CENTER FOR DRUG EVALUATION AND RESEARCH

APPROVAL PACKAGE FOR:

APPLICATION NUMBER

50-794

Clinical Pharmacology and Biopharmaceutics Review

CLINICAL PHARMACOLOGY/BIOPHARMACEUTICS REVIEW

NDA:	50-794
GENERIC NAME:	Azacitidine
BRAND NAME:	Vidaza™
DOSAGE FORM/ STRENGTH	100 mg of Azacitidine and 100 mg of Mannitol in Vials for Subcutaneous Injection
INDICATION:	Myelodysplastic Syndrome (MDS)
SUBMISSION TYPE:	NDA-NME
SUBMISSION DATE:	26-Dec-2003
APPLICANT:	Pharmion Corporation, Boulder, CO
OND DIVISION:	Division of Oncology Drug Products (HFD-150)
OCPB DIVISION:	Division of Pharmaceutical Evaluation I (HFD-860)
OCPB REVIEWER:	Sophia Abraham, Ph.D.
OCPB TEAM LEADER:	Atiqur Rahman, Ph.D.

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I. EXECUTIVE SUMMARY

The Applicant submitted NDA 50-974 to seek approval of Vidaza™ (Azacitidine Injectable Suspension) in the treatment of patients with myelodysplastic syndrome (MDS).

In support of this NDA, three clinical studies were submitted: one pivotal Phase 3 (Study 9221) and two supportive Phase 2 studies (Studies 8921 and 8421). In Studies 9221 and 8921, patients received 75 mg/m^2 azacitidine subcutaneously (SC) daily for 7 days on a 28-day cycle for a minimum of 4 cycles. In Study 8421, patients received the same dosing schedule with azacitidine given intravenously (IV). The selection of this dose and dosing schedule was based on a 30-year clinical experience with azacitidine with the intravenous administration. No pharmacokinetic/pharmacodynamics studies were conducted with the SC formulation to optimize azacitidine dose. The Applicant has not made any attempt to explore the relationships between degree of DNA hypomethylation and response rate and/or adverse events or between drug exposure and response rate and/or adverse events.

The human pharmacokinetics and bioavailability section of NDA 50-794 consists of a pharmacokinetic study (AZA-2002-BA-002) with single subcutaneous dose of 75 mg/m^2 and a single intravenous dose of 75 mg/m^2 of azacitidine given to six MDS patients.

The Applicant did not conduct a mass-balance study in cancer patients. However, published studies indicate that azacitidine and its metabolites are primary excreted by the kidneys and the label cautions against the use of Vidaza™ in renally and hepatically impaired patients.

The Applicant did not evaluate the effect of intrinsic factors such as age, gender, race, renal impairment or hepatic impairment on the pharmacokinetics of azacitidine. Based on the sub-group analyses of safety data, the Applicant included some specific dosing recommendations in the package insert regarding the use of Vidaza™ in elderly patients, in patients with renal impairment, and in patients with liver metastases or severe hepatic impairment. These recommendations are acceptable at this time from the clinical pharmacology perspectives. However, because azacitdine causes renal toxicity and because azacitidine and its metabolites are primary eliminated by the kidneys, we recommend that the Applicant should conduct a study in patients with varying degrees of renal impairment to properly provide dosing recommendations in this patient population (see Phase 4 Commitment). A [redacted] statement was added in the label for the use of Vidaza™ in patients with severe renal impairment (estimated creatinine clearance < 30 ml/min) and in patients with underlying hepatic cirrhosis.

The Applicant did not conduct any *in vivo* drug-drug interaction studies with azacitidine. The results of *in vitro* inhibition study are inconclusive and the study should be repeated (see Comments to the Applicant).

RECOMMENDATION

The NDA 50-794 Application submitted for Vidaza™ (Azacitidine Injectable Suspension) is acceptable from the Clinical Pharmacology and Biopharmaceutics perspectives. The Applicant should address the Phase 4 Commitment and the Comments below and incorporate the OCPB Labelling Recommendations as outlined in section IV of this Review (pp. 32) in the proposed package insert for Vidaza™

A PHASE 4 COMMITMENT

- As azacitidine and its metabolites are primarily excreted by the kidneys, we recommend that you conduct a formal pharmacokinetics and safety study in patients with varying degrees of renal impairment. The results of this study will help making dosing recommendations for this patient population especially during the first course of therapy. In study, you may also address the dose- and time-dependent kinetics of the drug at doses ranging from 25-100 mg/m^2. Please submit study protocol for Agency's review.

COMMENTS TO THE APPLICANT

1. The *in vitro* inhibition Study DXNI1001 had not been adequately conducted. The results of this studies showed some enzymatic activation and high variability which could not be explained. Some experiments were done only in duplicates which results in an unreliable mean values. Therefore, we recommend that you repeat this study with the emphasis of studying the stability of azacitidine in microsomal systems before incubation experiments with selected substrates, performing the experiments at least in triplicates, defining the controls and inhibition controls, and providing an adequate assay method and validation for each enzyme system.

2. We remind you of your commitment in the pre-NDA minutes of meeting on 14-Apr-2003 that you will characterize the disposition of azacitidine and determine the plasma levels and activity of its major metabolites in MDS patients following subcutaneous administration.

Please forward the above Recommendation, the Phase 4 Commitment, Comments to the Applicant, and OCPB Labeling Recommendations (pp. 32 of this review) to the Applicant.

II. SUMMARY OF CLINICAL PHARMACOLOGY AND BIOPHARMACEUTICS FINDINGS

The Applicant developed Vidaza™ (azacitidine for Injectable Suspension) for the treatment of patients with myelodysplastic syndromes (MDS).

Azacitidine (4-amino-1-β-D-ribofuranosyl-1,3,5-triazin-2(1*H*)-one) is a ring analog of the naturally occurring pyrimidine nucleoside cytidine, and differs from cytidine by having nitrogen in the 5 position of the heterocyclic ring. The proposed mechanism of action is that azacitidine acts as an inhibitor of DNA methylation (i.e., hypomethylation) which results in reverting the epigenetic changes and establishing the antiproliferative signals that were diminished by hypermethylation in malignant cells.

In support of this NDA, three clinical studies were conducted by the Cancer and Leukemia Group B (CALGB) in MDS patients under the sponsorship of the National Cancer Institute (NCI): one pivotal Phase 3 (Study 9221) and two supportive Phase 2 studies (Studies 8921 and 8421). In Studies 9221 and 8921, patients received 75 mg/m^2 azacitidine subcutaneously (SC) once daily for 7 days on a 28-day cycle for a minimum of 4 cycles. In Study 8421, patients received the same dosing schedule with azacitidine given intravenously (IV). The primary clinical endpoint used in all three studies was the overall response rate [Complete (CR) and Partial Response (PR)]. According to the Applicant, the overall response rate (CR+PR) in MDS patients ranged from 14-18%.The most frequently observed adverse events were nausea, anemia, thrombocytopenia, neutropenia, vomiting, pyrexia, leukopenia, diarrhea, fatigue, injection site erythema, and constipation (according to the Applicant). The safety data from Studies 9221, 8921, and 8421 and from the entire NCI experience with azacitidine since 1975 did not reveal any QT_c prolongation during azacitidine treatment (according to the Medical Reviewer).

The Applicant proposes a starting dose of 75 mg/m^2 to be given subcutaneously (SC) once daily for 7 days, every 28 days (1 cycle) for a minimum of four cycles. This dose could be modified based on either response to the drug or hematological toxicities (neutrophil, platelet, and white blood cells counts). The selection of this starting dose was based on published clinical data reported for the IV formulation over 30 years. No dose-response or exposure-response data are available for azacitidine. The Applicant did not make any attempt to explore the relationships between degree of DNA hypomethylation and response rate and/or adverse events or between drug exposure and response rate and/or adverse events.

Vidaza™ (azacitidine for Injectable Suspension) is supplied as a sterile lyophilized powder intended for reconstitution with sterile Water for Injection to form a suspension for subcutaneous (SC) injection. The drug product contains a 100 mg of azacitidine and 100 mg of mannitol in a 30-ml vial. Each vial is

reconstituted with 4 mL of sterile Water for Injection prior to SC administration. The proposed commercial SC formulation was used in the pivotal Phase 3 Study 9221. In addition, the proposed commercial SC formulation was compared to the IV formulation in the bioavailability Study AZA-2002-BA-002. This study was an open-label, randomized, two-treatment, two-period, crossover study in which six MDS patients were administered either a single 75 mg/m^2 SC dose of azacitidine or a single 75 mg/m^2 IV infusion dose over 10 minutes on two different occasions separated by a 7-day washout period. Plasma samples were analyzed for azacitidine using an adequately validated [redacted] assay method with [redacted] Azacitidine was the only drug moiety measured in Study AZA-2002-BA-002. The bioavailability of azacitidine from both formulations was comparable, (geometric mean ratio of AUC_{SC}/AUC_{IV}=89%).

Pharmacokinetics: Single-dose pharmacokinetics of azacitidine were determined in Study AZA-2002-BA-002 following SC and IV administrations of 75 mg/m^2 azacitidine to six MDS patients. Azacitidine is rapidly absorbed following SC administration with a mean maximum plasma concentration (C_{max}) of 750±403 ng/ml attained in 0.5 hour. Azacitidine is widely distributed throughout the body. Mean volume of distribution is 76 L following IV administration which exceeds the volume of total body water (42 L) suggesting extensive tissue distribution. Applicant has not evaluated the protein binding of azacitidine. Azacitidine is rapidly eliminated from the body; plasma concentrations were detectable up to 2 hours after the IV dose and up to 4 hours after the SC dose. The mean half-life after IV administration was 22±1.2 minutes, while that after SC administration was 41±8 minutes. The inter-subject variability in pharmacokinetic parameters is less than 50%. No information is available to determine the effect of dose or treatment cycle on the pharmacokinetics of azacitidine.

Metabolism: No information is provided in the NDA submission on the metabolic pathways of azacitidine in humans. *In vitro* studies conducted with human hepatic S9 fractions indicate that azacitidine may undergo hepatic metabolism. Published data indicate that azacitidine may undergo deamination by cytidine deaminase; however, the identity and activity of the metabolites have not been characterized in humans.

Excretion: The Applicant did not conduct a mass balance study in cancer patients. Published studies indicate that azacitidine and its metabolites are primary excreted by the kidneys. Following IV administration to 5 cancer patients, the cumulative urinary excretion is 85% of the radioactive dose while fecal excretion is only < 1% over three days. Mean excretion of radioactivity in urine following receiving SC administration of ^{14}C-azacitidine is 50%. The mean elimination half-lives of total radioactivity (azacitidine and its metabolites) are 3.5 hours and 4.2 hours after IV and SC administrations, respectively.

Special Populations: The Applicant did not evaluate the effect of intrinsic factors such as age, gender, race, renal impairment or hepatic impairment on the pharmacokinetics of azacitidine. Based on the sub-group analyses of safety data, the Applicant include in the product label some specific dosing recommendations regarding the use of Vidaza™ in elderly patients, in patients with renal impairment, and in patients with liver metastases or severe hepatic impairment. These recommendations are acceptable from the clinical pharmacology perspectives. However, because azacitidine cause renal toxicity and because azacitidine and its metabolites are primary eliminated by the kidneys, we recommend that the Applicant should conduct a study in patients with varying degrees of renal impairment to properly provide dosing recommendations in this patient population (see Phase 4 Commitment). A —— statement was added in the label for the use of Vidaza™ in patients with severe renal impairment (estimated creatinine clearance < 30 ml/min) and in patients with underlying hepatic cirrhosis. There are no clinical studies conducted with Vidaza™ in pediatric patients or pregnant women.

