1.7.3 Adjudicated Baseline Diagnosis of AML

According to the protocol, if the baseline diagnosis of AML was made either by the site or by the CALGB central hematopathologist, the diagnosis of AML was assigned to the subject. None of the subjects had a diagnosis of AML on entry. On review at the site, one subject was diagnosed as having AML (in the Observation arm). Adjudication by the central CALGB hemopathologist resulted in 18 additional subjects having AML. These 18 additional subjects are not identified in the CRFs as having AML.

Reviewer's Note: Of the 19 subjects diagnosed with AML by CALGB central pathologist, their initial diagnoses at the site were 13 RAEB, 3 RAEB-T, and 3 RA. On review at the site, the diagnoses changed to 11 RAEB, 7 RAEB-T and 1 AML. Some of these subjects had very long survivals, the longest being 1273 days after study entry (3.5 years). The median survival was 413 days and the mean survival was 535 days (range 38 to 1273 days). These data on survival are more characteristic of MDS than of AML. Causes of death in these subjects were complications of AML and bone marrow failure in 12, cardio-pulmonary arrest in 4, intracerebral hemorrhage in one, and subdural hematoma in one.

1.7.4 Populations Analyzed

- ITT population: all 191 subjects enrolled and randomized were analyzed for efficacy
- First subgroup: 19 patients with adjudicated AML were excluded
- Second subgroup: 76 subjects with major protocol violations plus 6 subjects with adjudicated AML were excluded
- Third subgroup: Because the Observation Group consisted of subjects who crossed over to the azacitidine arm and subjects who remained in observation only arm, FDA asked the sponsor to divide the groups as follows after excluding the adjudicated AML patients:
  - Group 1 - Azacitidine (N=89)
  - Group 2 - Observation Only (N=36)
  - Group 3 - Azacitidine after Observation, i.e. Crossover (N=47)
  - Group 4 - Azacitidine after Observation Who Met the Original Entry Criteria (N=41)
  - Groups 1 + 4 Combined – All Azacitidine Qualifiers (N=130)
- Safety population: all 191 subjects, since all subjects randomized to azacitidine received at least one dose of study medication

1.7.5 Demographic and Other Baseline Characteristics[1]

Demographic	Azacitidine N=99	Observation N=92	Azacitidine after Observation N=51	All Azacitidine N=150
Gender: Male (%)	72 (72.7)	60 (65.2)	31 (60.8)	103 (68.7)
Female (%)	27 (27.3)	32 (34.8)	20 (39.2)	47 (31.3)
Race: White (%)	93 (93.9)	85 (92.4)	47 (92.2)	140 (93.3)
Black (%)	1 (1)	1 (1)	1 (2)	2 (1.3)
Hispanic (%)	3 (3)	5 (5.4)	2 (3.9)	5 (3.3)
Asian (%)	2 (2)	1 (1)	1 (2)	3 (2)
Age: Mean±SD	67.3±10.4	68.0±10.2	67.0±9.3	67.2±10.0
<65 years (%)	36 (36.4)	33 (35.9)	17 (33.3)	53 (35.3)
65 – 74 years (%)	39 (39.4)	33(35.9)	22(43.1)	61 (40.7)
≥75 years (%)	24 (24.2)	25 (27.2)	12 (23.5)	36 (24.0)

[1]From Sponsor's Table 11.2-1.

Mean height was about 171±9.9 cm in all groups. Mean weight was about 78±16.5 cm in all groups. Mean BSA was about 1.90±0.23 m^2 for all groups.

Male preponderance in this study (69% males and 31% females) are characteristic for MDS, as is the older age of subjects. Preponderance of whites, in contrast to subjects of other races, in this study is not known to be a characteristic of MDS. The paucity of blacks, Hispanics and Asians/Orientals made analyses by race not reliable.

Subject Demographics at the Time of Randomization Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry, by FDA-Suggested Groups*

Demographic	Group 1 Azacitidine N=89	Group 2 Observation Only, N=36	Group 3 Azacitidine After Observation N=47	Group 4 Azacitidine After Observation Qualifiers N=41	Group 1 + 4 All Azacitidine Qualifiers (130)
Gender, M:F (%)	73%:27%	72%:28%	60%:40%	61%:39%	69%:31%
Race, % White	93%	92%	94%	95%	94%
Age, mean	67.2 years	69.7 years	67.1 years	67.0 years	67.1 years

*Data from Table 7.2-1, February 19, 2004 submission. Data on height, weight and BSA are not shown, but were closely similar in all groups

Reviewer's Table of MDS Subtypes As Diagnosed at Study Site[1] (ITT Population)

Site MDS type At study entry	Azacitidine N=99	Observation N=92	Azacitidine after Observation N=51	All Azacitidine N=150
RA	21 (21.2%)	18 (19.6%)	14 (27.5%)	35 (23.3%)
RARS	6 (6.1%)	5 (5.4%)	3 (5.9%)	9 (6%)
RAEB	42 (42.4%)	44 (47.8%)	25 (49%)	67 (44.7%)
RAEB-T	22 (22.2%)	17 (18.5%)	7 (13.7%)	29 (19.3%)
CMMoL	8 (8.1%)	7 (7.6%)	2 (3.9%)	10 (6.7%)
AML	0	1 (1.1%)	0	0

[1]From Sponsor's Table 11.2-2.

Reviewer's Table of MDS Subtypes as Adjudicated by the CALGB Central Laboratory[1]

Adjudicated MDS type at study entry	Azacitidine N=99	Observation N=92	Azacitidine after Observation N=51	All Azacitidine N=150
RA	21 (21.2%)	18 (19.6%)	14 (27.5%)	35 (23.3%)
RARS	6 (6.1%)	5 (5.4%)	3 (5.9%)	9 (6%)
RAEB	38 (38.4%)	39 (42.4%)	25 (45.1%)	61 (40.7%)
RAEB-T	16 (16.2%)	14 (15.2%)	5 (9.8%)	21 (14.0%)
CMMoL	8 (8.1%)	7 (7.6%)	2 (3.9%)	10 (6.7%)
AML	10 (10.1%)	9 (9.8%)	4 (7.8%)	14 (9.3%)

[1]From Sponsor's Table 11.2-2.

Reviewer's Table on Performance Status[1]

Performance Status (n,%)	Azacitidine N=99	Observation N=92	Azacitidine after Observation N=51	All Azacitidine N=150
0 Normal	35 (35.4)	26 (28.3)	13 (25.5)	48 (32.0)
1 Fatigue	34 (34.3)	39 (42.4)	24 (47.1)	58 (38.7)
2 Impaired	8 (8.1)	6 (6.5)	2 (3.9)	10 (6.7)
3 Bedrest	1 (1.0)	0	0	1 (0.7)
Unknown/Not Done	21 (21.2)	21 (22.8)	12 (23.5)	33 (22.0)

[1]From Sponsor's Table 11.2-2.

Reviewer's Table on Use of Transfusion Products During 3 Months Before Study Entry[1]

Product	Azacitidine N=99	Observation N=92	Azacitidine after Observation N=51	All Azacitidine N=150
Any Product	70 (70.7%)	59 (64.1%)	36 (70.6%)	106 (70.7%)
PRBC	66 (66.7%)	55 (59.8%)	34 (66.7%)	100 (66.7%)
Platelets	15 (15.2%)	12 (13%)	5 (9.8%)	20 (13.3%)
Plasma	1 (1%)	0	0	1 (0.7%)
Hetastarch	0	1 (1%)	1 (2%)	1 (0.7%)
Unknown	2 (2%)	2 (2%)	1 (2%)	3 (2%)

[1]From Sponsor's Table 11.2-2.

Reviewer's Comments on demographic and baseline disease data:

- *Subjects in the Azacitidine and Observation Groups were well matched by gender, race, age, height, weight, and BSA.*
- *Subjects in the Azacitidine and Observation Groups were well matched by MDS subtype.*
- *Cytogenetic analyses were not performed in the diagnostic work-ups.*
- *Performance status was unknown or not determined in about 20% of subjects in each group. About 70% of subjects in each group had performance status of 0 or 1 (normal or fatigue).*
- *Transfusion product use during the 3 months prior to the entry into the study was similar in the Azacitidine and the Observation Groups.*
- *Baseline bone marrow slides were reviewed locally for 190/191 (99%) of subjects, centrally for 188/191 (98%) of subjects, and by the blinded independent reviewer in 43/99 (43%) subjects in the Azacitidine Group and 45/92 (49%) subjects in the Observation Group. Adjudication by the central review resulted in the diagnosis of AML in 19 subjects, 10 in the Azacitidine Group and 9 in the Observation Group.*
- *Distribution of the MDS subtype based on both site diagnosis and adjudicated baseline diagnosis was similar for the Azacitidine and the Observation Groups, even though numbers of subjects in various categories changed.*
- *Medical conditions and previous surgeries were common in this study of mainly elderly subjects and there were no large differences between groups. Small numbers of subjects had prior chemotherapy (5% in the Azacitidine and 4% in the Observation Groups) and radiotherapy (10% in the Azacitidine and 5% in the Observation Groups).*
- *About 50% of subjects in each group had a variety of chest X-ray or ECG abnormalities.*
- *Concomitant medications were used more commonly by subjects in the Azacitidine Group than in the Observation Group: antiemetics and antinauseants (84% vs. 22%); analgesics (76% vs. 60%); systemic antibacterials ((76% vs. 59%); antihistamines (64% vs. 40%); psycholeptics (58% vs. 37%); and steroids [prohibited] (38% vs. 23%).*

1.7.6 Study Treatment and Treatment Compliance

All 99 subjects randomized to azacitidine received azacitidine for period as shown in Reviewer's Table below (data from sponsor's Table 4). In addition, 51 subjects who crossed over from the observation arm were treated with azacitidine. The remaining 41 subjects in the observation arm did not receive any doses of azacitidine.

Dose adjustments (increases and decreases) were specified in the protocol. One-half of all subjects treated with azacitidine received less than 75 mg/m^2/day, and one-half received 75 mg/m^2/day or more. There were 18 subjects who received 100 mg/m^2/day; no subjects received a higher dose.

Reviewer's Table. Dose and Duration of Exposure to Azacitidine

Average Azacitidine Dose	Azacitidine as Randomized N=99	Azacitidine After Observation N=51	All Azacitidine N=150
<75 mg/m^2/day	N=54 16.7±16.53 (SD) 28-day months	N=21 17.5±20.40 (SD) 28-day-months	N=75 16.9±17.56 (SD) 28-day months
≥75 mg/m^2/day	N=45 8.7±8.44 (SD) 28-day months	N=30 4.9±3.62 (SD) 28-day months	N=75 7.1±7.14 (SD) 28-day months

Azacitidine was administered SC either by the subject or the subject's caregiver. Dosing information, including total daily dose and the number of drug the drug administered each cycle was recorded on the CALGB Drug/Blood Sheet. With few exceptions, subjects received all 7 azacitidine doses during each treatment cycle.

1.7.7 Efficacy Analysis

1.7.7.1 Primary Endpoint: Overall Response Rate of CR + PR

Overall Response Rates: The overall response rate (CR + PR) and the best response rates (CR + PR + responses other than CR + PR) are shown in shown in Reviewer's Tables below (from sponsor's Table 11.4-1).

- Best responses rates were calculated for subjects randomized to azacitidine, and for subjects randomized to observation during the period before crossover. Then, best response rates are presented for the subset of observation subjects who did not cross over and for the subset of subjects who did cross over to azacitidine treatment. Finally, best response rates are presented for all subjects treated with azacitidine.
- The sponsor also analyzed response rates excluding subjects, who were adjudicated to have a diagnosis of AML at study entry. These data are presented in the Reviewer's Tables below. As noted above in Study Populations, there were 19 subjects who were adjudicated by the central CALGB laboratory to have had AML at study entry, 10 of them randomized to azacitidine treatment and 9, to observation. Their exclusion changes the results only slightly. One of the AML patients randomized to azacitidine had a CR and

one had a PR, the other 8 did not have a response. None of the 9 AML subjects in the observation group had a response.

- The overall response rate in subjects randomized to azacitidine was 16.2% (15.7%, when subjects subsequently diagnosed as having AML at study entry are excluded). This response rate was statistically significantly higher than the 0% response rate in the Observation Group ($p<0.0001$).
- None of the 6 subjects with CR in the azacitidine group had a major protocol violation; 3 of 10 subjects with PR had major protocol violations (single doses of IV or IM corticosteroids). The overall response rate in subjects randomized to azacitidine, excluding subjects with AML at study entry or with a major protocol violation, was 20.4%.
- CR of 6.1% (5.6% with the exclusion of the AML patient group, 9.3% with the exclusion of the AML patient group and of patients with major protocol violations) in the azacitidine group was also significantly higher than 0% in the observation group ($p=0.03$).
- The overall response rate in subjects who crossed over from the observation to azacitidine treatment was 11.8% (12.8% with the exclusion of patients with AML at study entry, 19.2% with exclusion of the AML patient group and of patients with major protocol violations), which was similar (given the small number of patients) to the overall response rate in the group that was randomized to azacitidine.
- The overall response rate in all the subjects treated with azacitidine was 14.7%.
- The overall response rate in all the subjects treated with azactidine, excluding subjects with adjudicated diagnosis of AML at study entry, was 14.7% (20/136).
- The overall response rate in all the subjects treated with azacitidine, excluding adjudicated AML at study entry subjects and subjects with major protocol violations, was 20.0% (16/80).

