4.
Overall Incidence of Eosinophilic Inclusions in the Respiratory and Olfactory Epithelium (Rat Carcinogenicity Study)

STATISTICAL COMPARISON WITH THE AIR CONTROL (GROUP 1)										
	Male					Female				
Group	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
Dosage level	**0**	**Lac.**	**Low**	**Mid**	**High**	**0**	**Lac.**	**Low**	**Mid**	**High**
Goblet cell hyperplasia										
Total	1	0	4	4	12**	0	3	6*	9**	8**
Trace	0	0	4	4	5*	0	3	6*	9**	8**
Minimal	1	0	0	0	7*	0	0	0	0	0
Eosinophilic inclusions-respiratory epithelium										
Total	15	29**	51**	54**	52**	11	38**	54**	50**	52**
Trace	13	29**	26**	19	9	10	36**	17	12	17
Minimal	2	0	24**	32**	37**	1	2	37**	38**	34**
Moderate	0	0	1	3	6*	0	0	0	0	1
Eosinophilic inclusions-olfactory epithelium										
Total	15	17	52**	55**	52**	7	20**	51**	50**	51**
Trace	7	10	11	5	2	4	17**	8	7	7
Minimal	4	6	21**	13*	8	1	2	20**	16**	19**
Moderate	4	1	18**	35**	30**	2	1	18**	18**	24**
Marked	0	0	2	2	12**	0	0	5*	9**	1

STATISTICAL COMPARISON WITH THE VEHICLE CONTROL (GROUP 2)										
	Male					Female				
Group	1	2	3	4	5	1	2	3	4	5
Dosage level	0	Lac.	Low	Mid	High	0	Lac.	Low	Mid	High
Goblet cell hyperplasia										
Total	1	0	4	4	12**	0	3	6	9	8
Trace	0	0	4	4	5*	0	3	6	9	8
Minimal	1	0	0	0	7**	0	0	0	0	0
Eosinophilic inclusions-respiratory epithelium										
Total	15	29	51**	54**	52**	11	38	54**	50**	52**
Trace	13	29	26	19	9	10	36	17	12	17
Minimal	2	0	24**	32**	37**	1	2	37**	38**	34**
Moderate	0	0	1	3	6*	0	0	0	0	1
Eosinophilic inclusions-olfactory epithelium										
Total	15	17	52**	55**	52**	7	20	51**	50**	51**
Trace	7	10	11	5	2	4	17	8	7	7
Minimal	4	6	21**	13	8	1	2	20**	16**	19**
Moderate	4	1	18**	35**	30**	2	1	18**	18**	24**
Marked	0	0	2	2	12**	0	0	5*	9**	1
Number nasal passages examined	55	55	55	55	55	55	55	55	55	55

*Fisher's Exact Test; * $p<0.05$ ** $p<0.01$ one-sided. Low, Middle and High Doses: 7.6, 15.1 and 30.2 mg/kg/day (Weeks 1-17); 14.2, 27.4 and 53.1 mg/kg/day (Weeks 17-104).*

OVERALL RESPIRATORY TRACT PATHOLOGY: ***NOAEL and Margin of Safety.***

The NOAEL dose for histopathology findings in the respiratory tract (e.g., epithelial hyperplasia and loss of cilia) in the dog may be set at the low dose (0.93 mg/kg/day), which had a daily drug exposure of 1.28-2.22 ug.h/ml. In the rat, the NOAEL for these effects could be set approximately at the low dose (14.2 mg/kg/day) that had exposure levels around 4.23 - 4.98 ug.h/ml. In comparison with human daily exposure (0.4 ug.h/ml), a margin of safety of 3.2-5.6 (dog vs. human) or 10.6-12.5 (rat vs. human) existed for these effects.

Clinical significance of the information on non-neoplastic hyperplasia and other cellular changes in the respiratory tract may become important, and of further concern when the sponsor is ready to seek approval on the prophylactic use of this drug for influenza infections. Under the current indication for a short-term treatment of influenza, it may carry less impact on risk/assessment of this NDA.