Drug-Drug Interactions: The Applicant did not conduct any *in vivo* drug-drug interactions studies with azacitidine. Azacitidine undergoes deamination and cytochrome P450 enzymes may not be involved in the biotransformation of the drug. The Applicant conducted *in vitro* studies with human liver microsomes to investigate the inhibitory potential of azacitidine on the cytochrome P450 enzymes (CYP) 1A2, 2C9, 2C19, 2D6, 2E1, or 3A4; however, the results of this study are inconclusive and the study should be repeated (see Comments to the Applicant). *In vitro* studies in cultured human hepatocytes indicate that azacitidine is not an inducer of CYP 1A2, 2C19, or 3A4/5 at the *in vitro* concentrations close to those seen clinically.

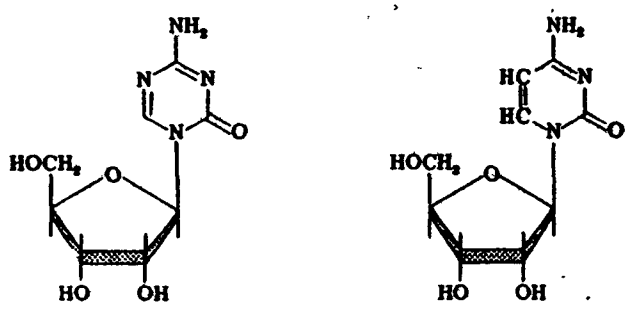
The Applicant has not evaluated the effect of azacitidine on the efflux pump transporter, P-glycoprotein. No, current data are available indicating that other metabolic/transporter pathways may be important.

III. QUESTION-BASED REVIEW

A. General Attributes

1. What are the highlights of the chemistry and physicochemical properties of azacitidine drug substance and drug product?

Azacitidine (4-amino-1-β-D-ribofuranosyl-1,3,5-triazin-2(1*H*)-one) is a ring analog of the naturally occurring pyrimidine nucleoside, cytidine. It differs from cytidine in having nitrogen in the 5-position of the heterocyclic ring. This substitution renders the ring chemically unstable and leads to rapid decomposition in aqueous solution. The structural formulas of azacitidine and cytidine are shown in Fig. 1.



Azacitidine **Cytidine**

Fig. 1. Structures of azacitidine and cytidine

Azacitidine has a molecular formula of $C_8H_{12}N_4O_5$ and a molecular weight of 244. The pKas of azacitidine determined at 25°C are 2.8±0.04 and 10.2±0.02. The octanol/water partition coefficient for azacitidine determined at 25°C is 1.7. Azacitidine is sparingly soluble in water, insoluble in ethanol, slightly soluble in propylene glycol, and polyethylene glycol, and freely soluble in dimethylsulfoxide.

Azacitidine is not stable in aqueous solutions. The degradation of azacitidine was in water at 5°C and 41% in water at 25°C over 24 hours. The degradation of azacitidine at 37°C in a pH 7.4 phosphate buffer solution was over 2 hours.

The Applicant identified and for azacitidine. The was

The drug product is as a sterile lyophilized powder intended for reconstitution with sterile Water for Injection to form a suspension for subcutaneous (SC) injection. It contains a 100 mg of azacitidine and 100 mg of mannitol in a 30-ml vial.

2. What is the proposed mechanism of action of azacitidine?

It is believed that hypermethylation of DNA is frequently associated with myelodysplastic syndromes (MDS)*. Azacitidine acts as an inhibitor of DNA methylation (i.e., hypomethylation) which results in reversing the epigenetic changes in the malignant cells and establishing the antiproliferative signals that were diminished by hypermethylation in malignant cells. Azacitidine is a pyrimidine analog of cytidine that readily incorporates into replicating DNA in place of cytidine. Like cytidine, azacitidine goes through three intracellular phosphorylation steps before incorporation into DNA. Azacitidine inhibits the methylation of newly synthesized DNA strands by inhibiting DNA methyl-transferase. First, the methyltransferase enzyme forms a covalent bond between its cysteine residue and the 5-position of the cytosine ring. A methyl group is then

transferred from S-adenosylmethionine to cytosine resulting in 5-methylcytosine in place of cytosine on the DNA chain. According to Jones and Taylor*, the concentration of azacitidine required for maximum inhibition of DNA methylation *in vitro* ranged from 2 to 5 µM.in mouse embryo cell (C3H/10T½CL8) cultures treated with azacitidine (mean C_{max}=3.1 µM after a single 75 mg/m^2 SC dose).

*Jones PA, Taylor SM, and Wilson VL, Inhibition of DNA by azacitidine. Cancer Res. 1983, 84:202-211.
*Jones PA and Taylor SM, Cellular differentiation, cytidine analogs and DNA methylation. Cell 1980, 20:85-93.

3. What is the proposed indication?

Vidaza™ is indicated for treatment of patients with myelodysplastic syndrome (MDS). It is effective for all five subtypes: refractory anemia (RA) or refractory anemia with ringed sideroblasts (RARS) requiring transfusions, with thrombocytopenia or significant clinical hemorrhage, or with neutropenia and infection requiring treatment with antibiotics; refractory anemia with excess blasts (RAEB), refractory anemia with excess blasts in transformation (RAEB-T), and chronic myelomonocytic leukemia (CMMoL).

4. What are the proposed dose and dosing regimens for azacitidine?

The recommended starting dose for MDS patients is 75 mg/m^2 once daily for 7 days every 28 days (1 cycle) for a minimum of four cycles. The dose may be increased to 100 mg/m^2 if no beneficial effect is seen after two treatment cycles and if no toxicity other than nausea and vomiting has occurred. Patients will be also monitored for hematological toxicities and azacitidine dose will be adjusted accordingly (Tables 1a and 1b). For patients with baseline WBC 3x10^9/L, ANC 1.5x10^9/L, and platelets 75x10^9/L, the dose is adjusted as follows (based on nadir counts for any given cycle):

Table 1a

Nadir Counts		% Dose in the Next Course	Azacitidine Dose (mg/m^2)
ANC (x10^9/L)	Platelets (x10^9/L)		
<0.5	<25	50%	37.5
0.5 –1.5	25-50	67%	50.25
>1.5	>50	100%	75.0

ANC=Absolute neutrophil count

For patients whose baseline counts are WBC <3x10^9/L, ANC<1.5 x10^9/L, or platelets <75x10^9/L, the dose is adjusted is based on nadir counts and bone marrow biopsy at the time of the nadir as in Table 1b.

Table 1b

WBC or Platelet Nadir % decrease in counts from baseline	Bone Marrow Biopsy at Time of Nadir (%)		
	30-60%	15-30%	<15%
	% Dose in the Next Course (Dose, mg/m^2)		
50 – 75%	100% (75.00)	50% (37.50)	33% (24.75)
> 75%	75% (56.25)	50% (37.50)	33% (24.75)

WBC=White blood cells

B. General Clinical Pharmacology

5. What are the design features of the pivotal clinical studies?

Azacitidine has been studied for almost 30 years as a cytotoxic agent to treat acute leukemias and other types of neoplasms. In 1985, the Cancer and Leukemia Group B (CALGB) began clinical studies with azacitidine in patients with myelodysplastic syndromes (MDS) under the sponsorship of the National Cancer Institute (NCI). The CALGB conducted one pivotal Phase 3 clinical study (Study 9221) and two supportive Phase 2 studies (8921 and 8421) to determine the efficacy and safety of azacitidine in patients with MDS. The primary endpoint determined in these studies was overall response rate [Complete (CR) and Partial Response (PR)].

Phase 3 Pivotal Clinical Study:

CALGB 9221: An open-label, multi-center, controlled, randomized, comparative, Phase 3 study conducted in 191 MDS patients in USA and Puerto Rico. Patients ranged in age from 31 to 92 years (132 males/59 females). Patients received either azacitidine 75 mg/m^2 SC for 7 days plus supportive care (n=99) or best supportive care only (n=92), repeated every 28 days (1 cycle) for a minimum of four cycles. The dose was increased to 100 mg/m^2 if no beneficial effect was seen after two treatment cycles. The overall response rate (CR+PR) in this study was 16.2% (n=16/99) for azaciditine arm and 0.0% (0/92) for the comparative arm ($p<0.0001$).

Supportive Clinical Studies:

CALGB 8921: An open-label, multi-center, uncontrolled, non-comparative, Phase 2 study conducted in 72 MDS patients in USA and Canada. Patients ranged in age from 23 to 82 (49 males/23 females). Patients received azacitidine 75 mg/m^2 SC for 7 days, repeated every 28 days (1 cycle) for a minimum of four cycles. The dose was increased to 100 mg/m^2 if no beneficial effect was seen after two treatment cycles. The overall response rate (CR+PR) in this study was 13.9% (n=10/72).

CALGB 8421: A pilot, open-label, multi-center, uncontrolled, non-comparative, Phase 2 study conducted in 48 MDS patients in USA and Canada. Patients ranged in age from 35 to 81 years (31 males/17 females). Patients received azacitidine 75 mg/m^2 as a continuous intravenous (IV) infusion for 7 days, repeated every 28 days (1 cycle) for a minimum of four cycles. The dose was increased to 100 mg/m^2 if no beneficial effect was seen after two treatment cycles. The overall response rate (CR+PR) in this study was 18.8% (n=9/48).

6. What is the basis for selecting the response endpoints, i.e., clinical or surrogate endpoints, or biomarkers (also called pharmaco-dynamics, PD) and how are they measured in clinical pharmacology and clinical studies?

The three CALGB Studies 9221, 8921, and 8421 used a surrogate clinical endpoint of "overall response rate" (CR+PR). In the pre-NDA meeting on December 2001, the FDA has agreed on the accelerated approval of Vidaza™ based upon overall response rate provided that a confirmatory study be conducted using the surrogate clinical endpoint of "survival".

This overall response rate endpoint is based on the assessment of the criteria of improvements in bone marrow blast counts, cellular morphology, and peripheral blood counts, which is similar to the assessment of acute leukemia (AL) response that commonly used by the International Working Group for MDS.

7. Are the active moieties in the plasma (or other biological fluid) appropriately identified and measured to assess the pharmacokinetic parameters and exposure-response relationships?

Azacitidine was the only drug moiety measured in the bioavailability Study AZA-2002-BA-002.

Exposure-Response Evaluations

8. What are the characteristics of the exposure-response relationship for efficacy?

The dose used in the three CALGB studies (Studies 9221, 8921, and 8421) was a 75 mg/m^2 dose daily for 7 days every 28 days. This dose was either increased or decreased based on the patient's response. Few subjects required a higher dose (≥ 100 mg/m^2) in order to achieve a response and a similar number of patients also required a lower dose (< 75 mg/m^2) to achieve a response (Table 2).