Reviewer's Table. Response to Azacitidine (ITT Population)*

Response	Azacitidine N=99	Observation Before Crossover N=92	Observation Without Crossover N=41	Azacitidine After Observation N=51	All Azacitidine N=150
CR + PR	16 (16.2%)	0	0	6 (11.8%)	22 (14.7%)
CR	6 (6.1%)	0	0	3 (5.9%)	9 (6.0%)
PR	10 (10.1%)	0	0	3 (5.9%)	13 (8.7%)

*Data from sponsor's Table 9, p. 40, ISE.

Reviewer's Table. Response to Azacitidine Excluding Subjects With Adjudicated Diagnosis of AML at Study Entry*

Response	Azacitidine N=89	Observation Before Crossover N=83	Observation Without Crossover N=36	Azacitidine After Observation N=47	All Azacitidine N=136
CR + PR	14 (15.7%)	0	0	6 (12.8%)	20 (14.7%)
CR	5 (5.6%)	0	0	3 (6.4%)	8 (5.9%)

PR	**9 (10.1%)**	**0**	**0**	**3 (6.4%)**	**12 (8.8%)**

*Data from sponsor's Table 13, p. 49, ISE.

Reviewer's Table. Response to Azacitidine Excluding Subjects With Adjudicated Diagnosis of AML at Study Entry or With a Major Protocol Violation*

Response	Azacitidine N=54	Observation Before Crossover N=48	Observation Without Crossover N=22	Azacitidine After Observation N=26	All Azacitidine N=80
CR + PR	11 (20.4%)	0	0	5 (19.2%)	16 (20.0%)
CR	**5 (9.3%)**	**0**	**0**	**3 (11.5%)**	**8 (10.0%)**
PR	**6 (11.1%)**	**0**	**0**	**2 (7.7%)**	**8 (10.0%)**

*Data from sponsor's Table 14, p. 50, ISE.

- The overall response rate in the groups of subjects requested by FDA is only slightly different (Reviewer's Table shown below). Six of the subjects in the azacitidine after crossover from observation group did not meet study entry criteria. Among these 6 subjects were 2 PR responders. Therefore, the azacitidine after observation group had a 9.8% response rate, and all azacitidine-treated subjects had a 13.8% response rate.

Reviewer's Table. Response to Azacitidine by FDA-Requested Groups*

Response	Group 1 N=89	Group 2 N=36	Group 3 N=47	Group 4 N=41	Groups 1 + 4 N=130
CR + PR	**14 (15.7%)**	**0**	**5 (10.6%)**	**4 (9.8%)**	**18 (13.8%)**
CR	**5 (5.6%)**	**0**	**4 (10.6%)**	**3 (7.3%)**	**8 (6.2%)**
PR	**9 (10.1%)**	**0**	**1 (2.1%)**	**1 (2.4%)**	**10 (7.7%)**

***Groups: 1 – Azacitidine, 2 – Observation only, 3 – Azacitidine after Observation, 4 – Azacitidine after Observation Who Qualified for Entry Criteria, 1 + 4 – All Azacitidine Qualifiers.**

Reviewer's Comments:

Group 2 subjects were comparable to azacitidine groups in gender, race, age and performance status. However, Group 2 was not comparable to Group 1 by MDS subtype. Group 2 had a lower proportion of subjects with RA and a higher proportion of subjects with RAEB-T and CMMoL than the other groups, as shown below in sponsor's Table 7.2-2. Since, generally, RAEB-T and CMMoL patients have shorter survivals than RA patients, Group 2 may not be an appropriate control for the azacitidine groups. The Reviewer explored this question by determining survival data by adjudicated MDS subtype in this trial and then comparing the survival data in Group 2 patients to the survival data for all subjects, irrespective of treatment.

Table 7.2-2: Baseline Disease Characteristics Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

	Number (%) of Subjects				
Baseline Disease Characteristic	Group 1 Azacitidine (N=89)	Group 2 Observation Only (N=36)	Group 3 Azacitidine After Observation (N=47)	Group 4 Azacitidine After Observation Qualifiers (N=41)	Group 1 + 4 All Azacitidine Qualifiers (N=130)
Site MDS diagnosis at study entry (Group 1 and 2) or at crossover (Group 3 and 4)					
RA	21 (23 60)	4 (11.11)	14 (29 79)	13 (31 71)	34 (26.15)
RARS	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
RAEB	38 (42.70)	16 (44 44)	20 (42 55)	19 (46 34)	57 (43 85)
RAEB-T	16 (17 98)	9 (25 00)	7 (14 89)	5 (12 20)	21 (16 15)
CMMoL	8 (8 99)	5 (13 89)	2 (4 26)	2 (4 88)	10 (7 69)
AML	0 (0 00)	0 (0 00)	2 (4 26)	0 (0 00)	0 (0 00)
Performance Status[a]					
0 Normal	30 (33 71)	10 (27 78)	13 (27.66)	11 (26 83)	41 (31 54)
1 Fatigue	32 (35 96)	13 (36 11)	21 (44 68)	20 (48 78)	52 (40.00)
2 Impaired	6 (6 74)	4 (11 11)	2 (4 26)	1 (2 44)	7 (5 38)
3 Bedrest	1 (1 12)	0 (0 00)	0 (0 00)	0 (0 00)	1 (0 77)
Unknown/Not Done	20 (22 47)	9 (25 00)	11 (23.40)	9 (21 95)	29 (22 31)

[a] Reported at time of randomization.

Reviewer's Table. Survival Data for All Subjects in CALGB 9221 Trial, by MDS Subtype

MDS Subtype and AML	Number of Subjects, N=191	Median Survival (days)	Mean Survival and Range (days)
RA	39	833	1,016 (17 to 2,751)
RARS	11	812	913 (270 to 1,719)
RAEB	77	531	723 (47 to 2,820)
RAEB-T	30	389	509 (68 to 2,319)
CMMoL	15	375	603 (70 to 2,518)
AML	19	413	535 (38 to 1,273)

The longest median and mean survivals were in RA patients and the shortest were in RAEB-T and CMMoL patients (as well as in AML patients). However, there was enormous spread within each subtype and among AML patients. The shortest survival was 17 days in a RA patient and the longest almost 8 years in a RAEB patient.

The survival data for some patients diagnosed as having AML, rather than MDS, by the central CALGB laboratory is more typical for MDS than for AML. The longest survivor among this group of patients was a 44-year old woman, who died 3.5 years after entry into the study. She was treated with azacitidine but was a non-responder.

Survival data for Observation Only patients (Group 2) by MDS type is shown in Reviewer's Table below. As compared to the data for the entire trial population

above, median and mean survivals were markedly shorter among Group 2 RA, RAEB, and RAEB-T (but not by CMMoL) patients. These findings suggest that Group 2 is not negatively impacted by greater percentages of RAEB-T and CMMoL patients and lower percentages of RA patients. Rather, factors other than MDS subtype affected Observation Only group survival and Group 2 patients appear not to be appropriate controls for Group 1 patients in survival analysis.

Reviewer's Table. Survival Data for Observation Only Patients by MDS Subtype

MDS Subtype	Number of Subjects, N=36	Median Survival (days)	Mean Survival and Range (days)
RA	**4**	**111**	**278 (17 to 873)**
RARS	**2**	**805**	**805 (370, 1240)**
RAEB	16	214	435 (63 to 2,677)
RAEB-T	**9**	**257**	**304 (75 to 647)**
CMMoL	**5**	**375**	**760 (74 to 2,518)**

Primary Endpoint: Non-Responses (Improvement, Stable Disease, Relapse, and Disease Progression)

- There were notable differences among treatment groups in non-responding subjects.
  - About 33% of subjects treated with azacitidine (both in randomized to azacitidine subjects and in azacitidine after observation subjects) were classified as having Improvement as compared to about 12.2%-19.6% in the observation without crossover subjects. These results are important, as some of the subjects lost their need for transfusions or increased their blood cell counts. This issue is discussed below.
  - Approximately similar proportions of azacitidine-treated and observation subjects had Stable Disease.
  - Disease Progression occurred in a larger proportion of observation subjects (before crossover and without crossover) than in azacitidine-treated subjects (10.9%-19.5% vs. 2.0%-3.9%).
  - A larger proportion of observation subjects were unevaluable as compared to azacitidine-treated subjects (14.1%-29.3% vs. 8.1%-10.7%).
  - These differences remain approximately the same after adjustments for AML patients and after reclassifying patient groups as suggested by the Agency, as shown by Reviewer's Tables below. Group 2 (Observation Only) had a far lesser proportion of patients with Improvement and a greater proportion with Disease Progression than the azacitidine-treated patient groups.

Reviewer's Table. Non-Response (No CR/PR) to Azacitidine (ITT Population)

	Azacitidine N=99	Observation Before Crossover N=92	Observation Without Crossover N=41	Azacitidine After Observation N=51	All Azacitidine N=150

Improvement, but not CR or PR	33 (33.3%)	18 (19.6%)	5 (12.2%)	17 (33.3%)	50 (33.3%)
Stable disease	40 (40.4%)	51 (55.4%)	16 (39.0%)	17 (33.3%)	57 (38.0%)
Relapse	0	0	0	1 (2.0%)	1 (0.7%)
Disease progression	2 (2.0%)	10 (10.9%)	8 (19.5%)	2 (3.9%)	4 (2.7%)
Unevaluable	8 (8.1%)	13 (14.1%)	12 (29.3%)	8 (15.7%)	16 (10.7%)
Total non-response	83 (83.8%)	92 (100%)	41 (100%)	45 (88.2%)	128 (85.3%)

Reviewer's Table. Non-Response to Azacitidine Excluding Subjects With Adjudicated Diagnosis of AML at Study Entry

	Azacitidine N=89	Observation Before Crossover N=83	Observation Without Crossover N=36	Azacitidine After Observation N=47	All Azacitidine N=136
Improvement, not CR or PR	31 (34.8%)	18 (21.7%)	5 (13.9%)	16 (34.0%)	47 (34.6%)
Stable disease	36 (40.4%)	48 (57.8%)	15 (41.7%)	15 (31.9%)	51 (37.5%)
Relapse	0	0	0	1 (2.0%)	1 (0.7%)
Disease progression	1 (1.1%)	7 (8.4%)	6 (16.7%)	2 (4.3%)	3 (2.2%)
Unevaluable	7 (7.9%)	10 (12.0%)	10 (27.8%)	7 (14.9%)	14 (10.3%)
Total non-response	75 (84.3%)	83 (100%)	36 (100%)	41 (87.2%)	116 (85.3%)

Reviewer's Table. Non-Response Azacitidine by FDA-Requested Groups*

Response	Group 1 N=89	Group 2 N=36	Group 3 N=47	Group 4 N=41	Groups 1 + 4, N=130
Improvement, not CR or PR	31 (34.8%)	5 (13.9%)	18 (38.3%)	17 (41.5%)	48 (36.9%)
Stable Disease	36 (40.4%)	15 (41.7%)	18 (38.3%)	16 (39.0%)	52 (40.0%)
Relapse	0	0	0	0	0
Disease Progression	1 (1.1%)	6 (16.7%)	0	0	1 (0.8%)
Unevaluable	7 (7.9%)	10 (27.8%)	6 (12.8%)	4 (9.8%)	11 (8.5%)
Total non-response	75 (84.3%)	36 (100%)	42 (89.4%)	37 (90.2%)	112 (86.2%)

*Groups: 1 – Azacitidine, 2 – Observation only, 3 – Azacitidine after Observation, 4 – Azacitidine after Observation Who Qualified for Entry Criteria, 1 + 4 – All Azacitidine Qualifiers.

Subset analyses by gender and age:

- The male to female ratio of the 16 azacitidine responders was 3:1, similar to the ratio in the overall azacitidine group, suggesting that the response was not affected by gender.
- Younger subjects appeared to respond less well than older subjects in this study. Only 5.6% (2/36) of subjects under age 65 responded, compared to 22.2% (14/63) of subjects 65 years or older. A subset of the latter subjects, those 75 years or older, responded similarly (16.7%, 4/24) to subjects between 65 and 74 years of age.

Characteristics of response:

- Among the subjects randomized to azacitidine, the initial positive effect (sponsor's term, not defined in the CALGB protocol), which is defined as the first day of achievement of target for ≥4 weeks for at least 1 abnormality at baseline, was seen as early as Day 29 (i.e. after 1 cycle). Three-quarters (12/16) of the responders showed an effect within 1 to 4 cycles of treatment; of the remaining 4 responders, 3 showed a response after 5 to 6 cycles and one showed the initial positive effect after 17 cycles (Day 477).
- In the crossover patients the onset of the initial positive effect occurred as early as after the 1st cycle of treatment (Day 31) to the 7th cycle (Day 197).
- About 80% of subjects experienced the initial positive effect preceding the achievement of CR or PR within 4 cycles of treatment with azacytidine. The remaining responses occurred over an extended priod of time.
- Reviewer's Table below summarizes the onset of the beginning of responses for all azacitidine-treated subjects (data from Sponsor's Tables 11.4-2 and 11.4-3).