REPRODUCTIVE SYSTEM: ***Fertility and General Reproductive Performance,***

The fertility and general reproductive performance were investigated in male (dosed for 10 weeks prior to mating, and throughout mating, gestation/lactation, and shortly after weaning) and female Sprague-Dawley rats (dosed for 3 weeks prior to mating through to day 19 of pregnancy, or day 21 post-partum). Embryo/fetal development studies were conducted in rats (dosed from days 6 to 15 of pregnancy) and New Zealand White rabbits (dosed from days 7 to 19 of pregnancy). Pre- and post-natal developmental studies were performed in rats (dosed from day 16 of pregnancy till

Embryo/Fetal, and Pre-/ Postnatal Development.

litter day 21 to 23). In all studies, iv instead of inhalational route of administration, and a maximally soluble iv dosage of 90 mg/kg/day were used.

The results showed that zanamivir was not teratogenic, had no significant effect on fertility, reproductive performance, parturition or development of the F_1 generation, except that (1) A slight reduction in mean performance times in the accelerating rotarod test was observed in F_1 males at 90mg/kg/day (week 4), and at 9 and 90mg/kg/day (week 6) in the fertility study and (2) a slight reduction in forelimb (but not hindlimb) grip strength and a slight reduction in the level of arousal at 90mg/kg/day were observed. These findings are considered to be little relevance to the clinical use of zanamivir.

The maximum drug concentration measured in these reprotoxicity studies ranged from 201-281 ug/ml in rats and 382-637 in rabbits. Because of insufficient blood sampling time points in both rat and rabbit reprotoxicity studies, AUC values were not available. However, a subchronic study in Wistar rat using the 90 mg/kg/day iv dose produced very high AUC values ranged from 8510-11940 ug.min/ml (or 141.8-199 ug.hr/ml).

TOXICOLOGY OF IMPURITIES

The impurity profile for batches of zanamivir used in the toxicity studies demonstrates that animals received total doses of these impurities far in excess of clinical exposure.

TOXICO-KINETICS

As stated in the beginning paragraph, because of zanavimir's short half-life and its straightforward urinary clearance, the drug exposure as measured by the toxicokinetics in all the repeat-dose toxicity studies (including the reprotoxicity studies and those using iv infusion techniques) did not show any accumulation of the drug, even though the area underneath the concentration curve yield significantiy higher figures than human's. The drug accumulation as reflected by the successful maintenance of a significant trough level over the entire study period is important for eliciting meaningful toxicities, and is often achieved in other drug studies in which the toxicity profile and target organ of toxicity have been fully explored.

[redacted]

DNA AND CHROMOSOME SYSTEMS

Zanamivir tested negative in the following genotoxic testing systems: AMES test, fluctuation test, yeast gene conversion assay, mouse lymphoma assay, in vitro chromosome aberration assay in human peripheral lymphocytes, and a micronucleus assay in mouse bone marrow.

CARCINO-GENICITY

Carcinogenicity studies of lifetime duration (104 weeks) were performed by the inhaled route in the B6C3F1 mouse (male: 26.6, 47.8 and 102 mg/kg/day; female: 28.0, 50.9 or 108 mg/kg/day) and Han Wistar rat (7.6, 15.1 and 30.2 mg/kg/day, Weeks 1-17; 14.2, 27.4 and 53.1 mg/kg/day, Weeks 17-104). An increase in lymphoblastic/lymphocytic lymphomas was observed in male rats exposed to 53.1mg/kg/day. The lymphomas were found wide-spread in various lymph nodes (e.g., cervical, tracheobronchial, mesenteric, axillary), and organs (e.g., lungs, liver, spleen). The distribution pattern suggests a highly metastatic nature of this tumor. The increase in lymphoma incidence was statistically significant when comparison was made with lactose controls ($p<0.017$), instead of with air controls ($p<0.084$). According to the FDA guidance document, for a common tumor (incidence$\cong$1%) such as this one, the outcome was not considered to be significant and thus zanamivir is not considered carcinogenic.