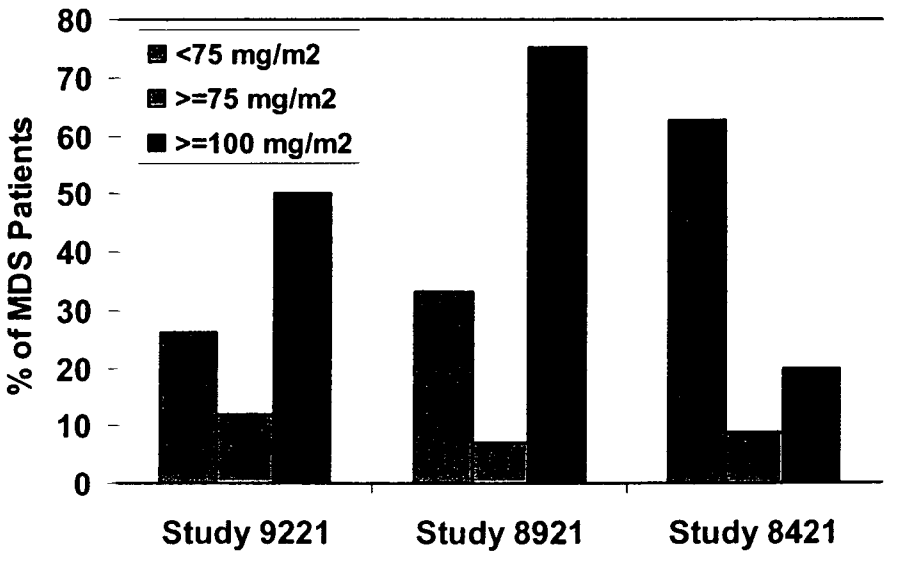
Table 2. Analysis of response rates by dose (Applicant's)

	Number (% of Subjects)											
	8421 Azacitidine (N=48)			8921 Azacitidine (N=70)			9221 All Azacitidine (N=150)			8921/9221 All Azacitidine (N=220)		
	<75 mg/m² (N=8)	≥75 to <100 mg/m² (N=34)	≥100 mg/m² (N=5)	<75 mg/m² (N=9)	≥75 to <100 mg/m² (N=55)	≥100 mg/m² (N=4)	<75 mg/m² (N=27)	≥75 to <100 mg/m² (N=101)	≥100 mg/m² (N=6)	<75 mg/m² (N=36)	≥75 to <100 mg/m² (N=156)	≥100 mg/m² (N=10)
Response												
Overall (CR+PR)	5 (62.5)	3 (8.8)	1 (20.0)	3 (33.3)	4 (7.3)	3 (75.0)	7 (25.9)	12 (11.9)	3 (5.0)	10 (27.8)	16 (10.3)	6 (60.0)
Complete (CR)	1 (12.5)	2 (5.9)	0 (0.0)	1 (11.1)	1 (1.8)	2 (50.0)	4 (14.8)	4 (4.0)	1 (16.7)	5 (13.9)	5 (3.2)	3 (30.0)
Partial (PR)	4 (50.0)	1 (2.9)	1 (20.0)	2 (22.2)	3 (5.5)	1 (25.0)	3 (11.1)	8 (7.9)	2 (33.3)	5 (13.9)	11 (7.1)	3 (30.0)
Improvement, not CR or PR	2 (25.0)	5 (14.7)	4 (80.0)	5 (55.6)	6 (10.9)	1 (25.0)	11 (40.7)	38 (37.6)	1 (16.7)	16 (44.4)	44 (28.2)	2 (20.0)
Stable Disease	1 (12.5)	26 (76.5)	0 (0.0)	1 (11.1)	45 (81.8)	0 (0.0)	8 (29.6)	48 (47.5)	1 (16.7)	9 (25.0)	93 (59.6)	1 (10.0)
Relapse	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (3.7)	0 (0.0)	0 (0.0)	1 (2.8)	0 (0.0)	0 (0.0)
Disease Progression	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	3 (3.0)	1 (16.7)	0 (0.0)	3 (1.9)	1 (10.0)

KEY: CR=Complete Response, PR=Partial Response, CALGB=Cancer and Leukemia Group B

Because of the few number of patients in the dosing groups of < 75 mg/m^2 and ≥ 100 mg/m^2, no obvious dose-response relationship among the three dosing groups could be drawn. In the pivotal study (Study 9221), the % of patients who achieved overall response was 25.9%, 11.9%, and 50% at doses of < 75 mg/m^2, ≥ 75-< 100 mg/m^2 and ≥100 mg/m^2, respectively.

Fig. 2. Percent of patients/dose group with Overall Response (CR+PR)



9. What are the characteristics of the exposure-response relationship for safety?

According to the Applicant, the most frequently observed treatment-emergent adverse events (TEAEs) were nausea, anemia, thrombocytopenia, neutropenia, vomiting, pyrexia, leukopenia, diarrhea, fatigue, injection site erythema, and constipation. The most frequently occurring adverse events (i.e., 20% of patients in the 8921/9221 all azacitidine group) summarized by dose are shown in Table 3.

Table 3. Most frequently occurring[a] TEAEs by dose (Applicant's)

	Number (%) of Subjects								
	Subcutaneous								
	8921			9221			8921,9221		
	Azacitidine (N=70)			All Azacitidine[c] (N=150)			All Azacitidine[c] (N=220)		
Preferred Term[b]	< 75 mg/m² (N=24)	≥ 75 mg/m² (N=69)	≥ 100 mg/m² (N=18)	< 75 mg/m² (N=76)	≥ 75 mg/m² (N=149)	≥ 100 mg/m² (N=19)	< 75 mg/m² (N=100)	≥ 75 mg/m² (N=218)	≥ 100 mg/m² (N=37)
At least 1 TEAE	24 (100.0)	68 (98.6)	18 (100.0)	76 (100.0)	149 (100.0)	18 (94.7)	100 (100.0)	217 (99.5)	36 (97.3)
Nausea	7 (29.2)	50 (72.5)	10 (55.6)	27 (35.5)	88 (59.1)	10 (52.6)	34 (34.0)	138 (63.3)	20 (54.1)
Anemia NOS	11 (45.8)	42 (60.9)	11 (61.1)	41 (53.9)	100 (67.1)	12 (63.2)	52 (52.0)	142 (65.1)	23 (62.2)
Thrombocytopenia	15 (62.5)	40 (58.0)	5 (27.8)	48 (63.2)	90 (60.4)	8 (42.1)	63 (63.0)	130 (59.6)	13 (35.1)
Vomiting NOS	4 (16.7)	44 (63.8)	5 (27.8)	13 (17.1)	65 (43.6)	3 (15.8)	17 (17.0)	109 (50 0)	8 (21.6)
Pyrexia	5 (20.8)	31 (44.9)	4 (22.2)	28 (36 8)	57 (38.3)	11 (57.9)	33 (33.0)	88 (40.4)	15 (40.5)
Leukopenia NOS	9 (37.5)	22 (31 9)	9 (50.0)	38 (50.0)	71 (47.7)	7 (36 8)	47 (47.0)	93 (42.7)	16 (43.2)
Diarrhea NOS	5 (20.8)	21 (30.4)	4 (22.2)	13 (17.1)	40 (26.8)	6 (31.6)	18 (18.0)	61 (28.0)	10 (27.0)
Fatigue	4 (16.7)	15 (21.7)	2 (11.1)	21 (27.6)	46 (30.9)	7 (36.8)	25 (25.0)	61 (28.0)	9 (24.3)
Injection site erythema	3 (12.5)	25 (36.2)	2 (11.1)	10 (13.2)	43 (28.9)	6 (31.6)	13 (13.0)	68 (31.2)	8 (21.6)
Constipation	2 (8.3)	13 (18.8)	2 (11.1)	11 (14.5)	49 (32.9)	6 (31.6)	13 (13.0)	62 (28.4)	8 (21.6)
Neutropenia	1 (4.2)	19 (27.5)	5 (27.8)	20 (26.3)	47 (31.5)	5 (26.3)	21 (21.0)	66 (30.3)	10 (27.0)
Ecchymosis	3 (12.5)	19 (27.5)	4 (22.2)	12 (15.8)	32 (21.5)	5 (26.3)	15 (15.0)	51 (23.4)	9 (24.3)
Cough	1 (4.2)	17 (24.6)	1 (5.6)	15 (19.7)	35 (23.5)	2 (10.5)	16 (16.0)	52 (23.9)	3 (8.1)
Weakness	0	17 (24.6)	3 (16.7)	12 (15.8)	31 (20.8)	6 (31.6)	12 (12.0)	48 (22.0)	9 (24.3)
Dyspnea NOS	2 (8.3)	13 (18.8)	3 (16.7)	13 (17.1)	34 (22.8)	7 (36.8)	15 (15.0)	47 (21.6)	10 (27.0)
Rigors	1 (4.2)	16 (23.2)	4 (22.2)	12 (15 8)	26 (17 4)	3 (15.8)	13 (13.0)	42 (19.3)	7 (18.9)
Petechiae	4 (16.7)	18 (26.1)	4 (22.2)	11 (14.5)	23 (15.4)	2 (10.5)	15 (15.0)	41 (18 8)	6 (16.2)
Injection site pain	1 (4.2)	11 (15.9)	3 (16.7)	9 (11 8)	28 (18.8)	6 (31.6)	10 (10.0)	39 (17.9)	9 (24.3)
Arthralgia	1 (4.2)	10 (14.5)	3 (16 7)	8 (10.5)	26 (17.4)	4 (21.1)	9 (9 0)	36 (16.5)	7 (18 9)
Headache NOS	1 (4.2)	11 (15.9)	2 (11.1)	13 (17.1)	22 (14.8)	5 (26 3)	14 (14 0)	33 (15.1)	7 (18.9)
Anorexia	1 (4 2)	9 (13.0)	4 (22 2)	11 (14.5)	22 (14.8)	3 (15 8)	12 (12 0)	31 (14.2)	7 (18.9)
Pharyngitis	1 (4 2)	10 (14 5)	2 (11 1)	14 (18.4)	18 (12.1)	4 (21.1)	15 (15 0)	28 (12.8)	6 (16.2)
Pain in limb	3 (12.5)	6 (8.7)	2 (11.1)	9 (11.8)	26 (17.4)	2 (10.5)	12 (12.0)	32 (14.7)	4 (10.8)
Edema peripheral	2 (8.3)	11 (15.9)	1 (5 6)	10 (13.2)	22 (14.8)	3 (15.8)	12 (12.0)	33 (15.1)	4 (10.8)
Dizziness	0	14 (20.3)	1 (5 6)	10 (13.2)	17 (11.4)	3 (15.8)	10 (10.0)	31 (14.2)	4 (10.8)
Contusion	3 (12.5)	6 (8.7)	2 (11.1)	11 (14 5)	21 (14.1)	3 (15 8)	14 (14.0)	27 (12.4)	5 (13.5)
Erythema	1 (4.2)	12 (17.4)	1 (5.6)	3 (3.9)	20 (13 4)	2 (10.5)	4 (4 0)	32 (14.7)	3 (8.1)
Epistaxis	1 (4.2)	11 (15.9)	0	5 (6.6)	19 (12.8)	3 (15.8)	6 (6.0)	30 (13.8)	3 (8.1)
Rash NOS	1 (4.2)	12 (17.4)	1 (5 6)	4 (5.3)	14 (9.4)	1 (5 3)	5 (5 0)	26 (11.9)	2 (5.4)
Injection site bruising	1 (4.2)	14 (20 3)	0	1 (1.3)	15 (10.1)	0	2 (2.0)	29 (13.3)	0
Anxiety	3 (12 5)	12 (17.4)	1 (5 6)	4 (5.3)	11 (7.4)	2 (10.5)	7 (7.0)	23 (10.6)	3 (8.1)
Hypokalemia	0	8 (11.6)	0	7 (9.2)	15 (10.1)	0	7 (7.0)	23 (10.6)	0
Appetite decreased NOS	1 (4.2)	8 (11.6)	0	9 (11.8)	13 (8.7)	0	10 (10.0)	21 (9.6)	0
Insomnia	0	5 (7.2)	1 (5 6)	5 (6.6)	14 (9.4)	3 (15.8)	5 (5.0)	19 (8.7)	4 (10.8)
Rales	0	7 (10.1)	1 (5.6)	3 (3.9)	9 (6.0)	0	3 (3.0)	16 (7.3)	1 (2.7)
Cellulitis	2 (8 3)	1 (1 4)	2 (11 1)	3 (3 9)	10 (6.7)	0	5 (5.0)	11 (5 0)	2 (5.4)

[a] ≥ 20.0% frequency in any treatment group.