Reviewer's Table. Day of the Initial Positive Effect and Cycle, Type of Response, Cumulative Percent of Response in All Azacitidine-treated Subjects (ITT Population)

Cycle #	Day of the Initial Positive Effect and Type of Response	# of subjects	Cumulative %
1 (28 days)	29, 31*, 33* (CR), 36 (CR)	5	23%
2 (56 days)	55 (CR), 57 (CR), 57, 57, 58 (CR), 63, 68*	7	55%
3 (84 days)	84	1	59%
4 (112 days)	113 (CR), 113* (CR), 114 (CR), 114*, 125	5	82%
5 (140 days)	141	1	86%
6 (168 days)	164	1	91%
7 (196 days)	197* (CR)	1	95%
17 (476 days)	477	1	100%

Patients had a PR unless noted to have had a CR.
***Crossover patients.**

- According to protocol, patients who achieved a CR were supposed to be withdrawn from the study after 3 further cycles. There were 2 such patients, the others received azacitidine until withdrawn from study for other reasons (see Patient Disposition). The number of cycles administered to the above patients with CR or PR is shown in the Reviewer's Table below.

Number of Treatment Cycles and On Study Days for PR and CR Subjects (ITT Population)

Number of Treatment Cycles (# of subjects)	Range of Days on Study	Percent of All CR + PR Subjects/Cumulative %
7 – 10 (mean 9.0), 7 subjects (4 CR, 3 PR)	182 – 331 (mean, 265)	32%
11 – 19 (mean 14.0), 10 subjects (3 CR, 7 PR)	313 – 553 (mean, 432)	45%/77%
22-28 (mean 24.3), 3 subjects (1 CR, 2 PR)	623 – 827 (mean, 727)	14%/91%
68, 1 subject (1 CR)	1932	4.5%/95.5%
89, 1 subject (1 PR)	2581	4.5%/100%

- All subtypes of MDS responded to azaciditine, even though the numbers of responders were too small to draw reliable conclusions. The denominators indicate the number of subjects in each group. The following data are shown in abbreviated form from Sponsor's Table 11.4-4.

Reviewer's Table. Response Rates of All Subjects by MDS Subtype at Baseline

MDS Subtype	Azacitidine N=99	Azacitidine Crossover N=51	All Azacitidine N=150	% CR + PR Response, All Azacitidine
RA	5=2 CR+3 PR/ (N=21)	2=1 CR+1 PR/ (N=14)	7=3 CR+4 PR/ (N=35)	20.0%
RARS	1 PR/ (N=6)	0/ (N=2)	1 PR/ (N=8)	12.5%
RAEB	5=2 CR+3 PR/ (N=38)	3=2 CR+1 PR/ (N=20)	8=4 CR+4 PR/ (N=58)	13.8%
RAEB-T	2 PR/ (N=16)	0/ (N=7)	2 PR/ (N=23)	8.7%
CMMoL	1 CR/ (N=8)	1 PR/ (N=2)	2=1 CR+1PR/ (N=10)	20.0%
AML	1 CR + 1 PR/ (N=10)	0/ (N=6)	2=1 CR+1 PR/ (N=16)	12.5%

- The first initial positive effect of azacitidine therapy was on decreasing the peripheral blood or marrow blasts in 7 subjects (5 with RAEB and 2 with RAEB-T), on increasing platelet counts in 6, on increasing Hgb and/or decreasing transfusion requirements in 6, and on increasing WBC in 3.

- The median duration of response (PR or better) for the 22 responders was 166 days (5.5 months); the range was from 15 days to 2011 days. Reviewer's Table

below graphically illustrates the range of response durations for CRs and PRs (data from Sponsor's Tables 11.4-2 and 11.4-3). However, failure of responses (PR or CR) did not indicate complete loss of beneficial effects. The duration of a positive effect (defined as the number of days from the first day of achievement of target for at least one baseline abnormality until intervention [transfusion], relapse [blasts >30%], or last observed value on study, whichever occured first) was much longer than the duration of PR or CR. This is evident in the Reviewer's Table below. The subject with the shortest PR (15 days) still had a benefit for at least 329 days.

- MDS type is shown for each response. There does not appear to be a relationship between MDS types and duration of responses, although the numbers of each type, especially of RARS, CMMoL and RAEB-T, are too small for a statistical analysis. The durations in the Table are grouped by hundred day-intervals.

Reviewer's Table. Duration of Clinical Response, Duration of Positive Effect, and MDS Type

Duration of CR or PR (days)	Duration of Positive Effect (days)	MDS Type
15 (PR)	329+	RAEB-T
43 (PR)	275+	RAEB-T
43+ (PR)	99+	RA
65 (PR)	281	RAEB-T
78 (PR)	771	RA
78+ (PR)	190+	RAEB
88 (PR)	432	RAEB
92+ (CR)	105+	RAEB
141 (PR)	274+	RA
163+ (CR)	177+	RAEB
163+ (CR)	283+	RAEB
168+ (CR)	285+	RAEB
170+ (CR)	246+	CMMoL
190 (PR)	252	RAEB
231+ (PR)	329+	CMMoL
279+ (PR)	351+	RAEB-T
280+ (PR)	323+	RAEB
332	451	RARS
449+ (CR)	449+	RA
450+ (CR)	483+	RA
1029 (CR)	1707	RA
2011 (PR)	2530+	RAEB

Reviewer's Comments:
Review of PATIENT PROFILES by the Reviewer yielded different results from the sponsor's results:

- *Duration of responses could be only estimated as minimum duration, since 75% of the patients were still in response status at the time of withdrawal from the study. The median duration was >330 days, the mean duration was >512 days, the range of duration was from >110 days to 2400 days.*

Crossovers from Observation to Azacitidine: The 6 responders to azacitidine after crossing over from the Observation Group all had Study Chair approval to cross over for the following reasons:

- need for RBC transfusions (3 subjects: 59306, 60366, and 61455);
- platelet count <20,000/mm^3 in Weeks 7 and 8 (Subject 58246);
- bone marrow blasts increasing from <5% to ≥15% on 2 aspirates (Subject 58246); and
- decrease in Hgb to ≤80 g/L on two consecutive values at least 2 weeks apart (Subject 61285).

Protocol Violations in Responders: None of the 6 CR responders in the azacitidine group had a major protocol violation; 3 of the 10 subjects with PR had a major protocol violation (one had a single hydrocortisone injection for shoulder tendonitis; one had one and the other two doses of IV hydrocortisone as pre-treatment before transfusions).

1.7.7.2 Evidence of Clinical Benefit (Improvement)

Reviewer's Note: This section was a primary efficacy endpoint in the CALGB 9221 study, but was removed from the primary endpoints by the sponsor. However, it is still described under the primary rather than under secondary endpoints in the NDA.

Subjects who did not meet the criteria for PR were assessed as Improvement if, for at least 4 weeks, at least one peripheral blood cell line showed a ≥50% restoration toward normal values, or if there was a ≥50% decrease in RBC or platelet transfusion requirement.

There were 33 subjects of the 99 randomized to the azacitidine group and 5 subjects of the 41 who remained in the Observation without crossover group that demonstrated Improvement. The bases for the Improvement are shown in the Reviewer's Table below (data summarized from Sponsor's Table 11.4-7).

Reviewer's Table. Evidence of Clinical Benefit in Subjects with Improvement

Peripheral blood abnormality	Abnormal at Baseline All Azacitidine N=38	Improvement All Azacitidine	Abnormal at Baseline Observation N=5	Improvement Observation
Anemia	32/33 (97%)	19/38 (59%)	5/5 (100%)	0/5 (0%)
Thrombocyto-penia	28/33 (85%)	22/28 (79%)	5/5 (100%)	4/5 (80%)
Neutropenia	24/33 (73%)	18/24 (75%)	3/5 (60%)	1/3 (33%)

In the azacitidine group, of the 33 patients who improved, 32 were anemic, 28 were thrombocytopenic, 24 were neutropenic, and 24 were RBC or platelet or both RBC and platelet transfusion-dependent at baseline.

- The improvement of anemia occurred in 19 of 32 anemic subjects, who became transfusion-free for 73 to 505 days. Eleven had ≥50% increase in Hgb, 8 had ≤50% increase.
- The improvement of thrombocytopenia occurred in 22 of 28 thrombocytopenic subjects, who became platelet transfusion-free for 57 to 1548 days. Twelve had ≥50% increase in platelet counts, ten had ≤50% increase.
- The improvement of neutropenia occurred in 18 subjects. Five subjects had ≥50% increase in neutrophils, 13 had ≤50% increase. (None of the subjects in the study had WBC transfusions).

There were only 5 subjects in the Observation group who met the criteria for Improvement.

- None of the 5 subjects with anemia showed improvement.
- Two of the 5 thrombocytopenic subjects had ≥50% increase in platelet counts, two had ≤50% increase.
- One of 3 neutropenic subjects had an increase in neutrophils (≤50%).

The patients with the best Improvement response had 2 or 3 cell line abnormalities at baseline and were RBC or platelet transfusion-dependent (24/33 in the azacitidine group; 4/5 in the observation group). The loss of transfusion-dependence was long-lasting in the 24 patients azacitidine group (range 38 to 970 days) compared to 0/4 subjects in the observation group.

Reviewer's Note:

- *Review of the PATIENT PROFILES by the reviewer showed that 30 patients (20.0% of 150) in the azacitidine-treated group met the criteria for Improvement. Patients who had and patients who had not experienced Improvement are listed in the Appendix. The median duration of Improvement was 200 days, the mean duration was 340 days. The range was 45 days to 1300 days. Only 3 patients continued in Improved status at the time of withdrawal from the study; 26 relapsed to pre-Improvement status.*
- *None of the patients in the observation group met the criteria for Improvement. The 5 subjects that were identified by the sponsor as showing Improvement maintained Improved status for only short periods of time (one was diagnosed with AML 11 days later, one died from respiratory failure 8 days later, one was diagnosed with myelofibrosis 52 days later, one was diagnosed with colon cancer 63 days later, and one withdrew from the study 65 days later and died 129 after withdrawal).*

Observation subjects who had a best response of Stable Disease or Disease Progression generally remained in the study for less than 6 months. Fifteen of these 24 subjects developed AML or progression of MDS, another 5 were

withdrawn due to death, adverse events, to seek alternative therapy, or did not wish to continue. Only one subject remained in the study until the last contact.

1.7.7.3 Secondary Endpoints

The following endpoints are summarized in this section:

- Time to events (death, disease progression, relapse, transformation to AML, and death or transformation to AML)
- RBC transfusion rates
- Platelet transfusion rates
- Antibiotic therapy rates
- Hemorrhage rates
- Hematologic changes, and
- Bone marrow analyses.

The times to events are summarized from the time of study entry to last follow-up. Subjects in the study were followed after withdrawal from the study until malignancy or death. Follow-up was available for 163 subjects for as long as 6 years after withdrawal from the study.

The sponsor also notes that the number of subjects in the Observation Group dropped considerably between Months 5 and 6 and became very small by Month 8 due to protocol design that allowed these subjects to receive azacitidine if specified criteria were met. Therefore, trends after that time should be interpreted with caution.

Reviewer's Note: The sponsor summarized rates of RBC transfusions, platelet transfusions, antibiotic therapy and hemorrhages by month for the group randomized to azacitidine (N=99) and for group randomized to observation (N=92). The data for the group randomized to azacitidine is further subdivided into data for responders (CR + PR) and data for non-responders. The data for the observation group is not further subdivided into data for a subgroup that crossed over to azacitidine treatment and data for a subgroup that did not cross over. The reviewer sees several problems in these by month summaries:

- *The above noted crossover of a portion of the observation group to azacitidine treatment, which is still included as control for the azacitidine group. Hence, there are responses in the observation group (N=92) that are due to responses among the crossover patients.*
- *The deletion of the Improvement group from responders and including this group among non-responders. These constituted one-third of the azacitidine group (33/99) and most of them became RBC and platelet transfusion-free and increased their WBC. For example, 35 subjects (35.4%) out of the original azacitidine group (99) became RBC transfusion free, 16 of them as a result of achieving CR or PR and 19 of them as a result of Improvement. The non-responders had 0% rate of becoming RBC transfusion-free. However, due to*

deletion of Improvement group from responders, the non-responder group will have been 22.9% (19/83) transfusion-free.

- *Analysis by the month from the start of enrollment does not take into account the time of onset of responses, which were early in some responders and late in others (see Reviewer's table above).*
- *For all of the above reasons, the statistical analyses were both difficult to interpret and failed to reach statistical significance. The sponsor then submitted additional analyses according to suggestions by FDA as to how the groups should be divided.*

1.7.7.3.1 Secondary Endpoint: Time to Death from Any Cause

Sponsor's Table 11.4-8 presents an analysis of time to death from any cause for the group randomized to azacitidine, the group randomized to observation, and the sub-group of azacitidine treatment after observation. This table is reproduced below, as well as a Kaplan-Meier plot of Time to Death.