However, because there is a tendency to an increased incidence in lymphomas, zanamivir might be immunosuppressive. The sponsor should perform an immunotoxicity study to determine whether the drug has any immunosuppressive toxicity.

ADME:

General PK Parameters.

Following iv administration to the rat, plasma clearance of zanamivir is rapid, showing a monophasic elimination with a half-life of approximately 15 minutes. In the dog, the half-life of zanamivir after intravenous administration is approximately 50 minutes. In both species, almost all of the drug is eliminated unchanged in the urine ($\cong$95%) and therefore, renal clearance accounts for almost the total clearance of zanamivir. The renal clearance in the rat and dog is consistent with the fact that the drug has low protein binding. Low volumes of distribution in the rat and dog indicate that zanamivir distributes poorly and is unlikely to penetrate cell membranes to a significant extent. Human pharmacokinetics shared similar clearance and distribution profiles with the rat and dog.

Gender Difference in Exposure and Bioavailability.

No difference in the pharmacokinetics of zanamivir between male and female rat or dog following a single dose was seen (following repeat iv dose in the dog, exposure appeared to be higher in females than males at all dosages, but there were no gender-related differences in toxicity.) Following oral administration, zanamivir is poorly absorbed with a bioavailability of 3% in the rat and 10% in the dog (human=3%). Data from studies with radiolabelled zanamivir (iv) in the rat and dog show that plasma drug levels account for all of the radioactivity in the plasma, indicating that zanamivir does not undergo metabolism.

Distribution.

Radiolabelled zanamivir is widely distributed throughout the tissues with levels in the blood, kidney and bladder being the highest. Radioactive material is cleared rapidly from most tissues, although very low levels persist in the gastrointestinal tract contents. Low levels also appeared to persist in the eyes of pigmented animals.

Chromatographic profiling of urine samples indicated that drug-related material consists entirely of unchanged zanamivir, with no evidence of any metabolites.

Placenta Transfer. In pregnant rabbits, ^{14}C-zanamivir and drug-related material crossed the placental barrier and widely distributed throughout fetal tissues. Drug-related levels in the fetus were higher on day 12 of pregnancy that on day 20, indicating that the placental barrier is more permeable to drug-related material on day 12 than on day 20. The percentage of the administered dose recovered in the fetus was small, ranging from 0.0006-0.0032%.

Excretion in Milk. Following iv ^{14}C- zanamivir (10mg/kg) to lactating rats, limited amounts of drug-related material partition into milk (Cmax in milk= 1ug equiv/mL at 0.5 hours post-dose, Cmax in maternal plasma= 10.1/equiv/ml).

Plasma Protein Binding and Metabolism. Zanamivir has low plasma proteins binding in rats, dogs and human. Plasma protein binding and the association with red blood cells in these species are also negligible. Zanamivir has no effect on metabolic pathways mediated by isozymes CYP1A1, CYP1A2, CYP2A6, CYP2C8/9/10, CYP2C18/19, CYP2D6, CYP2E1 or CYP3A4, and no significant changes in the levels of hepatic cytochrome P-450 isozymes at the end of a 5-week intravenous toxicity study in the rat were reported.

RISK ASSESSMENT OF ZANAMIVIR BASED ON NON-CLINICAL TOXICOLOGY STUDIES

- ADEQUACY OF THE SUBMISSIONS AND LIMITATIONS OF PRECLINICAL TOXICITY STUDIES CONDUCTED ON ZANAMIVIR. The series of toxicity studies reports submitted under IND [redacted] in support of this NDA included (1) 7 acute toxicity studies in rats and mice, (2) 23 repeat-dose toxicity studies in rats and dogs and mice (including carcinogenicity studies), (3) 7 reproductive toxicity studies in rats and rabbits, (4) 5 *In vitro* and *in vivo* mutagenicity assays, and other irritation toxicity studies and ADME studies. Most of the studies, especially the primary ones, were conducted in England under GLP. The scope of topics covered by the animal toxicity investigation is considered sufficient to support the NDA.

- LIMITATIONS OF PRECLINICAL TOXICITY STUDIES CONDUCTED ON ZANAMIVIR. The key studies on zanamivir were limited by feasibility factors.