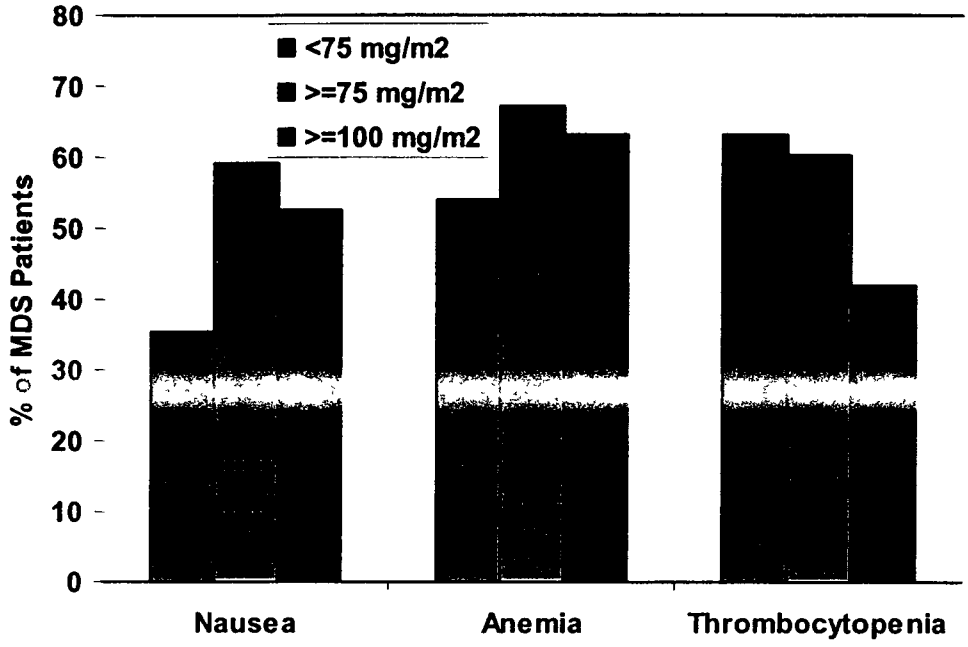
[b] Multiple reports of the same preferred term for a subject are only counted once within each dose category

[c] Includes all subjects exposed to azacitidine, including 9221 subjects after crossing over from observation.

Adverse events are counted for the last prior dose Adverse events before the first dose are excluded.

KEY: NOS=not otherwise specified, TEAE=treatment-emergent adverse event

Fig. 3. Percent of patients/dose group with most frequent adverse events (Study 9221)



In general, there is no obvious relationship between dose and toxicity; the incidence of adverse events was comparable among the dosing groups per each adverse event.

10. Does this drug prolong the QT_c interval?

According to the Medical Reviewer (HFD-150), the safety data from the three CALGB studies (Studies 9221, 8921, and 8421) and from the entire NCI experience with azacitidine since 1975 did not reveal any QT_c prolongation during azacitidine treatment. The electrocardiograms (ECGs) performed during the pivotal and supporting CALGB studies reported a variety of cardiac rhythm abnormalities that are consistent with the population's age, but there is no mention of QT_c prolongation.

11. Are the dose and dosing regimen of azacitidine consistent with the known relationship between dose-concentration-response, and are there any unresolved dosing or administration issues?

The Applicant has not conducted any formal, dose-rising, Phase 1 studies for azacitidine in MDS patients. The selection of azacitidine dose of 75 mg/m^2 given SC once daily for 7 days was based on published studies* using intravenous (IV) azacitidine. These studies were designed to determine the maximum tolerated dose (MTD) in other oncology indications. In an open-label, randomized, multi-center study, two azacitidine treatment regimens were compared in 27 subjects

with progressive cancer. Fifteen subjects received azacitidine 200-633 mg/m^2 as an IV bolus once a week for four weeks and the other 12 subjects received azacitidine 50-158 mg/m^2 daily as an IV bolus for 5 days followed by a 9-day rest period. The authors estimated an MTD of 150 mg/m^2 daily for 5 days every two weeks. In an another open-label, uncontrolled, multi-center study, Vogler evaluated the safety of azacitidine administered as a continuous IV infusion in doses ranging from 50 to 300 mg/m^2 daily for 5 days, with the cycle repeated every 2 to 4 weeks. The MTD determined in this study was 300 mg/m^2. The Applicant conducted CALGB studies (Studies 9221, 8921, and 8421) at a starting SC dose of 75 mg/m^2, which is much less than the MTD determined in the published Phase 1 dosing-rising studies.

*Shnider BI, Baig M, Colsky J. A phase I study of azacitidine (NSC-102816). J Clin Pharmacol 1976;16(4):205-212.

*Vogler WR, Miller DS, Keller JW. Azacitidine (NSC 102816): a new drug for the treatment of myeloblastic leukemia. Blood 1976;48(3):331-337.

Pharmacokinetic characteristics of Azacitidine

12. What are the single- and multiple-dose PK parameters following administration of azacitidine?

Single-Dose Administration:

In support of Human Pharmacokinetics/Bioavailability section (Item 6) of this NDA submission, the Applicant conducted only one clinical pharmacology/bio-availability study (Study AZA-2002-BA-002) to determine the single-dose pharmacokinetics and bioavailability of azacitidine following SC and IV administrations. Study AZA-2002-BA-002 was an open-label, randomized, two-treatment, two-period, crossover study in six MDS patients. The six MDS patients averaged (mean±SD) in age 71± 8.7 years and in weight 76.8 ± 14.4 kg. There were three males and three females; all were Caucasians and were nonsmokers. Each patient received either a single 75 mg/m^2 SC dose of azacitidine (the SC formulation proposed for marketing) or a single 75 mg/m^2 IV infusion dose of azacitidine administered over 10 minutes on two different occasions separated by a 7-day washout period. Pharmacokinetic (PK) sample collection was performed for up to 48 hours during each treatment period. Azacitidine concentrations in plasma samples were analyzed using [redacted], (see section III.F of this review for the assay validation).

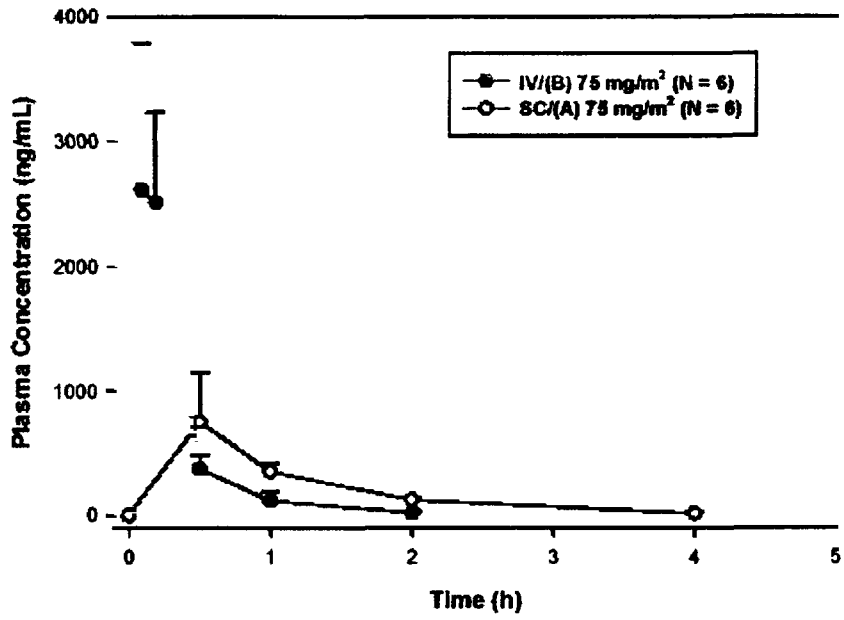
The single-dose pharmacokinetic (PK) parameters for azacitidine following SC and IV administrations to six MDS patients are summarized below (Table 4). The mean concentration-time profiles for azacitidine after SC and IV administrations are shown in Fig. 4.

Table 4. Arithmetic mean±SD (%CV) Single-dose PK parameters (n=6)

Treatment	C_{max} (ng/ml)	t_{max} (h)	$AUC_{0-\infty}$ (ng.h/ml)	$t_{½}$ (min)	CL (L/h)	Vd (L)
75 mg/m^2 SC	750±403 (54%)	0.5±0.0 (0%)	960±458 (47%)	41.4±8.4 (20%)	167±48 (29%)	--
75 mg/m^2 IV	2750±1069 (39%)	--	1044±285 (27%)	21.6±1.2 (5.7%)	146±47 (32%)	76±25 (33%)

t_{max} after SC dosing only; apparent clearance (CLsc) after SC dosing and systemic clearance (CL) following IV infusion; Vd following IV infusion only.

Fig. 4. Mean Azacitidine Concentration-Time Profiles after SC and IV Administrations (Applicant's)



Absolute bioavailability determined as $AUC_{0-\infty(SC)}/AUC_{0-\infty(IV)}$ averages 92±25%. Azacitidine is rapidly eliminated from the body; plasma concentrations were detectable up to 2 hours after the IV dose and up to 4 hours after the SC dose. The mean half-life after IV administration was 22±1.2 min, while that after SC administration was 41±8 min. The difference in elimination half-lives between SC and IV formulations may be an artifact due to the limited number of data points available to characterize the elimination phases for both formulations. The mean systemic (IV) clearance of azacitidine (147 L/h) exceeds the hepatic blood flow (90 L/h), suggesting that azacitidine may undergo extraheptic elimination.

Multiple-dose Administration:

The Applicant has not evaluated the steady state pharmacokinetics of azacitidine following the 75 mg/m^2 SC dose once daily for 7 days. As azacitidine is rapidly eliminated after SC administration (mean $t_{½}$=41 min), its accumulation upon chronic SC administration is not expected and multiple-dose kinetics are predictable of single-dose kinetics.

13. How do the pharmacokinetics of azacitidine in healthy volunteers compare to that in MDS patients?

Because azacitidine is a cytotoxic drug, it has never been administered to healthy volunteers.

14. What are the characteristics of azacitidine absorption?

Azacitidine is rapidly absorbed following SC administration with a mean maximum plasma concentration (C_{max}) of 750±403 ng/ml attained in 0.5 hour.