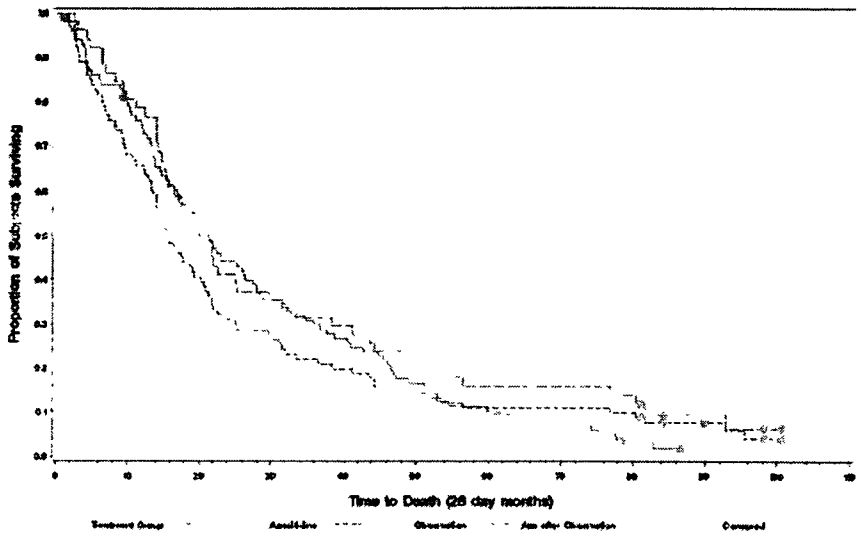
Table 11.4-8: Analysis of Time to Death of All Subjects From Any Cause

Treatment	Total N	Deaths n (%)	Censored[a] n (%)	Median Survival Time[b] (months)	95% CI on Median	Log-Rank Test[c] P-value
Azacitidine	99	95 (96 0)	4 (4 0)	20 1	(16 9 - 26 4)	0 6064
Responder	16	16 (100 0)	0 (0 0)	27 7	(21 9 - 32.9)	
Non-Responder	83	79 (95.2)	4 (4 8)	17 6	(13 6 - 22 9)	
Observation	92	86 (93.5)	6 (6 5)	15.4	(13 4 - 20 1)	
Azacitidine After Observation	51	47 (92.2)	4 (7 8)	21 3	(15 2 - 29 8)	
Responder	6	4 (66 7)	2 (33.3)	64 5	(43 6 - DNE)	
Non-Responder	45	43 (95.6)	2 (4 4)	18.6	(14 5 - 22.0)	

[a] Subjects who were alive were censored at date of last contact.
[b] The median, when it exists, provides a descriptive measure of the difference between groups
[c] Log-Rank test compares whether the azacitidine and observation groups follow the same survival curve
DNE = does not exist (due to small number of events).
Cross-reference. Table 14.2.7.1.1

The median survival was 20.1 months for the azacitidine group and 15.4 months for the observation group (including the crossover to azacitidine subjects); the difference was not statistically significant (p=0.61). The number of deaths and median survival is also given for Azacitidine after Observation Group (but not for subjects who remained in the Observation Only Group). Kaplan-Meier plots, shown in sponsor's Figure 11.4-1 for these populations showed three curves without significant differences (p=0.6064 by log-rank test for azacitidine and observation groups).

Figure 2: Kaplan-Meier Plot of Time to Death Comparing Azacitidine, Observation, and Azacitidine After Observation (CALGB 9221)



KEY CALGB=Cancer and Leukemia Group B
Data Source CALGB 9221 CSR Figure 14.2.7.6

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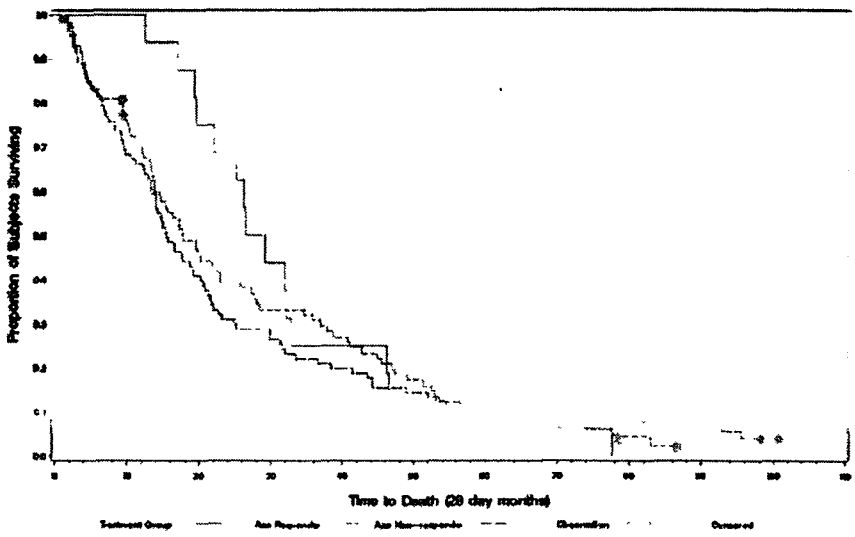
Figure 3: Kaplan-Meier Plot of Time to Death Comparing Azacitidine Responders and Non-responders, and Observation (CALGB 9221)



KEY: CALGB=Cancer and Leukemia Group B
Data Source: CALGB 9221 CSR Figure 14 2 7 5

The Kaplan-Meier plot in sponsor's Figure 11.4-2 shows time to death for azacitidine responders, azacitidine non-responders and the observation group. These plots show the difference between 50% survival times in the azacitidine responders and non-responders are reflected in the table above.

Time to death from any cause for all subjects by MDS subtype is presented by Reviewer's Table below (data from sponsor's Table 11.4-9). Median survival times (with CIs) in azacitidine group (further divided into responders and non-responders) and in the observation group are shown. In the reviewer's table below the median survival times (all within confidence intervals) are shown for the azacitidine and observation groups. The azacitidine responders' median survival times are presented in a separate table below.

Reviewer's Table. Time to Death from Any Cause of All Subjects by MDS Subtype

MDS Subtype	Treatment	Deaths/Total number of subjects*	Median Survival Time (months)
RA	Azacitidine	19/21	38.8 (17.1-52.9)
	Observation	14/18	22.0 (14.5-80.5)
RARS	Azacitidine	5/6	28.2 (14.5-47.3)
	Observation	5/5	44.3 (9.7-53.5)
RAEB	Azacitidine	37/38	21.8 (16.3-31.9)
	Observation	38/39	15.6 (10.5-20.9)
RAEB-T	Azacitidine	16/16	15.5 (10.2-20.1)
	Observation	14/14	11.1 (5.8-23.1)
CMMoL	Azacitidine	8/8	11.5 (4.0-27.8)
	Observation	6/7	13.4 (9.2-51.9)
AML	Azacitidine	10/10	25.5 (4.4-37.7)
	Observation	9/9	12.4 (6.5-25.5)

*Total number of subjects = number of deaths + number censored.

Although the numbers are small for some categories, the median survival times show large differences between MDS subtypes, from over 30 months for RA and RARS to about 13 for RAEB-T and CMMoL. With the exception of RARS and CMMoL subjects, azacitidine-treated subjects had longer survival times than subjects in the observation group. Azacitidine responders in all MDS categories had longer median survival times than non-responders as shown in Reviewer's table below (from sponsor's Table 11.4-9). *Reviewer's Note: Responder vs. non-responder analyses contain guarantee-time bias [the length of time to achieve a response] that favors responders [Anderson JR, Cain KC & Gelber RD. JCO 1983; 1:710-9].*

Reviewer's Table. Time to Death from Any Cause of Azacitidine Responders and Non-Responders by MDS Subtype

MDS Subtype	Treatment	Deaths/Total number of subjects*	Median Survival Time (months)
RA	Responder	5/5	46.2 (12.5-77.6)
	Non-responder	14/16	34.7 (13.4-51.3)
RARS	Responder	1/1	29.0
	Non-responder	4/5	27.4 (9.6-DNE)
RAEB	Responder	5/5	31.9 (19.5-46.5)
	Non-responder	32/33	19.9 (13.6-28.5)
RAEB-T	Responder	2/2	22.7 (19.3-26.2)
	Non-responder	14/14	14.9 (5.9-20.1)
CMMoL	Responder	1/1	16.9
	Non-responder	7/7	6.0 (4.0-27.8)
AML	Responder	2/2	25.8 (25.2-26.4)
	Non-responder	8/8	17.5 (4.0-40.6)

*Total number of subjects = number of deaths + number censored.

Analysis by groups as suggested by the FDA reinforces these conclusions. As shown in the Reviewer's Table below (from the Sponsor's Table 14.2.7.1.3A, prepared on January 26, 2004. Percentages are omitted),

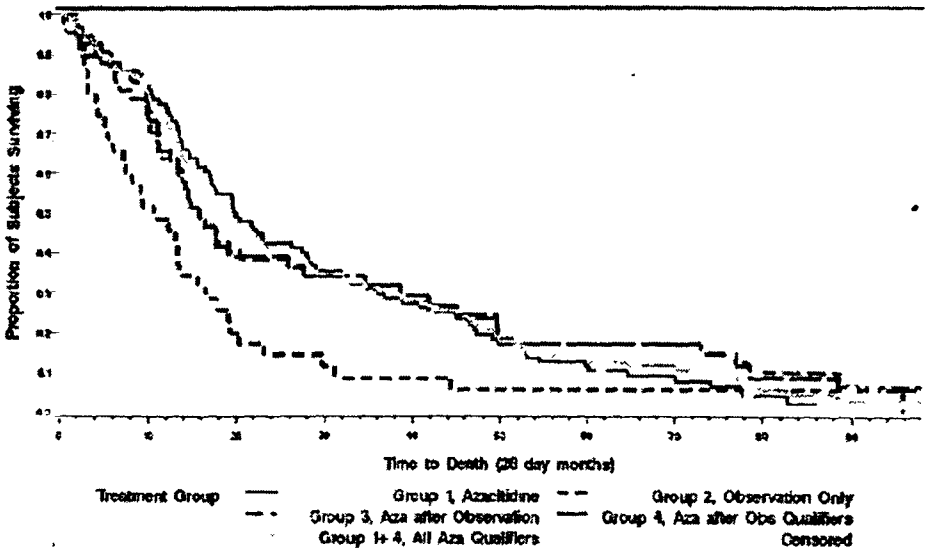
- Azacitidine responders had about three times the median survival of subjects in the observation group without crossover (30.4 months vs. 10.5 months).
- Azacidine responders had 73% longer median survival than azacitidine non-responders (30.4 months vs. 17.6 months).
- Azacitidine after crossover from observation responders had about a four-fold longer median survival times than non-responders (61.3 months vs. 14.3 months).
- All azacitidine responders had about a two-fold longer median survival times than non-responders (32.5 months vs. 16.4 months).
- Thus, azacitidine responders had 3-fold longer median survivals than observation subjects, and azacitidine non-responders had 56% longer survival than observation subjects. *Reviewer's Note: See above cautionary note and reference to Anderson's et al.*

Reviewer's Table. Analysis of Time to Death from Any Cause Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

Treatment	Subjects (Number)	Deaths (Number)	Censored (Number)	Median Survival Time (months), 95% CI on median
Group 1 (azacitidine)	89	85	4	19.9 (16.9-27.8)
Responder	14	14	0	30.4 (19.5-46.2)
Non-responder	75	71	4	17.6 (13.6-22.9)
Group 2 (Observation only)	36	34	2	10.5 (6.9-15.6)
Responder	0	0	0	
Non-responder	36	34	2	10.5 (6.9-15.6)
Group 3 (Azacitidine after Observation)	47	43	4	15.6 (12.4-25.8)
Responder	5	3	2	72.9 (34.4-DNE)
Non-responder	42	40	2	14.1 (10.9-19.0)
Group 4 (Azacitidine after Observation Qualifiers)	41	38	3	15.6 (12.4-27.5)
Responder	4	3	1	61.3 (34.4-DNE)
Non-responder	37	35	2	14.3 (10.9-19.0)
Groups 1 + 4	130	123	7	19.0 (15.6-22.9)
Responder	18	17	1	32.5 (26.2-46.5)
Non-responder	112	106	6	16.4 (13.8-19.9)
Log-Rank Rest p-values				
Group 1 vs. Group 2	0.0059			
Group 1 vs. Group 3	0.7066			
Group 1 vs. Group 4	0.6805			
Groups 1 + 4 vs. Group 2	0.0037			

A Kaplan-Meier plot demonstrates the difference between Group 2 (Observation Only) and the groups treated with azacitidine. However, these groups were not pre-specified and were not matched during randomization.

Figure 7.3-1: Kaplan-Meier Plot of Time to Death Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry



Analysis of MDS subtypes by group are shown below (from sponsor's Table 14.1.4.2A, prepared on January 22, 2004).

Reviewer's Table. MDS Diagnoses by Treatment Groups

Treatment Groups	RA	RARS	RAEB	RAEB-T	CMMoL
1 (N=89)	29 (33%)	7 (8%)	38 (43%)	8 (9%)	7 (8%)
2 (N=36)	3 (8%)	4 (11%)	16 (44%)	8 (22%)	5 (14%)
3 (N=47)	19 (40%)	5 (11%)	17 (36%)	4 (9%)	2 (4%)
4 (N=41)	18 (44%)	4 (10%)	17 (42%)	0	2 (5%)
1 + 4 (N=130)	47 (36%)	11 (%)	55 (42%)	8 (6%)	9 (7%)

While the MDS types were fairly well matched at original randomization (see Reviewer's Table below), comparison of MDS types in Group 1 vs. Group 2 shows that RA type was more prevalent in the azacitidine group and RAEB-T was more prevalent in the Observation Only group. Generally, in published literature, RAEB-T is thought to have a much worse prognosis than RA (see Background). In the present study, irrespective of treatment arm, RA patients had a median survival of 833 days and RAEB-T patients a median survival of 389 days (see Reviewer's Table below).