  - Intravenous bolus injection studies in the dog were limited by a maximum dose of 90 mg/kg because of the solubility (18 mg/ml) and maximum volume that the sponsor believed to be reasonable for administration. The duration and total dose of an iv infusion study in the dog was also limited by the maximum amount of fluid deliverable to animals as claimed by the sponsor (at the infusing rate of 3 ml/min and 5 ml/kg for a maximum daily dose of 90 mg/kg/day). The feasibility factor on the

infusion rate and volume in the dog had been debated by the Agency, because much higher volume of fluid over a longer infusion period had been administered by other laboratories. The sponsor's could have escalated the drug exposure in dogs to the extent that was achieved by the same type of iv infusion study conducted in the rat (432, 864 and 1728 mg/kg/day). More in-depth toxicology information in dogs than concluding that the drug has a minimal toxicity profile and no target organ of toxicity in this species might have been possible.

- The sponsor indicated that the inhalational carcinogenicity studies were maximally feasible in their daily exposure duration. The agency (both the reviewing pharmacologist and the Exec CAC) felt that longer terms of exposure had been regularly conducted in other laboratories and could have been administered without jeopardizing the studies. Higher exposure levels clearly might yield a more in-depth tumorigenicity profile in both the rat (e.g., lymphomas) and mouse.

- SYSTEMIC TOXICITY PROFILE. Based on the limited data obtained from the rat, zanamivir's target organ of systemic toxicity in this species is identified as the kidney, at which a dose-related, vacuolation of the proximal convoluted tubules in the renal cortex was elicited. This toxicity occurred at very high exposure levels and a sufficient margin of safety existed when compared with human clinical exposure.

- LOCAL TOXICITY PROFILE. An increase in incidence of epithelial hyperplasia, enlarged, diffusely distributed alveolar macrophages, loss of ciliated cells, and presence of eosinophilic inclusions was noted in respiratory linings following long-term inhalation of zanamivir. Although it may be considered an adaptive response of the respiratory system to the inhaling particulate matter, the findings in both rats and dogs were statistically significant and the effects were treatment related. The margin of safety ranged from 3.2-12.5. Because these toxicities emerged after long-term (≥26 weeks) inhalation of the drug, they may become more clinically relevant when the drug is indicated for prophylactic uses.

- TUMORIGENICITY AND TERATOGENICITY. The drug is neither carcinogenic nor teratogenic (*Pregnancy Category B)* at the doses tested in the animals. The borderline increase (non-significant) in the incidence in lymphoblastic/lymphocytic lymphomas in the rats had raised concerns about the immunosuppressive potential of this drug.

CONCLUSION

This NDA in its present form has provided adequate preclinical safety information in support of the approval of zanamivir for use in the treatment of influenza infection. The sponsor has employed reasonable levels of dosage and number of animals of both sexes in their toxicity

studies. The sponsor has explored the clinically relevant toxicity of the drug and provided assurance that adequate margin of safety existed in using the drug in reference to the indication proposed. It is thus concluded that the NDA has provided sufficient preclinical safety information to allow for prediction of potential toxicity in humans with the proposed indication in humans. The following regulatory requests should be sent to the sponsor for additional studies as a phase IV commitment:

REGULATORY REQUESTS:

[redacted]

/S/

Kuei-Meng Wu, Ph.D.
Reviewing Pharmacologist
DAVDP

Concurrences:
HFD-530/DepDir/WDempsey /S/ 3/7/99
DAVDP/HFD-530/PTL/JFarrel /S/ 3/4/99
Wu/Pharm/2/10/99 /S/ 4/99

Disk: JFarrelly

APPEARS THIS WAY
ON ORIGINAL

HFD-530 NDA 21-036 (000)
HFD-530/Division File
HFN-340
HFD-530/CSO
HFD-530/MO
HFD-530/Chem
HFD-530/Micro
HFD-530/Pharm

APPENDIX

DRAFT

LABELING

DRAFT

LABELING

APPEARS THIS WAY
ON ORIGINAL

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