15. What are the characteristics of azacitidine distribution?

Azacitidine is widely distributed throughout the body. Following IV administration to six MDS patients, azacitidine volume of distribution averages 76 L (volume of total body water= 42 L). The Applicant has not evaluated the plasma protein binding of azacitidine. However, as azacitidine has a volume of distribution exceeding total body water, it may bind more to tissue proteins than to plasma proteins.

16. Does the mass-balance study suggest renal or hepatic as the major route of azacitidine elimination?

The Applicant did not conduct a mass-balance study in cancer patients. Patients enrolled in the pivotal clinical study (Study 9221) had their ALT ranging from 4.0-113.0 U/L, AST from 8.0-112.0 U/L, total bilirubin from 3.4-59.9 µmol/L, and estimated creatinine clearance ranging from 21.4-177.6 ml/min.

Published mass-balance studies* revealed that azacitidine and its metabolites are primary excreted by the kidneys. For example, Israili et al. found that following IV administration to 5 cancer patients, cumulative urinary excretion was 73-98% of the total radioactive dose while fecal excretion was only < 1% over three days. However, it is not known how much parent drug excreted unchanged in urine or feces. During the pre-NDA meeting on 14-Apr-2003, the Applicant committed to characterize the disposition of azacitidine and determine the plasma levels and activity of azacitidine major metabolite(s) following SC administration to MDS patients (see Comments to the Applicant).

* Israili ZH, Vogler WR, et al, The disposition and pharmacokinetics in humans of azacitidine administered intravenously as a bolus by continuous infusion. Cancer Res 1976, 36:1453-1461.
* Troetel WM, Weiss AJ, et al, Absorption, distribution, and excretion of azacitidine in man. Cancer Chemother Rep, Part 1, 1972, 56: 405-411.
*Vogler R, arkin S and Velez-Garcia F, Phase 1 study of twice weekly azacitidine Cancer Chemother Rep, Part 1, 1974, 58:895-899.

17. What are the characteristics of azacitidine metabolism?

The Applicant did not evaluate the metabolism of azacitidine in humans. The Applicant evaluated the *in vitro* disappearance of azacitidine after incubation at 37° C with human hepatic S9 fractions and potassium phosphate buffer, pH 7.4 at azacitidine concentration of 64 µM and 20 µM, respectively (Study QKAN-2003-0199-BIO). At 90 minutes, the percent of unchanged azacitidine remained in the reaction mixture was 4.3% after incubation with human hepatic S9 fractions and 83% after incubation with potassium phosphate buffer, pH 7.4, suggesting that azacitidine may be metabolized in the liver. No metabolites were identified in the human hepatic S9 reaction mixtures. An *in vitro* ^{14}C- study is ongoing to characterize azacitidine metabolites.

*Chabner and Collins (1990) indicate that azacitidine may undergo deamination by cytidine deaminase, an enzyme found in high concentrations in liver, granulocytes, and intestinal epithelium and in lower concentrations in plasma. The identity of metabolites and their pharmacological activity have not been characterized in humans.

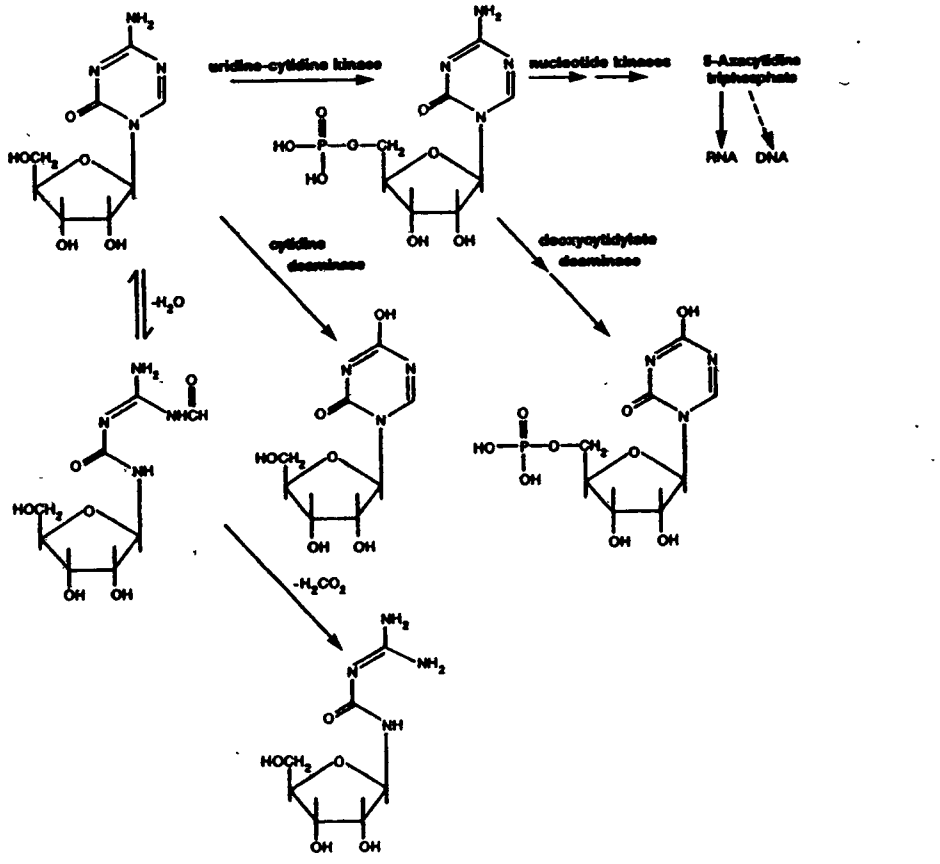


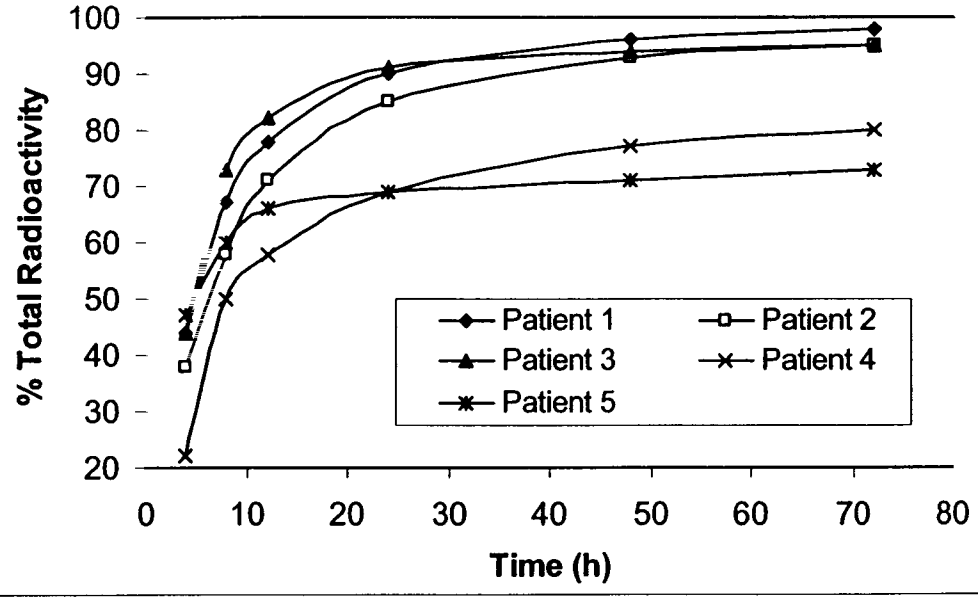
Fig. 5. Metabolic activation and degradation of azacitidine (Chabner and Collins 1990)

*Chabner BA and Collins JM, Cancer Chemotherapy: Principles and Practice. J. B. Lippincott Company Philadelphia, 1990, pp. 168-173.

18. What are the characteristics of azacitidine excretion?

Published mass-balance studies* indicated that azacitidine and its metabolites are primary excreted by the kidneys; however, it is not known how much unchanged parent drug excreted in urine or feces. Israili et al (1976) found that following single IV bolus doses of ^{14}C-azacitidine to five cancer patients (150, 150, 200, 200, and 250 mgm^2), cumulative urinary excretion accounted for 73-98% of the ^{14}C-dose over three days (Fig. 6) while fecal excretion was only < 1%. The majority of urinary radioactivity was present as parent drug plus unknown metabolites.

Fig. 6. Cumulative urinary excretion of total radioactivity after a single IV bolus dose



[data blotted by the reviewer from Israili et al Study, 1976]

Troetel et al. (1972) found that patients receiving SC azacitidine excreted less radioactivity in the urine (50% of total dose, n=5) than did those receiving IV azacitidine (85% of total dose, n=6). The mean plasma half-life of total radio-activity (azacitidine and its metabolites) was 3.5 hours and 4.2 hours after IV and SC administrations, respectively. Patients received 60 mg/m^2 doses (SC or IV), except for one patient who received a 35 mg/m^2 IV dose.

Vogler et al. (1974) found that approximately 90% of the radioactivity was present in the urine within 24 hours in three cancer patients after a 200 mg/m^2 IV dose of ^{14}C-azacitidine as part of a Phase 1 study. Little amounts of radioactivity were detected in the feces.

* Israili ZH, Vogler WR, et al, The disposition and pharmacokinetics in humans of azacitidine administered intravenously as a bolus by continuous infusion. Cancer Res 1976, 36:1453-1461.
* Troetel WM, Weiss AJ, et al, Absorption, distribution, and excretion of azacitidine in man. Cancer Chemother Rep, Part 1, 1972, 56: 405-411.
*Vogler R, arkin S and Velez-Garcia F, Phase 1 study of twice weekly azacitidine Cancer Chemother Rep, Part 1, 1974, 58:895-899.

19. Based on pharmacokinetic (PK) parameters, what is the degree of linearity or nonlinearity in the azacitidine dose-concentration relationship?

No information is available in the NDA submission whether azacitidine exhibits linear or non-linear kinetics. Azacitidine PK parameters were determined in Study AZA-2002-BA-002 after single-dose administration of 75 mg/m^2 of azacitidine. The product label recommends a starting dose of 75 mg/m^2 given subcutaneously once daily for 7 days. This starting dose is adjusted based on either patient's response or hematological toxicities during the course of therapy from 25 mg/m^2 to 100 mg/m^2. The Applicant should establish the linearity of azacitidine within this dosing range (see the Phase 4 Commitment).

20. How do azacitidine pharmacokinetic (PK) parameters change with time following chronic dosing of azacitidine?

No chronic dosing information is available to determine the time-dependent pharmacokinetics of azacitidine. Azacitidine PK parameters were determined in Study AZA-2002-BA-002 after single-dose administration. Accumulation of azacitidine following chronic SC administration is not expected since it is rapidly eliminated from the body (t½=41 min).

21. What is the intra- and inter-subject variability of pharmacokinetic parameters in patients, and what are the major causes of variability.

From Study AZA-2002-BA-002, the inter-subject variability in the PK parameters is less than 50%; this may be due to the inherent variability in azacitidine disposition or to the variability in the bioanalytical measures or due to the limited sample size (n=6).

C. Intrinsic Factors

22. What intrinsic factors influence exposure or response to azacitidine? What is the impact of these factors on exposure and response?