MDS Diagnoses by Treatment Arms in the Original Randomization

Treatment Groups	RA	RARS	RAEB	RAEB-T	CMMoL
Azacitidine (N=99)	21 (21%)	6 (6%)	42 (42%)	22 (22%)	8 (8%)
Observation (N=92)	18 (20%)	5 (5%)	44 (48%)	17 (19%)	7 (8%) + 1 AML

Reviewer's Analysis of Median Survivals:

The reviewer performed independent calculations from sponsor's data (Listings 16.2.5.2.1, 16.2.7.1.2, 16.2.8.7.1A, and 16.2.8.7A) of median survivals by treatment groups, responses, and MDS types, since CALGB had this information on all 191 enrolled patients, who were followed until death after withdrawal from the trial.

Reviewer's Table. Median Survivals in Azacitidine-treated, in Observation Only and in AML Subjects

Treatment Arm and No. of Subjects	Median Survival	Range (Min.-Max.)
Azacitidine, all N=136	591 days (19.7 months)	47 – 2,820 days
Responders (CR + PR), N=20	1,257 days (41.9 months)	351 – 2,820 days
• CR Responders, N=8	1,637 days (54.6 months)	613 – 2,359 days
• PR Responders, N=12	897 days (29.9 months)	351 – 2,820 days
Non-Responders, N=116	507 days (16.9 months)	47 – 2,751 days
Observation Only, N=36	275 days (9.2 months)	17 – 2,677 days
AML, N=19	413 days (13.8 months)	38 – 1,273 days
Responders (CR + PR), N=2	722 days (24.1 months)	705 – 738 days
Non-Responders, N=17	347 days (11.6 months)	38 – 1,273 days

Reviewer's Comments:

- *In general, the reviewer's conclusions are in agreement with those of the sponsor. Specifically,*
  - *All azacitidine-treated MDS subjects had a 2.15-fold longer median survival than Observation Only MDS subjects*
  - *Azacitidine-responders had a 2.13-fold longer median survival than non-responders*
  - *Complete responders had a 1.82-fold longer survival than partial responders*
  - *AML subjects had long median survivals, responders about 2-fold longer median survivals than non-responders*
- *However, the enormous range of survivals in both responders and non-responders suggests that factors other than azacitidine treatment may influence survival.*
- *Given the response rate of about 15% and the median survivals of responders, non-responders and observation only subjects, in order to show a survival advantage of azacitidine-treated subjects a minimum of 570 subjects should have been randomized to each treatment arm and no crossover permitted.*

Reviewer's Table below describes survivals by MDS subtype without reference to treatment arm.

Reviewer's Table. Survivals by MDS Subtypes and AML

MDS Subtype	Number of Subjects	Median Survival (days)	Mean Survival (days)	Range (days)
RA	39	833	1,016	17 – 2,751
RARS	11	812	913	270 – 1,719
RAEB	77	531	723	47 – 2,820
RAEB-T	30	389	509	68 – 2,319
CMMoL	15	375	603	70 – 2,518
AML	19	413	535	38 – 1,273

Reviewer's Comments:

- *The range of survivals in this trial population is remarkable, and it would be difficult to match patients in comparable groups without a very large number of patients.*
- *While median survivals were longer in RA and RARS subjects than in RAEB, RAEB-T and CMMoL subjects, there was extensive overlap between MDS groups.*
- *Median survival in AML subjects was longer than in RAEB-T and CMMoL subjects, and was more typical of MDS than of AML.*

1.7.7.3.2 Secondary Endpoint: Time to Disease Progression and Time to Relapse: Median times could not be estimated in these two endpoints due to low numbers of subjects experiencing these events.

1.7.7.3.3 Secondary Endpoint: Time to Transformation to AML, and Time to Death or Transformation to AML

Similar percentages of subjects transformed to AML in the azacitidine arm (35%; 31/89) and in the observation arm (40%; 33/83). The median time to transformation to AML was longer for the azacitidine group (45.8 months) than for the observation group (23.5 months). These data are shown in the Reviewer's Table below (from sponsor's Table 11.4-10) and in the Kaplan-Meier plot (Sponsor's Figure 5). Log-rank test p-value for the two groups was 0.1555, i.e. the difference was not statistically significant.

The indicator of Time to Death or Transformation to AML similarly showed no significant differences between the azacitidine and the observation group (17.7 months vs. 13.8 months), as shown in the Reviewer's Table below (from Sponsor's Table 11.4-11) and in the Kaplan-Meier plot (Sponsor's Figure 6). Log-rank test for the two groups showed a p-value of 0.48. The median time to death or transformation to for azacitidine responders was almost twice that of the observation group (25.8 months vs. 13.8 months).

Figure 5: Kaplan-Meier Plot of Time to Transformation to AML (CALGB 9221)

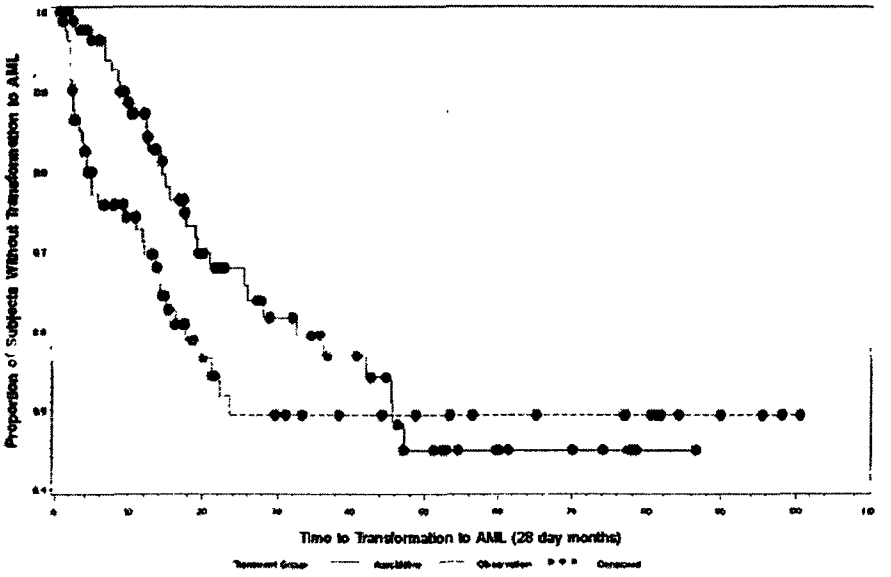
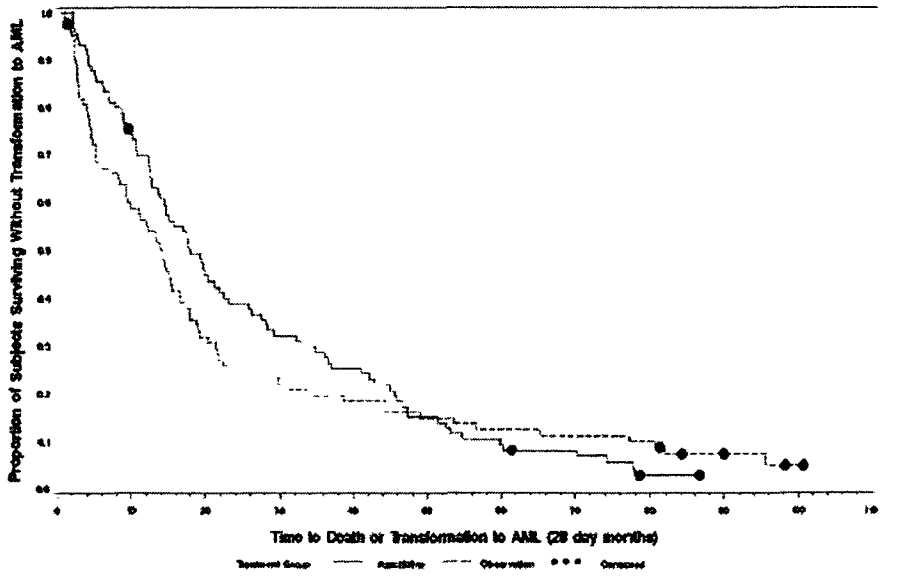


Figure 6: Kaplan-Meier Plot of Time to Death or Transformation to AML (CALGB 9221)



Reviewer's Table. Time to Transformation to AML, Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

Treatment	Total No. of Subjects	Transformed to AML	Censored	Median Time (months, CI)
Azacitidine	89	31(35%)	58	45.8 (28.3-DNE)
• Responder	14	7(50%)	7	35.9 (21.1-DNE)
• Non-Responder	75	24(32%)	51	47.2 (32.7-DNE)
Observation	83	33(40%)	50	23.5 (16.4-DNE)

Reviewer's Table. Time to Death or Transformation to AML, Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

Treatment	Total No. of Subjects	Transformed to AML	Censored	Median Time (months, CI)
Azacitidine	89	85 (96%)	4	17.7 (14.2-22.9)
• Responder	14	14 (100%)	0	25.8 (17.7-45.8)
• Non-Responder	75	71 (95%)	4	15.0 (12.3-21.7)
Observation	83	77 (93%)	6	13.8 (9.3-16.4)

As noted before, the original observation group comprised both observation group only subjects, and crossover to azacitidine group subjects, leading to confounding results. When the analysis of time to transformation to AML was carried out by FDA-suggested groups, it showed significant differences between subjects randomized to treatment with azacitidine (Group 1, Group 1 + 4) and observation only subjects (Group 2). Median time to transformation to AML was only 5 months in the observation only group, while it was about 46 months in subjects treated with azacitidine (Reviewer's Table below with data from sponsor's Table 7.3-6, February 19, 2004 submission). These differences were highly statistically significant.

Log-rank p-values were:
1) for Group 1 vs. Group 2, p=0.0007 and
2) for Group 1+ 4 vs. Group 2, p=0.0002.

Reviewer's Table. Time to Transformation to AML, Excluding Subjects With Adjudicated Diagnosis of AML at Study Entry by Groups, by FDA-Suggested Groups

Treatment	Total No. of Subjects	Transformed to AML	Censored	Median Time (months, CI)
Group 1, Azacitidine	89	31 (35%)	58	45.8 (28.3-DNE)
• Responder	14	7 (50%)	7	35.9 (21.1-DNE)
• Non-Responder	75	24 (32%)	51	47.2 (32.7-DNE)
Group 2, Observation Only	36	17 (47%)	19	5.0 (3.8-DNE)
Group 1+ 4, All azacitidine qualifiers	130	43 (33%)	87	47.2 (32.7-DNE)
• Responder	18	8(44%)	10	45.8(21.1-DNE)
• Non-Responder	112	35(31%)	77	DNE(36.3-DNE)

DNE = does not exist (due to small number of events)

Similarly, the median time to death or transformation to AML was only 4.2 months in the observation group, while it was 15-18 months in the azacitidine groups, with responders having median times of about 27 months. The differences were highly statistically significant.

Log-rank p-values were:
1) for Group 1 vs. Group 2, p=0.0013 and
2) for Group 1 + 4 vs. Group 2, p=0.0005.

Reviewer's Table. Time to Death or Transformation to AML, Excluding Subjects With Adjudicated Diagnosis of AML at Study Entry, by FDA-Suggested Groups

Treatment	Total No. of Subjects	Deaths or Transformed to AML	Censored	Median Time (months, CI)
Group 1, Azacitidine	89	85 (96%)	4	17.7 (14.2-22.9)
• Responder	14	14 (100%)	0	25.8 (17.7-45.8)
• Non-Responder	75	71 (95%)	4	15.0 (12.3-21.7)
Group 2, Observation Only	36	34 (94%)	2	4.2 (3.3-9.2)
Group 1+ 4, All azacitidine qualifiers	130	123 (95%)	7	15.0 (13.2-19.6)
• Responder	18	17 (94%)	1	27.4 (20.1-45.8)
• Non-Responder	112	106 (95%)	6	13.8 (12.0-16.1)

The two Kaplan-Meier plots for the FDA-suggested groups are shown below.