The Applicant did not evaluate the effect of intrinsic factors such as age, gender, race, renal impairment or hepatic impairment on the pharmacokinetics of azacitidine. However, the Applicant performed sub-group analyses on the safety data from CALGB Studies 9221 and 8921 following the SC administration. More than 90% of the patients enrolled in these studies were Caucasians; therefore, sub-group analyses of adverse events by race could not be performed. The age

of enrolled patients ranged from 23 to 92 years. Approximately two-thirds of enrolled patients were males. Patients enrolled in the pivotal Study 9221 had ALT ranging from 4.0-113.0 U/L, AST from 8.0-112.0 U/L, total bilirubin from 3.4-59.9 µmol/L, and estimated creatinine clearance ranging from 21.4-177.6 ml/min. Results from these sub-group safety analyses are shown below:

Safety Sub-Group Analyses by Age:

From Table 5, it is seen that in the all 8921/9221 azacitidine group (N=220), the incidence of neutropenia is about 1.5-fold higher in elderly patients (≥ 65 years and ≥ 75 years). Conversely, the incidence of injection site erythema, pharyngitis, injection site pain, night sweats, and hematoma tend to be higher in the younger group (< 65 years) compared to the elderly groups. In the 9221 observation group, no trends are noted in the incidence of neutropenia, pharyngitis, or night sweats based on age.

Table 5. TEAEs With a > 10.0% Difference in Frequency Across Age Groups

	Number (%) of Subjects					
	Subcutaneous					
	9221 Observation (N=92)			8921/9221 All Azacitidine[b] (N=220)		
	< 65 Years	≥ 65 Years	≥ 75 Years	< 65 Years	≥ 65 Years	≥ 75 Years
Preferred Term[a]	(N=33)	(N=58)	(N=25)	(N=79)	(N=137)	(N=52)
At least 1 TEAE	33 (100%)	56 (96.6%)	23 (92%)	79 (100%)	137 (100%)	52 (100%)
Neutropenia	4 (12.1 %)	6 (10.3%)	3 (12%)	20 (25.3%)	50 (36.5%)	19 (36.5%)
Injection site erythema	0	0	0	33 (41.8%)	43 (31.4%)	13 (25.0%)
Pharyngitis	3 (9.1%)	4 (6.9%)	1 (4.0%)	22 (27.8%)	22 (16.1%)	10 (19.2%)
Injection site pain	0	0	0	20 (25.3%)	29 (21.2%)	7 (13.5%)
Night sweats	1 (3.0%)	2 (3.4%)	1 (4.0%	12 (15.2%)	7 (5.1%)	3 (5.8%)
Hematoma NOS	0	0	0	11 (13.9%)	8 (5.8%)	2 (3.8%)

[a] Multiple reports of the same preferred term for a subject are only counted once within each treatment group.
[b] Includes all subjects exposed to azacitidine, including 9221 subjects after crossing over from observation.
Sorted by decreasing frequency in the 8921/9221 all azacitidine 75 years age group.
KEY: NOS=not otherwise specified, TEAE=treatment-emergent adverse event

Safety Sub-Group Analyses by Gender:

From table 6, it is seen that in the all 8921/9221 azacitidine group (n=220), the incidence of vomiting (1.3-fold), diarrhea (1.5-fold), headache (1.9-fold), arthralgia (1.6-fold), erythema (2-fold), injection site bruising (2.6-fold), tachycardia (2.9-fold), and post procedural hemorrhage (4.8-fold) is higher in

females than in males, whereas thrombocytopenia is more frequent in males than in females (2.6-fold higher in males than in females).

Table 6. TEAEs With a > 10.0% Difference in Frequency Between Males and Females

	Number (%) of Subjects			
	Subcutaneous			
	9221 Observation (N=92)		8921/9221 All Azacitidine[b] (N=220)	
	Male	Female	Male	Female
Preferred Term[a]	(N=60)	(N=32)	(N=150)	(N=70)
At least 1 TEAE	58 (96.7%)	31 (96.9%)	149 (99.3%)	70 (100 %)
Vomiting NOS	5 (8.3%)	0	73 (48.7%)	46 (65.7%)
Thrombocytopenia	28 (46.7%)	14 (43.8%)	104 (69.3%)	40 (57.1%)
Diarrhea NOS	10 (16.7%)	3 (9.4%)	47 (31.3%)	33 (47.1%)
Headache NOS	5 (8.3%)	5 (15.6%)	25 (16.7%)	23 (32.9%)
Arthralgia	2 (3.3%)	1 (3.1%)	28 (18.7%)	21 (30.0%)
Erythema	4 (6.7%)	0	19 (12.7%)	18 (25.7%)
Injection site bruising	0 (0.0%)	0	14 (9.3%)	17 (24.3%)
Tachycardia NOS	5 (8.3%)	1 (3.1%)	8 (5.3%)	11 (15.7%)
Post procedural hemorrhage	0	1 (3.1%)	4 (2.7%)	9 (12.9%)

[a] Multiple reports of the same preferred term for a subject are only counted once within each treatment group.
[b] Includes all subjects exposed to azacitidine, including 9221 subjects after crossing over from observation.
Sorted by decreasing frequency in the 8921/9221 all azacitidine female group.
KEY: NOS=not otherwise specified, TEAE=treatment-emergent adverse event

Safety Sub-Group Analyses by Renal Function:

From Table 7, in the 9221 all azacitidine group, in general the incidence of adverse events is higher in Quartile 1 (estimated creatinine clearance=21.4-57.1 ml/min) than in Quartiles 2-4 (estimated creatinine clearance=59.4-177.6 ml/min) For example, the incidence of arthralgia, gingival bleeding, post procedural hemorrhage cardiac failure congestive is 34.4%, 21.9%, 15.6%, and 12.5%, respectively in Quartile 1 versus 18.2%, 9.1%, 3.4%, and 2.3%, respectively in Quartiles 2-4.

Table 7. Baseline Estimated Creatinine Clearance: TEAEs in the 9221 All Azacitidine Group With a Higher Frequency in Quartile 1 vs. Quartiles 2-4 (> 10.0% Difference)

	Number (%) of Subjects			
	Subcutaneous			
	9221 Observation (N=92)		9221 All Azacitidine[b] (N=150)	
	Quartile 1	Quartiles 2-4	Quartile 1	Quartiles 2-4
Preferred Term[a]	(N=20)	(N=52)	(N=32)	(N=88)
At least 1 TEAE	19 (95%)	52 (100%)	32 (100%)	88 (100%)
Vomiting NOS	1 (5.0%)	1 (1.9%)	19 (59.4%)	39 (44.3%)
Rigors	3 (15.0%)	5 (9.6%)	11 (34.4%)	21 (23.9%)
Arthralgia	1 (5.0%)	1 (1.9%)	11 (34.4%)	16 (18.2%)
Pain NOS	0	0	9 (28.1%)	7 (8.0%)
Weight decreased	1 (5.0%)	7 (13.5%)	8 (25.0%)	10 (11.4%)
Fatigue aggravated	0	4 (7.7%)	8 (25.0%)	8 (9.1%)
Gingival bleeding	2 (10.0%)	1 (1.9%)	7 (21.9%)	8 (9.1%)
Post procedural hemorrhage	0	1 (1.9%)	5 (15.6%)	3 (3.4%)
Cardiac failure congestive	2 (10.0%)	2 (3.8%)	4 (12.5%)	2 (2.3%)

[a] Multiple reports of the same preferred term for a subject are only counted once within each treatment group.
[b] Includes all subjects exposed to azacitidine, including 9221 subjects after crossing over from observation.
Sorted by decreasing frequency in the 9221 all azacitidine Quartile 1 group.
Estimated creatinine clearance: Quartile 1 = 21.4-57.1 mL/min, Quartiles 2-4 = 59.4-177.6 mL/min.
Baseline was defined as the last lab value before the date of randomization or, if no value was available prior to randomization, the earliest value following randomization up to and including the day of first dose of study medication for azacitidine subjects, and on the day of randomization for observation subjects
KEY: NOS=not otherwise specified, TEAE=treatment-emergent adverse event

Safety Sub-Group Analyses by Hepatic function:

From Table 8, it is seen that with the exception of neutropenia, dizziness, or lymphadenopathy, which appear to increase in patients with increased transaminases, it does not appear to be any consistent trend for an increased incidence of adverse events patients with higher baseline liver function values (Quartile 4*) than those with lower baseline values (Quartiles 1-3*) in the all azacitidine group (n=150).

*Quartiles 1-3: ALT = 4-32 U/L, AST=8-31 U/L, Total bilirubin =3.4-17.7 µmol/L
*Quartile 4: ALT = 33-113 U/L, AST=32-112 U/L, Total bilirubin =18.8-59.9 µmol/L

Table 8. Baseline Liver Function Tests (ALT, AST, Total Bilirubin): TEAEs in the 9221 All Azacitidine Group With a Higher Frequency in Quartile 4 vs. Quartiles 1-3 (> 10.0% Difference)

		Number (%) of Subjects			
		Subcutaneous			
		9221 Observation (N=92)		9221 All Azacitidine[b] (N=150)	
Liver Function Test					
Preferred Term[a]		**Quartiles 1-3**	**Quartile 4**	**Quartiles 1-3**	**Quartile 4**
ALT[c]	**n**	**54**	**10**	**84**	**29**
At least 1 TEAE		53 (98.1%)	10 (100%)	84 (100%)	29 (100%)
Neutropenia		7 (13.0%)	1 (10.0%)	21 (25.0%)	14 (48.3%)
Ecchymosis		8 (14.8%)	0	23 (27.4%)	11 (37.9%)
			0		
Dizziness		5 (9.3%)	0	10 (11.9%)	9 (31.0%)
Febrile neutropenia		1 (1.9%)	0	9 (10.7%)	7 (24.1%)
Abdominal pain upper		2 (3.7%)	0	5 (6.0%)	7 (24.1%)
Lymphadenopathy		2 (3.7%)	0	4 (4.8%)	6 (20.7%)
Cellulitis		2 (3.7%)	1 (10.0%)	5 (6.0%)	5 (17.2%)
Pleural effusion		3 (5.6%)	0	4 (4.8%)	5 (17.2%)
Sinusitis NOS		3 (5.6%)	0	3 (3.6%)	4 (13.8%)
AST[d]	**n**	**54**	**14**	**88**	**31**
At least 1 TEAE		53 (98.1%)	14 (100%)	88 (100%)	31 (100%)
Neutropenia		7 (13.0%)	1 (7.1%)	26 (29.5%)	14 (45.2%)
Weight decreased		6 (11.1%)	2 (14.3%)	10 (11.4%)	8 (25.8%)
Dizziness		3 (5.6%)	2 (14.3%)	13 (14.8%)	8 (25.8%)
Lymphadenopathy		2 (3.7%)	0	5 (5.7%)	6 (19.4%)
Cardiac murmur NOS		7 (13.0%)	1 (7.1%)	5 (5.7%)	5 (16.1%)
Rash macular		1 (1.9%)	0	2 (2.3%)	4 (12.9%)
Total bilirubin[e]	**n**	**50**	**19**	**92**	**27**
At least 1 TEAE		50 (100%)	18 (94.7%)	92 (100%)	27 (100%)
Fatigue		18 (36.0%)	1 (5.3%)	33 (35.9%)	13 (48.1%)
Weakness		10 (20.0%)	5 (26.3%)	25 (27.2%)	11 (40.7%)
Anorexia		3 (6.0%)	0	17 (18.5%)	9 (33.3%)
Edema peripheral		3 (6.0%)	3 (15.8%)	11 (12.0%)	9 (33.3%)
Epistaxis		6 (12.0%)	1 (5.3%)	13 (14.1%)	7 (25.9%)
Cardiac murmur NOS		8 (16.0%)	0	3 (3.3%)	7 (25.9%)
Pneumonia NOS		2 (4.0%)	2 (10.5%)	4 (4.3%)	5 (18.5%)
Mouth hemorrhage		0	1 (5.3%)	1 (1.1%)	3 (11.1%)

[a] Multiple reports of the same preferred term for a subject are only counted once within each treatment group.
[b] Includes all subjects exposed to azacitidine, including 9221 subjects after crossing over from observation.
[c] ALT. Quartiles 1-3 = 4.0-32 0 U/L; Quartile 4 = 33.0-113.0 U/L.
[d] AST: Quartiles 1-3 = 8.0-31.0 U/L; Quartile 4 = 32.0-112.0 U/L.
[e] Total bilirubin: Quartiles 1-3 = 3.4-17.1 µmol/L; Quartile 4 =18.8-59.9 µmol/L.
Sorted by decreasing frequency in the 9221 all azacitidine Quartile 4 group.
Baseline was defined as the last lab value before the date of randomization or, if no value was available prior to randomization, the earliest value following randomization up to and including the day of first dose of study medication for azacitidine subjects, and on the day of randomization for observation subjects.
KEY: NOS=not otherwise specified, TEAE=treatment-emergent adverse event, ALT=alanine aminotransferase, AST=aspartate aminotransferase

23. Based on what is known about exposure-response relationships, what dosage regimen adjustments, if any, are recommended for each subgroup listed below?

Elderly Patients

The product label indicates that because azacitidine and its metabolites are primary excreted by the kidneys and because elderly patients are more likely to have decreased renal function, care should be taken in selecting azacitidine dose and the renal function should be monitored. This is acceptable from the clinical pharmacology perspectives.

Pediatric Patients

The product label indicates that safety and effectiveness of Vidaza™ have not been evaluated in pediatric patients.

Patients with Renal Impairment

The Applicant did not evaluate azacitidine pharmacokinetics in patients with renal impairment. Published data* indicate that patients taking azacitidine had their serum creatinine progressively rising from 1.2 mg/dl to 2.7 mg/dl and eventually died from sepsis.

The product label indicates that because of the renal toxicity of azacitidine, patients should be monitored and if there is an elevation in blood urea nitrogen or serum creatinine values, Vidaza™ dose should be held until these values return to normal or baseline and then reduced by 50% in the next treatment cycle. This is acceptable from the clinical pharmacology perspectives. However, the dosing recommendation that patients with renal impairment may be administered the starting dose of 75 mg/m^2 in the first cycle and this dose is reduced in subsequent cycle based on hematological and renal toxicities is not acceptable and was deleted from the label. Because azacitidine and its metabolites are primary excreted by the kidneys and because azacitidine causes renal toxicity, we recommend that the Applicant should conduct a study in patients with varying degrees of renal impairment to properly provide dosing recommendations in this patient population (see the Phase 4 Commitment). We added a statement under the [redacted] of the package insert for the use of Vidaza™ in patients with severe renal impairment (estimated creatinine clearance of < 30 ml/min).

*Greenberg MS, Reversible renal dysfunction due to azacitidine. Cancer Treat Rep 1979, 63:806.

Patients with Hepatic Impairment

The Applicant did not evaluate azacitidine pharmacokinetics in patients with hepatic impairment. In a study by Bellet et al (1972), four patients who had

abnormal total bilirubin and SGOT levels died from hepatic coma following azacitidine SC at doses of 0.8-2 mg/kg/day for 10 days (Table 9).

Table 9. Summary of patients died in hepatic coma

Patient	Sex	Age (Yr)	Tumor	Metastases	Dose mg/kg/d X10 days	#Bilirubin (mg/dl)	#SGOT (I.U.)	#Alkaline Phosphatase (Bod. U.)	#Albumin (g/dl)	Day of death
1	M	67	Colon	liver lung pleura	1.4	1.5/6.8	70/210	7.0/7.0	2.2	20
2	M	63	Melanoma	liver pleura skin	1.3	†0.5/1.5	16/168	1.9/8.0	2.8	20
3	F	61	Ovary	Liver skin	1.3	0.78/10.2	28/167	7.3/6.7	2.4	29
4	M	32	Melanoma	Liver Lung skin	1.6	†0.5/0.6	48/345	9.5/17.8	2.6	9

M=male, F=Female #(baseline value/post-treatment value) †patients clinically jaundiced

Liver biopsies of these 4 patients revealed evidence of significant hepatic tumor burden.

The product label indicates that due to hepatotoxicity, Vidaza™ should be contraindicated in patients with advanced malignant hepatic tumors and used with caution in patients with severe hepatic impairment. This is acceptable from the clinical pharmacology perspectives. However, the dosing recommendation

[redacted] is not acceptable and was deleted from the label. We added a statement under the [redacted] of the package insert for the use of Vidaza™ in patients with underlying hepatic cirrhosis.

*Ballet RE, Mastrangelo MJ, Engstrom RP, Hepatotoxicity of azacitidine (a clinical and pathologic study). Neoplasma 20:303-308.

24. What pregnancy and lactation use information is there in the application?

There are no clinical studies conducted in pregnant women with Vidaza™. The product label indicates because azacitidine presents a significant risk to the embryo, fetus, and male reproduction system, Vidaza™ should not be administered to pregnant women or women of childbearing potential who do not employ appropriate birth control practices.

D. Extrinsic factors

Drug-drug interactions

25. Is there any *in vitro* basis to suspect in vivo drug-drug interactions?

There are no reliable *in vitro* data to predict any *in vivo* drug-drug interactions.

26. Is azacitidine a substrate of CYP enzyme(s)?

Azacitidine undergoes deamination and CYP P450 enzymes are not expected to have an impact on the biotransformation of the drug.

27. Is azacitidine an inhibitor and/or an inducer of CYP enzymes?

Enzyme inhibition:

The Applicant conducted *in vitro* studies with human liver microsomes to investigate the inhibitory potential of azacitidine (0.1-100 µM) on the cytochrome P450 enzymes (CYP) 1A2 (7-ethoxyresorufin, 0.4 µM), 2C9 (tolbutamide, 100 µM), 2C19 (S-mephenytoin, 100 µM), 2D6 (bufuralol, 10 µM), 2E1 (chlorzoxazone, 40 µM), and 3A4 (testosterone, 65.2 µM) (Study DXNI1001). The positive controls used in the study were: furafulline, 10 µM (1A2), sulphaphenazole, 0 µM (2C9), Tranylcypromine, 50 µM (2C19), quinidine, 1 µM (2D6), diethyldithiocarbamate, 50 µM (2E1), and ketoconazole, 1 µM (3A4). The results are shown in Table 10.

Table 10. Mean % inhibition of CYP activity by azacitdine (duplicates or triplicates)

	Mean % Inhibition					
Inhibitor	**1A2**	**2C9**	**2C19**	**2D6**	**2E1**	**3A4**
Azacitidine (Conc., µM)						
0.1	-28.4	8.5	-23.4	14.3	-44.1	2.2
0.25	-29.1	7.2	-27	13.8	-30.4	-14.6
0.5	2.2	10.2	-34.5	17.2	-31.7	-51.1
0.7	ND	12.9	-17.5	ND	-22.4	-51.1
1.0	-24.3	ND	ND	21.5	-23.0	-48.0
2.5	-40.3	3.7	-26.5	23.9	-31.0	-50.2
5	-5.9	8.7	-27.6	9.76	-27.6	-25.7
7	-58.2	3.1	-27.5	20.7	-23.5	-23.4
10	-83.6	ND	ND	8.36	ND	ND
25	-25.4	0.39	-21.7	-4.7	-18.3	-18.6
50	-38.1	-0.29	-26.0	12.9	-13.7	3.9
70	0.0	-7.6	-9.2	-8.05	-3.8	-10.2
100	19.4	-9.05	-7.15	-20.9	27.1	-8.7
Positive Controls	53.7	80.4	51.8	80.8	75.0	70.8

ND=Not Determined

It appears that azacitidine may inhibit the activities of 1A2 and 2E1 at the 100 µM concentration; % inhibition was 19.4% and 27.1%, respectively. No obvious inhibition is noted at azacitidine concentrations close to its clinically relevant concentrations (mean C_{max} =3.1 µM after a single 75 mg/m^2 SC dose). In the

above results, some enzymatic activation is noted. In addition, data are highly variable with no trend of enzyme inhibition versus azacitidine concentrations. In a teleconference with the Applicant on 29-Mar-2004, no explanation was provided in regard of this high variability or enzymatic activation. The results of this study are inconclusive and are not acceptable from clinical pharmacology and biopharmaceutics perspectives. Therefore, we recommend that the Applicant should repeat this study (See Comments to the Applicant).

Enzyme induction:

In vitro studies with cultured human hepatocytes indicate that azacitidine (0.1, 10, and 100 µM) does not increase the activities of CYP 1A2 (7-ethoxyresorufin O-dealkylation), 2C19 (S-mephenytoin 4'-hydroxylation), or 3A4/5 (testosterone 6β-hydroxylation) (Study XT023018-3).

28. Is azacitidine a substrate and/or inhibitor of P-glycoprotein transporter processes?

The Applicant did not evaluate the effect of azacitidine on the efflux pump transporter, P-glycoprotein.

29. Are there other metabolic/transporter pathways that may be important?

No, current date are available indicating that other metabolic/transporter pathways may be important.

30. Does the product's label specify co-administration of another drug, and, if so, has the interaction potential between these drugs been evaluated?

None

31. What other co-medications are likely to be administered to the target patients population? What drug-drug interaction information is available for these comedications?

According to the Medical Reviewer of this NDA submission, during clinical studies (Studies 9221 and 8921), about 15% of patients in the all 8921/9221 azacitidine group concomitantly received CYP inducers (such as omeprazole, phenobarbital, rifampicin, sulfinpyrazone, sulphaphenzole, dexamethasone, isoniazid, ethanol, and St John's Wort). About 64% of patients in the all 8921/9221 azacitidine group concomitantly received CYP inhibitors (such as selective serotonin-reuptake inhibitors, fluoroquinolones, thiotepa, rifampicin, ketoconazole, omeprazole, quinidine, codeine, yohimbine, disulfiram, macrolide antibiotics, nefazodone, sertraline, and cimetidine). Other common concomitant medications received in the 8921/9221 all azacitidine group included analgesics

(73%), antibacterials (71%), antihistamines (58%), and psycholeptics (55%). No drug-drug interaction information is available for any of these comedications.

32. Are there any *in vivo* drug-drug interaction studies that indicate the exposure alone and/or exposure-response relationships are different when drugs are coadministered?

The Applicant did not conduct any *in vivo* drug-drug interactions studies for azacitidine.

33. Are there any medications that should be contraindicated in patients receiving azacitidine?

No, there are not.

34. Are there other drugs that may have a significant pharmacokinetic interaction when coadministered with azacitidine?

In vitro studies with human hepatocytes indicated that azacitidine is not an inducer of CYP enzymes. It is not known if azacitidine is an inhibitor of CYP enzymes; the *in vitro* inhibition studies conducted with human liver microsomes are inconclusive and should be repeated. The likelihood that azacitidine may inhibit the metabolism of other drugs that are substrates of CYP enzymes is not known.