Figure 7.3-3: Kaplan-Meier Plot of Time to Transformation to AML Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

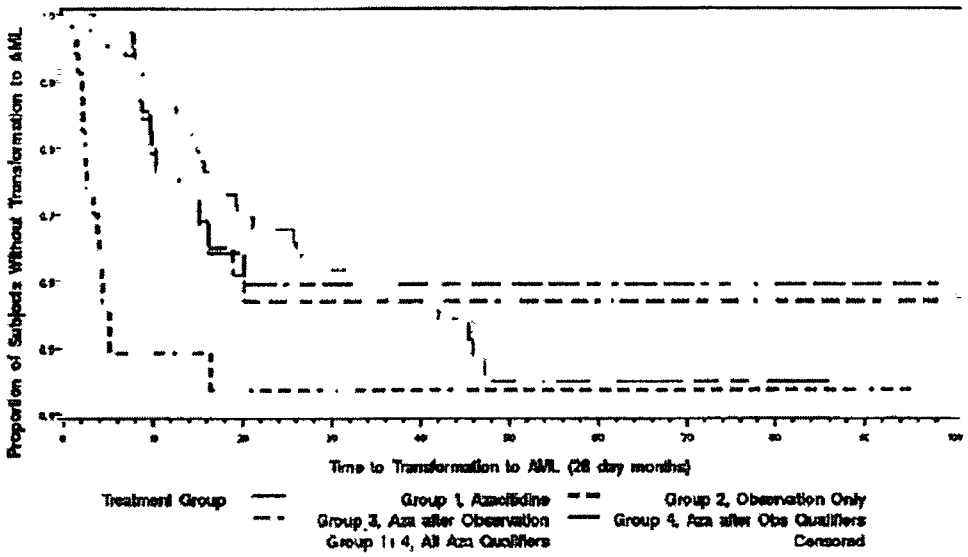
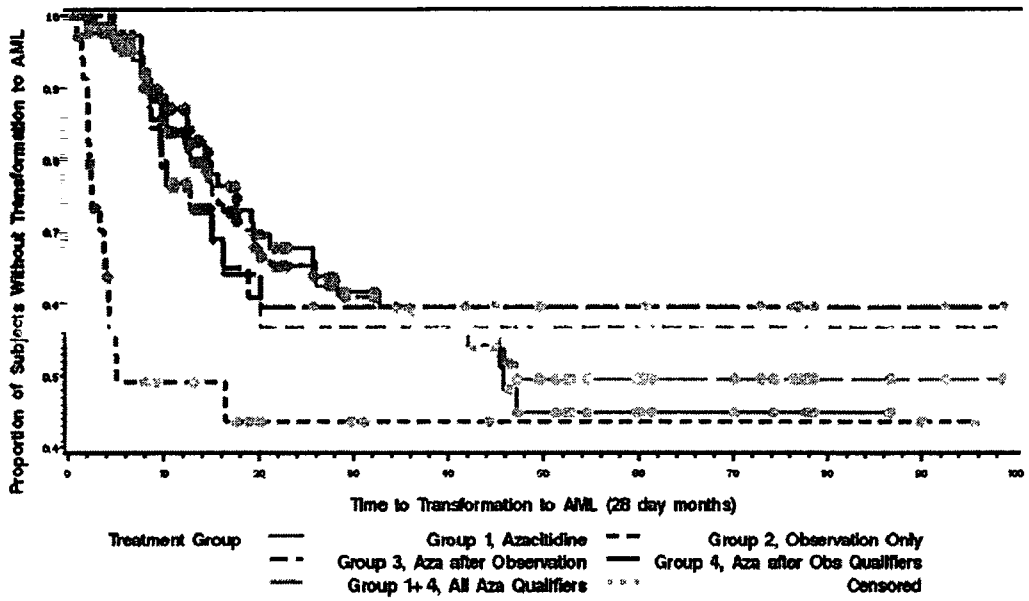


Figure 7.3-3: Kaplan-Meier Plot of Time to Transformation to AML Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry



1.7.7.3 Secondary Efficacy Endpoints

1.7.7.3.1 Secondary Endpoint: RBC Transfusion Rates

There were no significant differences in mean numbers of RBC units transfused to subjects in the original azacitidine group and in the original observation group, except that azacitidine responders had lower rates, as shown in the Reviewer's Table below (from sponsor's table 11.4-12). *Reviewer's Note: However, this endpoint is difficult to evaluate, as the numbers of subjects decreases with time in each group.* The sponsor presented the data by month.

- In the azacitidine group, the numbers of subjects decreased from 99 in Month 1 to 43 in Month 12, during which time the number of RBC units decreased from 3.45 in Month 1 to 0.77 in Month 12.
- The azacitidine responder group, the numbers of subjects remained steady at 16 through Month 11, when it decreased to 15. The numbers of RBC units transfused decreased steadily from 2.38 in Month 1 to 0.13 in Month 6 and to no transfusions from Month 7 to Month 11.
- In azacitidine non-responders group, the numbers of subjects decreased from 83 in Month 1 to 28 in Month 12. The numbers of RBC units transfused per subject per month decreased from 3.66 in Month 1 to 1.11 in Month 12. *Reviewer's Note: Thus, these data could be interpreted as showing that subjects using higher number of transfusions died and the remaining subjects had lesser transfusion needs, or that there may be some benefit of azacitidine to patients with Improvement, who did not reach PR criteria.*

- In the observation group, the numbers of subjects decreased from 92 in Month 1 to 6 in Month 12. *Reviewer's Note: The number of RBC units transfused decreased from 2.40 to 0.83 per subject per month, but these data are difficult to interpret because of the attrition of over 90% of the original subjects and the crossover subjects.*

Reviewer's Table. Mean Numbers of RBC Units Transfused per Subject per Month

Time	Azacitidine N=99	Azacitidine Responders N=16	Azacitidine Non-Responders N=83	Observation N=92
Baseline	1.99	1.25	2.13	1.95
Post-Baseline Average (12 months)	2.39	0.42	2.77	2.40

Analysis of RBC transfusions by FDA suggested groups is shown below in Reviewer's Table (from sponsor's Table 7.3-8, February 19, 2004 submission). The data are presented not by month as in the original submission but by 28-day treatment cycle, and transfusion rates are presented by Frequency of Transfusions per Cycle instead of the numbers of RBC units transfused. The numbers of subjects in each group appear not to be consistent with the numbers in the original submission, as described below.

Reviewer's Table. Mean Frequency of RBC Transfusions per Subject per 28-Day Cycle in FDA-Suggested Groups, with the Exclusion of AML Subjects

Group	Baseline (SD not shown)	Overall Post-Baseline (12 Cycles)(SD not shown)
Group 1: Azacitidine, N=89	1.08	1.43
• Responders, N=14	1.14	0.23
• Non-Responders, N=75	1.07	1.65
Group 2: Observation Only, N=36	1.31	1.30
Group 4: Azacitidine after Observation Qualifiers	1.12	1.56
Group 1 + 4: All Azacitidine	1.09	1.47
• Responders	1.17	0.24
• Non-Responders	1.08	1.67

- The number of subjects in Group 1 decreased from 89 in Cycle 1 to 26 in Cycle 12. The mean frequency of transfusions per subject per cycle decreased from 1.84 in Cycle 1 to 0.35 in Cycle 12.
- The number of subjects in Azacitidine responders decreased from 14 in Cycle 1 to 9 in Cycle 12. The mean frequency of transfusions decreased from 1.30 in Cycle 1 to 0.14 in Cycle 5 and to none or few (0.03 – 0.08) thereafter.
- The number of subjects in azacitidine non-responders decreased from 75 in Cycle 1 to 17 in Cycle 12. The mean frequency of transfusions decreased from 1.95 in Cycle 1 to 0.36 – 0.81 in Cycles 5 through 12.

- The number of subjects in Observation Only group decreased from 36 in Cycle 1 to 5 in Cycle 12. The mean frequency of transfusions fluctuated during the cycles from none to 1.43.
- The number of subjects in azacitidine after Observation qualifiers decreased from 41 in Cycle 1 to 7 in Cycle 12. The mean frequency of transfusions decreased from 2.06 in Cycle 1 to 0.28 –0.39 in Cycles 7 through 12.
- The numbers of subjects in all azacitidine qualifiers (Group 1 + 4) decreased from 130 (18 responders, 112 non-responders) in Cycle 1 to 33 (11 responders, 22 non-responders) in Cycle 12. Transfusion frequencies in this group followed those of subjects in Group 1 (both responders and non-responders showing comparable differences).

Reviewer's Note: The transfusion data show lower transfusion rates in azacitidine responders as compared to Observation Only group, but appear not to show benefit in the azacitidine non-responder group as compared to Observation Only group. These conclusions stem from transfusion analysis by treatment cycle (SD not indicated) from sponsor's Table 7.3-8.

The Frequency of Transfusions in Azacitidine Responders and Non-Responders and in Observation Only Subjects, by Treatment Cycle

Cycle	Group 1: Azacitidine responders	Group 1: Azaciticine Non-Responders	Group 2: Observation Only
1	1.30, n=14	1.95, n=75	0.85, n=36
2	0.76, n=14	1.48, n=64	1.43, n=33
3	0.68, n=14	1.40, n=62	1.43, n=27
4	0.30, n=14	1.01, n=57	1.12, n=20
5	0.14, n=14	0.78, n=39	0.71, n=16
6	0.00, n=14	0.81, n=35	0.00, n=8
7	0.00, n=14	0.66, n=30	0.62, n=7
8	0.00, n=14	0.66, n=27	0.33, n=6
9	0.03, n=14	0.54, n=22	1.40, n=5
10	0.00, n=14	0.46, n=19	0.80, n=5
11	0.07, n=10	0.36, n=18	0.00, n=5
12	0.08, n=9	0.49, n=17	0.80, n=5
Average	0.23	1.65	1.30

1.7.7.3.5 Secondary Endpoint: Platelet Transfusion Rates

The overall conclusions regarding platelet transfusions appear to be that platelet transfusions decreased in azacitidine responders, increased in azacitidine non-responders, and remained stable in the observation only subjects. However, these data (from sponsor's Table 11.4-13) do not take into account the large subject changes during the 12-month period described above. Furthermore, the differences in transfusion needs between responders and non-responders were present at baseline.

Reviewer's Table. Mean Numbers of Platelet Units Transfused per Subject per Month (ITT Population)

Time	Azacitidine N=99	Azacitidine Responders N=16	Azacitidine Non-Responders N=83	Observation N=92
Baseline	1.73	0.31	2.01	2.60
Post-Baseline Average (12 months)	4.02	0.18	4.76	2.48

Analysis by FDA-suggested groups suggests that platelet transfusions increased in all azacitidine treated patients and decreased in the Observation Only patients (data from sponsor's Tables 7.3-9). However, this analysis also does not take into account the large decreases in the number of subjects during the 12-month period. Platelet transfusions do not appear to be a reliable indicator of treatment efficacy.

Reviewer's Table. Mean Frequency of Platelet Transfusions per Subject per 28-Day Cycle in FDA-Suggested Groups, with the Exclusion of AML Subjects

Group	Baseline (SD not shown)	Overall Post-Baseline (12 Cycle average)(SD not shown)
Group 1: Azacitidine, N=89	0.35	1.15
• Responders, N=14	0.14	0.26
• Non-Responders, N=75	0.39	1.32
Group 2: Observation Only, N=36	1.22	0.66
Group 4: Azacitidine after Observation Qualifiers, N=41	0.68	1.08
Group 1 + 4: All Azacitidine, 130	0.45	1.13
• Responders, N=18	0.17	0.20
• Non-Responders, N=112	0.50	1.28

1.7.7.3.4 Secondary Endpoint: Rates of Antibiotic Therapy

The number of courses of antibiotic therapy was examined as a surrogate measure of the frequency of infections. One dose was counted as a treatment course, and courses had to be separated by at least 5 days. Antibiotics administered pre-procedurally or prophylactically were not included in this analysis. The following two Reviewer's Tables are from sponsor's data in Tables 11.4-14 and 7.3-10. (SDs not included). The differences between groups were not statistically significant. Most subjects received only one or two courses of antibiotics throughout the study; hence, monthly infection rates were very low.

Reviewer's Table. Rates of Courses of Antibiotic Therapy per Subject per Month (Six Months' Data)

Time	Azacitidine N=99	Azacitidine Responders N=16	Azacitidine Non-Responders N=83	Observation N=92
Baseline	0.06	0.07	0.06	0.09
Post-Baseline Average (6 months)	0.22	0.07	0.25	0.30

Reviewer's Table. Rates of Antibiotic Therapy per Subject per 28-Day Cycle in FDA-Suggested Groups, with the Exclusion of AML Subjects (12 Cycle Data)

Group	Baseline (SD not shown)	Overall Post-Baseline (12 Cycle average)(SD not shown)
Group 1: Azacitidine, N=89	0.07	0.19
• Responders, N=14	0.08	0.07
• Non-Responders, N=75	0.07	0.22
Group 2: Observation Only, N=36	0.15	0.42
Group 4: Azacitidine after Observation Qualifiers,N=41	0.15	0.20
Group 1 + 4: All Azacitidine, N=130	0.10	0.20
• Responders, N=18	0.09	0.06
• Non-Responders, N=112	0.10	0.22

1.7.7.3.7 Secondary Endpoint: Hemorrhage Rates

Hemorrhage rates are presented in Reviewer's Tables from sponsor's data in Tables 11.4-15 and 7.3-11. Hemorrhage rates were very low, and even though the differences between the azacitidine and the observation groups (0.26 and 0.46) were statistically significant (p=0.002), they probably were not clinically relevant. Analysis by FDA-suggested groups shows lower rates by azacitidine responders than by non-responders or by subjects in the Observation Only group.

Reviewer's Table. Hemorrhage Rates per Subject per Month (Twelve Months' Data)

Time	Azacitidine N=99	Azacitidine Responders N=16	Azacitidine Non-Responders N=83	Observation N=92
Baseline	0.00	0.00	0.00	0.01
Post-Baseline Average (12 months)	0.26	0.03	0.30	0.46

Reviewer's Table. Hemorrhage Rates of per Subject per 28-Day Cycle in FDA-Suggested Groups, with the Exclusion of AML Subjects (12 Cycle Data)

Group	Baseline (SD not shown)	Overall Post-Baseline (12 Cycle average)(SD not shown)
Group 1: Azacitidine, N=89	**0.00**	**0.27**
• **Responders, N=14**	**0.00**	**0.04**
• **Non-Responders, N=75**	**0.00**	**0.31**
Group 2: Observation Only, N=36	**0.00**	**0.20**
Group 4: Azacitidine after Observation Qualifiers, N=41	**0.08**	**0.15**
Group 1 + 4: All Azacitidine, N=130	**0.03**	**0.23**
• Responders, N=18	0.00	0.03
• **Non-Responders, N=112**	**0.03**	**0.27**

1.7.7.3.8 Secondary Endpoint: Hematologic Changes

Hemoglobin concentration changes between baseline and overall post-baseline averages were not significant between azacitidine and observation groups (p=0.07), and between Groups 1, 2, 4, and 1 + 4.

WBC changes were normal at baseline in azacitidine and observation groups and in Groups 1, 2, 4 and 1 + 4. Several high values at various months in both azacitidine and observation groups (and in Groups 1, 2, and 1 + 4) were probably due to peripheral blood blasts, as absolute neutrophil counts (ANC) did not show these changes.

Mean baseline platelet counts were low in all groups (94 – 113 G/L), and remained low in the overall post-baseline average values (104 – 117 G/L), with month-by-month fluctuations. Azacitidine responders had the highest platelet counts (average post-baseline 235 G/L).

ANC were in the lower range of normal values in all groups.

Bone marrow blasts at baseline were 10.2% in the azacitidine group and 10.9% in the observation group. The average change from baseline in the azacitidine group was a decrease in blasts by 2.2%, and a 5.2% increase in the observation group. When FDA-recommended groups were analyzed, azacitidine-treated groups had decreases in blasts, and Observation Only group had increases. These data (from sponsor's Tables 11.4-20 and 7.3-16) are shown in Reviewer's Tables below.

Reviewer's Table. Bone Marrow Blasts (%) in All Subjects

Time	Azacitidine N=76	Azacitidine Responders N=16	Azacitidine Non-Responders N=60	Observation N=69
Baseline	10.2%	8.4%	10.7%	10.9%
Post-Baseline Average (10 months)	-2.2%	-3.4%	-1.8%	+5.2%

Reviewer's Table. Bone Marrow Blasts (%) by Groups, Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

Group	Baseline (SD not shown)	Overall Post-Baseline (12 Cycle average)(SD not shown)
Group 1: Azacitidine, N=67	9.6%	-3.1%
Group 2: Observation Only, N=22	13.7%	+10.3%
Group 4: Azacitidine after Observation Qualifiers, N=36	8.8%	-1.0%
Group 1 + 4: All Azacitidine, N=130	9.3%	-2.4%

2.0 Title of Supporting Study: CALGB Study 8921. A Phase II Study of Subcutaneous 5-Azacitidine in Myelodysplastic Syndromes

2.1 Study Period: July 14, 1989 to March 14, 2003

2.2 Study Centers: 30 centers, all in the U.S. Of these, 23 centers enrolled 1 or 2 subjects each. Four sites enrolled 6 to 9 subjects per site and accounted for 40% (29/72) of the total enrolled population.

2.3 Study Objectives: The original prospective study objectives were

- To determine those myelodysplastic syndromes that respond optimally to subcutaneous azacitidine, and
- To evaluate azacitidine toxicity.

Data from the study was retrospectively re-collected for this NDA submission.

Primary and Secondary efficacy endpoints were redefined as in the CALGB 9221 Study. The primary endpoint was Overall Response Rate (CR + PR). The secondary endpoints were 1) time-to-event summaries of death, disease progression, transformation to AML, and death or transformation to AML; 2) requirements for RBC and platelet transfusions, rates of infection and of hemorrhages; and 3) changes in Hgb, WBC, platelet, and neutrophil (ANC) counts, and in percentages of bone marrow blasts.

2.4 Study Design: A Phase 2, open-label, uncontrolled, multicenter study, which evaluated SC azacitidine in subjects with MDS subtypes of RAEB, RAEB-T, and CMMoL. Subjects were to receive 75 mg/m^2 azacitidine SC daily for 7 days on a 28-day cycle for a minimum of 4 cycles. The dose could be adjusted (increased, decreased, or delayed) at the beginning of any cycle based on predefined hematology and renal laboratory results relating to the well-being of the subject. Response and safety assessments were to be performed during the performance of the study. After withdrawal from the study, subjects were to be followed for relapse or survival. Subjects who achieved a CR were to receive an additional 3 cycles of azacitidine treatment.

2.5 Study Plan: Assessments as described in CALGB 9221, bone marrow assessments at every 28-day cycle.

2.6 Study Population: 72 subjects enrolled and analyzed (planned enrollment was 50 subjects). Main inclusion criteria included: age >15 years old, diagnosis of RAEB, RAEB-T, or CMMoL as defined by FAB classification, informed consent, performance status 0-2 and life expectancy at least 2 months, liver and renal function tests with 1.5-2 ULN and serum $CO_2 \geq 19$ mEq/L. Exclusion criteria included: Any other illness that limited survival to <2 years; >30% blasts in the bone marrow, prior treatment with azacitidine, prior cytotoxic therapy for MDS, uncontrolled/severe CHF, and pregnancy. Subjects previously treated for cancer were eligible if they did not receive treatment within 6 months before study entry and were free of any evidence of malignancy for the previous 6 months.

2.7 Azacitidine Dose, Dose Adjustment and Method of Administration: As in CALGB 9221 trial.

2.8 Criteria for Response, Stable Disease, Relapse, and Disease Progression: As in CALGB 9221 trial.

2.9 Subject Disposition:

Subject disposition is summarized in sponsor's Table 32, which is shown below. The Table describes the number of subjects who received azacitidine, the number of subjects who withdrew from the study and the reasons for withdrawal, and the follow-up after study withdrawal.

- Completion status was available for 71/72 subjects. The remaining subject is still enrolled and appeared to be well at the time of last contact; the subject had received azacitidine for 6.5 years. Two subjects of 72 did not receive azacitidine, because the drug was not available for administration on Day 1 and they were diagnosed with AML within 6 days after enrollment into the study.

- The reasons for withdrawal from the study are mainly self-explanatory.
- Follow-up status was obtained as late as 13 years after study entry and 11 years after study withdrawal (range 0 to 3961 days). Under follow-up status, the numbers reflect the incidence after study withdrawal, with the exception of progression to AML, which includes the total incidence both on study and after study.

Table 32: Subject Disposition and Completion Status (CALGB 8921)

	Number (%) of Subjects	
Disposition Status	Azacitidine (N=72)	
Received study medication	70	(97.2)
Withdrew from therapy/study	71	(98.6)
Reason for Withdrawal[a]		
Achieved complete remission and therapy stopped	5	(6.9)
Completed treatment per protocol	1	(1.4)
Development of AML	12	(16.7)
Development of Relapse after PR or CR or Improvement	4	(5.6)
No response to therapy after 4 cycles of treatment	9	(12.5)
Adverse event	18	(25.0)
Poor compliance	1	(1.4)
Subject did not wish to continue in the study	5	(6.9)
Investigator discretion	3	(4.2)
Other[b]	6	(8.3)
Subject died	8	(11.1)
Follow-up status[a]		
Follow-up contact made	64	(88.9)[c]
Subject died	62	(86.1)
Subject progressed to AML[d]	32	(44.4)
Subject diagnosed with another malignancy	4	(5.6)
Subject received subsequent radiation therapy	3	(4.2)
Subject received subsequent chemotherapy	26	(36.1)

[a] Subjects may be categorized to more than 1 reason for withdrawal and follow-up status
[b] Specified reasons for other are listed by subject and treatment in CALGB 8921 CSR Appendix 16.2.9.13.2.
[c] Includes 1 subject (44415) who was still in the study at the time of last contact.
[d] Total of subjects who progressed to AML includes subjects who were withdrawn from the study due to development of AML.
KEY: AML=acute myelogenous leukemia, CR=Complete Response, PR=Partial Response, CALGB=Cancer and Leukemia Group B

2.10 Protocol Violations: 26% of the subjects took systemic steroids during the study, mainly pre-transfusion and for treatment of TEAEs such as fever, vomiting and rash. (Note: Steroid use was not prohibited in the protocol except to treat nausea). The other violations were few and included 2 patients who did not receive azacitidine, 3 who did not meet eligibility criteria, 2 who took cytotoxic therapy for MDS within 6 months, and 1 who did not receive azacitidine immediately after reconstitution.

2.11 Subject Demographics: Shown in Reviewer's Tables (data from sponsor's Tables 34 and 35). Demographic characteristics of the subjects appear to be similar to those of the subjects in CALGB 9221 study. MDS subtypes differ, as there were no RA and RARS subjects.

Baseline diagnoses were adjudicated by site, central, or blinded independent review of bone marrow and peripheral blood assessments. Baseline bone marrow slides were reviewed centrally for 86% (62/72) of subjects. At the time of retrospective re-collection of data, baseline bone marrow slides for review by the blinded independent reviewer were available for 61% (44/72) of subjects. One

subject was found to have AML at the site. Adjudication by central or blinded review resulted in a baseline diagnosis of AML for 17 subjects, including the 1 subject diagnosed with AML by the site. In the independent blinded review, 80% concordance was achieved with the site (35/44) and 73% concordance was achieved with the central pathology reviewer (27/37). The most common site diagnosis that was adjudicated to AML was RAEB-T (31%; 9/29), followed by RAEB (21%, 7/33).

Performance statuses and transfusion histories are stated in the Table. Other medical conditions and previous surgeries were common in this study of mostly elderly subjects.

Reviewer's Table. Subject Demographics

Demographic	Number of Subjects N=72	%
Gender: Male	49	68%
Female	23	32%
Race: White	68	94%
Black	2	3%
Hispanic	1	1%
Other	1	1%
Age: Mean (range)	64.6 (23 – 82)	
<65	26	36%
65 – 74	25	35%
>75	16	22%
Missing	5	7%
Height (cm): Mean (range)	170 (142 – 191)	
Weight (kg)	78 (46 – 156)	
BSA (m^2)	1.87 (1.40 – 2.48)	

Reviewer's Table. Baseline Disease Characteristics

Disease Characteristic	Number of Subjects N=72	%
Site diagnosis at study entry		
• RAEB	33	46%
• RAEB-T	29	40%
• CMMoL	9	13%
• AML	1	1%
Adjudicated diagnosis at study entry		
• RAEB	26	36%
• RAEB-T	20	28%
• CMMoL	9	13%
• AML	17	24%
Performance Status		
• 0 Normal	19	26%
• 1 Fatigue	34	47%
• 2 Impaired	6	8%
• Unknown/Not Done	13	18%

Transfusion product used in 3 months before study entry		
• **Any**	**54**	**75%**
• **RBC**	**53**	**74%**
• **Platelets**	**21**	**29%**

2.12 Primary Efficacy Results

2.12.1 Overall Response Rate

The primary endpoint was defined as the overall response rate of CR + PR. The best response attained during the study was used to categorize each subject, as shown in Reviewer's Table below (data from sponsor's Table 36).

Reviewer's Table. Response Rates of All Subjects (ITT Population)

Response	Number of Subjects N=72	%
Response		
• **Overall (CR + PR)**	**10**	**13.9%**
• **CR**	**4**	**5.6%**
• **PR**	**6**	**8.3%**
Non-Response		
• **Improvement, not CR or PR**	**12**	**16.7%**
• **Stable Disease**	**46**	**63.9%**
• **Disease Progression**	**0**	**0**
• **Unevaluable**	**4**	**5.6%**
Total Non-Response	**62**	**86.1%**

Four of the subjects had achieved CR, which occurred from the 6th to the 60th treatment cycle (162 to 1757 days on study). Six subjects achieved PR, which occurred from the 1st to 8th treatment cycle (22 to 232 days on study). The majority had either 2 or 3 cell line abnormalities at baseline (9/10). All responders were transfusion-free at the time of best response (5/10 were transfusion-dependent at baseline).

2.12.2 Duration of CR and PR

Sponsor's Table 37 lists the initial positive effect of responses (8 had decreases in blasts, 1 increase in WBC, 1 increase in platelet count), day and duration of the positive effect, best response and duration of best response, as well as MDS subtypes and abnormalities at baseline. Some of these data are shown in Reviewer's Table below.