35. Are there any unresolved questions related to metabolism, active metabolites, metabolic drug interactions or plasma protein binding?

The Applicant did not determine the plasma protein binding of azacitidine. Plasma Protein binding is not an issue as azacitidine has a large volume of distribution and may bind more to tissue proteins than to plasma proteins. There are unresolved issues related to metabolism, activity of the metabolite(s), and drug interactions. The following unresolved issues should be addressed (see Comments to the Applicant):

- The Applicant should adequately characterize the metabolism of azacitidine in MDS patients.
- The cytotoxic activity of any identified metabolite(s) should be determined. Plasma, urinary, and fecal levels of parent drug as well as identified metabolites should be measured.
- The specific CYP P450 that involved in azacitidine metabolism should be determined.
- The *in vitro* inhibition studies conducted with human liver microsomes are inconclusive and should be repeated.

36. What issues related to dose, dosing regimens or administration are unresolved, and represent significant omissions?

The Applicant did not provide any information regarding the selection of dose or dosing regimen in the submission. The selection of a 75 mg/m^2 SC dose once daily for 7 days in the treatment of MDS patients was based on a 30-year clinical experience with azacitidine in other cancer indications. This dose may be modified from 25-100 mg/m^2 depending on either patient's response or observed hematological toxicities experienced by the patient.

E. General Biopharmaceutics

37. Based on BCS principles, in what class is this drug and formulation? What solubility, permeability and dissolution data support this classification?

NOT APPLICABLE

38. What is the composition of the to-be-marketed formulation?

Azacitidine is supplied as a sterile lyophilized powder intended for reconstitution with sterile Water for Injection to form a suspension for subcutaneous (SC) injection. The drug product contains a 100 mg of azacitidine and 100 mg of mannitol in a 30-ml vial. Each vial is reconstituted with 4 mL of sterile Water for Injection prior to SC administration (25 mg/ml). It is recommended that the reconstituted azacitidine suspension should be stored for up to one hour at 25°C or for up to 8 hours between 2-8°C. The qualitative and quantitative formulation of the drug product proposed for marketing is described in Table 11.

Table 11. Qualitative/quantitative composition of azacitidine for Injectable Suspension (Applicant's)

Component	Reference to Quality Standard	Function	Amount
Azacitidine	In-house Standard	Drug Substance	—
Mannitol	USP / Ph.Eur.	—	100 mg
Water for Injection	USP / Ph.Eur.	—	4 mL
		Total Weight	—

39. What is the *in vivo* relationship of the proposed to-be-marketed formulation to the pivotal trial formulation in terms of comparative exposure?

The proposed commercial SC formulation was used in the pivotal Phase 3 study (Study 9221). In addition, the proposed commercial SC formulation was compared to the IV formulation used in early clinical studies in the bioavailability Study AZA-2002-BA-002. Study AZA-2002-BA-002 was an open-label,

randomized, two-treatment, two-period, crossover study in six MDS patients. Patients ranged in age from 57 to 83 years; there were three males and three females; all were Caucasians and nonsmokers. Each patient received the following two treatments on two different occasions separated by a 7-day washout period):

Treatment A: a single 75 mg/m^2 SC dose of azacitidine.

Treatment B: a single 75 mg/m^2 IV infusion dose of azacitidine administered over 10 minutes.

Results of this study are shown in Table 12.

Table 12. Geometric mean (%CV) exposure measures, point estimate, and 90% CI

PK parameter	Formulation (Treatment)	Geometric mean (n=6)*	% point estimate [90% CI]
C_{max} (ng/ml)	SC (A)	687.3	26.6 [19 – 37] %
	IV (B)	2580.3	
$AUC_{0-\infty}$ ng.h/ml)	SC (A)	896.4	88.6 [70 –112] %
	IV (B)	1011.2	

*The sample size was based on the recommendation from Food and Drug Administration (FDA) on 02-Oct-2002 to complete an azacitidine bioavailability study consisting of 6 to 10 patients.

The ratio of the SC geometric least-square (LS) mean to the IV geometric LS mean was 89% for log-transformed $AUC_{0-\infty}$, with a 90% CI of 70-112%, indicating that the bioavailability of azacitidine from the SC formulation and IV formulation is comparable. The label clearly indicates that Vidaza™ is marketed as SC Injectable Suspension. Although the IV formulation is approved in Europe, it will not used for marketing in USA.

40. What is the effect of food on the bioavailability of azacitidine and what dosing recommendations should be made regarding administration in relation to meals?

NOT APPLICABLE

41. Has the applicant developed an appropriate dissolution method and specification that will assure *in vivo* performance and quality of the product?

NOT APPLICABLE

F. ANALYTICAL SECTION

42. Were relevant metabolite concentrations measured in the clinical pharmacology and biopharmaceutics studies?

Azacitidine was the only drug-related species measured in the bioavailability study (Study AZA-2002-BA-002).

43. For all moieties measured, was free, bound, or total measured? What is the basis for that decision, and is it appropriate?

Protein binding has not been evaluated. Total azacitidine (free+bound) was the only species measured in plasma.

44. Were the analytical procedures used to determine azacitidine concentrations in this NDA acceptable?

Plasma samples from the bioavailability study (Study AZA-2002-BA-002) were analyzed for azacitidine using assay method with (Study QKAN-2002-0697-BIO). Azacitidine in whole blood samples was only stable for 30 minutes in ice bath. Due to this instability, plasma was separated and stored in an acetonitrile residue form ( ) at -70°C and then shipped to the analytical laboratory.

The method was adequately validated with respect to linearity, precision (%CV), accuracy, reproducibility, specificity, and sensitivity (LOQ) (see Table 13).

Table 13. Validation of LC/MS/MS analytical method for analysis of azacitidine in plasma Samples

LOQ* (ng/mL)	Linear Range (ng/mL)	Inter-batch precision of calibration curve (%CV)	Inter-batch accuracy of calibration curve (%)	Inter-batch precision of QCs (% CV)	Inter-batch accuracy for QC samples (%)	Intra-batch precision of QC samples (% CV)	Intra-batch accuracy of QC samples (%)
—	—	3.9-7.3%	98-102%	9.6-16.5%	90-103%	3.8-19%	86-118%

*LOQ Limit of quantitation QC quality control

This assay was partially validated to extend the quantitation range to — ng/ml.

LOQ* (ng/mL)	Linear Range (ng/mL)	Intra-batch accuracy of calibration curve (%)	Intra-batch precision of QC samples (% CV)	Intra-batch accuracy of QC samples (%)
—	—	94.5-105.2%	2.7-5.2%	86-95%

*LOQ Limit of quantitation QC quality control

Plasma samples with concentrations above the upper limit of the calibration curve (. — ng/ml) were diluted 1:10 to achieve concentrations within the linear range of — ng/ml.

Stability of azacitidine in acetonitrile/plasma samples was evaluated after — freeze-thaw cycle, at room temperature (for at least hours), and for a long-term. Azacitidine was stable for at least days after — freeze-thaw cycle, for hours at room temperature, and for 6 months at -70° C.

In conclusion, the Applicant has adequately documented the assay method and its validation.

IV. OCPB's LABELING RECOMMENDATIONS

[Note: Statements to be added are in red, italic and bold. Statements to be deleted are double-strikeout]

CLINICAL PHARMACOLOGY

Pharmacokinetics

4 pages redacted from this section of the approval package consisted of draft labeling

APPENDICES

18 pages redacted from this section of the approval package consisted of draft labeling

I. *Office of Clinical Pharmacology and Biopharmaceutics*

New Drug Application Filing and Review Form

General Information About the Submission

	Information		Information
NDA Number	**50-794**	**Brand Name**	**Vidaza™**
OCPB Division (I, II, III)	**DPE 1**	**Generic Name**	**Azacitidine**
Medical Division	**HFD-150**	**Drug Class**	**Antimetabolite**
OCPB Reviewer	**Sophia Abraham, Ph.D.**	**Indication(s)**	**Mutliple myeloma**
OCPB Team Leader	**Atiqur Rahman, Ph.D.**	**Dosage Form**	**100 mg Inj Suspension**
		Dosing Regimen	**75 mg/m^2 SC daily for 7 days every 28-day cycle**
Date of Submission	**26-Dec-2004**	**Route of Administration**	**SC**
Estimated Due Date of OCPB Review	**01-May-2004**	**Sponsor**	**Pharmion**
PDUFA Due Date	29-Jun-2004	Priority **Classification**	p
Division Due Date	**01-July-2004**		

Clin. Pharm. and Biopharm. Information

	"X" if included at filing	**Number of studies submitted**	**Number of studies reviewed**	**Critical Comments If any**
STUDY TYPE				
Table of Contents present and sufficient to locate reports, tables, data, etc.				
Tabular Listing of All Human Studies				
HPK Summary				
Labeling				
Reference Bioanalytical and Analytical Methods	**x**	[redacted]	[redacted]	
I. Clinical Pharmacology				
Mass balance:				
Isozyme characterization:	**x**	**1**	**1**	
Blood/plasma ratio:				
Plasma protein binding:				
Pharmacokinetics (e.g., Phase I) -				
Healthy Volunteers-				
single dose:				
multiple dose:				
II. Patients-				
single dose:				
multiple dose:				
Dose proportionality -				
fasting / non-fasting single dose:				
fasting / non-fasting multiple dose:				
Drug-drug interaction studies -				
In-vivo effects on primary drug:				
In-vivo effects of primary drug:				

In-vitro:	**x**	**2**	**2**
Subpopulation studies -			
ethnicity:			
gender:			
pediatrics:			
geriatrics:			
renal impairment:			
hepatic impairment:			
PD:			
Phase 2:			
Phase 3:			
PK/PD:			
Phase 1 and/or 2, proof of concept:			
Phase 3 clinical trial:			
Population Analyses -			
Data rich:			
Data sparse:			
II. Biopharmaceutics			
Absolute bioavailability:			
Relative bioavailability -	[redacted]	[redacted]	[redacted]
solution as reference:			
alternate formulation as reference:			
Bioequivalence studies -			
traditional design; single / multi dose:			
replicate design; single / multi dose:			
Food-drug interaction studies:			
Dissolution:			
(IVIVC):			
Bio-wavier request based on BCS			
BCS class			
III. Other CPB Studies			
Genotype/phenotype studies:			
Chronopharmacokinetics			
Pediatric development plan			
Literature References			
Total Number of Studies			

Filability and QBR comments				
	"X" if yes	**Comments**		
Application filable ?	x			
Comments sent to firm ?		We remind you of the post-approval Commitments regarding this application as outlined in the pre-NDA meetings on 19-Dec-2001.		
QBR questions (key issues to be considered)				
Other comments or information not included above				
Primary reviewer Signature and Date			**Sophia Abraham**	
Secondary reviewer Signature and Date				

CC: NDA 50-794, HFD-860 (Electronic Entry), HFD-150 (Baird), HFD-860 (Mehta, Sahajwella, Rahman, Abraham), CDR (Biopharm)

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

/s/

Sophia Abraham
4/30/04 04:37:39 PM
BIOPHARMACEUTICS

Atiqur Rahman
4/30/04 04:48:53 PM
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