Reviewer's Table. Duration of Positive Effect and of Response in Responders

Initial Positive Effect (Study Day)*	Duration of Positive Effect (days)	Best Response	Duration of CR or PR (days)**
Day 30	212	PR	99
Day 52	4752+ (13.0 years)	CR	4460 (12.2 years)
Day 57	465+ (1.3 years)	PR	50
Day 15	222+	PR	120+
Day 24	846+ (2.3 years)	PR	288
Day 32	318+	CR	209+
Day 15	515 (1.4 years)	PR	0***
Day 17	407+	CR	204+
Day 22	211+	CR	108+
Day 57	534+ (1.5 years)	PR	114

***First day of achievement of target for ≥4 weeks for at least 1 abnormality at baseline. Duration is number of days until transfusion, relapse, or last observed value.**
****Number of days from first achievement of target for ≥4 weeks for all baseline abnormalities until sustained loss of target values.**
*****This subject was assessed as PR by site investigator; however, diagnosis was adjudicated as AML.**

The initial positive effect was seen within 1 – 3 cycles of treatment, and was most frequently seen in bone marrow blasts (8/10). The mean duration of positive effect was 848 days (2.3 years); the median duration, 436 days (1.2 years). The mean duration of CR + PR was 565 days (1.5 years); the median duration, 117 days. The actual durations of positive effect or of response may have been artificially shortened by subjects withdrawing from the study, including subjects with CR after 3 additional cycles of treatment *(Reviewer's Note: and by subject adjudicated to have AML instead of PR).*

Reviewer's Notes: Review of PATIENT PROFILES showed that:

- *Duration of response could only be estimated, as 80% (8/10) of patients continued in response status at the time of withdrawal from the study.*
- *The median duration was 430 days+, the mean duration was 810 days +, the range was 195 days to 4481 days.*

2.12.3 Response Rate by MDS Subtype

Response rates by subtype are shown in Reviewer's Table below (sponsor's Table 38). Overall responses were observed across all subtypes; rates were 10% to 15%. Response rate in adjudicated AML subjects was 18%.

Response Rates of All Subjects by MDS Subtypes and AML at Baseline

MDS Subtype and Response	Number of Subjects	% of Subjects with Subtype
RAEB	**N=26**	**100%**
• **CR + PR**	**4**	**15.4%**
• **PR**	**4**	
RAEB-T	**N=20**	**100%**
• **CR + PR**	**2**	**10%**
• **CR**	**2**	
CMMoL	**N=9**	**100%**
• **CR + PR**	**1**	**11%**
• **CR**	**1**	
AML	N=17	100%
• **CR + PR**	**3**	**18%**
• **CR**	**1**	**6%**
• **PR**	**2**	**12%**

2.12.4 Responses Rate Excluding Subjects with Baseline Diagnosis of AML

Reviewer's Table below shows all Responses and Non-Responses in all subjects After exclusion of subjects that were adjudicated to have a diagnosis of AML (data from sponsor's Table 39).

Reviewer's Table. Response Rates Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

	Number of Subjects, N=55	% of Subjects 100%
Response		
• **Overall (CR + PR)**	**7**	**12.7%**
• **CR**	**3**	
• **PR**	**4**	
Non-Response		
• **Improvement, not CR or PR**	**10**	**18%**
• **Stable Disease**	**36**	**66%**
• **Disease Progression**	**0**	**0**
• **Unevaluable**	**2**	**4%**
Total Non-Response	**48**	**87%**

The sponsor also provides response data excluding 17 subjects with AML and 16 subjects with protocol violations, leaving 39 subjects. Six of these 39 subjects (15.4%) had CR + PR, 8 had improvement, 23 had stable disease and 2 were unevaluable. As described above, most of the protocol violations were the use of corticosteroids before transfusions.

2.12.5 Improvement

There were 12 subjects (ITT population) with best response of Improvement. All had either 2 or 3 cell line abnormalities, and 9/12 were transfusion-dependent.

- 11/12 were anemic at baseline, 9 experienced ≥50% increase in Hgb and were transfusion-free, and 1 experienced <50% in Hgb and was transfusion free.
- 12/12 had decreased platelets, 7 experienced ≥50% increase and were transfusion-free, and 2 experienced had <50% increase in platelets and were transfusion-free.
- 8 had WBC abnormalities at baseline and 5 had WBC increases.
- All Improvement subjects who were transfusion-dependent at baseline (9/9) were free of transfusions for 61 to 442 days.

Reviewer's Notes: Review of PATIENT PROFILES showed that

- *6 patients (8.3% of 72) had met the criteria for Improvement (see Appendix for patients with Improvement according to the sponsor and according to the Reviewer),*
- *4 patients continued in Improvement status at the time of withdrawal from the study, 2 had relapsed to pre-Improvement status,*
- *The median duration could be estimated as 209 days +, the mean duration as 300 days + with the range from 95 days to 693 + days.*

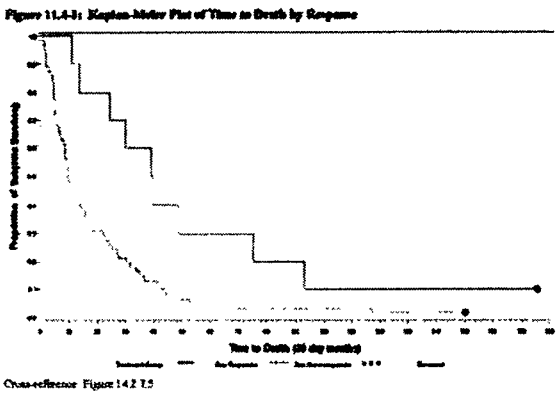
1.13.0 Secondary Efficacy Endpoints

1.13.1 Secondary Endpoint: Time to Death from Any Cause

Analysis of Time to Death is shown in Reviewer's Table below (from sponsor's Table 42) and in a Kaplan-Meier plot of Responders and Non-Responders.

Reviewer's Table. Time to Death from Any Cause (ITT Population)

Response to Azacitidine	No. of Subjects	Deaths, Number (%)	Censored, Number	Median Survival Time (months), 95% CI
Responder (CR + PR)	10	9 (90%)	1	39.3 (23.9 – 75.4)
Non-Responder	62	61 (98%)	1	9.6 (7.8 – 15.6)
TOTAL	72	70 (97%)	2	11.6 (8.7 – 17.8)



The median survival time of the 10 responders was more than 4 times longer than that of the non-responders.

1.13.2 Secondary Efficacy Endpoint: Survival Time by MDS Subtype

The data for this endpoint is shown in Reviewer's Table below (from sponsor's data in Table 43).

Time to Death from Any Cause of All Subjects by MDS Subtype

MDS Subtype	Response, N	Deaths, N	Censored, N	Survival Time (months), 95% CI
RAEB, Total	**26**	**25**	**1**	**19.9 (9.3 – 34.0)**
Responder	**4**	**4**	**0**	**34.9 (13.5 –48.5)**
Non-Responder	**22**	**21**	**1**	**12.9 (4.3 – 34.0)**
RAEB-T, Total	**20**	**20**	**0**	**8.4 (5.9 – 15.6)**
Responder	**2**	**2**	**0**	**49.6 (23.9 – 75.4)**
Non-Responder	**18**	**18**	**0**	**8.4 (5.9 – 12.5)**
CMMoL, Total	**9**	**9**	**0**	**25.0 (9.6 – 52.8)**
Responder	**1**	**1**	**0**	**93.3**
Non-Responder	**8**	**8**	**0**	**21.0 (8.7 – 52.8)**
AML, Total	**17**	**16**	**1**	**10.4 (5.2 – 22.8)**
Responder	**3**	**2**	**1**	**38.9 (10.6 – DNE)**
Non-Responder	**14**	**14**	**0**	**8.0 (4.8 – 17.0)**

Reviewer's Comments:

Several conclusions can be drawn from these data, however, these interpretations are preliminary since the numbers of subjects in each group were small.

- *Responders had 3 – 5-fold longer median survival times than non-responders in each MDS subtype.*
- *Responders' median survival times were quite long, from 3 to almost 8 years.*
- *Non-responders' median survival times decreased in the following order: CMMoL>RAEB>RAEB-T>AML.*
- *Long survival times were seen even in AML.*

When subjects with an adjudicated baseline diagnosis of AML were excluded from analyses, the median survival time was not substantially altered for either the entire group or subsets of responders and non-responders.

4.13.3 Secondary Efficacy Endpoint: Time to Transformation to AML

Transformation to AML in subjects who did not have an adjudicated diagnosis of AML at study entry occurred later and in smaller proportion of subjects among the responders than among non-responders, as shown in Reviewer's Table below (data from) sponsor's Table 44.

Reviewer's Table. Time to Transformation to AML, Excluding Subjects With Adjudicated Diagnosis of AML at Study Entry

Response to Azacitidine	Transformation to AML, N (%)	Censored N (%)	Median Time (months)	95% CI
Total, N=55	22 (40%)	33 (60%)	28.4	(16.7 – 117.2)
Responder, N=7	2 (29%)	5 (71%)	59.1	(32.3 – DNE)
Non-Responder, N=48	20 (42%)	28 (58%)	21.8	(12.5 – 117.2)

Transformation to AML occurred most often in RAEB-T patients (60%, 12/20) with median time to transformation of 11.0 months, and in CMMoL patients (56%, 5/9) with median time to transformation of 15.0 months.

2.13.4 Secondary Efficacy Endpoint: Time to Death or Transformation to AML

The combined endpoint of time to death or transformation to AML is shown in Reviewer's Table below (from sponsor's Table 45). The median time was more than 3 times longer in responders than in non-responders.

Reviewer's Table. Time to Death or Transformation to AML, Excluding Subjects with Adjudicated Diagnosis of AML at Study Entry

Response to Azacitidine	Death or Transformation to AML, N (%)	Censored N (%)	Median Time (months)	95% CI
Total, N=55	54 (98%)	1 (2%)	10.1	(5.9 – 15.7)
Responder, N=7	7 (100%)	0	32.3	(23.9 – 59.1)
Non-Responder, N=48	47 (98%)	1 (2%)	9.0	(5.0 – 12.0)

Death or transformation to AML occurred earlier in RAEB-T subjects (6.8 months) than in RAEB subjects (18.7 months) or in CMMoL subjects (9.5 months). These findings are consistent with the natural history of these subtypes.

2.13.5 Secondary Efficacy Endpoints: Rates of RBC and Platelet Transfusions, Antibiotic Therapy Courses, and Hemorrhagic Events

RBC transfusion rates are presented by number of units transfused per month. The mean baseline averages were 1.98 units for all 72 subjects, 0.97 units for the 10 responders and 2.15 units for 62 non-responders. Transfusion rates decreased over time for all subjects, so that by Month 10 all, 21 in number, became transfusion-free. Of these 21, 9 were responders and 12 were non-responders. These findings indicate that transfusion-dependent non-responders died or transformed to AML, while responders developed decreased transfusion requirement. The mean monthly number of units decreased from 2.80 in Month 1 to 1.50 in Month 2, to 1.00 in Month 3, to 0.40 in Month 4 and to 0.20 in Months 5 through 9, while the number of subjects remained steady at 10.

Platelet transfusions stopped in responders after Month 3, while they continued in non-responders, although decreasing from 8.77 units in Month 1 to 1.50 units in Month 12.

Antibiotic therapy course rates were very low, both in responders and non-responders.

Bleeding episodes rates were low and did not occur after Month 4 in responders and Month 7 in non-responders.

2.13.6 Secondary Endpoints: Peripheral Blood Cell Counts and Bone Marrow Blast Percentages

Mean Hgb values were within mild anemia range (90-110 G/L), WBC and ANC counts were within expanded normal limits, and platelet counts were decreased to below normal (with the exception of some very high values in the first 3 months that were probably a reporting error).

Baseline bone marrow blast percentage was 12.8%. It decreased in all subjects, more so in responders (mean change -7.1% in responders and –2.7% in non-responders).

2.13.7 Response Rates by Age and Gender

These analyses are presented in sponsor's Tables 55 and 56, and are summarized in the Reviewer's Table below. Only response rates are presented; non-response rates in sponsor's tables are not reproduced.

Reviewer's Table. Response Rates by Age and Gender

Parameter		Overall CR + PR	CR	PR
Age:	<65 years	3/26 (12%)	2	1
	65 – 74	7/41 (17%)	2	5
	≥75	2/16 (13%)	1	1
Gender:	Male	6/49 (12%)	3	3
	Female	4/23 (17%)	1	3

The 10 responders had a mean age of 65.1 years, with a range from 38 to 76 years. The mean age and the mean were similar to those of all subjects. Responses by age group were similar, suggesting that response was not affected by age.

The male-to-female ratio of the 10 responders was 3 to 2, which was similar to the ratio of all subjects, suggesting that responses were not affected by gender.

3.0 Supporting Study CALGB 8421. Title: 5-Azacitidine to Induce Differentiation in Myelodysplastic Syndromes: A Phase I – II Pilot Study

3.1 Study Period: June 20, 1985 to May 31, 1994

3.2 Study Centers: 17 centers, 49 subjects enrolled. Twelve sites enrolled 1 or 2 subjects each. Three sites enrolled 5 to 11 subjects per site; these 3 sites accounted for 52% of the enrolled population.

3.3 Study Objectives: The original prospective study objectives were as stated below.

- To test the effect of azacitidine given in repeated continuous low dose infusions on the differentiation of myelodysplastic syndromes.
- To determine an appropriate dose and regimen of azacitidine as a feasibility pilot for eventual application.
- To determine those myelodysplastic syndromes that respond optimally to differentiation treatment.
- To determine if azacitidine would affect the natural history and outcome of myelodysplastic syndromes.

During retrospective analysis the primary efficacy endpoint was defined as Overall Response Rate (CR + PR). The best response attained during the study was used to categorize each subject.

3.4 Study Design: A Phase 2, open-label, multicenter, uncontrolled study, which evaluated IV azacitidine in subjects with the MDS subtypes of RAEB and RAEB-T.